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# CONTENTS / İÇİNDEKİLER

- Research Article*
- Proximate assessment and bioassay of breakfast cereals produced from blends of acha (*Digitaria exilis*), mungbean (*Vigna eadiata*) and cashew nut (*Anarcadium occidentale* Linn) flours 1
- Inainfe Woripre & Ifeoma Mbaeyi-Nwaoha & Deborah Chinwendu Ofoegbu*
- Research Article*
- Enrichment of wheat bread with carob molasses pulp, a dietary fiber source: Impact on bread quality and acceptability 17
- Sema Aydın & Yüksel Özdemir & Müge Keçeli*
- Research Article*
- Sensory profile and physicochemical composition of premixed and post mixed fruit wine from blends of pineapple and watermelon juice 27
- Azuka Irene Ugo & Mbaeyi-Nwaoha Ifeoma & Uvere Peter Ofoegbu & Deborah Chinwendu*
- Research Article*
- Assessment of aroma profiles and mineral content of buffalo yogurt marketed in Cukurova region of Turkey 35
- Nuray Güzeler & Çağla Özbek & Murat Kalender*
- Research Article*
- Quality evaluation of formulated instant noodles from wheat, rice (*Oryza sativa*) and mushroom (*Agaricus bisporus*) flour blends 55
- Ifeoma Elizabeth Mbaeyi-Nwaoha & Chioma Gloria Mgbemere & Ngozi Chioma Okoronkwo*
- Review Article/Derleme Makale*
- Meyve ve sebzelerde bulunan biyoaktif bileşenlerin sağlık üzerine etkileri 77
- Health effects of bioactive components in fruits and vegetables*
- Gül Karadağ & Ayşe Demet Karaman & Serdal Öğüt*
- Review Article/Derleme Makale*
- Kardiyovasküler hastalıklarda bağırsak mikrobiyotasının rolü 91
- The role of microbiota in cardiovascular diseases*
- Ebrar Kökcü & Üla Akil & Esmâ Asil*
- Review Article/Derleme Makale*
- Alternatif ve sürdürülebilir bir gıda kaynağı olarak algler 101
- "Algae as an alternative and sustainable food source"*
- Derya Deniz Şirinyıldız & Aslı Yorulmaz*
- Clinical Research / Klinik Araştırma*
- Yoğun bakım ünitesinde yatan hastaların beslenme durumları ve bası yarası oluşma riski arasındaki ilişkinin değerlendirilmesi 43
- Evaluation of the relationship between the nutritional status and the risk of pressure sore formation of patients hospitalized in the intensive care unit*
- Özlem Özpak Akkuş & Betül Gülşen Atalay & Eda Parlak*
- Review Article*
- Dietary analysis of traditional food cultures in India: An overview of 2600 BCE to the 21<sup>st</sup> century 119
- Sudhanand Prasad Lal & Abhishek Mahendra & Akshay Singh*




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Research Article

# Proximate assessment and bioassay of breakfast cereals produced from blends of acha (*Digitaria exilis*), mungbean (*Vigna eadiata*) and cashew nut (*Anarcadium occidentale* Linn) flours

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## Article info

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## ABSTRACT

Breakfast cereals were formulated from blends of acha, fermented mungbean and cashew nut (undefatted and defatted) flours. The undefatted and defatted cashew nut flours were used at different levels of substitution (10, 20, 30, 40 and 50 %) with the best blend of acha and mungbean (80:20) flour which was determined by sensory evaluation. Breakfast cereals were produced by toasting (170 °C) a dry heat treatment process to gelatinize and semi-dextrinize the starch in order to generate dry ready to eat products. The samples were subjected to proximate and bioassay analyses. The breakfast cereal was used for a four week bioassay study using eighty four (84) healthy male albino rats weighing 60 - 90 g. The proximate composition showed: crude protein 10.24-20.10 %, moisture content 6.01-8.07 %, crude fat 3.60-24.51 %, crude fibre 4.02-7.07 %, ash 3.39-4.80 % and carbohydrate 46.4-68.08 %. Feed intake was measured daily while weight gain was measured weekly. Results after the study showed; feed intake 234.22 – 344.55 g, weight gain 34.14 – 80.25 g, feed efficiency 0.14-0.25 and protein efficiency 0.99-1.89. Feeding experiment showed positive increase in weight when the rats were fed the formulated breakfast products. This implies that the



products could support growth. No adverse growth rate was observed. Feed intake was also acceptable and compared well with the stable diet for rats. Feed efficiency of the samples compared well with the control diet.

## INTRODUCTION

Breakfast is the nutritional foundation or the first meal of the day. It is considered the most vital meal of the day. According to Sharma and Caralli (2004), breakfast cereal is defined as any food obtained by swelling, roasting, grinding, rolling or flaking any cereal. Despite being consumed dry in the early hours of the morning, breakfast cereals provide a good source of energy, which is a vital requirement of the human body. Nonetheless, the value of breakfast cereal is undeniable in this age of fast living, rapid urbanization, and above all, a health-conscious society (Janvekar, 2010). Ready-to-eat breakfast cereals are gradually displacing most conventional breakfast diets in most developing countries. They are popular among urban dwellers because of their convenience, nutritional value, increased income, status symbol, and job demands (Usman, 2012). Breakfast cereals now offer a variety of options for meeting today's recommendations for people of all ages to improve their whole grain intake. Cereals, on the other hand, are the most common breakfast products (Sharma and Caralli, 2004).

Acha (*Digitaria exilis*) is a West African grass also known as fundi, fonio, hungry rice, fonio blanc and petit mil. Acha, though neglected, is probably the oldest African cereal (Ayo and Nkama, 2004). Acha could not keep up with the latest international cereals, which were made particularly convenient for consumers by the use of mills and processing, due to a lack of interest and support from authorities (mostly non-African colonial authorities, missionaries, and agricultural researchers). The old grains languished and were primarily used as a food source for the poor and in rural areas (Ayo and

Nkama, 2004). The acha cereal grain has been named one of the world's most fascinating plants. The proteins in acha grains (8–11 %) are not easily extractable. Their digestibility, on the other hand, outperforms sorghum and millet. Due to its rich in methionine and cysteine content, acha is considered one of the most nutritious grains (Jideani, 1997; Jideani et al., 1994). It may have essential functional properties due to the high levels of residue protein in it. Acha is also a delicious cereal that is regarded as one of the best in the world (NRC, 1996). The acha can benefit greatly from this combination of nutrition and flavour.

Mung bean (*Vigna radiata*), also known as green gram, is a tropical legume that is primarily grown in Asia and also Nigeria. Mungbean is a great source of high-quality protein and one of the cheapest and richest plant protein sources (Akaerue and Onwuka, 2010). It is high in amino acids, especially lysine and thus can be used to complement human diets that are primarily focused on cereals.

Cashew nut meal has recently been approved for use in poultry, especially layers, in addition to human consumption. Cashew seeds' main products are the kernels, which have nutritional and economic value as confectionary nuts (Ogungbenle, 2014). The nuts have long played an important part of the meals of many cultures and civilisations, because of their great nutritional content, their broad variety of taste and distinct flavours and high energy value.

Researchers and policymakers overlook many of the underutilized food crops native to third-world countries, especially Nigeria. These underutilized crops, on the other hand, may have a lot of potential particularly in terms of improving food quality and thus people's nutrient intake. One of the best ways to minimize nutritional, environmental and financial vulnerability in times of change is to increase the use of underutilized crops (Pasicznik and Jaenicke, 2009).

From the foregoing, it is clear that some of these locally available cereals and legumes, which are cultivated in large quantities, can be used to

formulate products, thus highlighting the raw materials' other utility potentials (Mbaeyi, 2005). As a result, the aim of this project was to formulate a breakfast cereal with acha, mungbean and cashew nut flour in its defatted and undefatted form. This would be accomplished by fermenting the legume (mungbean) in order to impart those desirable qualities. This project aims to raise public awareness about the nutritional benefits of defatted and undefatted cashew nut flour and to provide useful information on how to use it effectively in a variety of food applications

## MATERIALS AND METHODS

### Procurement of raw materials

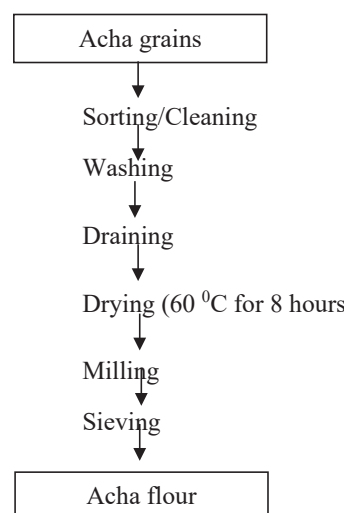
The grains of acha (*Digitaria subtilis*) were purchased at the Ogbete market in Enugu State. Mungbean (*Vigna radiata*), cashew nut (*Anacardium occidentale* Linn) were purchased from Obollo-afor market while salt, and sugar were purchased from Ogige Market in Nsukka, Enugu State, Nigeria.

### Sample preparation

At the beginning acha grains, mungbean seed and cashew kernels were properly sorted and cleaned to extract stones, weeviled seeds, and other foreign matter.

### Processing of acha into flour

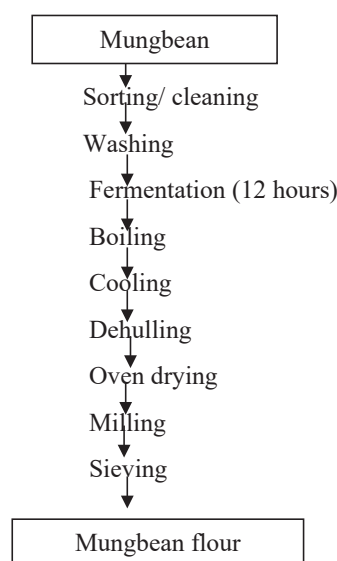
With a slight modification, the procedure mentioned by Agu et al., (2015) was used. 5 kilograms of cleaned and sorted acha were thoroughly washed and drained. To make the flour, the acha was dried in a hot air oven, milled, and sieved through a 1mm pore size sieve. The flour was stored in a transparent airtight container until it was required. Figure 1 depicts the flow diagram for the development of acha flour.



**Figure 1.** Flow diagram for acha flour production. (Agu et al., 2015)

### Processing of mungbean into flour

The procedure described by Enwere (1998) was used with minor modifications. 5 kilograms of mungbeans were cleaned, sorted, and thoroughly washed with clean water before being fermented for 12 hours, then boiled for 30 minutes, cooled and dehulled. To make flour, dehulled mungbeans were dried in a hot air oven, milled and sieved through a 1mm pore size sieve. The flour was held in a clear, airtight container until it was required. Figure 2 depicts the flow diagram for the processing of mungbean flour.



**Figure 2.** Flow diagram for production of mungbean flour. (Enwere, 1998)

### Processing of cashew nut into flour

The modified methods of Badje et al., (2018) was used. A total of 10 kilograms of cashew kernels were cleaned, sorted for discolored kernels and insect damage, and divided into two equal parts. The cashew kernels were milled separately using a blender. One portion of the flour was sieved through a 0.4mm sieve and left as undefatted flour. The defatted flour was made from the remaining part. By continuous maceration for 30 minutes, the flour was de-oiled twice using an apolar solvent (hexane) and the flour/solvent ratio (w:w). The supernatant containing the oil and hexane mixture was removed after 24 hours of incubation, and the remaining cakes were collected in a muslin cloth and pressed. The defatted oil cakes were air dried to remove the solvents, then oven dried for 1 hour at 65 °C to remove all of the solvents. The defatted flour is then held at room temperature in an airtight jar. The flow diagram for the production of cashew nut flour is shown in Figures 3 and 4.

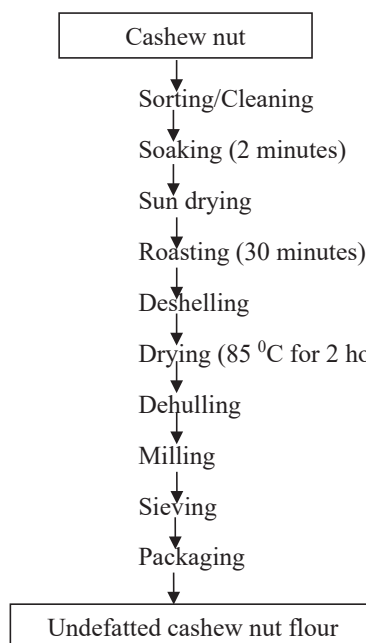


Figure 3. Flow Chart for production of undefatted cashew nut flour. (Badje et al., 2018)

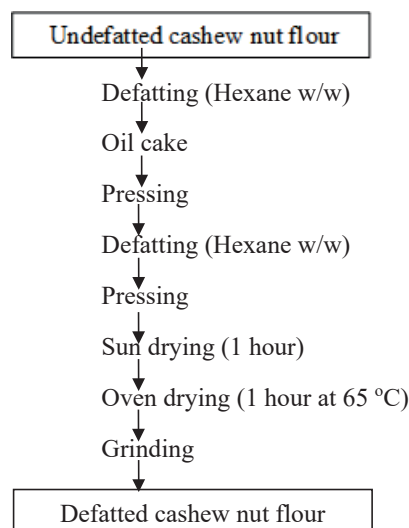


Figure 4. Flow chart for production of defatted cashew nut flour. (Badje et al., 2018)

### Product formulation and production of breakfast cereals

The best blend of acha and mungbean flour was determined using sensory evaluation after composite flour was formulated by mixing acha and mungbean flour. The composite flour (made of acha: mungbean flours) was mixed with graded levels of undefatted and defatted cashewnut flour (100:0; 90:10; 80:20; 70:30; 60:40; 50:50), sugar, salt, and water to make 11 breakfast cereal samples and toasted at 170 °C with continuous stirring till dried products were obtained (Figure 5). A control sample was produced from 100 % acha and mungbean composite flour as shown in Table 1.

#### Production of Breakfast Cereals

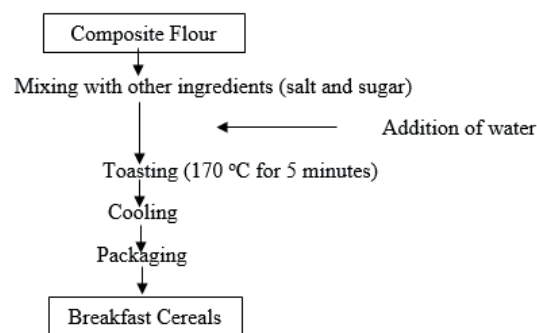


Figure 5. Flow chart for the production of breakfast cereal. (Usman, 2012)

## Analysis of Samples

The proximate composition of the blended samples were examined using AOAC, 2010. A bioassay was done using albino rats to determine the bioavailability of protein in comparison with a control (commercial product).

### Proximate Analysis of the Samples

#### Determination of Moisture Content

Clean crucibles were dried for 1 hour in a hot air oven at 100 oC and then cooled in a desiccator to achieve a constant weight. Two grams of each of the samples were weighed into the different crucibles and dried for four hours at 105oC to ensure that the weights were constant. Loss in weight of the samples were recorded and the moisture content was calculated as:

$$\% \text{ Moisture Content} = \frac{\text{weight loss}}{\text{Sample weight}} \times \frac{100}{1}$$

#### Determination of crude fat

A Soxhlet extractor was mounted, along with a reflux condenser and a 500 ml round bottom flask. A labeled thimble was used to measure two grams of the sample. The round bottom flask was filled with petroleum ether (300 ml), and the extractor thimble was plugged with cotton wool. After allowing the Soxhlet apparatus to reflux for about 6 hours, the thimble was

removed. Petroleum ether was collected for re-use. The flask was dried for 1 hour at 105 °C in an oven, then cooled in a dessicator before being weighed. The percentage fat was calculated as:

$$\% \text{ Fat} = \frac{\text{Weight of fat}}{\text{Sample weight}} \times \frac{100}{1}$$

#### Determination of crude protein

After putting 2 g of the sample in a Kjeldhal flask, the flask was filled with anhydrous sodium sulphate (5 g of Kjeldhal catalyst). In addition to a few boiling chips, concentrated sulphuric acid (H<sub>2</sub>SO<sub>4</sub>; 25 ml) was added. In a fume chamber, the flask was heated until a clear solution was obtained. The clear solution was moved into a 250 ml volumetric flask and filled up to the mark with distilled water after cooling at room temperature.

Distillation: The apparatus was set up after cleaning the distillation unit volume. A 100 ml conical flask was filled with five milliliters (5 ml) of 2 % boric acid solution and three drops of methyl red indicator. 5 ml of the sample digest was pipetted into the apparatus and washed down with distilled water after placing the conical flask under the condenser. In addition, 5 ml of 60 % NaOH was added to the digest. 100 ml of the sample was stored in the receiving flask after it was heated. The content was titrated with

**Table 1.** Composite flour formulations for breakfast cereals made from acha, mungbean, undefatted and defatted cashew nut flour blends

Sample	Sample Code	Code Ratio	Percentage
ACMB	AC+MB	100: 0	100% AC + MB, 0%DCN/UCN
AMDN <sub>1</sub>	AC+MB: DCN	90: 10	90 % AC+ MB, 10 % DCN
AMDN <sub>2</sub>	AC+MB: DCN	80: 20	80 % AC + MB, 20 % DCN
AMDN <sub>3</sub>	AC+MB: DCN	70: 30	70 % AC + MB, 30 % DCN
AMDN <sub>4</sub>	AC+MB: DCN	60: 40	60 % AC + MB, 40 % DCN
AMDN <sub>5</sub>	AC+MB: DCN	50: 50	50 % AC + MB, 50 % DCN
AMUN <sub>1</sub>	AC+MB: UCN	90: 10	90 % AC + MB, 10 % UCN
AMUN <sub>2</sub>	AC+MB: UCN	80: 20	80 % AC + MB, 20 % UCN
AMUN <sub>3</sub>	AC+MB: UCN	70: 30	70 % AC + MB, 30 % UCN
AMUN <sub>4</sub>	AC+MB: UCN	60: 40	60 % AC + MB, 40 % UCN
AMUN <sub>5</sub>	AC+MB: UCN	50: 50	50 % AC + MB, 50 % UCN

AC= Acha; MB= Mungbean; DCN= Defatted Cashew nut Flour; UCN= Undefatted Cashew nut Flour



0.04 M H<sub>2</sub>SO<sub>4</sub> and the desired end point, which was pink in color was achieved. The nitrogen percentage was determined using the following formula:

$$\% = \frac{\bar{T} \times 14.01 \times 0.01 \times \text{dilution factor}}{2.0 \times 1000} \times \frac{100}{1}$$

Where, T= Titre value;

2.0 g = Weight of the sample;

0.01 = Molarity of HCl;

14.01 = Atomic mass of nitrogen;

% protein = % N × 6.25

Where; 6.25 = Conversion factor of protein.

#### Determination of total ash

Two grams of well-blended samples were weighed into a shallow ashing dish (crucible) that has been ignited, cooled in a desiccator, and weighed after reaching room temperature. The crucibles and their contents were heated to 550 oC in a muffle furnace. The ashing took 8 hours to complete. The ashed samples were removed from the muffle furnace, moistened with a few drops of water to expose un-ashed carbon, dried in the oven at 100 oC for four hours. The crucibles were removed from the muffle furnace, cooled in a desiccator, and weighed after reaching room temperature. Percentage ash was calculated using the expression:

$$\% \text{ Ash} = \frac{\text{Weight of Ash}}{\text{Weight of sample}} \times \frac{100}{1}$$

#### Determination of crude fibre

3 grams of the sample was placed in a 50 ml beaker, fats were extracted with petroleum ether by stirring, settling, and decanting three times. Before being transferred to a 60 ml dried beaker, the sample was air dried. After adding 200 ml of 1.25 H<sub>2</sub>SO<sub>4</sub>, a few drops of anti-foaming agent were added to the beaker. This beaker was placed on the digestion apparatus using a pre-adjusted hot plate and boiled for 30 minutes with frequent rotating of the beaker in order to prevent solid adhesion to the sides of the beaker. The mixture was allowed to settle for 1 minute before being filtered through a Buchner funnel.

The insoluble substance was washed away with boiling water without breaking the suction until it was acid free. The residue was then rinsed back into the original flask with a wash bottle containing 200 ml NaOH solution. It was quickly boiled for another 30 minutes with the same precautions as before. It was filtered under suction after 30 minutes after being allowed to stand for 1 minute. The residue was washed with boiling water first, then 1 % HCl acid, and then boiling water again until it was acid-free. It was then moved to an ash dish and dried at 100 °C to a constant weight after being washed twice with alcohol and three times with ether. This dried residue was incinerated at 600 oC for 30 minutes before cooling in a desiccator and being weighed. The fibre content was calculated as a difference between the incinerated residue and the oven dried residue and expressed in percentage as shown:

$$\% \text{ Crude fibre} = \frac{\text{weight after oven dried} - \text{weight after incineration}}{\text{Total weight of sample taken}} \times \frac{100}{1}$$

#### Determination of carbohydrate content (by difference)

The carbohydrate content was calculated using the difference method.

$$\% \text{ Carbohydrate} = 100 - \% (\text{moisture} + \text{protein} + \text{ash} + \text{crude fibre})$$

#### Bioassay

The breakfast cereal was used for 28 days bioassay study using 84 healthy male albino rats weighing 60 - 90 g. The rats were divided into twelve groups (seven per group) including a control group. The albino rats were housed in well ventilated cages containing wood shaving for bedding. The rats were allowed to acclimatize for seven days and were maintained with the breakfast cereal and tap water under room temperature.

#### Determination of feed and water intake

Feed intake was determined using the slightly modified method described by Kamau et al.,

(2017). The quantity of feed and water consumed was calculated daily based on quantity of feed and water supplied the day before and the quantity left after 24 hours.

### Determination of weight gain/growth rate

Weight gain was determined by slightly modifying the method described by Kamau et al., (2017). Body weights were taken before starting dosing, once every seven days, and on the last day of the study. A digital top loader balance was used to determine the body weights. The difference between the original and final body weight was used to calculate weight gain.

### Feed Efficiency Ratio (FER)

The feed efficiency ratio was calculated using the formula suggested by FAO (2011). Feed efficiency ratio was expressed as:

$$\text{FER} = \frac{\text{weight gain (g)}}{\text{feed intake (g)}}$$

### Protein Efficiency Ratio (PER)

The Protein Efficiency Ratio (PER) was calculated using the formula suggested by FAO (2011). Protein efficiency ratio was expressed as:

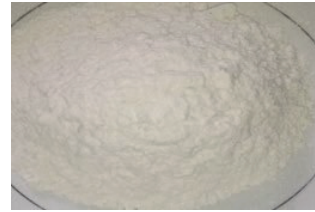
$$\text{PER} = \frac{\text{feed intake (g)}}{\text{gram of protein in diet (g)}}$$

### Experimental design and data analysis

Experimental design and data analysis was carried out in accordance with the method described by Onuh et al., (2019). The experiment was designed using a totally randomized method (CRD). Statistical Product for Service Solution (SPSS) version 23.0 was used to analyze the data using one-way analysis of variance (ANOVA) and Duncan multiple range test ( $p=0.05$ ) to separate the means.

## RESULTS AND DISCUSSION

Plates 1 – 4 show the single flours from acha, mungbean and cashew nut (defatted and undefatted) flours. The formulated breakfast cereals are presented in Plates 5 – 10.



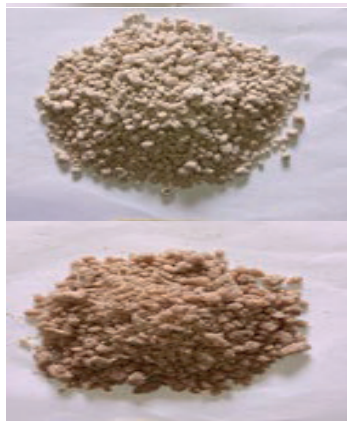


Plate 1: Acha flour

Plate 2: Mungbean

Plate 3: Undefined cashew nut flour

Plate 4: defatted cashew nut flour

Plate 5: Sample ACMB

Plate 6: Sample AMDN2

Plate 7: Sample AMDN3

Plate 8: Sample AMDN5

Plate 9: Sample AMUN1

Plate 10: Sample AMUN5

#### KEY:

ACMB = 80 % acha and 20 % mungbean flour

AMDN2 = 80 % ACMB and 20 % defatted cashew nut flour

AMDN3 = 70 % ACMB and 30 % defatted cashew nut flour

AMDN5 = 50 % ACMB and 50 % defatted cashew nut flour

AMUN1 = 90 % ACMB and 10 % undefined cashew nut flour

AMUN5 = 50 % ACMB and 50 % undefined cashew nut flour

#### **Proximate composition of the formulated breakfast cereals from acha, mungbean, defatted and undefined cashew nut flours**

Tables 2 and 3 show the proximate composition of the raw materials and formulated samples. The results showed some significant changes at  $p < 0.05$ .

The proximate composition of acha, mungbean and cashew nut flours is shown in Table 2. The moisture content varied between 4.41 and 8.05 %. The moisture content of the different flours

were significantly different ( $p > 0.05$ ). Mungbean flour had the highest moisture value (8.05 %) and is lower than the values obtained by Mbaeyi-Nwaoha and Odo (2018) and that obtained by Oburuoga and Anyika (2012); 12.33 and 10.74%, respectively. The moisture content of acha flour is 7.22 %, and is significantly different from the moisture content of the defatted and undefined cashew nut flours. The lowest moisture content was found in defatted cashew nut flour (4.41 %). This was consistent with the values obtained by Badje et al., (2018) and Emelike et al., (2015) for defatted cashewnut flour (4.2 and 4.4 %). This indicated that defatting reduced the moisture content of raw materials. The different flours with lower moisture content (less than 14%) could have a lower risk of bacterial activity and mould growth, which could cause undesirable changes (Ihekoronye and Ngoddy, 1985).

The amount of ash in the samples ranged from 2.10 to 4.60 %. The highest value (4.60 %) was found in undefined cashew nut flour, while the lowest (2.10 %) was found in acha flour. The ash content for mungbean and acha flour was comparable to the values (3.23 and 2.25 respectively) reported by Oburuoga and Anyika (2012). The ash content of defatted cashew nut flour (2.71 %) was significantly lower ( $p < 0.05$ ) than that of undefined flour (4.60 %).

The percentage of protein in the samples ranged from 7.05 to 31.04 %. The protein content of acha flour was the lowest (8.05 %). This was comparable to the value obtained by Ayo and Johnson (2018). Mungbean flour had a protein content of 22.08 %. This was higher than the values reported by Setyaningsih et al., (2019) who reported 18.42 % and Mbaeyi-Nwaoha and Odo (2018) who reported 18.92 %, but lower than that reported by Oburuoga and Onyika (2012) which was 31.31 %.



When compared to the undefatted sample (20.78 %), the protein content of the defatted flour (31.04 %) is significantly ( $p < 0.05$ ) higher.

According to Aremu et al., (2005), the crude protein content of raw cashew kernels was higher than that of bambara ground nut (11.6 %) and kersting's groundnut (12.9 %). Omosuli et al., (2009) recorded a higher value (27.31 %) for roasted and defatted cashew nut flour. The disparity between these reports may be a result of methods used in processing the cashew nut and the type of raw material.

The crude fat content of acha and mungbean flours were 2.07 and 2.51 %, respectively. This low-fat content could be probably due to the fact that acha and mungbean flours were obtained from cereal and legume which are known to contain little fat. Defatting significantly ( $p < 0.05$ ) decreased the crude fat content of cashew nut flour to 19.69 % from 41.37 % in the undefatted sample. These values were comparable with those observed by Badje et al., (2018); Emelike et al., (2015) and Ogungbenle (2014). It is possible that the differences in these reports are due to the processing methods used, variations in raw materials and chemicals used for evaluation.

Crude fibre content ranged from 1.74 and 6.36 %. Acha flour had a crude fibre content of 4.29 % while mungbean flour had a crude fibre content of 6.11 %. When compared to the undefatted sample, the crude fiber content of defatted flour (6.36 %) was significantly ( $p < 0.05$ ) different (1.73 %). Owing to the removal of fat, defatted samples have a higher flour volume per unit weight than

undefatted samples, leading to the rise in fibre content of the defatted samples (Omowaye-Taiwo et al., 2014).

The carbohydrate content of acha flour was 77.25 % and therefore significantly ( $p < 0.05$ ) different than the other flours. Mungbean flour had 57.72 % carbohydrate content. Setyaningsih et al., (2019) reported similar results for mungbean flour (66.25 %) and Ayo and Johnson, (2018) reported similar results for acha flour (81.60 %). The carbohydrate content of the defatted flour 35.78 % and the undefatted flour 26.07 % differed significantly ( $p < 0.05$ ). This indicates that the various flours are healthy sources of energy and are capable of meeting the body's daily energy requirements.

#### Proximate composition of the formulated breakfast cereals from acha, mungbean, defatted and undefatted cashew nut flours

Table 3 shows the proximate composition of formulated breakfast products made from acha, mungbean, defatted, and undefatted cashew nut flours.

The moisture content of the formulated breakfast cereals ranged from 6.01 to 8.07 %. The highest value (8.07 %) was observed in the product containing 10 % undefatted cashew nut flour (AMUN1) while the least value (6.01 %) was observed in the sample containing 50 % defatted cashew nut flour (AMDN5). The moisture content of the samples were significantly different ( $p < 0.05$ ). A decrease in moisture content was observed as the addition of undefatted and

**Table 2.** Proximate composition (%) of acha, mungbean, defatted and undefatted cashew nut flours (dry basis)

Sample	Moisture	Ash	Fat	Protein	Crude Fibre	Carbohydrate
ACH	7.23 <sup>c</sup> ±0.19	2.10 <sup>a</sup> ±0.25	2.07 <sup>a</sup> ±0.07	7.05 <sup>a</sup> ±0.11	4.29 <sup>b</sup> ±0.05	77.25 <sup>d</sup> ±0.03
MBN	8.05 <sup>d</sup> ±0.09	3.52 <sup>c</sup> ±0.13	2.51 <sup>b</sup> ±0.08	22.03 <sup>c</sup> ± 0.19	6.11 <sup>c</sup> ± 0.18	57.72 <sup>c</sup> ± 0.02
UCN	5.45 <sup>b</sup> ±0.10	4.60 <sup>d</sup> ±0.04	41.37 <sup>d</sup> ±0.18	20.78 <sup>b</sup> ±0.21	1.74 <sup>a</sup> ±0.06	26.07 <sup>a</sup> ±0.48
DCN	4.41 <sup>a</sup> ±0.16	2.71 <sup>b</sup> ±0.11	19.69 <sup>c</sup> ±0.10	31.04 <sup>d</sup> ±0.27	6.36 <sup>c</sup> ±0.16	35.78 <sup>b</sup> ±0.27

Values are means ± standard deviation of duplicate determinations. Values in the same column with different superscripts are significantly ( $p < 0.05$ ) different. ACH = Acha flour, MBN = Mungbean flour, UCN = Undefatted cashew nut flour, DCN = Defatted cashew nut flour.

defatted cashew nut flours increased. This was in agreement with the works done by Badje et al., (2018) and Ojinnaka and Agubulom (2013) who made bread from defatted cashew nut/wheat composite flour and wheat/cashew paste cookies, respectively.

The fat content of the formulated breakfast cereals ranged from 3.60 (ACMB) to 24.51 % (AMUN5). There were significant ( $p<0.05$ ) differences in the fat content of the samples. The lowest value (3.60 %) for fat was recorded in the sample (ACMB) without cashew nut flour addition. This could be due to low fat values observed in acha and mungbean flours. The highest value (24.51 %) was recorded in the sample with 50 % undefatted cashew nut flour (AMUN5). As the amount of undefatted and defatted cashew nut flours was increased, the fat content of the samples increased. This could be due to the high fat

content of the undefatted (41.37 %) and defatted (19.69 %) cashew nut flours.

Crude protein content of the samples ranged from 10.24 to 20.10 %. The crude protein content of the samples was significantly ( $p<0.05$ ) different. The protein content of sample AMUN1 (10 % undefatted cashew nut flour) was the lowest (10.24 %), while sample AMDN5 (50 % defatted cashew nut flour) had the highest. With the addition of undefatted cashew nut flour, the protein content of the formulated products increased. This could be due to the fact that cashew nuts contain a significant amount of protein. With the addition of defatted cashew nut flour, the protein content of the samples increased as well. This increase in protein was, however, significantly ( $p<0.05$ ) higher than that observed in the undefatted cashew nut flour samples. This could indicate that the presence of

**Table 3.** Proximate composition (%) of the formulated breakfast cereals from acha, mungbean, defatted and undefatted cashew nut flour blends

Sample	Moisture	Ash	Fat	Protein	Crude Fibre	Carbohydrate
ACMB	7.58 <sup>f</sup> ±0.21	3.39 <sup>a</sup> ±0.39	3.60 <sup>a</sup> ±0.39	11.61 <sup>c</sup> ±0.05	5.74 <sup>d</sup> ±0.08	68.08 <sup>k</sup> ±0.26
AMDN <sub>1</sub>	7.32 <sup>ef</sup> ±0.27	3.39 <sup>a</sup> ±0.25	6.11 <sup>b</sup> ±0.06	13.89 <sup>f</sup> ±0.04	5.82 <sup>de</sup> ±0.06	63.48 <sup>i</sup> ±0.49
AMDN <sub>2</sub>	7.23 <sup>dc</sup> ±0.05	3.61 <sup>ab</sup> ±0.11	7.92 <sup>c</sup> ±0.06	14.98 <sup>g</sup> ±0.15	5.96 <sup>ef</sup> ±0.08	60.31 <sup>h</sup> ±0.00
AMDN <sub>3</sub>	6.92 <sup>c</sup> ±0.13	3.82 <sup>bcd</sup> ±0.02	9.92 <sup>d</sup> ±0.23	17.01 <sup>h</sup> ±0.03	6.06 <sup>f</sup> ±0.05	56.28 <sup>f</sup> ±0.16
AMDN <sub>4</sub>	6.54 <sup>b</sup> ±0.06	4.01 <sup>cd</sup> ±0.04	11.82 <sup>e</sup> ±0.01	18.61 <sup>i</sup> ±0.03	6.78 <sup>g</sup> ±0.10	52.24 <sup>d</sup> ±0.02
AMDN <sub>5</sub>	6.01 <sup>a</sup> ±0.04	4.15 <sup>dc</sup> ±0.04	13.09 <sup>e</sup> ±0.13	20.10 <sup>j</sup> ±0.03	7.07 <sup>h</sup> ±0.05	49.81 <sup>b</sup> ±0.19
AMUN <sub>1</sub>	8.07 <sup>g</sup> ±0.03	3.48 <sup>ab</sup> ±0.02	10.85 <sup>c</sup> ±0.03	10.24 <sup>a</sup> ±0.04	5.70 <sup>d</sup> ±0.09	61.40 <sup>i</sup> ±0.47
AMUN <sub>2</sub>	7.86 <sup>g</sup> ±0.06	3.71 <sup>abc</sup> ±0.03	13.68 <sup>b</sup> ±0.14	11.04 <sup>b</sup> ±0.09	5.05 <sup>c</sup> ±0.05	58.66 <sup>g</sup> ±0.13
AMUN <sub>3</sub>	7.40 <sup>ef</sup> ±0.08	4.12 <sup>dc</sup> ±0.08	17.09 <sup>i</sup> ±0.23	11.61 <sup>c</sup> ±0.03	4.77 <sup>b</sup> ±0.11	55.00 <sup>e</sup> ±0.26
AMUN <sub>4</sub>	7.03 <sup>cd</sup> ±0.13	4.46 <sup>c</sup> ±0.04	20.71 <sup>j</sup> ±0.09	12.99 <sup>d</sup> ±0.11	4.15 <sup>a</sup> ±0.05	50.68 <sup>c</sup> ±0.05
AMUN <sub>5</sub>	6.89 <sup>c</sup> ±0.11	4.80 <sup>f</sup> ±0.04	24.51 <sup>k</sup> ±0.02	13.39 <sup>c</sup> ±0.04	4.02 <sup>a</sup> ±0.06	46.40 <sup>a</sup> ±0.01

Values are means ± standard deviation of duplicate determinations. Values in the same column with different superscripts are significantly ( $p<0.05$ ) different. ACMB= Breakfast cereal made from 80% Acha and 20% Mungbean flour, AMDN<sub>1</sub>= Breakfast cereal made from 90% ACMB + 10% Defatted cashew nut flour, AMDN<sub>2</sub>= Breakfast cereal made from 80% ACMB + 20% Defatted cashew nut flour, AMDN<sub>3</sub>= Breakfast cereal made from 70% ACMB + 30% Defatted cashew nut flour, AMDN<sub>4</sub>= Breakfast cereal made from 60% ACMB + 40% Defatted cashew nut flour, AMDN<sub>5</sub>= Breakfast cereal made from 50% ACMB + 50% Defatted cashew nut flour, AMUN<sub>1</sub>= Breakfast cereal made from 90% ACMB + 10% Undefatted cashew nut flour, AMUN<sub>2</sub>= Breakfast cereal made from 80% ACMB + 20% Undefatted cashew nut flour, AMUN<sub>3</sub>= Breakfast cereal made from 70% ACMB + 30% Undefatted cashew nut flour, AMUN<sub>4</sub>= Breakfast cereal made from 60% ACMB + 40% Undefatted cashew nut flour, AMUN<sub>5</sub>= Breakfast cereal made from 50% ACMB + 50% Undefatted cashew nut flour.

oil in the sample causes some protein globules to demobilize (Ogungbenle, 2014). As a result, the removal of fat contributed to an increase in other nutritional parameters like crude protein (Ogungbenle, 2014). This suggests that the sample should be taken defatted, particularly for adults and children who require less fat and more protein.

The ash content of the formulated breakfast cereals showed significant ( $p < 0.05$ ) differences with values ranging from 3.39 to 4.80 %. Kanu et al., (2009) observed lower values (1.3-2.3 %) in a porridge-type breakfast cereal made from pigeon pea and sesame seed.

The crude fibre content of formulated breakfast cereals ranged from 4.02 to 7.07 %. The crude fiber content of the samples differed significantly

( $p < 0.05$ ). As the amount of undefatted cashew nut flour added increased, the crude fiber content decreased. This could be due to the low crude fiber content observed in the undefatted cashew nut flour. The crude fiber content of the formulated breakfast cereals, on the other hand, increased as addition of defatted cashew nut flour increased. This could be due to the increased crude fiber content of the cashew nut flour as a result of defatting.

The carbohydrate content of the formulated breakfast cereals differed significantly ( $p < 0.05$ ), varying from 46.40 to 68.08 %. The carbohydrate content of sample ACMB (80 % acha and 20 % mungbean flour) was the highest (68.08 %). This could be because acha flour, which is a cereal, has a high starch content. Sample AMUN5 (50

**Table 4.** Effect of the formulated breakfast cereal on the feed intake

Sample	Week 1	Week 2	Week 3	Week 4	Total Feed Intake(g)
RCHW	81.44 <sup>bcd</sup> ± 11.27	76.46 <sup>efg</sup> ± 18.67	74.78 <sup>bcd</sup> ± 13.57	90.52 <sup>ef</sup> ± 6.98	323.20
ACMB	74.76 <sup>bc</sup> ± 18.72	64.67 <sup>bcd</sup> ± 7.93	66.31 <sup>abc</sup> ± 11.99	78.12 ± 9.48	283.86
AMDN <sub>1</sub>	91.99 <sup>cd</sup> ± 8.57	85.79 <sup>e</sup> ± 16.40	78.85 <sup>cd</sup> ± 17.51	87.92 <sup>ef</sup> ± 8.75d	344.55
AMDN <sub>2</sub>	91.62 <sup>cd</sup> ± 11.23	75.29 <sup>defg</sup> ± 16.22	64.09 <sup>ab</sup> ± 11.26	81.45 <sup>cdef</sup> ± 13.43	312.45
AMDN <sub>3</sub>	86.20 <sup>bcd</sup> ± 15.85	69.75 <sup>cdef</sup> ± 5.10	82.28 <sup>d</sup> ± 9.51	83.17 <sup>cdef</sup> ± 13.82	321.40
AMDN <sub>4</sub>	93.14 <sup>d</sup> ± 9.81	82.12 <sup>fg</sup> ± 10.65	71.99 <sup>abcd</sup> ± 12.33	91.90 <sup>f</sup> ± 7.18	339.15
AMDN <sub>5</sub>	79.74 <sup>bcd</sup> ± 15.89	49.25 <sup>a</sup> ± 7.49	66.86 <sup>abc</sup> ± 8.05	77.47 <sup>bcd</sup> ± 13.37	272.71
AMUN <sub>1</sub>	74.82 <sup>bc</sup> ± 21.09	62.44 <sup>abcd</sup> ± 11.83	76.42 <sup>bcd</sup> ± 11.07	68.70 <sup>ab</sup> ± 8.20	282.38
AMUN <sub>2</sub>	76.88 <sup>bcd</sup> ± 12.72	67.12 <sup>bcd</sup> ± 9.61	70.30 <sup>abcd</sup> ± 10.76	84.22 <sup>cdef</sup> ± 10.85	298.52
AMUN <sub>3</sub>	75.24 <sup>bc</sup> ± 12.99	65.11 <sup>bcd</sup> ± 4.70	63.18 <sup>ab</sup> ± 10.70	72.53 <sup>abc</sup> ± 12.31	276.06
AMUN <sub>4</sub>	70.0 <sup>ab</sup> ± 9.74	61.08 <sup>abc</sup> ± 10.16	58.89 <sup>a</sup> ± 11.79	67.69 <sup>ab</sup> ± 8.12	257.66
AMUN <sub>5</sub>	55.82 <sup>a</sup> ± 15.64	54.40 ± 8.21 <sup>ab</sup>	60.08 ± 8.85 <sup>a</sup>	63.92 ± 7.83 <sup>a</sup>	234.22

Values are means ± standard deviation of duplicate determinations. Values in the same column with different superscripts are significantly ( $p < 0.05$ ) different. ACMB= Breakfast cereal made from 80% Acha and 20% Mungbean flour, AMDN1= Breakfast cereal made from 90% ACMB + 10% Defatted cashew nut flour, AMDN2= Breakfast cereal made from 80% ACMB + 20% Defatted cashew nut flour, AMDN3= Breakfast cereal made from 70% ACMB + 30% Defatted cashew nut flour, AMDN4= Breakfast cereal made from 60% ACMB + 40% Defatted cashew nut flour, AMDN5= Breakfast cereal made from 50% ACMB + 50% Defatted cashew nut flour, AMUN1= Breakfast cereal made from 90% ACMB + 10% Undefatted cashew nut flour, AMUN2= Breakfast cereal made from 80% ACMB + 20% Undefatted cashew nut flour, AMUN3= Breakfast cereal made from 70% ACMB + 30% Undefatted cashew nut flour, AMUN4= Breakfast cereal made from 60% ACMB + 40% Undefatted cashew nut flour, AMUN5= Breakfast cereal made from 50% ACMB + 50% Undefatted cashew nut flour, RCHW= Rat chow

% undefatted cashew nut flour) had the lowest (46.40 %) carbohydrate content. This could be due to the high fat content observed in the sample.

### Effect of formulated breakfast cereals on feed intake

The effect of the formulated breakfast cereals on the feed intake of the rats is shown in Table 4 which shows the results of the average weekly feed intake of the experimental animals. The experimental animals consumed between 234.22 and 344.55 g of feed in total. It was observed that the groups fed with samples AMDN1 (90 % ACMB and 10 % defatted cashew nut), AMDN4 (60 % ACMB and 40 % defatted cashew nut) and

the commercial control diet consumed more food than the other groups. Rats fed samples AMUN4 (60 % ACMB and 40 % undefatted cashew nut) and AMUN5 (50 % ACMB and 50 % undefatted cashew nut) were observed to have consumed the least quantity of food. This could be due to the fact that samples AMUN4 and AMUN5 had higher portions of the undefatted cashew nut flour and cashew due to its high fat content could increase satiety thereby reducing rate and volume of food consumed. The quantity of food consumed and the composition of the food are factors that determine the nutrition of the consumer (Ikujenlola et al., 2015).

### Effect of formulated breakfast cereals on the weight gain/growth rate

**Table 5.** Effect of the formulated breakfast cereals on weight (grams) gain of the rats

Sample	Initial Weight (g)	Week 1	Week 2	Week 3	Week 4	Weight Gain
RCHW	71.23 <sup>a</sup> ±5.98	93.12 <sup>ab</sup> ±6.59	115.16 <sup>dc</sup> ±2.98	194.68 <sup>c</sup> ±9.64	151.48 <sup>c</sup> ±7.68	80.25
ACMB	73.84 <sup>ab</sup> ±5.26	86.49 <sup>a</sup> ±6.81	95.32 <sup>a</sup> ±3.29	102.47 <sup>a</sup> ±4.13	107.98 <sup>a</sup> ±2.73	34.14
AMDN <sub>1</sub>	72.31 <sup>ab</sup> ±9.08	94.62 <sup>b</sup> ±9.62	122.73 <sup>c</sup> ±13.33	136.42 <sup>d</sup> ±13.61	145.45 <sup>dc</sup> ±12.90	73.14
AMDN <sub>2</sub>	72.04 <sup>ab</sup> ±8.17	91.93 <sup>ab</sup> ±8.91	114.47 <sup>dc</sup> ±8.48	124.58 <sup>b</sup> ±9.76	132.92 <sup>bc</sup> ±9.32	60.88
AMDN <sub>3</sub>	73.03 <sup>ab</sup> ±8.47	96.91 <sup>b</sup> ±9.2	112.98 <sup>dc</sup> ±9.45	121.24 <sup>b</sup> ±9.20	127.27 <sup>b</sup> ±9.59	54.24
AMDN <sub>4</sub>	72.98 <sup>ab</sup> ±11.18	98.09 <sup>b</sup> ±10.06	123.05 <sup>c</sup> ±12.35	132.01 <sup>cd</sup> ±12.3	139.57 <sup>bc</sup> ±9.04	66.59
AMDN <sub>5</sub>	70.60 <sup>a</sup> ±5.54	92.20 <sup>ab</sup> ±7.87	116.14 <sup>dc</sup> ±10.26	124.68 <sup>bc</sup> ±9.00	132.37 <sup>bc</sup> ±9.04	61.77
AMUN <sub>1</sub>	68.73 <sup>b</sup> ±31.80	77.87 <sup>ab</sup> ±35.66	110.47 <sup>cd</sup> ±8.67	118.47 <sup>b</sup> ±8.23	123.23 <sup>b</sup> ±9.00	54.50
AMUN <sub>2</sub>	68.14 <sup>a</sup> ±2.69	83.79 <sup>a</sup> ±3.12	100.44 <sup>ab</sup> ±3.48	106.12 <sup>a</sup> ±3.41	109.85 <sup>a</sup> ±4.41	41.71
AMUN <sub>3</sub>	69.91 <sup>a</sup> ±7.37	87.64 <sup>ab</sup> ±7.39	108.38 <sup>bcd</sup> ±6.42	118.85 <sup>b</sup> ±11.55	125.82 <sup>b</sup> ±11.99	55.91
AMUN <sub>4</sub>	68.39±4.24 <sup>a</sup>	85.52±7.11 <sup>a</sup>	95.76±6.07 <sup>a</sup>	102.86±5.74 <sup>a</sup>	107.35±6.11 <sup>a</sup>	38.96
AMUN <sub>5</sub>	67.34±3.57 <sup>a</sup>	83.33±6.30 <sup>ab</sup>	101.64±6.01 <sup>abc</sup>	105.66±4.94 <sup>a</sup>	110.27±5.83 <sup>a</sup>	42.93

Values are means ± standard deviation of duplicate determinations. Values in the same column with different superscripts are significantly ( $p < 0.05$ ) different. ACMB= Breakfast cereal made from 80% Acha and 20% Mungbean flour, AMDN1= Breakfast cereal made from 90% ACMB + 10% Defatted cashew nut flour, AMDN2= Breakfast cereal made from 80% ACMB + 20% Defatted cashew nut flour, AMDN3= Breakfast cereal made from 70% ACMB + 30% Defatted cashew nut flour, AMDN4= Breakfast cereal made from 60% ACMB + 40% Defatted cashew nut flour, AMDN5= Breakfast cereal made from 50% ACMB + 50% Defatted cashew nut flour, AMUN1= Breakfast cereal made from 90% ACMB + 10% Undefatted cashew nut flour, AMUN2= Breakfast cereal made from 80% ACMB + 20% Undefatted cashew nut flour, AMUN3= Breakfast cereal made from 70% ACMB + 30% Undefatted cashew nut flour, AMUN4= Breakfast cereal made from 60% ACMB + 40% Undefatted cashew nut flour, AMUN5= Breakfast cereal made from 50% ACMB + 50% Undefatted cashew nut flour, RCHW = Rat chow

Table 5 shows the effect of the formulated breakfast cereals on the weight gain/growth rate of the rats. The mean weight gain of the animals ranged between 34.14 – 80.25 g. All the formulated diets supported positive weight gain throughout the feeding trial except the control diet (rat chow), which showed a decrease in weight in the last week of the feeding trial. This could mean that the formulated diets support growth as shown by the increase in weight.

The highest weight gain over the period of investigation was recorded for the control diet with an average total weight gain of 80.25 g while the second-best diet was sample AMDN1 (90 ACMB and 10 % defatted cashew nut) with an average weight gain of 73.14 % while sample ACMB (80 acha and 20 % mungbean) had the least weight gain (34.14 %).

In addition, it was observed that the samples incorporated with the defatted cashew nut flour showed higher weight gain than those incorporated with the undefatted cashew nut flour. This may indicate that removing the fat led to an improvement in other nutritional parameters such as crude protein (Ogungbenle, 2014) and, as a result, increased the growth rate of the test animals.

#### Effect of the formulated diet on the feed and protein efficiency of the diets

The Feed Efficiency Ratio (FER) of the diets are presented in Table 6. The values vary from 0.12-0.25. The feed efficiency of the formulated diets compared well with the control diet. The rats fed rat chaw had the highest feed efficiency. This could be due to the fact that rat chaw is a stable

**Table 6.** Food intake, protein intake and body weight gain of rats for the assessment of FER and PER

Samples	Feed Intake (g)	Protein Intake (g)	Weight Gain (g)	FER	PER
ACMB	283.86	32.95	34.14	0.25	1.03
AMDN <sub>1</sub>	344.55	47.89	73.14	0.21	1.53
AMDN <sub>2</sub>	312.45	49.63	60.88	0.19	1.22
AMDN <sub>3</sub>	321.40	54.64	54.24	0.17	0.99
AMDN <sub>4</sub>	339.15	63.08	66.59	0.20	1.06
AMDN <sub>5</sub>	272.71	54.81	61.77	0.23	1.13
AMUN <sub>1</sub>	282.38	28.80	54.50	0.19	1.89
AMUN <sub>2</sub>	298.52	32.84	41.71	0.14	1.27
AMUN <sub>3</sub>	276.06	32.02	55.91	0.20	1.75
AMUN <sub>4</sub>	257.66	33.43	38.39	0.15	1.15
AMUN <sub>5</sub>	234.22	31.36	42.93	0.18	1.37
RCHW	323.20	42.02	80.25	0.25	1.19

ACMB= Breakfast cereal made from 80% Acha and 20% Mungbean flour (ACMB), AMDN1= Breakfast cereal made from 90% ACMB + 10% Defatted cashew nut flour, AMDN2= Breakfast cereal made from 80% ACMB + 20% Defatted cashew nut flour, AMDN3= Breakfast cereal made from 70% ACMB + 30% Defatted cashew nut flour, AMDN4= Breakfast cereal made from 60% ACMB + 40% Defatted cashew nut flour, AMDN5= Breakfast cereal made from 50% ACMB + 50% Defatted cashew nut flour, AMUN1= Breakfast cereal made from 90% AMB + 10% Undefatted cashew nut flour, AMUN2= Breakfast cereal made from 80% ACMB + 20% Undefatted cashew nut flour, AMUN3= Breakfast cereal made from 70% ACMB + 30% Undefatted cashew nut flour, AMUN4= Breakfast cereal made from 60% ACMB + 40% Undefatted cashew nut flour, AMUN5= Breakfast cereal made from 50% ACMB + 50% Undefatted cashew nut flour, RCHW= Rat chow



food for rats. Sample AMDN5 (50 % ACMB and 50 % defatted cashew nut) had the highest (0.23) feed efficiency amongst the formulated diets while sample ACMB (80 % mungbean and 20 % acha flour) had the least (0.12) feed efficiency. The feed efficiency ratio measures the ability of a food to sustain growth.

Table 6 shows the Protein Efficiency Ratio (PER) of the formulated breakfast cereals and the control diet. The protein efficiency ratio of the formulated diets varies between 0.99 -1.91. Sample AMDN3 (70 % ACH and 30 % MBN) had the least (0.99) PER. This could be due to lower protein quality in the diet. Table 6: Protein Efficiency Ratio (PER) of the formulated breakfast cereals

The control diet had the highest (1.91) PER. The PER values of the formulated diets (0.99–1.89) did not match those recorded in the literature for casein (2.5), whole egg (3.8) and cow's milk (2.0) as reported by Okoye (1992). This might be due to the fact that proteins from animal sources have a higher biological value and, as a result, a higher PER than proteins from plant sources. Anti-nutritional factors in the diet may have hindered successful protein utilization (Nassar and Sousa, 2007).

## CONCLUSION

This study showed that an acceptable breakfast product of adequate nutritional value could be produced from graded portions of acha, mungbean and cashew nut (defatted and undefatted) flours. Producing breakfast cereals with cashew nut improved the proximate composition especially the protein content (up to 20.10 %). The fat content of the formulated breakfast products improved as the amount of cashew nut flour added was increased. This could lead to rancidity thereby affecting the shelf stability of the product. Feeding trial showed positive increase in weight when the rats were fed the formulated breakfast products, this implies that the products could support growth. No adverse growth rate was observed. Feed intake was also acceptable and compared well with the stable diet for rats. Feed efficiency of the samples compared well with the control diet.

Protein efficiency was not up to other protein standards but compared well.

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*Bu sayfa dizgiden dolayı boş bırakılmıştır*



*Research Article*

# Enrichment of wheat bread with carob molasses pulp, a dietary fiber source: Impact on bread quality and acceptability

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## ABSTRACT

Carob is one of the most important plant sources of dietary fiber, which is essential for human health and must be consumed daily. Carob molasses (pekmez) obtained from carob fruit contains many beneficial components for health. Although the molasses pulp that comes out as waste in the production of molasses contains a large amount of fiber, it is not evaluated. In this study; purification, drying and grinding of the crude carob fiber (CCF) from raw molasses pulp was carried out. The obtained CCF flour was added to the bread. After baking bread, the effects of the addition of 1 to 5 % CCF flour on chemicals (moisture, ash and protein) and also textural (hardness, color) and sensory properties (acceptability, taste, softness, appearance) of the bread samples were investigated. The results showed that the addition of CCF up to 4 % into the bread dough had no significant effect compared with the control group on these properties. Therefore, a bread formulation can be developed which is a fibrous bakery product with reduced fat for health and which has better sensory appreciation.

## 1. INTRODUCTION

The part of the carob fruit that can be eaten is rich in nutritive components like minerals (calcium, potassium, phosphorus, iron, etc.), carbohydrates (glucose, fructose, and sucrose), and polyphenolic compounds with an antioxidant activity and beneficial components like crude fiber (Ozcan et al., 2007; Kiroglu, 2001). The sweet, edible portion of the carob fruit can be eaten fresh, or it can be used in the production of molasses (known as pekmez in Turkish) via either traditional or industrial processes. The use of carob fruit directly in the form of flour (Aydın et al., 2017; Papaefstathiou et al., 2018) or its addition into various food products is not a preferable method because it causes a loss in the organoleptic characteristics of the foods. In recent years, there has been a rise in the awareness of the benefits that carob molasses has for human health, which has led to an increase in the commercial production of the carob molasses. Although the carob pulp that is left over after the production of carob molasses contains a sizable quantity of carob fibre in its raw form, it is not utilized in the production of carob-based foods (Ozdemir et al., 2022).

Functional foods are those that are consumed as part of a regular diet, contain no synthetic ingredients, have nutritive effects, and reduce the risk of disease formation due to multiple factors. (Biesalski, 2005) Dietary fiber is one of the functional food components. Dietary fiber is one of the major components of edible plant parts that are resistant to digestion and absorption in the human small intestine and undergo complete or partial fermentation in the large intestine (Harris and Ferguson, 1999). Although dietary fibers are subdivided into numerous subgroups, FAO and WHO have examined them as soluble and insoluble fibers based on their water solubility (Ramulu and Rao, 2003). Insoluble fibers cannot form a gel despite absorbing 20 times as much water as soluble fibers (Tamer et al., 2004). Soluble fibers bind water and form a gel when combined with water. Water-insoluble fibers; lignin, cellulose, and water-insoluble pentosans, whereas water-soluble fibers; pectins and gums (Knuckles et al., 1997).

Glucose and insulin metabolism are improved by soluble fibers. In addition, they reduce the concentration of low-density lipoprotein (LDL) cholesterol in the serum (Gül, 2007). However, water-insoluble fibers have beneficial effects on bowel movements (Kahlon et al., 2001). Dietary fibers have a sedative effect by increasing stool volume and water content. Due to the water-binding property of dietary fibers, stool volume has increased. This prevents constipation (Ekici and Ercoşkun, 2007). It has been discovered that dietary fiber prevents colon cancer in significant ways. It achieves this effect by altering the colon's bacterial flora, preventing the production of toxic metabolites, and shortening the time these metabolites spend in contact with intestinal cells (Anonymous, 1990). There is an inverse correlation between consuming water-insoluble fiber and developing colon cancer. (Anonymous, 1983) suggests increasing consumption of high-fiber foods like wheat and corn bran.

Diabetes is believed to be one of the diseases linked to a lack of dietary fiber. A high consumption of dietary fiber reduces serum glucose concentration. According to (Anderson, 1980), it reduces the need for insulin in diabetics, thereby providing health benefits (Villanueva-Suarez et al., 2003). Soluble fibers regulate blood sugar by allowing glucose to enter the bloodstream slowly, thereby regulating blood sugar levels.

Dietary fiber products are gaining prominence in the concept of healthy nutrition in the modern world. It is well known that dietary fibers play a crucial role in preventing the development of obesity, cardiovascular disease, diabetes, and certain types of cancer (Aksoy, 2000). Therefore, a comprehensive understanding of the chemical and nutritional properties of dietary fiber is essential for expanding its applications.

The addition of fibers to food products began in the 1970s with the production of a few white and wheat breads containing fiber addition (Burdurlu and Karadeniz, 2003). Since this type of bread contains fewer calories than standard products, this procedure is performed for the health benefit of weight loss. The functional aspects of dietary fibers have been identified and

developed during this time. As a result of the numerous advancements that have been made, it is now utilized in a wide variety of products. (Tamer et al., 2004) Numerous categories in the food industry now consider fiber to be a crucial and valuable ingredient. The possible use of an additive carob fibre to rye bread, which had a positive impact on human health, was first examined by Haber et al., 2002. They have shown that a 6% addition of carob fibre to rye bread has a positive effect by lowering cholesterol levels, especially LDL (Rozylo et al., 2017).

In this study, it was aimed to increase the nutritional value of bread consumed daily. For this purpose, carob crude fiber (CCF) was used in the production of traditional or industrially produced bread. The effects of CCF supplement on chemicals (moisture, ash and protein) texture, color and sensory properties of bread were investigated and new formulations developed by using different amounts of CCF instead of wheat flour

## 2. MATERIALS & METHODS

### 2.1. Materials

In this study, carob molasses pulp obtained from a company producing carob molasses in Mersin was used. Carob molasses pulps were dried, ground, and sieved through 450-micron mesh. Dietary fiber obtained from carob molasses pulp has been used in bread production. Wheat flour, salt, and yeast used in bread making were obtained from "Food Studies Application and Research Center" in Mersin University. Ingredients have been stored at +4 °C. Nitric acid, acetic acid, sulfuric acid, trichloroacetic acid, petroleum ether and acetone were obtained from Sigma (Sigma-Aldrich, St. Luis, Mo, USA).

### 2.2. Methods

#### 2.2.1. The process of drying and grinding the carob molasses pulp

The raw carob molasses pulp was dehydrated in an oven (J,P, Selecto, Spain) at temperatures ranging from 50 °C to 55 °C for a duration of seven hours. Following the step of drying, the products were milled using a laboratory mill

(IKA, M20, Labortechnik, Staufen, Germany), and the fraction of this flour was separated using sieves 450 µm in size. This flour was referred to as carob molasses pulp flour, and after it was produced, the flours were placed in airtight plastic containers and kept at a temperature of 4 °C until further analysis.

#### 2.2.2. Carob fiber purification from carob molasses pulp flour

The ISO 6541:1981 method for crude fiber isolation from carob molasses pulp flour was modified (ISO,1981). 10 grams of flour were weighed and placed in a flask of boiling water. 150 mL of boiling extraction solution (70 mL of 70 % acetic acid, 5 mL of concentrated nitric acid, and 2 g of trichloroacetic acid solution) was added to a boiling flask, and the mixture was stirred. In a perpendicularly connected refrigerated boiling flask, the mixture was boiled at 220 °C. for 90 minutes. After boiling, the resulting mixture was filtered through filter paper with a 40 m pore diameter (Whatman Grade). The residues that remained were then washed with 500 mL of hot distilled water. The residue was washed three times with 50 mL of pure acetone solution and once with 50 mL of pure petroleum ether after being cleaned with hot distilled water. After the final washing, the residue was dried in an oven for 10 hours at 105 °C. The powdered substance is known as crude carob fiber (CCF) flour (Keçeli, 2016).

### 1.3. Preparation of bread samples

The traditional method of making bread was used, but instead of flour, various quantities of CCF were incorporated into the bread-making process. The mixtures obtained in accordance with the formulations given in Table 1 were adjusted to mixer speed 2 and mixed for 3 minutes to obtain bread dough. After shaping the dough, it was left to rest for 10 minutes. The rested dough was left to ferment for 30 minutes in an incubator with a temperature of 35 °C and a relative humidity of 85 %. The doughs that came out of the incubator were baked at 220 °C for 20 minutes.

## 1.4. Proximate Evaluation

Official methods were used to determine the moisture, protein, and ash content of control samples and bread samples made with CCF flour (AOAC, 1990). Moisture was measured using a moisture analyzer (Mettler Toledo Moisture Analyzer, HX204, Switzerland), protein was measured using a protein analyzer (Kjeldatherm, Spain), and ash was measured using a gravimetric method (using furnace Carbolite Parsons Lane, England).

### 1.4.1. Color Analysis

Automatic colorimeter readings were taken of the bread samples to determine their outer crust color values (ColorQuest XE Hunter Lab., USA). The findings were reported using the CIELAB system with the parameters  $L^*$ ,  $a^*$ , and  $b^*$ , which indicated lightness (+ve) to darkness (-ve), redness (+ve) to greenness (-ve), and yellowness (+ve) to blueness (-ve), respectively. The color evaluations of the bread samples were performed thrice, with each run consisting of two repetitions.

### 1.4.2. Texture

For texture analysis, hardness values of bread samples were determined using the AACC 74-09 (AACC, 1999) method in a Texture Analyzer (Stable Micro Systems, England). The maximum force in the force-deformation curve obtained as a result of the test expresses the degree of hardness of the sample. Texture analysis was applied to both the outer and inner parts of the samples, which were kept at 25 °C for 1 hour after baking. Experiment 3 repetitions were performed in 2 parallels.

### 1.4.3. Volume and Weight Losses

The bread volume determination method was used for volume analysis (AACC, 1995). After the bread samples had been baked for one hour at room temperature, the test was conducted. The test is based on the principle of rapeseed displacement. The principle of the experiment; Rapeseed seeds are filled to the marked level in a measuring cylinder. It was then emptied again. The loaves were placed in the cylinder, and rapeseed was added once more. The level is populated until it reaches the size indicated. The remaining rapeseed seeds' volume was measured. Three repetitions of the experiment were conducted in two parallels. The weight loss of the bread samples was measured before and after baking

## 1.5. Acceptability in general and sensory qualities

A trained panel of 11 judges evaluated the acceptability and sensory qualities of bread samples using a seven-point hedonic scale (1: dislike to 7: extremely like) (Ekıcı et al., 2015). Staff members, graduate students from the department of food engineering, and experts in food sensory evaluation were chosen as the panelists. It was intended to identify the bread formulations, with various amounts of CCF added, that were sensory equivalent to the control sample. Between samples, water and bread were offered to help with palate cleaning.

## 1.6. Stastical Analysis

The findings of the study were evaluated with the help of the statistical analysis program SPSS version 16.0, which was developed by SPSS Inc. in Chicago, Illinois. In situations where

**Table 1.** Formulations of breads

Ingredients	Control	CCF18	CCF27	CCF35	CCF43	CCF65
CCF (g)	0	1	2	3	4	5
Flour (g)	100	99	98	97	96	95
Salt (g)	1.5	1.5	1.5	1.5	1.5	1.5
Bread yeast (g)	4	4	4	4	4	4
Water (ml)	60	60	60	60	60	60



there were more than two groups that needed to be compared with each other, the One-Way ANOVA was used to determine the difference in mean between the groups. For the purpose of carrying out statistical analysis on the fiber samples, a t-test was conducted.

### 3. RESULT & DISCUSSION

#### 3.1. Proximate Analysis of Bread Samples

Ash, moisture and protein values of bread samples with and without CCF added are given in Table 2. In general, an increase was observed in the ash, moisture and protein values of the bread samples with the addition of CCF, and this increase was found to be statistically significant ( $p \leq 0.05$ ). On the other hand, there is a 16% increase in moisture level compared to the control, especially according to the added CCF ratio. This increase is thought to be proportional to the water absorption capacity of the fiber. A much smaller increase was observed in the amount of protein and ash.

(Ozdemir et al., 2021) reported that carob molasses pulp crude fiber was purified, and some chemical parameters were analyzed. Accordingly, the percentages of moisture, ash, protein, lignin, and other insoluble fibers in crude fiber were calculated to be 6.79, 10.95, 3.03, 52.03, and 27.20 %, respectively. (Owena et al., 2003) determined the amounts of soluble fiber (pectin), insoluble fiber (cellulose, hemicelluloses, lignin, and water-insoluble polyphenols), and water-soluble polyphenols (condensed tannins) in the commercial carob fiber Caromax™ to be

6.2 %, 68.54 %, and 2.84 %, respectively. In a patent study, the carob fiber's lignin, cellulose, hemicelluloses, pectin, and tannins percentages were reported as 50-65 %, 15-25 %, 0.5-2 %, and 3-7 %, respectively.

(Ragae et al., 2011) examined the effects of fiber supplementation on bread's antioxidant capacity and nutritional value. According to the results, the addition of fiber increased the ash content. According to the Turkish Food Codex, the max. humidity value is 38%. However, in our study, the moisture content of the bread prepared with the traditional method was found to be 39.63%. It is thought that the reason for this is the use of the inside of the breads for moisture determination. This increase can be cause of is that the added fiber has a moisture content of 6.79%. It can be said that another reason for the increase in humidity is the storage conditions. (Gomez et al., 2003) analyzed bread with microcrystalline cellulose, pea, cocoa, coffee, orange and two types of wheat bran fiber. It was determined that when the fiber samples were added to the bread at the rate of 2% (except for the orange fiber), the other fibers did not cause a change on the moisture. It was observed that the moisture values increased as the amount of added fiber increased. (1): Different letters in the same column indicate that, there is a statistically significant difference between the values in the same column.

Table 2. Proximate analysis of bread samples with and without CCF

Sample No	% CCF	Ash (%)	Moisture (%)	Protein (%)
Control	0	0.96±0.01 <sup>f</sup>	39.63±0.26 <sup>e</sup>	7.75±0.04 <sup>b</sup>
CCF18	1	1.13±0.03 <sup>e</sup>	43.47±0.19 <sup>d</sup>	7.79±0.12 <sup>b</sup>
CCF27	2	1.26±0.04 <sup>d</sup>	44.88±0.06 <sup>c</sup>	7.82±0.03 <sup>b</sup>
CCF35	3	1.33±0.00 <sup>c</sup>	44.37±0.09 <sup>c</sup>	7.84±0.04 <sup>a</sup>
CCF42	4	1.43±0.02 <sup>b</sup>	45.62±0.28 <sup>b</sup>	7.85±0.11 <sup>a</sup>
CCF63	5	1.54±0.02 <sup>a</sup>	46.23±0.46 <sup>a</sup>	7.88±0.01 <sup>a</sup>

(1): Different letters in the same column indicate that, there is a statistically significant difference between the values in the same column.

(2): Results are calculated on dry basis. Values given are the mean ± standard deviation of three replicates.

lues in the same column that differ significantly ( $p \leq 0.05$ ) are indicated by different superscripts.

### 3.2. Color

CIELAB system color values for control and CCF added outer crust bread samples are given in Table 3. As can be seen in the table, there is a decrease in  $L^*$  (brightness),  $a^*$  (redness) and  $b^*$  (yellowness) values compared to the control sample. In general, the slight decrease in color values is in parallel with the increase in CCF % ratio. This decrease in breads with added CCF was found to be statistically significant ( $p \leq 0.05$ ). It is thought that crude fiber obtained from molasses pulp causes a decrease in  $L$  values because it contains polyphonic compounds, even if it are small. The type of flour and the additives added play an important role in the color values of the breads. (Seres et al., 2005); In a study they carried out, they added dietary fiber obtained from sugar beets to bread wheat. The researchers determined that there was a decrease in the  $L^*$ ,  $a^*$ ,  $b^*$  values of the breads depending on the fiber color.

### 3.3. Texture

The force values applied for the outer crust and crump hardness measurements of bread samples prepared with the addition of CCF are given in Table 4. As can be seen from the table, as the CCF ratio increases, the force applied to both the outer crust and the crump of the bread samples increases. When the outer crust hardness was added at the rate of 1 to 4%, the degree of hardness was not statistically significant compared to the control bread. However, if 5% is added, the outer crust of the bread forms a significant hard structure.

On the other hand, the force applied to the inner part of the seedlings was found to be statistically significant between the samples with CCF added. This difference was also found to be statistically significant with the increase in the amount of CCF. It is thought that the reason for applying more force in the crump is that it is more in the crump of the bread than in the outer crust (Ozdemir et al., 2021) reported that the crude fiber purified from carob molasses was found

**Table 3.** Color value of outer crust bread samples with and without CCF

Sample No	% CCF	$L^*$	$a^*$	$b^*$
Control	0	69.76±1.09 <sup>a</sup>	5.64±0.31 <sup>a</sup>	18.48±0.15 <sup>a</sup>
CCF18	1	61.25±1.16 <sup>b</sup>	4.85±0.29 <sup>b</sup>	16.00±0.37 <sup>b</sup>
CCF27	2	58.13±1.70 <sup>c</sup>	4.79±0.15 <sup>b</sup>	14.29±0.44 <sup>c</sup>
CCF35	3	56.42±0.79 <sup>d</sup>	4.76±0.31 <sup>b</sup>	14.02±0.09 <sup>c</sup>
CCF42	4	55.19±0.58 <sup>d</sup>	4.70±0.15 <sup>b</sup>	12.95±0.37 <sup>d</sup>
CCF63	5	51.08±0.69 <sup>c</sup>	4.13±0.06 <sup>c</sup>	11.77±0.41 <sup>c</sup>

\*Values in the same column that differ significantly ( $p \leq 0.05$ ) are indicated by different superscripts.

**Table 4.** Texture analysis of crust and crumb samples with and without CCF

Sample No	% CCF	Max Force (N)	
		Crust	Crumb
Control	0	19.82 ± 1.31 <sup>c</sup>	12.66 ± 0.83 <sup>f</sup>
CCF18	1	22.80 ± 1.68 <sup>c</sup>	16.82 ± 1.01 <sup>e</sup>
CCF27	2	24.81 ± 4.91 <sup>b,c</sup>	24.06 ± 2.46 <sup>d</sup>
CCF35	3	25.71 ± 5.64 <sup>b,c</sup>	35.57 ± 2.48 <sup>e</sup>
CCF42	4	30.29 ± 5.31 <sup>b</sup>	42.51 ± 3.41 <sup>b</sup>
CCF63	5	63.42 ± 5.34 <sup>a</sup>	45.89 ± 2.74 <sup>a</sup>

\*Significant differences between values in the same column are indicated by various superscripts ( $p \leq 0.05$ ).

to be 52.03 % lignin and 27.20 % other insoluble fiber. Therefore, it can be said that the hardness in the internal structure of bread comes from lignin and insoluble fibers.

(Shehzad et al., 2011) observed the effect of adding dietary fiber to bread on the rheological properties of dough. According to the study, the addition of dietary fiber decreases the amount of water in the viscoelastic gluten network structure of dough. Since this prevents the expansion of gas bubbles at the interface, it results in the formation of breads with a firmer texture.

(Wang et al., 2002) investigated the effects of adding a combination of chickpea fiber, carob fiber, and inulin fiber to the flour used to make bread. According to the research, it has been determined that the mentioned ingredients cause softening of the bread crust.

(Gomez et al., 2003); added 1%, 2%, 3% and 10% of the fibers obtained from coffee and cocoa to wheat flour. When the textural properties of the

bread samples with dietary fiber were examined, no statistically significant difference could be determined within the samples themselves. However, it was determined that the sample containing 10% dietary fiber differed from the control sample, although it did not have a negative effect on its textural properties.

### 3.4. Volume and weight losses

Volume and weight losses of bread formulations with and without added CCF are given in Table 5. As the CCF ratio in bread increases, there is a decrease of up to 10% in volume compared to the control sample. As can be seen from the table, breads with 1 to 3% CCF additives differ significantly from control breads. In the case of adding 4 to 5%, the difference was found to be statistically significant ( $p \leq 0.05$ ) compared to both control and bread with up to 3% CCF added. On the other hand, there was no statistically significant ( $p \leq 0.05$ ) difference between all bread samples before and after baking.

**Table 5.** Volume and weight losses values of control sample and bread samples with CCF added

Sample No	% CCF	Volume (cm <sup>3</sup> )	Weight Losses, g
Control	0	485.50±11.26 <sup>a</sup>	22.41±0.57 <sup>a</sup>
CCF18	1	453.33±21.26 <sup>b</sup>	22.43±0.35 <sup>a</sup>
CCF27	2	450.00±0.00 <sup>b</sup>	22.46±0.30 <sup>a</sup>
CCF35	3	447.50±12.50 <sup>b</sup>	22.52±0.44 <sup>a</sup>
CCF42	4	421.67±6.29 <sup>c</sup>	22.61±0.40 <sup>a</sup>
CCF63	5	413.33±6.29 <sup>c</sup>	22.68±0.36 <sup>a</sup>

\*Different superscripts denote differences between values in the same column that are statistically significant ( $p \leq 0.05$ ).

**Table 6.** Sensory analysis of control sample and bread samples with CCF added

Sample No	% CCF	Appearance	Softness	Taste	Color	Adhesiveness	Overall acceptability
Control	0	5.20±0.84 <sup>a</sup>	5.20±0.84 <sup>a</sup>	5.80±0.84 <sup>a</sup>	5.20±1.30 <sup>a</sup>	5.40±0.55 <sup>a</sup>	5.80±0.45 <sup>a</sup>
CCF18	1	5.00±0.71 <sup>a</sup>	5.20±0.84 <sup>a</sup>	5.40±0.55 <sup>a</sup>	4.60±1.14 <sup>a</sup>	5.40±0.55 <sup>a</sup>	5.40±0.55 <sup>a</sup>
CCF27	2	4.60±0.55 <sup>a,b</sup>	5.00±0.71 <sup>a</sup>	5.40±0.55 <sup>a</sup>	4.60±1.14 <sup>a</sup>	5.20±0.84 <sup>a,b</sup>	5.00±1.22 <sup>a</sup>
CCF35	3	4.20±0.45 <sup>a,b</sup>	5.00±0.71 <sup>a</sup>	4.40±1.14 <sup>a</sup>	4.40±0.55 <sup>a</sup>	4.80±0.45 <sup>a,b,c</sup>	4.80±0.84 <sup>a</sup>
CCF42	4	3.80±0.84 <sup>b</sup>	4.40±0.89 <sup>a,b</sup>	4.00±0.00 <sup>a</sup>	4.20±0.84 <sup>a,b</sup>	4.20±1.09 <sup>b,c</sup>	3.60±0.55 <sup>b</sup>
CCF63	5	2.60±1.14 <sup>c</sup>	3.20±1.48 <sup>b</sup>	2.40±0.55 <sup>b</sup>	2.80±1.26 <sup>b</sup>	4.00±0.71 <sup>c</sup>	2.60±1.14 <sup>b</sup>

\*Different superscripts indicate statistically significant differences between values within the same column ( $p \leq 0.05$ ).

Water provides the ideal fermentation environment for dough. Moreover, dietary fiber has a high-water holding capacity. Additionally, a certain amount of water is required for the formation of the gluten network. Since the amount of water required for the formation of the gluten network structure was lacking in the medium, the gas bubbles that formed during fermentation were unable to be retained in the bread's structure. It is believed that this circumstance results in a reduction in bread volume due to the increased CCF ratio. In their research, (Miguel et al., 1999) added between 2-30% peach fiber to cake samples. It was determined that there was no difference in volume values when compared to the control group, as the addition of dietary fiber to samples containing 20% or less of gluten had no effect on the gluten structure.

### 3.5. Sensory Evaluation

The sensory properties of food have an important effect on the commercialization of that food. The scores of hedonic sensorial characteristics of bread in terms of appearance, softness, taste, color, adhesiveness, and acceptability were presented in Table 6. It was observed that bread enriched with CCF was not found statistically different than the control sample and CCF substitution had no significant effect on sensory properties ( $p>0.05$ ), except for 5 % CCF added.

(Sangnark and Noonhorm, 2004) extracted dietary fiber from sugarcane and substituted it for 0-15% of wheat flour. They identified that as the amount of sugar cane used increased, the bread's sensory appeal diminished. (Gomez et al., 2003) added 2% fiber derived from coffee and cocoa to wheat flour and compared it to control bread. Researchers have found that consumers have a low preference for breads containing fiber.

## 4. CONCLUSION

In this study, a functional product was produced by using crude fiber (CCF) purified from the waste from the production of carob molasses for the formulation of bread that is consumed frequently on a daily basis. Thus, a type of food waste was evaluated industrially, contributing both environmentally and economically. Thanks

to the fact that CCF is rich in dietary fibers, which are high in its structure, the composition of the bread is enriched and a product with higher nutritional value is obtained. Adding up to 4 % CCF to the bread formulation does not affect the sensory properties of bread such as appearance, softness, taste, color and adhesiveness. The results of this study indicated that carob crude fiber may be used in bread manufacture by bakery products to improve health parameters.

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Research Article

# Sensory profile and physicochemical composition of premixed and post mixed fruit wine from blends of pineapple and watermelon juice

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## ABSTRACT

Wines are mostly produced from grapes that are not grown in Nigeria, hence the need for alternative use of fruits for wine production. Tropical fruits have high perishability, thus, the production of wine from common fruits could help reduce the level of post-harvest losses and increase the variety of wine. In the research. The wine samples were produced from juices that were blended before and after fermentation known as premixed; coded as PWp and post mixed; coded as PWs at the ratio of pineapple to watermelon as follows 90:10, 80:20, 70:30, 60:40, and 50:50, and fermented for 7 days at 28±2°C and aged for two weeks, bottled and corked. Physicochemical analysis and sensory evaluation were carried. Experimental design used was a split plot in Completely Randomized Design and data obtained were statistically analyzed. pH of both premixed and post mixed wine decreased as total acidity increased which may be due to yeast metabolism. Total soluble solid decreased in premixed wine due to the level of water melon added to pineapple wine increased; while increase in post mixed wine increased as level of watermelon added to the pineapple wine increased. Moisture content of the premixed and post mixed wines decreased. Post mixed wines were preferred and had higher scores in terms of taste, mouth feel and overall acceptability. Blending of pineapple and watermelon after fermentation in the ratio of 80:20 was the best in physicochemical examination.



## INTRODUCTION

Fruits have living biological systems, begin to deteriorate immediately after harvest and have a short shelf life due to their high moisture and nutrient contents. Improper post-harvest handling and inadequate processing facilities have resulted in 20% to 50% loss of horticultural product (Kasso and Bekele, 2018). The shelf-life of these highly perishable fruits can be increased by fermentation of the fruit juices from fruits to make fruit wines which are high sources of energy, vitamins, minerals, etc.

Although, grape wine is the most widely consumed fruit wine in the world, due to the increasing diversification of consumers' needs, the variety of fruit wines in the market is becoming more and more abundant (Yang et al., 2020). Wine is a mild natural tranquilizer that serves to reduce anxiety and tension. As part of a normal diet, wine provides the body with energy, substances that aid digestion and small amounts of minerals and vitamins. Pineapple (*Ananas comosus*) is a tropical plant with edible multiple fruit consisting of coalesced berries. Pineapples are a good source of sugar as they have high sugar proportion which is suitable for making wine (Adaikan and Ganesan, 2004). Watermelon is a vine-like (scrambler and trailer) flowering plant which is thought to have originated in southern Africa, because it is found growing wild throughout the area. It produces a fruit that is about 93% water, hence the name "water" melon. Watermelon is a rich natural source of lycopene, a carotenoid of great interest because of its antioxidant capacity and potential health benefits (Rhodes and Zhang, 1999). The objective of this work was to evaluate the effects of blending on the sensory profile and physicochemical compositions of table wine from premixed and post-mixed pineapple and watermelon blends.

## MATERIAL AND METHOD

### Material

Mature and ripe pineapple and watermelon were procured from Ogige market in Nsukka, Enugu State, Nigeria. Other materials were

granulated sugar, instant dry bakers' yeast, sodium metabisulphite, distilled water, funnel, aluminum foil, stainless knives, electric juice extractor (Model MJ-SJ01WTZ Sheldon Manufacturing Incorporation, Oregon, USA), muslin cloth.

## METHOD

### Preparation of juice from water melon and pineapple

The procedures were carried out according to Ibegbulemet al. (2014) with modifications. All equipments were washed and sterilized with hot water. The pineapples and watermelon were washed, peeled, juice extracted and pressed.

### Inoculation of must

In a conical flask, 700 mL of the must (extracted juice) (500 mL pineapple and 200 mL watermelon) was added to 10 g of sugar and 10 g of brewers' yeast. The starter culture was inactivated for 2 days at room temperature before inoculation into the must. The must was inoculated with the wine yeast (brewer's yeast).

### Fermentation of the musts

After inoculation, the must was allowed to ferment in gallon at a temperature of  $28 \pm 2^\circ\text{C}$  for 7 days. The post mixed wine which was blended into various ratios (90:10, 80:20, 70:30, 60:40 and 50:50), was filled into sterilized bottle and sealed/corked and then allowed to age for 2 weeks to allow the development of characteristic flavor of the wines. This process was termed post fermentation and it involves the blending of the juices into various proportions (90:10, 80:20, 70:30, 60:40 and 50:50), fermentation, filtration, bottling and aging was also done.

### Physical and chemical analyses

The following analyses were carried out on the fresh juices and wines produced from them and their blends.

### Determination of percentage yield

The yield of the juice (%) was calculated using the method of Tressler and Joslyn (1961).

### Determination of moisture content

Moisture content of any wine influences the other components of the wine and also the storage stability of the final product. Moisture content was determined by the hot air oven method described by the AOAC. Stainless steel oven dishes were cleaned and dried in the oven (Fulton, Model NYC -101 Sheldon Manufacturing Incorporation, Oregon, USA) at 100°C for one hour. The oven dishes were cooled in a desiccator and then weighed. 10 mL of each of the sample was placed in the oven dish and dried at 100°C. The sample was removed from the oven and placed in a desiccator to cool to room temperature ( $27 \pm 2^\circ\text{C}$ ) before weighing.

The oven dishes were put back into the oven and weighed intermittently until a constant weight was recorded. The loss in weight from the original sample weight was calculated as the moisture content.

$$\text{Moisture Content (\%)} = \frac{W_2 - W_3}{W_2 - W_1} \times 100$$

Where;

$W_1$  = weight of empty oven dish,

$W_2$  = weight of oven dish + sample before drying,

$W_3$  = weight of oven dish + sample after drying.

### Determination of total soluble solids

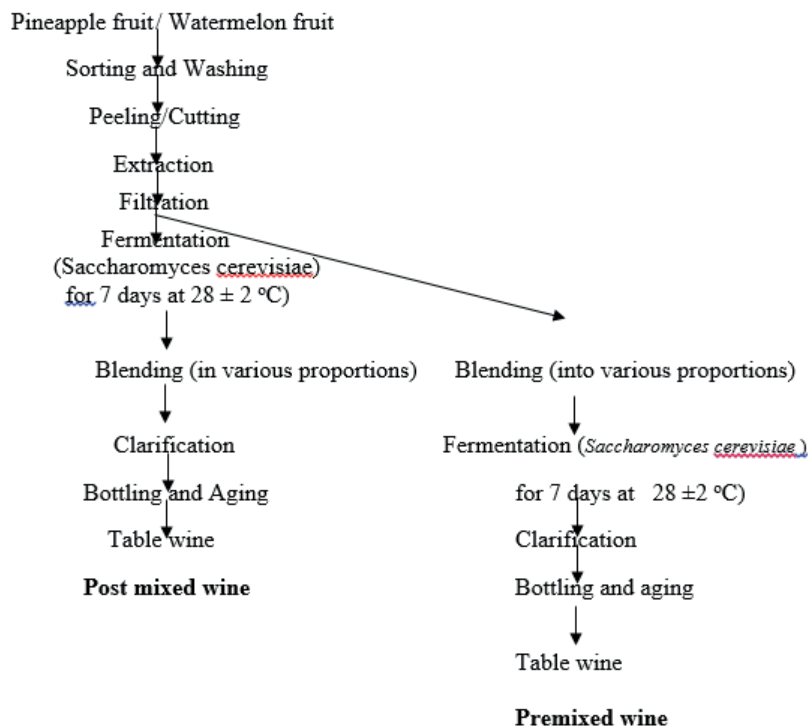
This was carried out with the method described by Pearson (1976). 10 mL of the sample was pipetted into a washed, dried and weighed crucible. The dish and the contents (crucible containing 10 mL of the sample) were put into an oven and dried at 70°C for 3 hours at pressure not exceeding 100 mm Hg. It was cooled in a desiccator and the weight of the solid determined.

Percentage (%) total solid was calculated as:

$$\begin{aligned} &\text{by mass of total solid (\%)} \\ &= \frac{\text{weight of dried solid}}{\text{Volume of sample}} \times 100 \end{aligned}$$

### Determination of pH

The pH was determined using a pH meter as described by the AOAC. 5 mL of sample was measured into the beaker and glass electrode was inserted inside the beaker and the reading was taken.



**Figure 1.** Flow chart for the production of table wine from postmixed and premixed blends of pineapple and watermelon



### Determination of titratable acidity

Determination of titratable acidity of the wine was carried out in accordance with the method described by AOAC (2010). 10 mL of the wine was diluted to 250 mL using distilled water and titrated with standardized 0.1N NaOH (Sodium Hydroxide) solution using 0.3 mL phenolphthalein for each 100 mL solution as an indicator to get a pink end point, which persisted for 30 seconds. This was expressed in terms of NaOH/100 mL of the sample.

### Determination of alcohol content

The alcohol content was determined using the method of difference in potential alcohol method (Jacobson, 2006). In this method, the alcohol contents were calculated based on the sugar contents of the must before fermentation and the final sugar level of the fermented must.

### Sensory evaluation

Sensory evaluation was carried out on the samples using a 9-point Hedonic scale (where '9' was graded extremely liked, while 1 was assigned extremely disliked). 20 members semi-trained panel of judges evaluated and scored the products based on flavor, taste, aftertaste, mouthfeel, color and overall acceptability. The sample were filled in disposable cups which were labeled as PWp1 to PWp5 and PWs1 to PWs5 for premixed and post mixed and the control was labeled as G and water was provided for rinsing of their mouth after each testing.

### Data analysis and experimental design

The experimental design that was used for this analysis was Split plot in Completely Randomized Design. The wine produced from two fruits (pineapple and watermelon) was the main plot while premixed and post mixed into various proportions were the sub plots. The data generated from all analyses and sensory evaluation were subjected to Statistical Analysis of variance (ANOVA) using the Statistical Package for Social Science (SPSS) version 16. Means were separated using the Duncan's Multiple Range Test and the significance was accepted at  $p < 0.05$  (Steel and Torrie, 1980).

## RESULTS AND DISCUSSION

Physicochemical composition of fresh pineapple (*Ananas comosus*) and watermelon (*Citrullus lunatus*) juice is given in Table 1

**Table 1.** Physicochemical composition of fresh pineapple (*A.comosus*) and (*C.lunatus*) juices

Parameters	PJ	WJ
Juice Yield (%)	65.0	74.0
M.C (%)	87.70 <sup>b</sup> ± 0.34	91.87 <sup>a</sup> ± 0.71
TSS (°Brix)	13.12 <sup>a</sup> ± 0.02	4.11 <sup>b</sup> ± 0.03
pH	4.1	5.2
T.A (%)	0.57 <sup>a</sup> ± 0.03	0.45 <sup>b</sup> ± 0.02
Alcohol (%)	Not detected	Not detected

Values are means ± standard deviation of triplicate determinations.

WJ = Watermelon Juice; PJ = Pineapple Juice; TSS = Total Soluble Solids, M.C= Moisture content, T.A= Titratable Acidity

The percentage yield of pineapple juice and watermelon juice were 65.0 and 74.0%, respectively and this could be attributed to the fact that watermelon had higher moisture content (91.87%) when compared to pineapple

juice (87.70%). The high moisture content of watermelon is in agreement with the work by Oyeleke et al. (2012) who stated that watermelon juice had 94.63% moisture content. High moisture content makes the juice suitable as a refreshing and thirst-quenching product which is characteristic of good juice. The pineapple juice had lower moisture content (87.70%) but higher total soluble solids (13.12°Brix). This was because the total soluble solids added body and improved the taste of the juice.

The pH of pineapple juice was 4.1 and watermelon was 5.2. The results on pH showed that standard pineapple juice is acidic (pH < 7.0) and that of watermelon is slightly acidic, (between 5.0 and 5.8) according to Akinosun (2010).

The titratable acidity of pineapple juice was 0.57% and that of watermelon was 0.45%. There exists a correlation between pH and acidity of the juice, the higher the acidity, the lower the pH of the juice. Both juices had no alcohol content.

There was a decrease in the pH of both the premixed and post mixed pineapple-watermelon



wine (4.0 to 3.8). This decrease is as a result of the increase in the quantity of watermelon juice which caused, the pH to reduce. This was as a result of the fact that watermelon fruit generally is of medium acidity. Thus, the pH gradually dropped from 5.2 before the fermentation to 3.4 after the fermentation. There was a significant decrease ( $p < 0.05$ ) in pH of the fresh juice after fermentation that is from 4.1 to 3.5 for pineapple wine and from 5.2 to 3.4 for watermelon wine. This might be due to increase in titratable acidity and pH are sometimes inversely proportional to each other though not in all cases. The drop in pH and corresponding increase in titratable acidity of must during the fermentation is attributed to yeast metabolism.

There was a decrease in the titratable acidity of PWP (0.54 to 0.48%) as the level of watermelon added to the pineapple juice increased while there was an increase in the PWS (0.48 to 0.55%) as the level of watermelon added to the pineapple wine increased. This could be a result of the fact that pineapple is more acidic than watermelon. There was no significant difference ( $p > 0.05$ ) among samples PWP4, PWP5 and PWS5 and among samples PWP2, PWP3, and PWS4 in their titratable acidity. The control sample had

the highest titratable acidity followed by wine produced from PJ and WJ and then post mixed pineapple and watermelon blends.

There was a decrease in moisture content of the sample PWP (93.10 to 88.65%) and PWS (91.02 to 88.08 %). Sample PWP5 had the highest moisture content (93.10%) followed by sample PWP2 (92.03%), that is, to say that moisture content of the mixed fruit wine was higher than the fruit wine and these values were higher than the control sample (87.10%).

The total soluble solids (TSS) increased with increase in the volume of watermelon added to the pineapple wine in the PWP (9.57 to 12.54 oBrix) as well as the PWS (8.93 to 10.90 oBrix) when compared to the fruit wine. Increase in total soluble solid is an indication that addition of pineapple juice had resulted in high sugar-to-acid ratio in the blends and this confirmed a high correlation between total soluble solids and sugar content as observed by Ravi et al. (2010). The increase in total soluble solids on addition of pineapple to watermelon could also be attributed to the conversion of polysaccharides and other constituents of the juice to sugar. These results were in agreement with that of

**Table 2.** Physicochemical composition of wine from pineapple and watermelon

Samples	pH	MC(%)	TSS (Brix)	T.A (%)	Alcohol(%)
P	3.5	87.25 <sup>a</sup> ±1.11	8.50 <sup>a</sup> ± 0.15	0.67 <sup>ab</sup> ±0.01	13.03 <sup>dc</sup> ±0.32
W	3.4	88.25 <sup>b</sup> ±0.87	9.60 <sup>b</sup> ± 0.15	0.51 <sup>a</sup> ±0.03	12.90 <sup>dc</sup> ±0.20
G	3.8	87.10 <sup>a</sup> ±1.08	12.20 <sup>d</sup> ±0.10	0.71 <sup>c</sup> ±0.06	13.93 <sup>c</sup> ±0.31
PWP <sub>P1</sub>	3.9	88.65 <sup>b</sup> ±0.10	10.82 <sup>c</sup> ±0.35	0.54 <sup>ab</sup> ±0.04	12.27 <sup>cd</sup> ±0.13
PWP <sub>P2</sub>	3.9	92.03 <sup>c</sup> ±0.21	12.54 <sup>d</sup> ±0.19	0.53 <sup>a</sup> ±0.04	8.50 <sup>a</sup> ±2.33
PWP <sub>P3</sub>	3.9	91.66 <sup>c</sup> ±0.51	9.57 <sup>b</sup> ±0.36	0.50 <sup>a</sup> ±0.17	11.04 <sup>ab</sup> ±0.14
PWP <sub>P4</sub>	3.8	89.24 <sup>b</sup> ±0.21	10.70 <sup>c</sup> ±0.41	0.51 <sup>a</sup> ±0.05	12.73 <sup>dc</sup> ±0.02
PWP <sub>P5</sub>	3.8	93.10 <sup>c</sup> ±0.52	10.90 <sup>c</sup> ±0.18	0.48 <sup>a</sup> ±0.06	9.77 <sup>b</sup> ±0.45
PWS <sub>S1</sub>	3.8	89.70 <sup>b</sup> ±0.28	10.56 <sup>bc</sup> ±0.29	0.48 <sup>a</sup> ±0.04	12.44 <sup>c</sup> ±0.21
PWS <sub>S2</sub>	3.8	89.03 <sup>b</sup> ±0.52	10.90 <sup>bc</sup> ±0.39	0.50 <sup>a</sup> ±0.02	13.98 <sup>cd</sup> ±0.08
PWS <sub>S3</sub>	4.0	89.01 <sup>b</sup> ±0.44	8.93 <sup>a</sup> ±0.35	0.53 <sup>a</sup> ±0.06	12.73 <sup>cd</sup> ±0.46
PWS <sub>S4</sub>	3.8	91.02 <sup>c</sup> ±0.68	10.63 <sup>bc</sup> ±0.41	0.51 <sup>a</sup> ±0.02	9.16 <sup>a</sup> ±0.91
PWS <sub>S5</sub>	3.9	88.08 <sup>b</sup> ±0.64	10.64 <sup>c</sup> ±0.24	0.55 <sup>a</sup> ±0.08	11.82 <sup>b</sup> ±0.04

Values are means ± standard deviation of triplicate determinations. Means with different superscripts in the same column are significantly different at  $p < 0.05$

P = Pineapple wine; W = Watermelon wine; G = Commercial Grape wine; PWP<sub>P1</sub> (90:10), PWP<sub>P2</sub> (80:20), PWP<sub>P3</sub> (70:30), PWP<sub>P4</sub> (60:40), PWP<sub>P5</sub> (50:50) = Pre-mixed Pineapple-Watermelon wine; PWS<sub>S1</sub> (90:10) = Post mixed Pineapple-Watermelon wine, PWS<sub>S2</sub> (80:20), ; PWS<sub>S3</sub> (70:30), PWS<sub>S4</sub> (60:40), ; PWS<sub>S5</sub> (50:50) = Post mixed Pineapple-Watermelon wine; TSS = Total Soluble Solids. PJ = Pineapple juice; WJ = Watermelon

Ifie et al. (2012) who reported the increase in total soluble solids and pH, and increase in the yield of alcohol during the fermentation of roselle wine. However, roselle wine had lower ethanol content (9.6%), final TSS (4.8 °Brix) and pH-value (3.09). The PWS3 had lower total soluble solid (8.93 °Brix). The decrease in the total soluble solids of the different proportions after fermentation could also be attributed to the enzymatic, chemical, biological and physical alterations of the must after fermentation. There was no significant difference ( $p < 0.05$ ) between the TSS of the control sample (G) and PWP2.

There was an increase in alcohol content of the premixed wine (PWP) and post mixed wine (PWS) (8.50 to 12.75% and 9.16% to 13.98%) respectively. The alcohol content (8.50% to 13.98%) of the wine was at the acceptable range for table wine. The final alcohol content of the wine (13.98%) ranks it among good table wines. Based on Bisson and Butzke (2009), a good table wine must have alcohol content between 8 and 14%. Grape wine prepared in the study of Bindon et al. (2013) had alcohol content of 11.77 - 15.5% and pH of 3.46 - 3.62.

The sensory scores for the preximed and post mixed pineapple and watermelon wines are shown in Table 3.

Table 3 showed the mean sensory scores of the

blended wine for color, flavor, taste, aftertaste, mouthfeel and overall acceptability. There was a reduction in the level of acceptance of color in the premixed wine (6.75 to 5.35) and post mixed wine (6.75 to 5.60) on the addition of watermelon to pineapple wine.

There was a reduction in flavor, taste, aftertaste of the premixed wine and post mixed wine. Based on the mouthfeel, there was a decrease in the premixed wine (6.05 to 4.75) and (6.50 to 5.50) for post mixed wine. There was also a decrease in the overall acceptability of the premixed wine (6.50 to 5.15) on the addition of watermelon to pineapple wine and these was also seen on the post mixed wine (6.50 to 5.20).

The control sample (G) was most preferred with higher scores in color (7.55) and flavor (6.75). This might be as a result of familiarity and conversance of the panelist with grape wine. Sample PWS2 were more preferred and had higher score in terms of taste (6.50), mouthfeel (6.60) and overall acceptability (6.85). Generally, the mean sensory scores for the whole samples compared favorably well with the control (G) in taste, aftertaste, mouthfeel and overall acceptability as compared with Ningli et al. (2017) who reported that the pineapple wine has higher acceptance and there were significant ( $p < 0.05$ ) differences in the evaluated attributes.

**Table 3.** Sensory scores of the premixed and post mixed pineapple and watermelon wine

Sample	Color	Flavor	Taste	Aftertaste	Mouthfeel	O.A
G	7.55 <sup>c</sup> ±0.88	6.75 <sup>c</sup> ±1.57	6.05 <sup>bc</sup> ±1.76	5.90 <sup>ab</sup> ±1.68	5.75 <sup>abc</sup> ±1.80	6.45 <sup>b</sup> ±1.23
PWP <sub>P1</sub>	6.75 <sup>b</sup> ±1.25	6.15 <sup>bc</sup> ±1.38	5.85 <sup>a</sup> ±1.31	6.15 <sup>b</sup> ±0.14	6.05 <sup>b</sup> ±1.15	6.50 <sup>b</sup> ±1.32
PWP <sub>P2</sub>	6.50 <sup>b</sup> ±0.88	6.45 <sup>bc</sup> ±0.99	6.00 <sup>a</sup> ±1.03	6.20 <sup>b</sup> ±1.05	6.40 <sup>c</sup> ±0.94	6.50 <sup>b</sup> ±0.76
PWP <sub>P3</sub>	6.35 <sup>b</sup> ±1.03	5.90 <sup>ab</sup> ±1.25	5.40 <sup>a</sup> ±1.19	5.55 <sup>ab</sup> ±0.94	5.55 <sup>ab</sup> ±1.09	5.90 <sup>ab</sup> ±0.97
PWP <sub>P4</sub>	6.50 <sup>b</sup> ±1.10	5.75 <sup>ab</sup> ±1.41	5.60 <sup>a</sup> ±0.31	5.35 <sup>ab</sup> ±1.18	6.00 <sup>b</sup> ±0.79	5.95 <sup>b</sup> ±1.31
PWP <sub>P5</sub>	5.35 <sup>a</sup> ±1.32	5.04 <sup>a</sup> ±1.46	5.10 <sup>a</sup> ±1.51	5.05 <sup>a</sup> ±1.57	4.75 <sup>a</sup> ±1.80	5.15 <sup>a</sup> ±2.03
PWS <sub>S1</sub>	6.75 <sup>b</sup> ±1.29	5.70 <sup>ab</sup> ±1.59	6.20 <sup>c</sup> ±1.15	6.10 <sup>ab</sup> ±0.78	6.50 <sup>bc</sup> ±0.94	6.50 <sup>b</sup> ±0.94
PWS <sub>S2</sub>	6.55 <sup>b</sup> ±1.23	6.30 <sup>bc</sup> ±1.26	6.50 <sup>c</sup> ±1.00	6.35 <sup>b</sup> ±1.09	6.60 <sup>c</sup> ±0.99	6.85 <sup>b</sup> ±0.86
PWS <sub>S3</sub>	6.15 <sup>ab</sup> ±1.18	5.30 <sup>a</sup> ±1.30	5.25 <sup>ab</sup> ±1.37	5.75 <sup>ab</sup> ±1.52	5.70 <sup>abc</sup> ±1.30	6.40 <sup>b</sup> ±1.39
PWS <sub>S4</sub>	6.25 <sup>ab</sup> ±1.12	5.60 <sup>ab</sup> ±1.39	5.95 <sup>bc</sup> ±0.76	5.55 <sup>ab</sup> ±1.39	5.55 <sup>ab</sup> ±1.39	5.95 <sup>ab</sup> ±1.36
PWS <sub>S5</sub>	5.60 <sup>a</sup> ±1.23	5.45 <sup>ab</sup> ±1.23	5.05 <sup>a</sup> ±1.36	5.30 <sup>a</sup> ±1.69	5.00 <sup>a</sup> ±2.08	5.20 <sup>a</sup> ±2.04

Values are means ± standard deviation of triplicate determinations. Means with different superscripts in the same column are significantly different at ( $p < 0.05$ )

G =Grape wine, PWP = Post-mixed Pineapple-Watermelon wine, PWS = Post-mixed Pineapple-Watermelon wine, O. A= Overall acceptability

## CONCLUSION

Wine was produced from the 'must' prepared from blends of pineapple and watermelon fruit and was found to compare favorably with the wine produced from grape (control) in most of the physicochemical parameters (titratable acidity, total soluble solid among others) evaluated.

In terms of the time for fermentation, post-mixed wine was the best in the physicochemical properties examined. From the data obtained, the post-mixed wine sample in the ratio of 80:20 was the best physicochemically. With respect to the sensory attributes examined, there was slight significant difference ( $p < 0.05$ ) in the taste, flavor, appearance and overall acceptability of the different blends which made some samples more acceptable than the other. The formulated wine compared favorably with grape wine (control) since it had similar properties with it and it was organoleptically acceptable to the potential consumers and these were observed in the result of the sensory evaluation.

### Conflict of interest

The authors declare no conflict of interest

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*Bu sayfa dizgiden dolayı boş bırakılmıştır*

*Research Article*

# Assessment of aroma profiles and mineral content of buffalo yogurt marketed in Cukurova region of Turkey

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## ABSTRACT

The purpose of this study was identification of some chemical compositions, mineral content and aroma profile of buffalo yogurt samples which were collected from Çukurova markets. For this purpose, some chemical analyses of 20 buffalo yogurt samples obtained from Mersin and Adana provinces were performed. As a result of the chemical analysis, the average dry matter content of buffalo yogurts was 19.37%, fat content was 6.54%, protein content was 4.10% and ash content was 1.37%. Aroma profile was evaluated by GC/MS (Gas Chromatography/Mass Spectrometry) using SPME (Solid-Phase Microextraction) technique. A total of 35 volatile compounds were detected including four aldehydes, four ketones, six acids, seven alcohols, four esters and ten miscellaneous compounds. Alcohols were the largest class of volatile compounds while acid and ketone groups were the major compounds in buffalo yogurt. The analysis of minor mineral content was performed by LA-ICP/MS (Inductively Coupled Plasma/Mass Spectrometer) and major minerals were detected by AAS (Atomic Absorption Spectrometer). According to the results of mineral analysis, the average Ca, K, Mg, Na, Cu, Fe, Mn and Zn levels were found to be 900.52 mg/L, 1678.18 mg/L, 729.80 mg/L, 325.78 mg/L, 0.28 mg/L, 7.60 mg/L, 0.39 mg/L, and 24.64 mg/L, respectively.





## INTRODUCTION

Buffalo yogurt is an important product which reflects the main characteristics of buffalo milk ideally (Güzeler et al., 2019). It has a creamy structure and higher amount of dry matter, fat and protein contents than other yogurt types which are obtained from other animal milks. Most of the buffalo milk in Turkey is processed into yogurt (Bilgin and Kaptan, 2016). Buffalo yogurt is preferred by many consumers because of its aroma and high fat content (Ghoneem et al., 2018). The network of buffalo yogurt is more porous and it contains more and larger fat globules than bovine yogurt (Nguyen et al., 2015). Buffalo yogurt also has higher acid production rate, extent of proteolysis and flavor development by lactic cultures (Yadav et al., 2018). It is possible to obtain buffalo yogurt with high yield and low syneresis and there is no need to add milk powder or hydrocolloid to buffalo milk for yogurt production because of its high dry matter content (Khalifa and Zakaria, 2019).

In 2019, the total number of buffaloes in Turkey was 184.192 (head). 2.934 of them were in the Mediterranean region and this region had the minimum number of buffaloes in Turkey. The total numbers of buffaloes (head) in Adana and Mersin provinces in Çukurova region which is located in the Eastern Mediterranean were 425 and 56, respectively. The total amount of buffalo milk produced in Turkey in 2019 was 79341 tons, 1181 tons were produced in the Mediterranean region, 155 tons in Adana and 24 tons in Mersin (TÜİK, 2020).

Because of limited production of buffalo milk and dairy products in Çukurova region, it is not possible to reach these products easily. Therefore, buffalo milk products are very valuable in Çukurova region. It is well known that geographical location affects the compositions of buffalo dairy products, processing steps and overall quality (Akgün et al., 2016). There is no information about characteristics of buffalo dairy products in Çukurova region and limited published data about mineral composition and aroma profile of buffalo yogurt. Research on buffalo yogurt, especially on their mineral contents and aroma profile are still limited.

Only a few studies have been performed on this subject. Erkaya and Şengül (2012) investigated mineral contents of buffalo yogurt. Erkaya and Şengül (2011) and Emirmustafaoğlu et al. (2020) studied volatile compounds of buffalo yogurt. To fill this gap, the present investigation was performed to determine mineral compositions and aroma profiles of buffalo yogurt samples, which were collected from Çukurova markets.

## MATERIAL AND METHOD

In this research, 20 samples of buffalo yogurt were collected from local producers in Mersin and Adana provinces in Çukurova region. Some compositional properties, mineral contents and aroma profiles of yogurt samples were examined at Çukurova University, Faculty of Agriculture, Department of Food Engineering, Milk Technology Laboratory.

### Chemical composition

Dry matter content was determined gravimetrically by drying the samples at 100°C until constant weighing (IDF, 2005). Fat content was determined according to Gerber method (TSE, 2002). Micro Kjeldahl method was used for determining total nitrogen content and total nitrogen content was multiplied by factor 6.38 for determining protein content (IDF, 2014). Ash content was calculated by burning the samples at 550°C, cooling in the desiccator and weighing (Kurt et al., 2007).

### Aroma profile analysis

SPME fibers were placed on the injection port of Agilent 7890B gas chromatograph at 250°C for 5 minutes for preconditioning and placed on the gas phase flask for extraction for 60 minutes. Desorption was performed at 250°C for 3 minutes and a temperature programmed route was used for chromatography. The temperature was kept at 35°C for 3 minutes and then raised to 140°C at the rate of 4°C /min. The temperature was kept at 140°C for 1 minute and increased to 250°C in 3 minutes. The transfer line temperature was set at 250°C. Helium was used as carrier gas at a flow rate of 1.0 mL / min. For mass spectroscopy, electron ionization was performed at 70 eV and the ion source temperature was at 230°C,



mass scanning range was  $m/z$  33-450 AMU and emission current was 100  $\mu\text{A}$  (Dan et al., 2019).

### Mineral composition

The concentrations of Cu, Fe, Mn and Zn of the samples were determined by LA-ICP/MS (Perkin Elmer Nexion 2000 P) and as applied by Khan et al. (2014). For this purpose, 2 mg of sample was taken into Teflon tubes and 5 mL of 65%  $\text{HNO}_3$  and 1 mL of 30%  $\text{H}_2\text{O}_2$  were added to it. The samples were heated in microwave oven (Berghof speedwave MWS-2 Germany) at 170°C for 10 minutes, at 200 °C for 15 minutes and at 100 °C for 10 minutes. 1 mL of the distillate obtained by heating in the microwave oven was taken and completed to 10 mL with distilled water and mineral content was determined with LA-ICP/MS.

AAS (Perkin Elmer PinAAcle 900T) device was used to determine Ca, Mg, K and Na concentrations. 3 grams of sample was placed in Teflon cups and 5 mL of deionized water and 5 mL of concentrated  $\text{HNO}_3$  were added to it. Teflon cups were taken to the microwave mineralization device after shaking to mix the solution. After the microwave heating process, the samples were filtered through filter paper and completed to 50  $\text{cm}^3$  with deionized water. Measurements of the samples were carried out on AAS device at certain wavelengths (Capcarova et al., 2017).

## RESULTS AND DISCUSSION

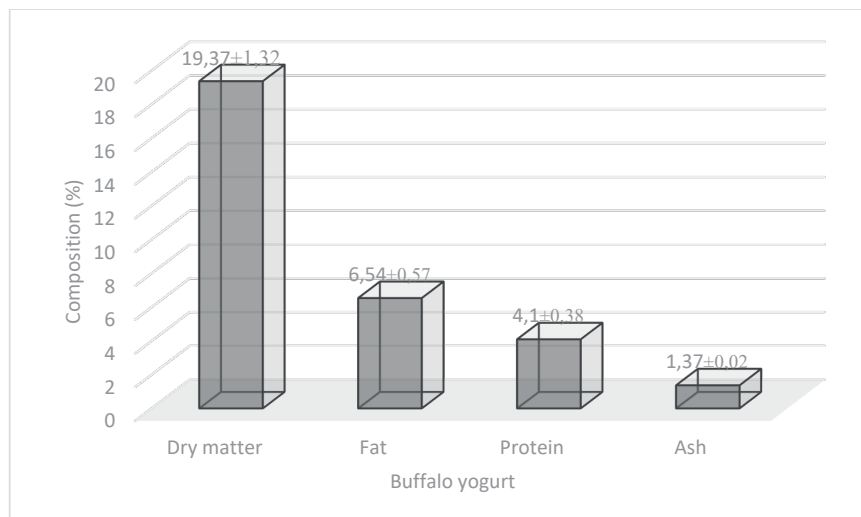
### Chemical Composition

The mean values for the chemical compositions of buffalo yogurt samples collected from Çukurova markets are given in Figure 1.

The average dry matter content of buffalo yogurt samples was 19.37%. It was determined that buffalo yogurt samples collected from Çukurova markets had higher dry matter content than other buffalo yogurt samples which were collected from other places such as Erzurum (17.87%) in Turkey, Cairo (14.91-16.61%) and Damietta Governorate (17.70-18.01%) in Egypt and Karnal (13.11-14.11%) in India (Erkaya and Şengül, 2011; El-Shibiny et al., 2018; Ghoneem et al., 2018; Yadav et al., 2018).

The average fat content of buffalo yogurt samples from Çukurova markets was found close to the results (6.10%) specified by Bezerra et al. (2012) from north-eastern Brazil, lower than the results (8.40% and 7.10-7.20%, respectively) specified by Erkaya and Şengül (2011) and Ghoneem et al. (2018) and higher than the results (3.25% and 3.50-5.90%, respectively) specified by Yadav et al. (2018) and El-Shibiny et al. (2018). These differences might be due to the different production methods of yogurt samples.

**Figure 1.** Chemical compositions of buffalo yogurt samples collected from Çukurova markets



The average nitrogen content of yogurt samples was found to be 0.64% and the average protein content was calculated as 4.10%. El-Shibiny et al. (2018) stated that the nitrogen contents of buffalo yogurts from Cairo in Egypt were between 0.68-0.73%. Some researchers stated the protein content of buffalo yogurt samples as 4.67% (Erkaya and Şengül, 2011), 3.10% (Bezerra et al., 2012) and 3.98-4.30% (Yadav et al., 2018). These findings were generally found nitrogen and protein content.

The average ash content of the yogurt samples was found as 1.37% and it was concluded that the ash content of buffalo yogurt samples from Çukurova region was higher than other researchers' findings (0.87%, 0.70%, 0.74-1.17%,

1.14-1.17% and 0.81-0.92%, respectively) from other places (Erkaya and Şengül, 2011; Bezerra et al., 2012; El-Shibiny et al., 2018; Ghoneem et al., 2018; Yadav et al., 2018).

### Volatile aroma profile

A total of 35 volatiles including four aldehydes, four ketones, six acids, seven alcohols, four esters and ten miscellaneous compounds were identified in buffalo yogurts from Çukurova region, as shown in Table 1.

Aldehydes were not major compounds in buffalo yogurt samples because of transforming to alcohols or corresponding acids. It is known that acetaldehyde, diacetyl and acetoin are main flavor compounds in yogurts (Tian et al.,

**Table 1.** Volatile compounds of buffalo yogurt samples collected from Çukurova markets

	Compounds	Relative peak area (%)	RT (min)	Molecular formula
Aldehydes	Octanal	0.244±0.023	3.283	C <sub>8</sub> H <sub>16</sub> O
	Acetaldehyde	0.401±0.066	3.489	C <sub>2</sub> H <sub>4</sub> O
	Benzaldehyde	1.104±0.360	30.092	C <sub>7</sub> H <sub>6</sub> O
	3-Hydroxybutanal	1.020±0.204	32.000	C <sub>4</sub> H <sub>8</sub> O <sub>2</sub>
Ketones	2,3-Pentanedione	1.579±0.293	10.938	C <sub>5</sub> H <sub>8</sub> O <sub>2</sub>
	2-Heptanone	3.093±0.479	16.330	C <sub>7</sub> H <sub>14</sub> O
	Acetoin	20.731±0.889	20.838	C <sub>4</sub> H <sub>8</sub> O <sub>2</sub>
	2-Nonanone	0.626±0.219	25.466	C <sub>9</sub> H <sub>18</sub> O
Acids	2-(Methylsulfonyl)acetic acid	0.376±0.067	11.411	C <sub>3</sub> H <sub>6</sub> O <sub>4</sub> S
	Acetic acid	35.249±2.995	27.518	C <sub>2</sub> H <sub>4</sub> O <sub>2</sub>
	Butanoic acid	4.742±0.561	34.239	C <sub>4</sub> H <sub>8</sub> O <sub>2</sub>
	Isovaleric acid	1.755±0.152	36.155	C <sub>5</sub> H <sub>10</sub> O <sub>2</sub>
	Hexanoic acid	3.047±0.443	42.043	C <sub>6</sub> H <sub>12</sub> O <sub>2</sub>
	Caprylic acid	0.648±0.160	47.463	C <sub>8</sub> H <sub>16</sub> O <sub>2</sub>
Alcohols	Cyclobutanol	0.211±0.061	2.994	C <sub>4</sub> H <sub>8</sub> O
	trans-1,2-Cyclopentanediol	0.270±0.022	16.918	C <sub>5</sub> H <sub>10</sub> O <sub>2</sub>
	Isoamyl alcohol	5.349±0.758	17.596	C <sub>5</sub> H <sub>12</sub> O
	3-Methyl-2-pentanol	2.629±0.338	24.032	C <sub>6</sub> H <sub>14</sub> O
	3-Methyl-1,2-cyclopentanediol	1.083±0.227	29.185	C <sub>6</sub> H <sub>12</sub> O <sub>2</sub>
	2,3-Butanediol	0.301±0.051	30.735	C <sub>4</sub> H <sub>10</sub> O <sub>2</sub>
	2-Phenylethanol	0.201±0.006	43.627	C <sub>8</sub> H <sub>10</sub> O
Esters	Ethyl Acetate	1.685±0.526	5.654	C <sub>4</sub> H <sub>8</sub> O <sub>2</sub>
	Vinyl formate	0.274±0.062	5.913	C <sub>3</sub> H <sub>4</sub> O <sub>2</sub>
	Vinyl acetate	4.224±0.614	7.982	C <sub>4</sub> H <sub>6</sub> O <sub>2</sub>
	Benzyl carbazate	0.421±0.125	10.156	C <sub>8</sub> H <sub>10</sub> N <sub>2</sub> O <sub>2</sub>
Miscellaneous	Carbon dioxide	2.026±0.410	2.897	CO <sub>2</sub>
	Dimethylphosphine	0.388±0.167	3.817	C <sub>2</sub> H <sub>7</sub> P
	Butanimidamide	0.629±0.189	4.465	C <sub>4</sub> H <sub>10</sub> N <sub>2</sub>
	Formamide	1.888±0.163	6.855	CH <sub>3</sub> NO
	D-Limonene	0.258±0.060	17.188	C <sub>10</sub> H <sub>16</sub>
	1-Nonyne	0.445±0.242	18.667	C <sub>9</sub> H <sub>16</sub>
	Tetramethyloxirane	3.062±0.417	23.347	C <sub>6</sub> H <sub>12</sub> O
	Dimethyl trisulfide	0.214±0.018	24.934	C <sub>2</sub> H <sub>6</sub> S <sub>3</sub>
	2-Imino-3-methylthiazolidine	0.283±0.028	30.330	C <sub>4</sub> H <sub>8</sub> N <sub>2</sub> S
	Oxime-, methoxy-phenyl-	0.289±0.007	39.696	C <sub>8</sub> H <sub>9</sub> NO <sub>2</sub>

RT: Retention time

The values were expressed by average ± standard deviation.

2019). However, acetaldehyde level was found to be low and diacetyl wasn't detected among ketone groups in buffalo yogurts. Acetoin which is a reduced form of diacetyl and produces mild creamy flavor in yogurts (Cheng, 2010) had the highest level among ketone groups in buffalo yogurts. *Lactococcus lactis* can generate acetoin during fermentation (Walsh, 2016). Other identified ketones in buffalo yogurts were responsible sweet, fruity, buttery and creamy flavor of samples (Sidira et al., 2017). Erkaya and Şengül (2011) detected acetaldehyde and hexanal as aldehydes, diacetyl, acetoin, 2,3-pentanedione, 2-heptanone, 2-nonanone and 2-undecanone as ketones in buffalo yogurts. Emirmustafaoğlu et al. (2020) also found that the main volatiles of buffalo yogurts were acetaldehyde, diacetyl, acetoin and acetone.

Acetic acid was the compound which had the highest level among all volatile compounds. It is produced by hetero fermentative lactic acid bacteria and produces excess vinegar flavor if its concentration is too high (Tian et al., 2019). Butanoic and hexanoic acids were other important acids in buffalo yogurts. Butanoic acid gives cheesy flavor while hexanoic acid contributes spicy, rancidity and floral flavor (Tian et al., 2019). Erkaya and Şengül (2011) detected acetic acid, butanoic acid, hexanoic acid, caprylic acid, capric acid and benzoic acid in buffalo yogurt samples.

Alcohols were the largest class of volatile compounds in the samples. The most dominant alcohol in buffalo yogurt samples was isoamyl alcohol. It contributes fruity flavor (Costa et al., 2019). Kavaz Yüksel and Bakırcı, (2015) and Costa et al. (2019) also determined isoamyl alcohol in cow milk yogurts. Erkaya and Şengül (2011) and Emirmustafaoğlu et al. (2020) found ethanol as the only alcohol in buffalo yogurts. However, ethanol was not identified in the samples collected from Çukurova region.

Among the esters, vinyl acetate had the highest level in buffalo yogurts. However, no study was found in the literature about presence of vinyl acetate in yogurt samples. Erkaya and Şengül (2011) identified ethyl acetate (pineapple and fruity flavor), ethyl octanoate and diethyl

phthalate in buffalo yogurts. Su et al. (2017), Zhao et al. (2018) and Fang et al. (2020) also found ethyl acetate in yogurt samples obtained from different milk types.

Tetramethyloxirane, carbon dioxide and formamide were the majors among miscellaneous compounds. However, no study was found in the literature about presence of tetramethyloxirane in yogurt samples. Carbon dioxide was observed in buffalo yogurt samples depending on the activities of yogurt starters (Rysstad and Abrahamsen, 1987). Formamide contributes ammonia-like flavor (Hohn, 1999). No information could be found about presence of formamide in yogurt samples in the literature.

### Mineral composition

Mineral concentrations of milk and dairy products vary depending on the animal species, breed, nutrition, lactation stage and health conditions (Paszczyk et al., 2019). The data about mineral compositions in terms of major and minor elements of buffalo yogurt are shown at Table 2.

Erkaya and Şengül (2012) examined mineral contents of yogurts which were made from different milk types collected from Erzurum province in Turkey. They determined the buffalo yogurts' Ca, K, Mg, Na, Fe, Mn and Zn were 1697 mg/kg, 1164 mg/kg, 156 mg/kg, 344 mg/kg, 13.57 mg/kg, 0.15 mg/kg and 63.95 mg/kg, respectively. When the mineral contents were compared, it was seen that Ca content of buffalo yogurt from Çukurova region was lower than Ca content of the samples in Erzurum. On the other hand, Mg levels of Çukurova buffalo yogurts were quite higher than those samples. The levels of Na, Fe and Zn were also found lower while K and Mn levels were higher.

Mineral composition analyses were generally performed in yogurts made by cow, goat or sheep milks by other researchers. According to literature data, mineral compositions vary between 430-1936 mg/kg Ca, 890-3508 mg/kg K, 83-406 mg/kg Mg, 390-2550 mg/kg Na, 0.09-1.68 mg/kg Cu, 0.29-19.00 mg/kg Fe, 0.02-0.16 mg/kg Mn and 2.56-60.68 mg/kg Zn for cow milk yogurt, between 1455-2178 mg/kg Ca, 511-2486

mg/kg K, 150-587 mg/kg Mg, 375-568 mg/kg Na, 0.15 mg/kg Cu, 0.75-16.79 mg/kg Fe, 0.14 mg/kg Mn and 4.46-75.03 mg/kg Zn for goat milk yogurt, between 1879-2352 mg/kg Ca, 1133-1944 mg/kg K, 186-197 mg/kg Mg, 410-567 mg/kg Na, 0.12 mg/kg Cu, 0.26-12.79 mg/kg Fe, 0.19 mg/kg Mn and 6.10-72.99 mg/kg Zn for sheep milk yogurt (Güler, 2007; Isleten and Karagul-Yuceer, 2008; Güler and Şanal, 2009; Abou Jaoude et al., 2010; Navarro-Alarcón et al., 2011; Erkaya and Şengül, 2012; Cano-Sancho *et al.*, 2015; Luis et al., 2015; Curti et al., 2017; Pimentel et al., 2018; Paszczyk et al., 2019; Souza et al., 2019). It was concluded that Mg content of buffalo yogurt was quite higher than cow, goat and sheep milk yogurts, while Na content of buffalo yogurt was quite lower. Buffalo yogurt had higher Cu and Mn contents and lower Ca content than goat and sheep milk yogurts. It was observed that the contents of other mineral compounds of buffalo yogurt were close to other animal milk yogurts.

## CONCLUSION

In this research, some chemical properties, aroma profile and mineral content of buffalo yogurt samples marketed in Çukurova region were determined. As a result of the research, it was concluded that dry matter and ash contents of buffalo yogurt from Çukurova region were higher than other buffalo yogurt samples from different regions. According to mineral composition analysis of buffalo yogurt samples collected from Çukurova region, the levels of Mg, K and Mn were higher and the levels of Ca, Na,

Fe and Zn were lower than buffalo yogurt from Erzurum province. When the mineral content of buffalo yogurt was compared with other animal milk yogurts, it was determined that the level of Mg was quite higher when the level of Na was quite lower than cow, goat and sheep milk yogurts. Additionally, Ca level of buffalo yogurt was lower and Cu and Mn levels were higher than goat and sheep milk yogurts. A total of 35 volatile compounds were detected in buffalo yogurt samples. These were determined as four aldehydes, four ketones, six acids, seven alcohols, four esters and ten miscellaneous compounds. The major volatile compound groups of buffalo yogurt were acid and ketone groups. Alcohols were the largest class of volatile compounds in the samples.

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**Table 2.** Mineral contents of buffalo yogurt samples collected from Çukurova markets

Minerals (mg/L)	Yogurt			
	Mean	SD	Min.	Max.
Ca	900.52	78.89	811.69	962.43
K	1678.18	179.45	1532.22	1878.54
Mg	729.80	45.73	685.17	776.56
Na	325.78	24.33	298.33	344.70
Cu	0.28	0.23	0.08	0.54
Fe	7.60	3.89	3.68	11.47
Mn	0.39	0.32	0.05	0.71
Zn	24.64	3.70	20.75	28.13



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Clinical Research/Klinik Araştırma

## Yoğun bakım ünitesinde yatan hastaların beslenme durumları ve bası yarası oluşma riski arasındaki ilişkinin değerlendirilmesi

*Evaluation of the relationship between the nutritional status and the risk of pressure sore formation of patients hospitalized in the intensive care unit*

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### ÖZET

**Amaç:** Bu çalışmanın amacı yoğun bakım ünitelerinde yatan hastaların bası yarası insidanslarını ve bası yaralarının ilişkili olabileceği risk faktörlerini araştırmaktır.

**Bireyler ve Yöntem:** Mersin Şehir Hastanesi Hastanesi Yoğun bakım ünitelerinde yatan hastalarda yapılan bu çalışmada 18-65 yaş arası, albümin değeri > 2.5 g/dL ve BKİ değeri 18.5-24.9 Kg/m<sup>2</sup> olan 200 hasta değerlendirilmiştir. Çalışmada hastaların tanımlayıcı özellikleri ve antropometrik ölçümleri sorgulanırken, hastaların hastalık ciddiyetlerini değerlendirmek için APACHE II, bası yarası risklerini değerlendirmek için Norton Bası Yarası ölçeği, malnutrisyon durumlarını saptamak için ise NRS2002 tarama testleri hastalara uygulanmıştır. Ayrıca hastaların beslenme durumları ve serum albümin değerleri diyetisyen tarafından günlük vizitelerle 30 gün boyunca izlenmiş ve kayıt altına alınmıştır.

**Bulgular:** Hastaların bası yarası durumlarına göre APACHE II (p<0.001), NRS-2002 skorları (p<0.001), takip sonu hedeflenen enerji (p<0.001) ve protein (p<0.001) gereksinimlerini karşılama yüzdeleri arasındaki farkın istatistiksel olarak anlamlı olduğu görülmüştür. Yapılan ileri analizlerde; bası yarası

olmayan hastalara göre; bası yarası yatışta olan ( $p<0.001$ ) ve hastanede gelişen hastaların ( $p<0.001$ ) APACHE II ve NRS-2002 skorlarının daha yüksek, takip sonunda hedeflenen enerji ve protein gereksinimlerini karşılama yüzdelerinin ise daha düşük olduğu bulunmuştur. Yatışında bası yarası olan hastaların başlangıç serum albümin değerlerinin  $<3$  g/dL olduğu ve bu grupla birlikte hastanede bası yarası gelişen hastaların da takip sonu serum albümin değerlerinin yatış değerlerine göre istatistiksel olarak azaldığı görülmüştür ( $p<0.001$ ).

**Sonuç:** Yoğun bakım ünitesinde yatan hastalarda yüksek malnutrisyon riski olanların, yatışları sırasında değerlendirilen düşük serum albümin değerlerinin, uygulanan beslenme destek tedavisinin etkinliğinin ve yüksek APACHE II skorlarının hastalarda bası yarası durumu ile ilişkilendirilebileceği düşünülmektedir.

## EXTENDED ABSTRACT

**Aim:** Pressure sores are localized damage to the skin and underlying tissue caused by pressure, friction, and/or a combination of these factors and usually occur on bony parts. In addition to external factors such as pressure, humidity, and friction, internal factors such as advanced age, malnutrition, decreased oral intake, edema/hypoalbuminemia, state of consciousness, decreased sensory perception, decreased mobilization, and incontinence play a vital role in the formation of pressure sores (Keller et al., 2002). Various scales, such as Norton, Braden, and Waterlow are used to evaluate patients at risk for pressure sores. It is argued that among these scales, Norton and Braden scales have the highest reliability (Magnan & Maklebus, 2009). Intensive care unit patients constitute one of the riskiest groups in terms of malnutrition and pressure sores. According to a recent systematic review, patients hospitalized in intensive care units have a high prevalence of malnutrition ranging from 38% to 78% (Munoz et al., 2022), which leads to increased morbidity and mortality rates, longer hospital stays (Havens et al. et al., 2018; Mogensen et al., 2018; Osooli et al., 2019), and hospital-acquired pressure sores ranging from 12% to 24.5% (Chaboyer et al., 2018). Based on these reasons, this study aimed to investigate the incidence of pressure sores and risk factors that may be associated with pressure sores in patients hospitalized in intensive care units.

**Methods:** In this cross-sectional study of patients hospitalized in intensive care units of Mersin City Educational and Research Hospital, 200 patients aged 18-65 years, with an albumin value  $> 2.5$  g/dL and a BMI value of 18.5-24.9 kg/m<sup>2</sup> were evaluated. Pregnant and lactating patients, patients not aged 18-65 years, with BMI  $<18.5$  kg/m<sup>2</sup> and  $>25$  kg/m<sup>2</sup>, and

who did not accept to participate in the study were not included. In the study, the patients' descriptive characteristics and anthropometric measurements (body weight, height, BMI values) were questioned. The APACHE II screening tests were used to assess the severity of their diseases, the Norton Pressure Sore Risk Assessment Scale was used to assess the risks of pressure sores, and the NRS2002 screening test was used to detect malnutrition conditions. In addition, the patients' nutritional status and serum albumin values were monitored and recorded by the dietitian at daily visits for 30 days. Ethics committee approval was obtained from the Scientific Research and Publication Ethics Committee of Toros University (Date: 25.09.2019 and #42), and verbal consent was obtained from the patients or their relatives.

The data obtained in the study were evaluated using SPSS for Windows 22 software. Appropriate descriptive values are presented for qualitative and quantitative variables, and the conformity of the variables to normal distribution was examined using visual (histogram and probability graphs) and analytical methods (Kolmogorov-Smirnov/Shapiro-Wilk tests). The non-parametric Wilcoxon test was used to determine the difference between two dependent groups and continuous variables. Non-parametric Mann-Whitney-U test was used between two independent groups and for continuous variables. Non-parametric Kruskal-Wallis analysis was used for the three groups. The significance level for statistical analysis was accepted as 0.05.

**Results:** In this study, the rate of patients with high-risk nutritional status according to the NRS-2002 score in patients hospitalized in intensive care units was 58% ( $n=116$ ). According to the Norton scale, the rate of patients with a high risk of developing pressure sores during admission to the intensive care unit was 62.5% ( $n=125$ ). The mean APACHE II score was  $13.94\pm 7.47$ . It was found that 21.5% ( $n=43$ ) of the patients had pressure sores during admission to the intensive care unit. Twenty percent ( $n=40$ ) of the patients occurred pressure sores in the hospital. During the follow-up, 11% ( $n=22$ ) of the patients died, and 89% ( $n=178$ ) of the patients were discharged. There was no statistically significant difference between the mortality rates of patients without pressure sores, patients with pressure sores at admission, and patients who occurred pressure sores in the hospital ( $p>0.05$ ). As expected, the patients' Norton scale scores were higher in those with pressure sores at admission and those who developed pressure sores in the hospital compared to those without pressure sores ( $p<0.001$ ). It was seen that the difference between the APACHE II ( $p<0.001$ ) and NRS-2002 scores ( $p<0.001$ ) and the

percentages of meeting the target energy ( $p<0.001$ ) and protein ( $p<0.001$ ) needs at the end of the follow-up according to the pressure sore status of the patients were statistically significant. In further analyses, the APACHE II and NRS-2002 scores of patients with pressure sores during admission ( $p<0.001$ ) and those who developed pressure sores in the hospital ( $p<0.001$ ) were higher, and the percentages of meeting their targeted energy and protein needs at the end of follow-up were lower than those without pressure sores. It was observed that the initial serum albumin values of the patients with pressure sores during admission were  $<3$  g/dL, and the serum albumin values of the patients who developed pressure sores in the hospital and this group were statistically decreased at the end of the follow-up compared to the admission values ( $p<0.001$ ).

Pressure sores are an important problem that increases both the burden of caregivers and the cost of care and significantly affects the mortality and morbidity rates by prolonging the hospital stay of intensive care patients. In order to evaluate this problem in a timely and accurate manner, it is crucial to identify risky patients with scales such as Norton that measure pressure sores, evaluate the patient's nutrition and general condition, and monitor and initiate nutritional support treatments. In this study, it was observed that the following could be associated with pressure sore status: those at high risk of malnutrition according to NRS-2002 in intensive care unit patients, low serum albumin values evaluated during admission, and high APACHE II score, assessing the efficacy of nutritional support therapy and disease severity of the patients. There are also limitations of the study that should be acknowledged. The first of the limitations of the study is that it was designed as cross-sectional, and therefore generalizations cannot be made. Another limitation is the inclusion of patients, regardless of age and diagnosis, in the intensive care unit patients. For this reason, further randomized controlled studies are needed to examine the relationship between pressure sores and nutritional status in intensive care unit patients.

## GİRİŞ

Bası yaraları baskı, sürtünme ve/veya bu faktörlerin bir araya gelmesinden kaynaklanan deri ve alttaki doku alanında meydana gelen lokalize hasarlar olup genellikle kemik çıkıntıları üzerinde oluşmaktadır. Bası yarası oluşumunda basınç, nem ve sürtünme gibi dışsal etkilerin yanı sıra ileri yaş, malnutrisyon, oral alımın azalması, ödem/hipoalbuminemi, bilinç durumu, duyuşsal algının azalması, mobilizasyonun azalması, inkontinans gibi içsel faktörler de önemli rol oynamaktadır (Keller ve ark., 2002). Bu faktörlerin ışığında bası yarası açısından riskli hastaların değerlendirilmesi için Norton, Braden, Waterlow gibi ölçekler kullanılmaktadır. Bu ölçekler arasında güvenilirliği yüksek olanların Norton ve Braden ölçekleri olduğu savunulmaktadır (Magnan ve Maklebus, 2009).

Beslenme, deri bütünlüğünü arttırmada ve bası yaralarının varlığında doku onarımını desteklemede önemli bir rol oynamaktadır. Kötü beslenme, düşük Beden Kütle İndeksi (BKİ) ve hipoalbuminemi ile ilişkilendirilen malnutrisyon, bası yarası gelişiminde etkili faktörlerden biri olarak kabul edilmektedir (Berlowitz, 2014). Malnutrisyon riski, Nutrisyonel Risk Screening 2002 (NRS 2002) gibi standartlaştırılmış tarama araçları kullanılarak hızlı ve etkili bir şekilde belirlenebilmektedir. Malnutrisyon riskinin saptanması için NRS2002 tarama testinin kullanıldığı bir çalışmada, NRS2002 testinin ön değerlendirmesinin bası yarası gelişme insidansının bir göstergesi olduğu sonucuna varılmıştır (Alhaug ve ark., 2017). Brezilya'da yapılan başka bir çalışmanın da bu sonuçları doğrular nitelikte olduğu görülmektedir (Serpa ve ark., 2020).

Malnutrisyon ve bası yarası açısından en riskli olan gruplardan birini yoğun bakım ünitesi hastaları oluşturmaktadır. Yakın zamanda yapılan sistematik bir incelemeye göre yoğun bakım ünitelerinde yatan hastalarının %38 ile %78 arasında değişen yüksek bir malnutrisyon prevalansına sahip olduğu (Munoz ve ark, 2022), bu durumun da artmış morbidite ve mortalite oranlarına, daha uzun süre hastanede kalış süresine (Havens ve ark., 2018; Mogensen ve ark., 2018; Osooli ve ark., 2019) ve %12 ile %24.5 arasında değişen hastane kaynaklı bası yaralarına ne-

den olduğu bildirilmektedir (Chaboyer ve ark., 2018). Norveç'te yapılan başka bir çalışmada ise hastanelerde bası yaralarının en fazla yoğun bakım ünitelerinde görüldüğü (%18) ve görülen bası yaralarının %14'ünün ise hastane kaynaklı olduğu vurgulanmaktadır (Bredesen ve ark., 2015). Yoğun bakım hastalarında beslenme gereksinimleri hastalığın ciddiyetine ve evrelerine göre değişkenlik göstermektedir. Bu hastalarda artan malnutrisyon ve bası yarası prevelansı, yetersiz enerji ve besin öğeleri alımının yanı sıra oluşan hipermetabolik yanıtta da kaynaklanabilmektedir. Hipermetabolik durumlarda artan enerji ihtiyacı önce glikojen depolarından ve ardından da visseral depolardan çekilmektedir. Bu durum da kas kaybına, nitrojen tutulumuna ve albümin sentezinin bozularak azalmasına neden olmaktadır. Düşük albümin düzeyleri ise interstiyel ödeme neden oldukları için yara iyileşmesini geciktirmektedir (Seres, 2021).

Deri bütünlüğünün sağlanması için yeterli enerji ve besin öğelerine ihtiyaç duyulmaktadır. Bu yüzden hastaların optimal enerji ve protein gereksinimleri sağlanarak beslenme durumlarının düzeltilmesi bası yarası gelişiminin önlenmesini ve tedavisini destekleyen önemli bir adım olarak görülmektedir. Yapılan araştırmalar, bası yarası riski taşıyan malnutrisyonlu yoğun bakım hastalarının günde 30-35 kkal/kg enerji ve 1.25-1.5 g protein/kg ihtiyacı olduğunu belirtirken (Bauer ve ark., 2013; Morley ve ark., 2010), Amerika ve Avrupa'da yayınlanan kılavuzlar tarafından ise bu hastaların 25-30 kkal/kg enerji ve 1.2 g protein/kg ihtiyacı olduğu belirtilmektedir (Singer ve ark., 2019; Mehta ve ark., 2017). Hastaların alımı beslenme ihtiyaçlarını karşılamaya yetmediğinde ise bağışıklığı modüle eden besin öğelerini (arginin, çinko ve antioksidanlar) içeren beslenme destek ürünleriyle takviyelerin yapılması önerilmektedir (EPUAP, 2019).

Bu çalışmanın amacı yoğun bakım ünitelerinde yatan hastalarda bası yarası gelişme insidanslarını ve bası yarasının ilişkili olabileceği risk faktörleri araştırmaktır.

## ANA METİN

Bu kesitsel çalışma bir Mersin Şehir Eğitim ve Araştırma Hastanesi yoğun bakım ünitelerinde yatan, serum albümin değeri >2.5 g/dL ve BKİ değeri 18.5-24.9 Kg/m<sup>2</sup> olan, 18-65 yaş arası 200 hasta ile yürütülmüştür. Çalışmaya gebe ve emziciler, 18-65 yaş aralığında olmayan, BKİ <18.5 Kg/m<sup>2</sup> ve >25 Kg/m<sup>2</sup> olan ve çalışmaya katılmayı kabul etmeyen hastalar dahil edilmemiştir. Hastaların demografik ve klinik özellikleri, beslenme durumları, beslenme yolu ve dozları, günlük enerji ve protein alımları, antropometrik ölçümleri (vücut ağırlığı, boy uzunluğu, BKİ) ve serum albümin değerleri diyetisyen tarafından günlük viziteler ve günlük tutulan hasta kayıtları ile değerlendirilmiştir. Hastaların vücut ağırlığı ve boy uzunluğu ölçümleri hasta dosyalarından alınmıştır. Hastaların BKİ değerleri, vücut ağırlığı (kg)/boy uzunluğu (m)<sup>2</sup> formülü ile hesaplanmıştır (WHO, 2022).

Yoğun bakım ünitelerinde, hastalık ciddiyetini değerlendirerek mortaliteyi tahmin eden "prognostik skorlama sistemleri" yaygın olarak kullanılmaktadır (Özdemir, 2014). Çalışmamızda yoğun bakım ünitelerinde sorumlu hemşireler tarafından değerlendirilen prognostik sorgulama sistemlerinden biri olan Akut Fizyoloji ve Kronik Sağlık Değerlendirmesi II (Acute Physiology and Chronic Health Evaluation APACHE II) verileri kayıt altına alınmıştır. APACHE II toplam skoru akut fizyoloji skoru, yaş ve kronik sağlık değerlendirme olmak üzere üç alt başlığın toplamından oluşmakta olup, en yüksek değer 71'dir. Toplam skor puanı 25 olduğunda mortalite oranı %25, 35 puan ve üzerinde olduğunda ise mortalite oranı %80'e yükselmektedir (Knaus ve ark., 1985).

Hastalarda beslenme durumunun değerlendirilmesi, hastaneye yatıştan sonraki ilk 24 saat içinde Nütrisyonel Risk Değerlendirme Skoru-2002 (NRS-2002) kullanılarak, servis hemşireleri tarafından yapılmıştır. NRS-2002 skoru ≥3 olan hastalar, nütrisyonel açıdan yüksek riskli hasta olarak kabul edilmektedir (Kondrup ve ark., 2003). NRS-2002'nin 2014 yılında Türkiye'de yapılan geçerlilik ve güvenilirlik çalışmasında, sensitivitesi %88, spesifitesi %92 olarak bulunmuş ve ülkemizde yatan hastalarda sıklıkla tercih edilen



bir tarama testi olarak kabul edilmektedir (Taş, 2020). Yoğun bakımda yatan hastalarda hemodinamik stabilizasyon sağlandıktan sonra 24-48 saat içinde hastaların beslenme desteğine başlanması ve hedef enerji alımına 48-72 saatte ulaşması hedeflenmektedir (Heyland ve ark., 2003). Bu çalışmada da hastalar hedeflenen enerji ve protein gereksinimlerine ulaşma süreleri 1 ay boyunca haftalık olarak kontrol edilmiş ve. Yoğun bakımda yatış süreleri ve mortalite durumları kayıt altına alınmıştır. Yoğun bakımda yatan hastanın beslenme desteği ihtiyacı için ilk tercih oral destek ve/veya enteral beslenme desteği olmuş ancak oral-enteral yoldan beslenme desteği sağlanamayacaksa gecikmeden beslenme desteği parenteral beslenme olarak önerilmiştir. Hastaların günlük enerji gereksinimleri 25-30 kkal/kg formülü kullanılarak hesaplanmıştır. Günlük hedeflenen protein gereksinimi ise 1.2-1.5 g/kg olarak hesaplanmış ve hedeflenen protein gereksinimi hastanın klinik özelliklerine göre gereken durumlarda yeniden düzenlenmiştir (Kreymann ve ark., 2006).

Bu çalışmada hastaların bası yarası oluşma risklerini değerlendirmek için Norton bası yarası değerlendirme ölçeği kullanılmıştır. Bu ölçek, bası yarası riskini tanılamak için literatürde yer alan ilk ölçek olup hastanede yatan hastaların sistematik olarak değerlendirilmesine dayanır (Fırat Kılıç ve ark., 2017). Ölçek fiziksel durum, mental durum, aktivite, hareketlilik ve inkontinans olmak üzere 5 maddeden oluşmaktadır. Her bir madde 1-4 arasında puanlandırılmaktadır ve ölçekten alınabilecek en düşük puan 5, en yüksek puan 20'dir (Norton 1989), toplamda 14 puan ve altında olan hastalar riskli olarak değerlendirilmektedir (Pritchard 1986). Ölçeğin Türkçeye uyarlaması Avşar ve Karadağ tarafından yapılmış ve geçerli-güvenilir bir ölçek olarak bulunmuştur (Avşar, 2012).

Bu çalışma için Toros Üniversitesi Bilimsel Araştırma ve Yayın Etiği Kurulundan 25.09.2019 tarih ve 42 karar sayısı ile etik kurul onayı alınmış olup, çalışmaya katılan hastalardan ya da yakınlarından sözlü onam alınmıştır.

## Verilerin İstatistiksel Analizi

Çalışmada elde edilen veriler, SPSS for Windows - 22 yazılımı kullanılarak değerlendirilmiştir. Nitel ve nicel değişkenler için uygun betimsel değerler verilmiştir. Nitel değişkenler, sayı (n) ve yüzde (%) olarak, nicel değişkenler ise ortalama ve standart sapma ( $\pm$ SD) olarak ifade edilmiştir. Değişkenlerin normal dağılıma uygunluğu görsel (histogram ve olasılık grafikleri) ve analitik yöntemler (Kolmogorov-Smirnov/Shapiro-Wilk testleri) kullanılarak incelenmiştir. Bağımlı iki grup ve sürekli değişkenler arasındaki farkın belirlenmesi için non-parametrik Wilcoxon testi, bağımsız iki grup arasında, sürekli değişkenler için non-parametrik Mann-Whitney-U testi, üçlü bağımsız grup arasındaki analizler için non-parametrik Kruskal-Wallis analizi kullanılmıştır. İstatistiksel analizler için anlamlılık düzeyi 0.05 olarak kabul edilmiştir.

## SONUÇLAR

Bu çalışmada, yoğun bakım ünitelerinde yatan hastalarda NRS-2002 skoruna göre beslenme durumları yüksek riskli olan hastaların oranı %58 (n=116), Norton ölçeğine göre yoğun bakıma yatışta bası yarası oluşma riski yüksek olan hastaların oranı ise %62.5 (n=125) olarak bulunurken, APACHE II skorları ise  $13.94 \pm 7.47$  olarak bulunmuştur. Hastaların %21.5 (n=43)'ünde yoğun bakıma yatış sırasında bası yarası olduğu, %20 (n=40)'sinde ise hastanede bası yarası olduğu tespit edilmiştir. Hastalar bası yarası bölgelerine göre sınıflandırıldığında, sakrum bası yarası olan hasta oranının %27.5 (n=55), femur %1 (n=2), topuk %6.5 (n=13), kalça %4.5 (n=9), koksiks %2 (n=4) olduğu görülmüştür. Hastalar bası yarası evrelerine göre sınıflandırıldığında ise bası yarası 1. evre hasta oranı %14.5 (n=29), 2. evre %11 (n=22), 3. evre %7.5 (n=15), 4. evre %9 (n=18) olduğu tespit edilmiştir. Takip süresince hastaların %11'inde (n=22) ölüm görülmüş ve hastaların %89'u (n=178) ise taburcu edilmiştir. Bası yarası olmayan, bası yarası olan ve bası yarası hastanede gelişen hastaların mortalite oranları arasında istatistiksel olarak anlamlı bir fark bulunmamıştır ( $p > 0.05$ ). Hastaların demografik ve klinik özellikleri Tablo 1'de gösterilmiştir.

Beklendiği üzere hastaların Norton ölçeği skor puanlarının hastane yatışında bası yarası olan ve bası yarası hastanede gelişen hastaların, bası yarası olmayan hastalara göre daha yüksek olduğu görülmüştür ( $p<0.001$ ). Bu çalışmada Norton ölçeğine göre yüksek riskli olarak değerlendirilen hastalar içinde ( $n=125$ ) malnütrisyonlu olmayan hastaların oranı ( $n=70$ ) daha yüksek bulunmuştur ( $p<0.001$ ). Hastaların bası yarası durumlarına göre APACHE II ( $p<0.001$ ) ve NRS-2002 ( $p<0.001$ ) skor puanları arasındaki farkın istatistiksel olarak anlamlı olduğu görülmüştür. Yapılan ileri analizde; bası yarası yatışında olan ( $p<0.001$ ) ve hastanede gelişen hastaların ( $p<0.001$ ) APACHE II ve NRS-2002 skor puanlarının bası yarası olmayan hastaların APACHE II ve NRS-2002 skor puanlarından daha yüksek olduğu görülmüştür (Tablo 2).

Yoğun bakımda beslenme desteği başlanan hastaların %15'i oral ve enteral ( $n=30$ ), %20'si ( $n=40$ )

oral ve parenteral, % 8.5'i ( $n=17$ ) yalnızca enteral, % 55.5'i ( $n=111$ ) yalnızca parenteral, %1'i ( $n=2$ ) enteral ve parenteral kombine beslenme desteği almışlardır. Hastaların bası yarası ve beslenme durumuna ait değişkenler arasındaki ilişki Tablo 2' de gösterilmiştir. Hastaların bası yarası durumları ile takip sonunda hedeflenen ortalama enerji ( $p<0.001$ ) ve protein ( $p<0.001$ ) gereksinimleri karşılama yüzdeleri arasındaki fark istatistiksel olarak anlamlı bulunmuştur. Yapılan ileri analizde, bası yarası olmayan hastaların enerji ve protein gereksinimlerini karşılama yüzdelerinin bası yarası yatışında olan ( $p<0.001$ ) ve hastanede gelişen hastaların ( $p<0.001$ ) gereksinimlerini karşılama yüzdelerine göre daha yüksek olduğu bulunmuştur (Tablo 2).

Yoğun bakıma yatışta NRS-2002 skoruna göre yüksek riskli olarak değerlendirilen hastalarda takip sonu serum albümin değerlerinin 3 g/dL'nin altına düştüğü ve yatıştaki albümin de-

**Tablo 1.** Demografik ve klinik bulgular

Değişken	Değer
Yaş, ( $\bar{X}\pm SD$ ), yıl	64.77±17.48
Cinsiyet (E/K), n (%)	109(54.5)/91(45.5)
Beden kütle indeksi, ( $\bar{X}\pm SD$ ) $kg/m^2$	25.33±4.63
Yüksek riskli hasta oranı (NRS-2002) n, (%)	116 (58)
Yatış sırasındaki tanı, n, (%)	
Nörolojik hastalıklar	47 (23.5)
Cerrahi hastalıklar	30 (15)
Dahili hastalıklar	123 (61.5)
Ko-morbid hastalıklar, n, (%)	
Diabetes Mellitus	14 (7)
Hipertansiyon	44 (22)
Kalp yetmezliği	29 (14.5)
Kronik obstrüktif akciğer hastalığı	23 (11.5)
Alzheimer	1 (0.5)
Akut böbrek yetmezliği	17 (8.5)
Kanser	16 (8.0)
Hastaların Bilinç Durumları n, (%)	
Açık	93 (46.5)
Stupor	100 (50)
Kapalı	7 (3.5)
APACHE-2 skoru, ( $\bar{X}\pm SD$ )	13.94±7.47
Bası yarası, n, (%)	83 (41.5)
Yok	117 (58.5)
Yatış sırasında bası yarası	43 (21.5)
Hastanede oluşan bası yarası	40 (20)
Norton skoru ( $\bar{X}\pm SD$ ),	12.21±4.71
Norton skoruna göre sınıflama, n, (%)	
Riskli $\leq 14$ puan	125 (62.5)
Normal $> 14$ puan	75 (37.5)



ğlerine göre aradaki farkın istatistiksel olarak anlamlı olduğu görülmüştür ( $p<0.001$ ). Yatışında bası yarası olan hastaların başlangıç serum albümin ortalama değerlerinin 3 g/dL'nin altında olduğu ve takip sonu değerinin düştüğü, yine hastanede bası yarası gelişen hastaların da takip sonu serum albümin ortalama değerlerinin 3 g/dL'nin altına düştüğü görülmüştür ( $p<0.001$ ) (Tablo 3).

Yoğun bakım üniteleri fiziksel durumu ağır olan hastaların yakından izlendiği ve yaşam fonksiyonlarının desteklendiği özel birimlerdir (Diker ve ark., 2009). Bu hastalarda malnütrisyon ve bası yaraları görülme olasılıkları oldukça yaygındır (Taş, 2020). Özellikle YBÜ'de yatış süreleri uzayan hastalarda bası yaraları daha sık gözlenmektedir (Kıraner E. ve ark., 2016). Malnütrisyon yara iyileşmesi ile birlikte bası yarası gelişiminde de önemli bir rol oynamaktadır (Bayır ve

ark., 2015; Taş, 2020). Bası yaralarının yoğun bakım ünitelerinde görülme insidansı (%56) yüksek olduğundan (Fırat Kılıç ve Sucudağ, 2017) malnütrisyon ve bası yaralarının önlenmesi, yoğun bakım ünitesinin amaçları arasında öncelikli olarak yer almalıdır (Taş, 2020; Kabalak ve ark, 2013; Uslu ve Terzioğlu, 2015). Bu nedenle bası yaralarına neden olabilecek risk faktörlerinin belirlenmesi ve önlenmesi gerekmektedir. Bası yaralarını değerlendirirken; güvenilir, geçerli ve hastanın içinde bulunduğu durumu tam olarak ortaya koyabilen risk değerlendirme ölçeklerinin kullanılmasına dikkat edilmelidir (Fırat Kılıç ve Sucudağ, 2017). Norton risk değerlendirme ölçeği, hastalarda bası yaraları riskini değerlendiren ilk ölçek olmasına rağmen, literatürde az sayıda çalışma bulunmaktadır (Pancorbo-Hidalgo ve ark., 2006). Bu çalışmada da beklendiği üzere Norton risk değerlendirme ölçeği ile bası yarası

**Tablo 2.** Hastaların bası yarası durumu ile beslenme durumlarına ait değişkenler arasındaki ilişki

	Bası yarası durumu			p
	Bası yarası yok (117)	Yatışında vardı (n=43)	Hastanede gelişti (n=40)	
Norton skoru	9.90±4.01	16.72±2.66	14.13±3.66	0.001*
NRS-2002	2.42±1.68 <sup>a,b</sup>	3.91±1.28 <sup>a</sup>	3.28±1.76 <sup>b</sup>	0.001*
APACHE II skoru	11.13±6.11 <sup>a,b</sup>	19.88±8.35 <sup>a</sup>	15.75±5.72 <sup>b</sup>	0.001*
Toplam enerji gereksinimi (kkal/gün)	1773.62±203.12	1836.63±237.01	1834.55±164.45	0.100
Toplam protein gereksinimi (g/gün)	84.92±9.93	88.12±11.35	87.90±7.68	0.080
Takip sonunda hedeflenen enerji gereksinimini karşılama yüzdesi	78.11±28.15 <sup>a,b</sup>	60.21±24.55 <sup>a</sup>	54.52±23.33 <sup>b</sup>	0.001*
Takip sonunda hedeflenen protein gereksinimini karşılama yüzdesi	63.54±25.73 <sup>a,b</sup>	49.70±22.31 <sup>a</sup>	44.80±21.89 <sup>b</sup>	0.001*

\* Kruskal Wallis testi,  $p<0.001$ , <sup>a,b</sup> Mann-Whitney U testi,  $p<0.001$

**Tablo 3.** Yoğun bakımda yatan hastaların giriş ve takip sonu ortalama serum albümin değerlerinin malnütrisyon ve bası yarası durumuna göre değerlendirilmesi

	Giriş serum albümin (g/L), ( $\bar{X}\pm SD$ )	Takip sonu serum albümin (g/L), ( $\bar{X}\pm SD$ )	P
<b>Norton Ölçeği</b>			
Riskli $\leq 14$ puan (n=125)	3.40±0.57	3.19±0.61	0.001*
Normal $> 14$ puan (n=75)	3.10±0.57 <sup>b</sup>	2.73±0.63 <sup>b</sup>	0.001*
<b>Bası yarası varlığı</b>			
Bası yarası yok (n=117)	3.50±0.52 <sup>a</sup>	3.33±0.53 <sup>a</sup>	0.001*
Yatışında vardı (n=43)	2.83±0.48 <sup>a,b</sup>	2.39±0.48 <sup>a,b</sup>	0.001*
Hastanede gelişti (n=40)	3.16±0.52 <sup>a,b</sup>	2.75±0.56 <sup>a,b</sup>	0.001*
<b>NRS-2002 Skoru</b>			
Yüksek riskli $\geq 3$ puan (n=116)	3.16±0.57	2.84±0.62	0.001*
Normal $< 3$ puan (n=84)	3.46±0.57	3.25±0.63	0.001*

\* Wilcoxon signed rank test  $p<0.001$ , <sup>a</sup>Kruskal Wallis testi,  $p<0.001$ , <sup>b</sup>Mann-Whitney-U test

durumu gelişmesi arasında bir ilişki saptanmıştır. Bası yarası olmayan hastalarda, Norton risk değerlendirme ölçeği ortalama değerleri bası yarası olan ve bası yarası hastanede gelişen hastalara göre daha düşük bulunmuştur ( $p<0.05$ ) (Tablo 2).

Literatürde, yoğun bakım ünitelerinde hastanın genel durumunu değerlendirmek, sağ kalımı tahmin etmek ve hastalık şiddetini belirlemek amacı ile yaygın olarak kullanılan APACHE II skoru ile bası yarası gelişmesi arasındaki ilişkiyi değerlendiren çalışmalar bulunmaktadır (Clough, 1994; Inman ve ark., 1993; Gül ve ark., 2016; Kelebek ve ark., 2007). Bu çalışmada da, bası yarası durumu ile APACHE II skoru arasında anlamlı bir ilişki bulunmuş olup, bası yarası yatışta var olan ve hastanede gelişen hastalarda, APACHE II skoru ortalama değerlerinin bası yarası olmayan hastalara göre daha yüksek olduğu tespit edilmiş ( $p<0.05$ ) ve bası yarası olan veya hastanede gelişen hastaların genel durumunun daha kötü olduğu sonucuna varılmıştır. Kelebek Girgin ve Kurhan Erarı (2007)'nin çalışmasında da bu çalışmayla benzer olarak anlamlı bir ilişki saptanmış ve APACHE II skoru arttıkça bası yarasının gelişme riskinin de artmış olduğu bulunmuştur. Gül ve ark. (2016)'nın çalışmasında ise, APACHE II skoru ile bası yarası gelişmesi arasında anlamlı bir ilişki bulunamamıştır.

Artan enerji açığı ile yoğun bakım ünitesinde sıklıkla görülen komplikasyonlardan biri olan bası yarası arasında güçlü bir ilişki olduğu rapor edilmektedir (Bayır ve ark., 2015). Bu nedenle, yaşlı ve yatağa bağımlı hastalarda beslenme problemlerinin yakından takip edilmesi ve gerekli önlemlerin alınması gerekmektedir (Taş, 2020). Enerji, protein ve diğer besinlerin yetersiz alınması veya artmış vücut ihtiyacının karşılanamaması nedeniyle organların boyut veya fonksiyonlarında kayıp ve klinik sonuçlarda ölçülebilir yan etkiye sebep olabilen malnutrisyon gelişebilmektedir. Buna istinaden bu hastalarda; sarkopeni, enfeksiyonlara eğilim, bası yarası, akut böbrek yetmezliği ve mortalite daha sık gözlenmektedir (Çınar ve ark., 2017). Bu yüzden; yoğun bakım ünitesi hastalarında beslenmenin değerlendirilmesi, gerektiğinde desteklenmesi ve sonuçların takip edilmesi tedavinin en önemli unsurlarından biri

olmalıdır (Taş, 2020). Nutrisyonel taramalar, beslenme durumunun değerlendirmesi ile malnutrisyonu olan hastaları ve malnutrisyon riski altında olan bireyleri saptamak amacı ile yapılmaktadır. Yoğun bakım ünitesinde yatan hastaların yeterince beslenip beslenmediklerini değerlendirmek amacıyla, hasta için uygun bir ölçme yöntemi kullanılmalı, değerlendirme sonucuna göre hastanın beslenme desteği alıp almayacağına, alacaksa besin ögesi gereksinimlerine karar verilmelidir (Diker ve ark., 2009). ESPEN (European society of Parenteral and Enteral Nutrition) tarafından kullanımı önerilen NRS 2002 skoru, hastanede yatan hastalarda malnutrisyon oluşma riskini saptama, malnutrisyon riski yüksek hastaları tanımlama ve bu hastalarda beslenme desteğinin yeterliliğini değerlendirme amacıyla yoğun bakım ünitesi hastaları için de kullanılan beslenme durumunu değerlendirme testi olarak kabul edilmektedir (Bayır H. ve ark., 2015). Literatürde yatan hastalarda NRS-2002 skoru ile beslenme durumunu değerlendiren birçok çalışma bulunmaktadır (Schuetz P. ve ark., 2019; Acehan ve ark., 2020; Anthony PS, 2008; Johansen N. ve ark., 2004; Starke ve ark., 2011). Bu yüzden; çalışmamızda da yoğun bakım hastalarının beslenme durumunun değerlendirilmesinde NRS-2002 tarama testi tercih edilmiştir. Aynı zamanda bu çalışmada, bası yarası gelişimi ile NRS-2002 skorları arasındaki ilişkiye de bakılmış ve bası yarası yatışta olan ve hastanede gelişen gruptaki hastalar beslenme açısından yüksek riskli ( $\geq 3$ ) bulunurken; bası yarası olmayan hastaların beslenme durumunun normal ( $<3$ ) olduğu saptanmıştır ( $p<0.05$ ) (Tablo 2). Literatürde bu çalışmaya benzer şekilde bası yarasının NRS-2002 skoru ile ilişkili olduğunu gösteren birçok çalışma da bulunmaktadır (Alhaug ve ark., 2017; Rashvand ve ark., 2019; Terekeci ve ark., 2009).

Yoğun bakım ünitesi hastalarında beslenme durumu değerlendirilmediğinde veya beslenme tedavisi takip edilmediğinde, enerji tüketimi için gereken besin öğeleri vücut tarafından temin edilemeyecek, zaman içinde ilerleyici vücut ağırlığı kaybı gelişecek ve hastanın yaşam kalitesi ve sağ kalımı olumsuz yönde etkilenecektir (Çınar ve ark., 2017). Bu hastalarda beslenme tedavisinin, bireye özel ayarlanmasına ve hastanın mevcut durumuna göre şekillendirilmesine

dikkat edilmelidir (Taş, 2020). Bu çalışmada da bası yarasının, yoğun bakım ünitesi hastalarında enerji ve proteinin karşılanması açısından olumsuz etkileri olduğu belirlenip, bası yarası olmayan hastalarda 1 aylık takip sonrasında enerji ve protein gereksinimlerinin karşılanma yüzdeleri daha yüksek bulunmuştur ( $p<0.05$ ) (Tablo 2). Literatürde de bu çalışmadaki sonucu destekler şekilde, bası yarasının hastaların enerji ve protein gereksinimlerini karşılaması üzerine olumsuz etkilerini gösteren birçok çalışma yer almaktadır (Roberts ve ark., 2013; Dupertuis ve ark., 2003; Mudge ve ark., 2011; Thibault ve ark., 2011). Bu çalışmada, bası yarası olan ve hastanede gelişen hastalarda oluşan artan katabolizmanın hastaların hedeflenen enerji ve protein gereksinimlerini karşılayamamalarına neden olabileceği düşünülmüştür.

Visseral proteinlerden albümin düzeyinin yarılanma ömrünün uzun olması ve enfeksiyon veya inflamasyon gibi durumlarda beslenme durumunu iyi yansıtmamasına rağmen; yoğun bakım hastalarında bu durumların haricinde beslenme durumunu değerlendirmede ve uygulanan beslenme tedavisinin etkinliğini göstermede sıklıkla tercih edilmektedir (Bayır ve ark., 2015). Literatürde albümin düzeyi ile bası yarası gelişmesi arasındaki ilişkiyi değerlendiren çalışmalarda düşük albümin düzeylerinin interstisyel ödeme neden olarak yara iyileşmesini olumsuz yönde etkilediği düşünülmektedir (Kelebek Girgin ve Kurhan Erarı, 2007; Gül ve ark., 2016; Tokgöz ve Demir, 2010; Ortaç Ersoy ve ark., 2013). Bu çalışmada da yoğun bakım ünitelerinde yatan hastaların albümin değerlerinin 1 aylık takip sonrasında, yatışta bası yarası olan ve hastanede bası yarası gelişen hastalarda takip sonrasında ilk değerlere kıyasla istatistiksel olarak anlamlı bir şekilde düştüğü gözlenmiştir ( $p<0.05$ ) (Tablo 3). Yoğun bakımda artmış katabolizma, inflamasyon gibi nedenlerin dışında yetersiz kalan beslenme tedavilerinden ötürü de hipoalbüminemi gelişebilmektedir (Kıraner ve ark., 2016). Bu çalışmada da NRS-2002 skoruna göre yüksek riskli olan hastalarda albümin değerlerinde 1 aylık takip sonrasında malnütrisyon göstergesi olan sınır değerlerin ( $<3$  g/dL) de altına düştüğü gözlenmiştir. Ayrıca, bası yarası oluşma riskini değerlendiren Norton ölçeğine göre de yüksek

riskli olarak değerlendirilen hastalarda takip sonunda hastanın ilk kabul edildiği değerlere kıyasla albümin değerlerinde yine istatistiksel olarak anlamlı bir düşüş elde edilmiştir ( $p<0.05$ ). Literatürde bası yarasını değerlendiren ölçeklerle albümin değerleri arasındaki ilişkiyi değerlendiren çalışmalarla benzer sonuçlar elde edildiği görülmüştür (Sung ve Park, 2011; Woo ve ark., 2022; Park, 2014).

Araştırmanın sınırlılıklarından biri çalışmanın kesitsel olmasıdır ve bu yüzden çalışmada genelleme yapılamamaktadır. Ayrıca, tüm yoğun bakım ünitesi hastalarında yaş ve tanı ayırt etmeksizin hastaların çalışmaya dahil edilmesi yine çalışmanın sınırlılıklarından birini oluşturmaktadır. Bu nedenle, farklı yaş ve tanılardaki hastaların genel durumları çalışmanın sonucu üzerinde etkili olmuş olabilir.

Bu çalışmada, yoğun bakım ünitesinde yatan hastalarda NRS-2002'ye göre yüksek malnütrisyon riskli olanların, hastaların yatışları sırasında değerlendirilen düşük serum albümin değerlerinin, uygulanan beslenme destek tedavisinin etkinliğinin ve hastalarının hastalık ciddiyetlerini değerlendiren yüksek APACHE II skorunun, hastalarda bası yarası durumu ile ilişkilendirilebileceği görülmüştür. Bası yaraları, yoğun bakım hastalarının hastanede kalış sürelerini uzatarak mortalite ve morbidite oranını önemli derecede etkileyen hem bakım verenlerin yükünü hem de bakımın maliyetini arttıran önemli bir sorundur. Bu sorunun zamanında ve doğru değerlendirilebilmesi için, Norton gibi bası yaralarını ölçen ölçekler ile riskli hastaların belirlenmesi, hastanın beslenme ve genel durumunun değerlendirilmesi ve gerekli beslenme destek tedavilerin başlanması ve izlenmesi son derece önemlidir. Yoğun bakım ünitesi hastalarında, bası yarası ile beslenme durumu arasındaki ilişkiyi irdeleyen randomize kontrollü daha fazla çalışmaya ihtiyaç bulunmaktadır.

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Research Article

# Quality evaluation of formulated instant noodles from wheat, rice (*Oryza sativa*) and mushroom (*Agaricus bisporus*) flour blends

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## ABSTRACT

The purpose of this study is to produce and evaluate qualitatively, instant noodles from wheat, rice and mushroom flour blends. A composite flour of wheat and rice flours (90:10) was obtained as the best blends after a preliminary sensory evaluation and substituted with 10, 20, 30, 40 and 50 % mushroom flour coded as WRM1 (90:10), WRM2 (80:20), WRM3 (70:30), WRM4 (60:40), WRM5 (50:50) and commercial noodles (Indomie) served as the control. The formulated blends were used to produce instant noodles. The instant noodles were analysed for proximate composition, micronutrients (vitamin B<sub>1</sub>, B<sub>2</sub> and B<sub>3</sub>, iron, potassium, and phosphorus) microbial quality, cooking characteristics, sensory qualities and functional properties of the flour blends using standard procedures. Results showed that mushroom flour increased the crude protein (9.49-15.39 %), ash (1.39-5.31 %), crude fiber (1.50-5.40 %), moisture content (7.92-14.48 %). It, however, decreased the fat (0.5-1.50 %) and carbohydrate content (58.42-77.45 %). Potassium and vitamin B<sub>3</sub> were identified as the predominant micronutrients in the instant noodles samples and increased with level of mushroom addition. Sample WRM1 (90:10) with 10% mushroom flour had the highest mean for all sensory attributes (taste, colour, appearance, texture and overall acceptability) compared to other samples. The total

viable count ranged from  $4.3 \times 10^2$  (control) to  $1.78 \times 10^3$  cfu/g in sample WRM4 (60:40). Thus, mushroom flour could be incorporated into instant noodles to obtain an acceptable product rich in dietary fiber, protein, ash, vitamin B<sub>3</sub> and potassium but low in fat and carbohydrate.

## INTRODUCTION

Noodles are narrow strips of unleavened dough which is stretched, extruded or rolled flat and cut into one of a variety of shapes (Anon, 2019). Noodle has been increasingly an important food commodity worldwide. In 2018, the World Instant Noodles Association reported that 103.620 million servings were consumed worldwide (World Instant Noodle Association, 2019). The success of noodles all over the world is due to its low and affordable prices, convenience and the minimum efforts the products require for cooking.

There are many types of noodles, but the "instant" types continue to show increasing popularity globally as these products offer ease in preparation while being economical and tasty (Akanbi et al., 2011). Noodles are produced basically from wheat flour. According to FAO estimate, world would require around 840 million tonnes of wheat by 2050 from its current production level of 642 million tonnes. To meet this demand, developing countries need to increase their wheat production by 77 % (FAO, 2009). However, wheat production in Nigeria has been unpredictable. Reports indicated that until 1985, domestic wheat production in Nigeria was about 66.000 tons (Olugbemi, 1991). In 1988/89 crop production season about 600,000 tons of wheat was produced from a total of 214.000 hectares with an average yield of 2 tons per hectare (Olugbemi, 1991). In 2011 the production was 165,000 metric tonnes which drastically dropped to 60.000 metric tonnes in 2016 (Olugbemi, 1991), thus noodle production depended on wheat importation mostly from the United States since wheat cannot perform

well under tropical climate. According to Weigand (2011), African wheat imports will soar from 22.3 million tonnes in 2010 to 51.4 million tonnes in 2050, while the Middle East's imports will double from the region's minor wheat producers, from 14.4 million tonnes to 29.5 million tonnes. Brazilian wheat imports will climb from 6.5 million tonnes in 2010 to 10.5 million tonnes in 2050 in order to fulfill domestic demand. This wheat importation, has resulted in an immense drain on the economy, suppressing and displacing of indigenous cereals, with a resultant detrimental effect on the agricultural and technological development, and cause of poverty in Nigeria (Kumara, 2015). In order to reduce the impact of wheat importation on the economy, the Federal Government released a policy mandating flour mills to partially or wholly substitute wheat flour (Ammar et al., 2009). This resulted in the adoption of alternative solutions by using flours obtained from other raw materials in combination with wheat flour. In addition to wheat importation, wheat as a major raw material for noodle production has some inherent limitations such as low mineral composition, fiber, protein and high cost compared to local cereals such as rice. Efforts have been put in place via research to develop composite flour from available tubers, cereals and legumes for partial replacement of wheat flour in noodle production. The use of readily available cereals and inexpensive legume flours to complement the wheat flour can enhance the nutrient content of noodles and decrease the demand for importation of wheat flour (Onwurafor et al., 2020). To reduce the demand for wheat flour, substitution of wheat flour with other local flour is needed. Rice-mushroom-wheat composite flours could be used as an alternative. Rice is regarded as one of the most appropriate cereal grains for producing gluten-free products owing to its benefits of high digestibility, hypoallergenic properties among others (Marco and Rosell, 2008). Rice-based noodles are prepared from rice cultivars containing high-amylose content, low-gelatinization temperature and high-gel consistency (Yoenyongbuddhagal et al., 2002). Textural and cooking properties of rice-based noodles are dependent upon flour swelling power, pasting properties and

gel hardness (Bhattacharya et al., 1999) which directly affects consumer acceptance (Horndok and Noomhorm, 2007).

In general, products made from cereals are low in sodium, amino acids and total fat. Noodles produced from wheat flour contain 11-15 % protein dry basis but are deficient in lysine and threonine (the first and second limiting amino acid), common to most cereal products (Tongpun, 2006). Cereals are rich in minerals but the bioavailability of these minerals is usually low due to the presence of anti-nutritional factors such as phytate, trypsin inhibitor and polyphenols. Phytic acid is the most important anti-nutrient because it is found in most of the cereals and has strong ability to complex multi-charged metal ions, especially zinc, calcium and iron and makes them unavailable for human body utilization. The absence of lysine makes it difficult for the body to synthesize protein, hormones, enzymes and antibodies which are needed for growth and other functions (Flodin, 1997). Food fortification is adopted to improve the nutritive value of noodles made from cereals.

Mushroom is a macro fungus with a distinctive fruiting body, which can be either epigeous or hypogeous and large enough to be seen with naked eye and to be picked by hand (Chang and Miles, 1992). Mushrooms according to Bano (1993), have about twice the protein content of vegetables four times that of oranges and significantly higher than that of wheat and are high in vitamins such as riboflavin, biotin and thiamine. Mushrooms also present high nutritional value, having some bioactive components, like dietary fibre, antioxidants, minerals, folates, essential amino acids (such as lysine) as well as vitamins B<sub>1</sub>, B<sub>2</sub>, B<sub>3</sub> and C (Li et al., 2015) and a wide spectrum of mineral substances. It represents a good source of biologically valuable substances for human nutrition. However, it is a perishable product due to its high moisture content. Okafor et al. (2012) showed that there was significant improvement in the bread protein content and nutritional quality of wheat flour utilized in bread production when mushroom powder was added to the wheat. Incorporation of mushroom

flour in the preparation of noodles will impart additional health benefits to it. Therefore, the purpose of this study is to produce and evaluate qualitatively, instant noodles from wheat, rice and mushroom flour blends so as to enhance the utilization of mushroom in food processing and improve the nutritional quality of instant noodles via its protein and mineral content and solve nutritional imbalance such as micro-nutrient deficiency and malnutrition.

## MATERIAL AND METHOD

Durum wheat (Crown flour mill) flour and vegetable oil (Power oil) were purchased from Ogige market, Nsukka Enugu State, Nigeria. Parboiled seeds of Rice (*Oryza sativa*) were obtained from Adani Rice Mill in Uzo Uwani Local Government Area, Enugu State, Nigeria. Mushroom (*Agaricus bisporu*) was procured from Afor Enyiogugu market, Mbaise, Imo State, Nigeria. The production and analyses of the developed product was carried out in the food processing laboratory of the department of Food Science and Technology, University of Nigeria, Nsukka.

The mushroom (*Agaricus bisporu*) was processed by modifying the method described by Bello et al. (2017). Fresh mushrooms were cleaned, cut into slices (about 3 mm thickness) and dried for 6 hours at a temperature of 50°C in a hot air laboratory oven (LABE 1201, Divine International, and Delhi). Dried mushroom samples were milled and sifted through an 80 mesh screen to obtain fine powders. The obtained powder was cooled, hygienically packed and stored in air tight container for further use.

The Rice flour was processed by modifying the method described by Iwe et al. (2016). Parboiled rice grains were cleaned, sorted and washed, then steeped in water for 12 hours, drained and dried in a hot air laboratory oven (LABE 1201, Divine International, Delhi) at a temperature of 60°C for 30 min. It was finely milled using a rice milling machine (Angel bergs, miller Germany) and sieved with a sieve (mesh size 300 µm) to obtain fine flour which was designated as DM/PRF (Dried or Milled Parboiled Rice Flour).

### Formulation of blends

The flour obtained from rice and wheat was blended in different percentages as shown in Table 1 to produce noodles (procedure written in the production of instant noodles below). The noodles produced from the blended flour were subjected to sensory evaluation in order to obtain the best blend. The noodles were cooked for 10 min, cooled and presented to 20 semi-trained panelists from among students of the Department of Food Science and Technology, University of Nigeria, Nsukka, who are familiar with noodles to assess for appearance, flavor, taste, texture and general acceptability on a 9-point Hedonic scale as described by Ihekoronye and Ngoddy (1985). The samples were presented in coded plastic plates. The order of presentation of samples to the judges was randomized. Clean water was supplied for the panelists to rinse

their mouths between evaluations. Noodle from the composite flour was compared with 100 % wheat noodle. Based on the sensory evaluation of the noodles, the blend of wheat and rice flour (90:10) was chosen as the best blend. The best blend was then blended with mushroom flour as shown in Table 2 to produce the final product.

### Production of instant noodles

The noodles were produced using the method described by Hou (2001). Four hundred grams (400 g) of each flour blend was mixed differently with 140 ml of water and 4 g of salt and kneaded until the flour forms dough sheets of about 3 mm thickness. The crumbly dough obtained was rested for 30 minutes to mature and then kneaded to uniformly distribute the ingredients and hydrate all the flour particles. The dough was then passed through rotating rollers of a hand operating extruder (LAMI LM-20, China) to

**Table 1.** Blending ratios of wheat and rice flour

Sample code	WF (%)	RF (%)
WF + RF <sub>0</sub> (100:0)	100	0
WF + RF <sub>00</sub> (0:100)	0	100
WF + RF <sub>1</sub> (90:10)	90	10
WF + RF <sub>2</sub> (80:20)	80	20
WF + RF <sub>3</sub> (70:30)	70	30
WF + RF <sub>4</sub> (60:40)	60	40
WF + RF <sub>5</sub> (50:50)	50	50

**Key:** WF + RF<sub>0</sub> = 100% wheat flour and 0% rice flour; WF + RF<sub>00</sub> = 0% wheat flour and 100% rice flour; WF + RF<sub>1</sub> = 90% of wheat flour and 10% rice flour; WF + RF<sub>2</sub> = 80% wheat flour and 20% rice flour; WF + RF<sub>3</sub> = 70% wheat flour and 30% rice flour; WF + RF<sub>4</sub> = 60% wheat flour and 40% rice flour; WF + RF<sub>5</sub> = 50% wheat flour and 50% rice flour

**Table 2.** Blending ratios of wheat/ rice flour and mushroom flour

Sample code	Wheat/ local rice flour blend (90:10 best blend) (%)	Mushroom flour (%)
WR/MF <sub>0</sub> (100:0)	100	0
WR/MF <sub>1</sub> (90:10)	90	10
WR/MF <sub>2</sub> (80:20)	80	20
WR/MF <sub>3</sub> (70:30)	70	30
WR/MF <sub>4</sub> (60:40)	60	40
WR/MF <sub>5</sub> (50:50)	50	50

**Key:** WR/MF<sub>0</sub> = 100 % of the best blend flour and 0% of mushroom flour; WR/ MF<sub>1</sub> = 90 % of the best blend flour and 10 % of mushroom flour; WR/MF<sub>2</sub> = 80 % of the best blend flour and 20 % of mushroom flour; WR/MF<sub>3</sub> (70:30) = 70 % of the best blend flour and 30 % of mushroom flour; WR/MF<sub>4</sub> = 60 % of the best blend and 40 % of mushroom flour; WR/MF<sub>5</sub> = 50 % of the best blend and 50 % of mushroom flour



produce a noodle sheet. The sheet was repeatedly folded and passed through the rollers to facilitate gluten development, which gives the noodles its stringy and chewy texture. The gap between the finishing rolls was adjusted to produce the desired thickness of the noodle belt that was then immediately cut in the cutting section of the same machine. The noodles were steamed at 98-100°C for 1-5 minutes, which gelatinizes the starch and improves the texture of the noodles, after which the noodles were dried by frying in oil at 135-145°C for 2 minutes. Small quantities of the developed products/flour in grams (0.1g, 1g, 2g, 5g, etc.) were used for analysis.

#### **Analysis of raw materials and noodles produced from wheat, local rice and mushroom**

The flours were analyzed for their proximate, functional properties and micro-nutrients while the noodles were analyzed for their proximate composition, cooking characteristics, microbial analysis for one month at seven (7) days interval, sensory attribute and micronutrients.

#### **Determination of proximate composition**

The flours and the developed instant noodles were analyzed for their moisture content, crude fiber, ash, protein and fat content using AOAC (2010) method while carbohydrate content was determined by their difference.

#### **Determination of selected functional properties of flour blends**

The methods described by Onwuka (2018) were used in the determination of the functional properties of the flour blends. The functional properties determined included the water and oil absorption capacities, bulk density and swelling capacity.

#### **Determination of water and oil absorption capacities**

The water absorption capacity of the flours was determined as follows. One (1) gram of sample was mixed with 10 mL of distilled water using a warring whirl mixer. The sample was allowed to stand at ambient temperature (30±2 °C) for 30 seconds and then centrifuged for 30 minutes at 5,000 rpm × g. Water absorption

was examined as per cent water bound per gram flour. For oil absorption capacity, one gram of sample was mixed with 10 mL soybean oil (Sp. Gravity: 0.9092) and allowed to stand at ambient temperature (30±2°C) for 30 minutes and then centrifuged for 30 min at 5,000 rpm × g. Oil absorption was examined as percent oil bound per gram flour (Onwuka, 2018).

Absorbed water/ oil = (total water/oil – free water/oil) × density of water/oil

#### **Determination of bulk density**

The bulk density was determined as follows. A graduated measuring cylinder of 10 ml capacity was weighed and gently filled with the sample, followed by gently tapping the bottom until there was no further diminution of the sample level after filling to the 10 ml mark (Onwuka, 2018).

The bulk density was calculated as:

Bulk density (g/ml) = Weight of sample (g)/  
Volume of sample (ml)

#### **Determination of swelling capacity**

The swelling capacity was determined by modifying the method of Prescott et al. (2005). The flour sample (0.1 g) was weighed into a test tube and 10 ml of distilled water added. The mixture was heated in a water bath at a temperature of 50°C for 30 minutes with continuous shaking. In the end, the test tube was centrifuged at 1500 rpm for 20 minutes in order to facilitate the removal of the supernatant which was carefully decanted and the weight of the starch paste taken. This was carried out over a temperature range of 50 – 100°C. The swelling power was calculated as follows:

$$\text{Swelling power} = \frac{\text{weight of starch paste}}{\text{weight of dry starch sample}}$$

#### **Determination of micronutrients, Determination of vitamins, Determination of vitamin B<sub>1</sub> (Thiamine)**

Thiamin was determined using AOAC (2010) procedure. Seventy five milliliter (75 ml) of 0.2 N HCl was added to 2 g of sample and the mixture boiled over a water bath (Stuart; RE300B, UK).



After cooling, 5 ml of phosphatase enzyme solution was then added and the mixture incubated at 37°C overnight. The solution was placed in a 100 ml volumetric flask and the volume made up with distilled H<sub>2</sub>O. The solution was filtered and the filtrate purified by passing through silicate column. Twenty-five (25) ml of the filtrate was put in a conical flask and 5 ml of acidic KCl exudate, 3 ml of alkaline ferricyanide solution, and 15 ml isobutanol were added, and shaken for 2 min. The solution was then allowed to separate and the alcohol layer taken. About 3 g of anhydrous sodium sulphate was added to the alcohol layer. A 5 ml of thiamine solution was accurately measured into another 50 ml stoppered flask. The oxidation and extraction of thiochrome as already carried out with the sample was repeated using thiamine solution. A 3 ml of 15 % NaOH was added to the blank instead of alkaline ferricyanide. The blank sample solution was poured into fluorescence reading tube and reading taken: Thiamine was calculated as follows:

$$\% \text{ thiamine} = \frac{X}{Y} \times \frac{1}{5} \times \frac{25}{V} \times \frac{100}{W}$$

Where W= weight of sample; X = reading of sample- reading of blank; Y = reading of thiamine standard – reading of blank standard; V = volume of solution used for test on the column.

#### Determination of vitamin B<sub>2</sub> (Riboflavin)

The method of AOAC (2010) was used. Two grams (2 g) of the food material was taken in a conical flask. Fifty milliliter (50 ml) of 0.2N HCl was added and boiled in a water bath (Stuart; RE300B, UK) for 1h. The solution was cooled and the pH adjusted to 6.0 using NaOH about 1 N HCl was added to lower pH (METER TOLED. Seven-Multi. pH MV/ORP-MTW 1.49/01.38. Schwerzenback) to 4.5, then filtered in a 100 ml measuring flask and used to make volume up to mark. To remove interference, two tubes was taken and labeled 1 and 2 for tube 1 and 10 ml of filtration, and 1 ml of water for tube 2 respectively. Then, 10 ml of filtrate was added to 1ml of riboflavin standard. One milliliter (1 ml) of acetic acid (glacial) was added to each test tube, mixed and then 0.5 ml of 3% KMnO<sub>4</sub> solution was added. The solution was kept away

for 2 min and then 0.5 ml of 0.3 % H<sub>2</sub>O<sub>2</sub> added and mixed well. The flourimeter was adjusted to excitation wavelength of 470 nm and emission wavelength of 525 nm. Also, the flourimeter was adjusted to zero deflection against 0.1 N H<sub>2</sub>SO<sub>4</sub> and 100 against tube 2. The fluorescence tube was measured. Twenty milligram (20 g) of sodium hydrogen sulphate was added to both tubes and fluorescence measured within 10 seconds and recorded as blank readings.

#### Calculation:

Wt = weight sample

X = (reading of sample 1) – (reading of sample blank)

Y = (reading of sample + standard tube 2) – (reading of sample + standard blank)

Riboflavin (mg per sample) = X/Y-X x 1/wt

#### Determination of vitamin B<sub>3</sub> (Niacin)

Five grams (5 g) of the sample was treated with 50 ml of 1 N sulphuric acid for 30 minutes. 0.5 ml ammonia solution was added to it and it was then filtered. To 10 ml of the filtrate, 5 ml of 0.5 % potassium cyanide was added. This was further acidified with 5 ml of 0.02 N sulphuric acids. The absorbance of the resultant solution was recorded at 420 nm. The absorbance obtained from the sample extract was converted to Niacin concentration by means of a calibration curve generated using different standard concentrations (AOAC, 2010).

#### Determination of minerals

Mineral analysis was determined using the method described by AOAC (2010). Two grams (2 g) of the sample were weighed and subjected to dry ashing for 5 hours in a well cleaned porcelain crucible at 550°C. The resultant ash was dissolved in 5ml of HNO<sub>3</sub>/HCl/H<sub>2</sub>O (1:2:3) and heated gently on a hot plate until brown fume disappears, remaining the mineral. Deionized water of 5 ml was added and heated until colorless solution is obtained. The solution in each crucible was filtered into 100 ml volumetric flask and the volume made up to 100 ml with the deionized water. The solution

was then used to analyze for magnesium, potassium and phosphorus. This was done using Atomic Absorption Spectrophotometer and the absorbance was read at maximum wavelength of absorbance of the respective minerals. The results were expressed as mg/100g.

### Cooking characteristics of noodles

Cooking quality of noodles is the most important aspect from the consumer's point of view, including optimal cooking time, swelling or water uptake during cooking, the texture of the cooked product, stickiness, aroma and taste. These cooking factors of noodles were related to the gelatinization rates and chemical composition of the noodles used. Cooking time, cooking quality, solid loss and water absorption were studied as per the methods described by American Association of Cereal Chemists (AACC, 2000).

### Optimum cooking time

The optimum cooking time of the noodles was evaluated according to the modified method of

Schoenlechner et al. (2010). One hundred grams of noodles was put into a beaker containing 1 L of boiling water (without salt addition). Every minute, some pieces were taken out and pressed between two glass plates (2.5 cm × 2.5 cm). The optimal cooking time (OCT) corresponded to the disappearance of the white center core.

### Determination of cooking yield

Cooking yield was determined according to the method of American Association of Cereal Chemists (AACC, 2000). Ten grams (10 g) of the noodles was added to a beaker containing about 10 ml boiling water. The beaker was covered and the noodles cooked for 10 minutes. The cooked noodles were drained and then weighed. The cooking yield was calculated using the equation:

$$\text{Cooking yield (\%)} = \frac{\text{weight of noodles after cooking} - \text{weight of noodles before cooking}}{\text{weight of noodles before cooking}} \times 100$$

$$\text{Cooking loss (\%)} = \frac{\text{weight of gruel and dish} - \text{weight after drying}}{\text{weight after drying}} \times 100$$

### Determination of cooking loss

The cooking loss was determined according to the method of American Association of Cereal Chemists (AACC, 2000). The gruel obtained after cooking the noodles was poured into a 200 ml volumetric flask and adjusted to volume with distilled water. Ten milliliter of the solution was pipetted into an aluminum dish and dried to a constant weight at 10°C. The cooking loss was calculated as follows:

### Microbial analysis

The microbial analysis was determined using the method of Prescott et al. (2005).

### Media preparation

Nutrient Agar powder (7 g) was dissolved in distilled water (250 ml). Thirteen grams (13 g) of sabourand Dextrose Agar (SDA) was dissolved in water (250 ml). The mixtures were stabilized by bringing them to boiling while homogenizing by shaking in whirl motion. The mixtures were sterilized by autoclaving for 15 minutes at the temperature of 121°C. The ringer solution was allowed to cool after sterilization to about 40-47°C.

### Ringer solution preparation

One ringer tablet was dissolved in distilled water (500 ml). The clear solution formed was sterilized by autoclaving for 15 minutes at the temperature of 121°C. The ringer solution was allowed to cool completely to a temperature of 28°C.

### Determination of total viable count

The total viable count was determined by the method of Pour Plate Count. The method involved weighing the sample (1 g) into a sterile test tube. A  $\frac{1}{4}$  strength Ringers solution (9 ml) was poured into it and also into other test tube arranged for serial dilution. The sample with

the solution was homogenized by shaking. The sample with solution was pipetted (1 ml) into test tube containing Ringers solution (9 ml).

Then, 1 ml of different dilution factor was transferred into the sterile petri dishes and sterile nutrient agar was poured into the same petri dish and was mixed by rocking. When they solidified, they were turned upside down and cultured by incubation for 24 h at temperature of 37°C. At the end of the incubation period, the colonies were counted using the colony counter (Gallenkamp colony counter, CWN 330- 010X) and the number of colonies recorded appropriately.

#### **Determination of mold count**

The mold count was determined using Sabouraud Dextrose Agar (SDA) as the plating medium. The sample (1 g) was weighed and put in a test tube prepared for serial dilution. The ringer solution (9 ml) was aseptically transferred serially into other test tubes. Serial dilution of  $10^{-1}$  was used for mould count determination. Appropriate diluent (1 ml) was transferred into the sterile petri dishes. Sabouraud Dextrose Agar was used for culturing the organism for 48 hours at room temperature. The mold colonies were enumerated and calculated as colony forming units (cfu)/g of the sample.

$Cfu/g = \text{Number of colonies} \times \text{reciprocal of dilution factor}$

#### **Sensory evaluation**

The noodles were cooked and assessed by a 20-man semi-trained panel selected randomly from among students of the Department of Food Science and Technology, University of Nigeria, Nsukka. The samples were evaluated for colour, appearance, mouth feel, aroma, taste, texture and general acceptability on a 9-point Hedonic scale ranging from 1-9 where 1:dislike extremely and 9: like extremely as described by Ihekoronye and Ngoddy (1985). The samples were presented in coded plates. The order of presentation of samples was randomized. Water was served to the panelist to rinse their mouths in-between sample evaluation.

#### **Data analysis and experimental design**

The experimental design that was used is Completely Randomized Design and the mean values were subjected to analysis of variance (ANOVA) using Duncan's Multiple Range Test (DMRT) and SPSS (Statistical Product for Service Solution) version 20 computer was used. Significance was accepted at  $p < 0.05$  (Steel and Torrie, 1980).

## **RESULTS AND DISCUSSION**

#### **Proximate Composition of Wheat, Rice and Mushroom Flours**

The proximate composition (%) of wheat, rice and mushroom flours used in the instant noodles production is as shown in Table 3. There were significant ( $p < 0.05$ ) differences among the flour samples in their protein, ash, fat, moisture and carbohydrate content.

The results for the protein content showed that the composite flour had significantly ( $p < 0.05$ ) higher protein content than the value for wheat flour. The protein content of the mushroom flour had the highest (20.88%), while the wheat flour had the least protein content (8.17%). This makes mushroom flour a good protein supplement. The protein content compared well with 11.91-22.60 % obtained by (Nwagu and Obiakor-okeke, 2014) in the analysis of nutritional profile of three different mushroom varieties A (white button mushroom), B (oyster mushroom) and C (crimini mushroom) consumed in Amaifeke, Orlu LGA, Imo State. It was reported that, with the exception of green peas and pulses, fresh mushrooms had higher protein content than most vegetables (Bora and Kawatra, 2014).

There was no significant ( $p > 0.05$ ) difference between samples RF (rice flour) and WF (wheat flour) in their ash content, while sample MF (mushroom flour) differed significantly ( $p < 0.05$ ) from the two flour samples. The high ash content of mushroom flour suggests that incorporation of mushroom flour will increase the mineral content of the blends. Ash content represents the total mineral content in foods. It is essential to determine the full nutritional value, quality and stability of the food. Higher ash content will

contribute to the flavor and nutrient quality of the product. The rice flour had the lowest ash content (0.5%).

The fat content of the rice flour had the highest value (1.79%), while the wheat flour had the least value (1.15%). The fat content of mushroom flour is low (1.25 %) compared to (2.0 %) obtained by Shah et al. (1997). Crude fat content of mushrooms is usually low, and is from 1.0 % to 6.7 % for certain species collected in China (Xue-mei et al., 2013). According to Liu et al. (2010), the % of total fatty acids for stearic, palmitic, linoleic and oleic acid in *Tricholoma matsutake* was 2%, 9%, 27% and 58%, respectively. Ratio of unsaturated to saturated (UFA/SFA) fatty acids is an important measure to judge stand or fall of fatty acid in mushrooms (Zhang and Ran, 2005).

The mushroom flour ranked highest in the fiber content (9.97%). The value was significantly ( $p < 0.05$ ) different from the other flours. The fiber contents of the mushrooms are reasonably high, suggesting that the mushrooms would be valuable in improving human health by quickening the excretion of wastes and toxins from the body. Crude fibre is a group of indigestible carbohydrates. It can improve the function of the alimentary tract and also lower blood glucose and cholesterol levels. The values obtained are similar to those obtained by (Nwagu and Obiakor-okeke, 2014). They ranged from 7.94 to 18.63% in the analysis of nutritional profile of three different mushroom varieties consumed in Amaifeke, Orlu LGA, Imo State.

Cereals are usually high in carbohydrate. From the findings, rice flour had the highest value (85.90 %) followed by the wheat flour (77.03%) and mushroom flour had the least carbohydrate value (47.15%). The moisture content of the flours

varied from 2.70- 11.92%. Wheat flour (11.92%), rice flour (2.70%) and mushroom flour (10.58%), respectively. All the flour blends had moisture content below 13% hence less susceptible to spoilage and prolonged shelf-life of the flours was assured. The moisture content of the wheat flour was below the maximum moisture content limit of 15.5% stipulated by FAO, (2006). The moisture content of the rice flour 2.70% was below the 10% stipulated standard for dry food products (SON, 1988). There were significant ( $p < 0.05$ ) differences among the flour samples.

### Micronutrient composition of wheat, rice and mushroom flours.

#### Vitamins

Table 4 shows the vitamin content of the wheat, rice and mushroom flours. There was no significant ( $p > 0.05$ ) difference between samples RF (rice flour) and MF (mushroom flour) in their vitamin content, whereas sample WF (wheat flour) differed significantly ( $p < 0.05$ ) from the two samples.

Mushrooms contain several primary vitamins including thiamine, riboflavin, niacin, tocopherol and vitamin D (Kalac, 2013). For other researchers using several species, the content of thiamine, riboflavin, niacin and ascorbic varied from 0.02–1.6, 0.3–4.5, 1.2–6.6 and 1.3–2.7 mg 100 g/1 dm, respectively (Quan et al., 2007; Wu et al., 2005; Xu et al., 2012; Zhou and Yin, 2008; Zhu et al., 2007). The values for vitamin B<sub>1</sub> (0.25 mg/100g), B<sub>2</sub> (0.07 mg/100g) and B<sub>3</sub> (1.26 mg/100g) obtained from this study were in line with the report. The B-complex vitamins, especially thiamin, riboflavin and niacin offered by natural brown rice promote youthful energy and nourishment to skin and blood vessels (Lloyd et al., 2000).

**Table 3.** Proximate composition of wheat, rice and mushroom flour

Sample	Protein (%)	Ash (%)	Fat (%)	Fiber (%)	Moisture (%)	Carbohydrate(%)
WF	8.17 <sup>a</sup> ± 0.02	1.29 <sup>a</sup> ± 0.73	1.15 <sup>a</sup> ± 0.03	0.96 <sup>b</sup> ± 0.02	11.92 <sup>c</sup> ± 0.02	77.03 <sup>c</sup> ± 0.04
RF	8.53 <sup>b</sup> ± 0.01	0.50 <sup>a</sup> ± 0.14	1.79 <sup>b</sup> ± 0.01	0.60 <sup>a</sup> ± 0.28	2.70 <sup>a</sup> ± 0.02	85.90 <sup>b</sup> ± 0.03
MF	20.88 <sup>c</sup> ± 0.03	10.15 <sup>b</sup> ± 0.01	1.25 <sup>c</sup> ± 0.01	9.97 <sup>b</sup> ± 0.02	10.58 <sup>b</sup> ± 0.02	47.15 <sup>a</sup> ± 0.01

Values are means ± standard deviation of duplicate replications. Means within a column with the same superscript were not significantly ( $p < 0.05$ ) different **Key:** WF= Wheat flour; RF= Rice flour; MF= Mushroom flour



### Mineral content of wheat, rice and mushroom flours

Table 5 shows the mineral content of the wheat, rice and mushroom flours. There was a significant ( $p < 0.05$ ) difference among the flour samples in their mineral content.

The mostly occurring microelements in mushroom are iron and potassium with phosphorus in significant quantity. Potassium is very important in the maintenance of osmotic balance between cells and the interstitial fluid in animal systems. The potassium content for the different flour samples was highest in mushroom flour (22.90 mg/100g). This value was higher

than the values obtained for the specie *C. ventricosum* (2.7 mg/100g) by Liu et al. (2012) although it was lower than that for the range of values reported by Afiukwa et al. (2013), which was 221.13 mg/100g. Iron, which is essential for the biosynthesis of the oxygen-carrying pigment of red blood cells and the cytochromes that function in cellular respiration, is also present in good amounts in the mushrooms (Wani et al., 2010). The iron content in the mushroom flour is higher than those in rice and wheat flour (4.77 mg/100g). The mushroom flour iron content was lower than that obtained for the specie *C. ventricosum* (6.73 mg/100g) as reported by Liu et

al. (2012). The lower value obtained could be as a result of the differences in species. Low levels of phosphorus were observed in three flour samples. The mushroom flour had the highest value (0.18 mg/100g) and wheat flour (0.05 mg/100g) with the least value.

### Functional properties of flour blends from wheat, rice and mushroom

The functional properties of flour blends of wheat, rice and mushroom flours are as shown in Table 6.

Water absorption capacity represents the ability of a product to associate with water under conditions where water is limited (Singh, 2001). Percent water absorption varied from 100% to 250% with the blend with no mushroom flour (100:0) having the least value and blend with 50% mushroom powder having the highest value. Water absorption capacity is a critical function of protein in various food products like soups, dough and baked products (Adeyeye and Aye, 1998). Water absorption capacity increased significantly ( $p < 0.05$ ) with increasing levels of mushroom flour. This could probably be due to the high protein and fiber content of mushroom flour. Kaur et al. (2013) also observed significant increase in water absorption on addition of more than 8% mushroom powder to semolina for

**Table 4.** Vitamin content (mg/100g) of wheat, rice and mushroom flours

Sample	vitamin B <sub>1</sub> (mg/100 g)	vitamin B <sub>2</sub> (mg/100 g)	Vitamin B <sub>3</sub> (mg/100 g)
WF	0.25 <sup>a</sup> ± 0.04	0.07 <sup>a</sup> ±0.02	1.26 <sup>a</sup> ±0.02
RF	0.36 <sup>b</sup> ±0.02	0.05 <sup>a</sup> ±0.02	2.50 <sup>b</sup> ± 0.14
MF	0.28 <sup>ab</sup> ±0.02	0.25 <sup>b</sup> ±0.02	3.00 <sup>b</sup> ±0.28

Values are means ± standard deviation of duplicate replications. Means within a column with the same superscript were not significantly ( $p < 0.05$ ) different. **Key:** WF= Wheat flour; RF= Rice flour; MF= Mushroom flour.

**Table 5.** Mineral content of wheat, rice and mushroom flours

Sample	Iron (mg/100g)	Potassium (mg/100g)	Phosphorus (mg/100g)
WF	1.10 <sup>a</sup> ± 0.01	1.78 <sup>a</sup> ±0.02	0.05 <sup>a</sup> ±0.02
RF	1.10 <sup>a</sup> ±0.04	4.18 <sup>a</sup> ±0.01	0.11 <sup>ab</sup> ±0.02
MF	4.77 <sup>b</sup> ±0.02	22.90 <sup>b</sup> ±0.02	0.18 <sup>b</sup> ±0.02

Values are means ± SD of duplicate replications. Means within a column with the same superscript were not significantly ( $p < 0.05$ ) different. **Key:** WF= Wheat flour; RF= Rice flour; MF= Mushroom flour.



pasta extrusion.

The highest value of oil absorption capacity was observed for flour blend of wheat and rice flour without mushroom flour (100:0) 372% and the lowest value (195.3%) for the 90% composite wheat-rice flour and 10% mushroom flour. Increase in the addition of the mushroom flour significantly ( $p < 0.05$ ) increased the oil absorption capacity. The increment of OAC was due to the high protein contents of the mushroom flour as it increased in the composite flour. This might be due to the presence of protein exposing more non-polar amino acids to the fat and enhancing hydrophobicity; as a result of this, flour absorbs more oil (Oluwamukomi et al., 2011). The water and oil binding capacity of food protein depends upon the intrinsic factors like amino acid composition, protein conformation and surface polarity or hydrophobicity (Kaushal, et al., 2012). The wheat and rice flour without mushroom flour (100:0) blend which had the highest oil absorption capacity could therefore, be a better flavor retainer. The ability of the proteins of these flours to bind with oil makes it useful in food system where optimum oil absorption is desired. This makes flour to have potential functional uses in foods such as sausage production. The oil absorption capacity also makes the flour suitable in facilitating enhancement in flavor and mouth feel when used in food preparation. Due to these properties, the protein probably could be used as functional ingredient in foods such as whipped toppings, sausages, chiffon dessert, angel and sponge cakes among other products.

The swelling capacity was found highest for 50% composite wheat-rice flour with 50% mushroom flour blend (28.0 ml) and lowest for 100 % composite wheat-rice flour blend without mushroom flour (100:0) (15.0 ml). The swelling capacity of flours depends on size of particles, types of variety and types of processing methods or unit operations.

According to Gustavo et al. (2005), the bulk density of food powders depends on the combined effect of interrelated factors such as the intensity of attractive inter-particles forces, particle size, and number of contact points. It is clear that a change in any of the powder characteristics might result in a significant ( $p < 0.05$ ) change in the powder bulk density. Bulk density is an indication of the porosity of a product which influences package design and could be used to determine the type of packaging material required. Low bulk density is desirable and required for infant foods (Omerie et al., 2015).

#### Proximate composition (%) of instant noodles formulated from wheat, rice and mushroom flour blends

Table 7 shows the proximate composition of the formulated instant noodles from wheat, rice and mushroom flour blends. Variations exist in the proximate composition of noodles from wheat and mushroom flour.

The protein content of the samples ranged from 9.49-15.39%. Sample WRM5 (50:50) had the highest protein content and sample WR0 (100:0)

**Table 6.** Functional properties of flour blends from wheat, rice and mushroom flours

Sample	Bulk density (g/cm <sup>3</sup> )	Water absorption (%)	Oil absorption (%)	Swelling capacity (ml)
A (100:0)	0.71 <sup>c</sup> ± 0.02	100 <sup>a</sup> ± 0.28	372 <sup>c</sup> ± 0.00	15.0 <sup>a</sup> ± 0.14
B (90:10)	0.69 <sup>c,d</sup> ± 0.01	120 <sup>b</sup> ± 0.00	195.3 <sup>a</sup> ± 0.28	20.0 <sup>b</sup> ± 1.44
C (80:20)	0.67 <sup>c,d</sup> ± 0.02	150 <sup>c</sup> ± 0.14	260 <sup>b</sup> ± 0.28	22.0 <sup>c</sup> ± 0.00
D (70:30)	0.63 <sup>b,c</sup> ± 0.02	200 <sup>d</sup> ± 0.00	269 <sup>c</sup> ± 0.28	25.0 <sup>d</sup> ± 0.28
E (60:40)	0.59 <sup>b</sup> ± 0.01	210 <sup>c</sup> ± 0.14	279 <sup>d</sup> ± 0.00	26.0 <sup>d</sup> ± 0.00
F (50:50)	0.53 <sup>a</sup> ± 0.02	250 <sup>f</sup> ± 1.97	279 <sup>d</sup> ± 0.56	28.0 <sup>e</sup> ± 0.14

Values are means ± standard deviation of duplicate replications. Means within a column with the same superscript were not significantly ( $p < 0.05$ ) different. **Key:** A (100:0): 90% wheat and 10 % rice flour + 0 % mushroom flour; B (90:10): 90% wheat and rice flour + 10 % mushroom flour; C (80: 20): 80% wheat and rice flour + 20 % mushroom flour; D (70:30): 70% wheat and local rice flour + 30 % mushroom flour; E (60:40): 60% wheat and local rice flour + 40 % mushroom flour; F (50:50): 50% wheat and local rice flour + 50 % mushroom flour.

had the least protein value. Substitution of wheat flour with an increasing quantity of mushroom flour increased the protein content of the noodles. The low protein contents of sample WR0 (100:0) is due to incorporation of more quantity of wheat flour and absence of mushroom flour in the blends. Mushroom protein includes all nine essential amino acids required by humans, although it can be limiting in sulphur-containing amino acids, such as cystine and methionine. In terms of the essential amino acid index, amino acid score and nutritional index, mushrooms are between low grade vegetable and high grade meats with values that are close to that of milk, some species even well above milk which is an animal product (Bora and Kawatra, 2014). As a result of this, FAO has recommended mushrooms as a supplementary food item in the context of the world protein shortage for the growing populations of the developing countries. The protein content was within the range (10.54-14.34%) obtained by Bindvi et al. (2017) in instant noodles supplemented with oyster mushroom (*P. ostreatus*). The high protein content of the noodles from most blends is significant in curbing protein-energy malnutrition which mostly occurs in regions that their staple food is low in protein but rich in carbohydrates.

There were significant ( $p < 0.05$ ) differences among the ash content of noodles from the blends. The ash content ranged from 1.39 to 5.31% with sample WRM5 (50:50) having the highest ash content while the CTRL (commercial sample) having the least ash content. The high ash content of the instant noodles might be attributed to the fact that mushroom has been reported to be a good source of minerals as there was a notable increase in the ash content with addition of mushroom. The ash content of a food material could be used as an index of minerals constituents of the food which is necessary for growth and development. The values obtained were higher than those reported by Bindvi et al. (2017) which ranged from 1.84 to 1.76%. This may be attributed to the nutritional varieties in the different species.

The crude fiber content of the samples ranged from 1.50 to 5.40%. Sample WRM5 (50:50) ranked

highest and CTRL (commercial product) the lowest value. Fiber content was more in noodles from the composite flour compared to the control and the difference was significant at  $p < 0.05$ . There was no significant ( $p > 0.05$ ) difference between samples WRM4 (60:40) and WRM5 (50:50). Increase in the incorporation of mushroom flour significantly increased the crude fiber content of the instant noodles. The high values obtained might be due to the fact that mushroom used in instant noodles production were rich sources of dietary fibre which therefore increased the fibre content of the product with increase in level of the composite flour incorporation. A high intake of dietary fibre is positively related to different physiological and metabolic effects (NRC, 1989). Fibre prevents constipation. Soluble fibre helps to reduce the cholesterol level in the blood, slows down digestion and sudden release of energy, thus making blood level stable. Food containing at least 3 g/100g dietary fibers (DF) can be referred to as a source of DF. It is high in DF when it contains at least 6 g/100g dietary fibers (FAO, 1995). The consumption of this product will meet the WHO recommendation for dietary fibre intake of about 25 g per day (Sandstead, 1995).

The fat content ranged from 0.5 to 1.50% with sample WR0 (100:0) which had the highest fat content and sample WRM5 (50:50) had the lowest value. Fat content decreased slightly with the addition of mushroom powder. However there was no significant ( $p > 0.05$ ) difference among samples WRM1 (90:10), WRM2 (80:20) and WRM3 (70:30). Fat plays a role in determining the shelf-life of foods (Brons et al., 2008). Crude fat content of mushrooms is usually low. It ranged from 1.0% to 6.7% for certain species collected in China as reported by Xue-mei et al. (2013). A high amount of fat could accelerate spoilage by promoting rancidity which could lead to the production of off flavors and odours. Also diet high in fat predispose consumer to different illness such as obesity, heart disease among other ailments. The values obtained from this study were comparable to the values reported by Yin and Zhou (2008) in Yunnan wild edible *Boletus*. Pedneault et al. (2006) reported that fat fraction in mushrooms is mainly composed of

unsaturated fatty acids. 1.0 %.

The moisture content ranged from 7.92 to 14.48 % with sample CTRL (commercial product) having the lowest value and WRM5 (50:50) the least value. The samples varied significantly ( $p < 0.05$ .) The moisture content of fresh mushrooms is about 70 to 95%, depending on the species, harvest time and environmental conditions, it falls to around 10 to 13% when dried (Breene, 1990). Moisture content is an index of the shelf-stability of food because microorganisms require moisture for their deteriorative activities within the food.

The carbohydrate values obtained ranged from 58.42 to 77.45% and the CTRL (commercial product) with the highest value, WRM5 with

50% mushroom powder incorporation the least value. The carbohydrate value reduced slightly with increase in mushroom flour incorporation. The samples varied significantly ( $p < 0.05$ ). The carbohydrate values were comparable to the values (74%) obtained by (Carneiro et al., 2013) and the range (58.11 to 61.98%) obtained by Bello et al. (2017). Carbohydrates in foods provide energy and digestible carbohydrates found in mushrooms are such as mannitol (0.3–5.5% dm) (Vaz et al., 2011), glucose (0.5 to 3.6% dm) (Kim et al., 2009) and glycogen (1.0–1.6 % dm), Diez and Alvarez (2001). Non-digestible carbohydrates form a large portion of the total carbohydrates of mushrooms, and major compounds are

oligosaccharides and non-starch polysaccharides such as chitin,  $\beta$ -glucans and mannans (Cheung, 2010). Polysaccharides are the best known and most potent mushroom derived substances with antitumor and immunomodulation properties. The results indicate that noodles from composite especially with a higher content of mushroom flour have enhanced nutrients than those of 100% wheat flour and may benefit consumers more than the control.

### Microbial counts of instant noodles from wheat, rice and mushroom flour blends

Table 8 shows the total viable and mold count of instant noodles from blends of wheat, rice and mushroom flours for four weeks' storage. Total viable counts are one of the microbial tests carried out to ensure safety of food products. It is used to determine hygienic conditions during food processing for both raw materials and equipment. It is used to evaluate effectiveness of processing methods, heat treatment and storage conditions for products. Total viable count gives a quantitative idea about the presence of microorganisms in the sample. The total viable count of the noodles samples for the first week ranged from  $1.0 \times 10^3$  cfu/g to  $8.3 \times 10^2$  cfu/g. Sample WRM1 (90:10) had the highest value while sample WRM2 (80:20) had the lowest value. For the second week TVC ranged from  $1.2 \times 10^4$  cfu/g to  $9.2 \times 10^3$  cfu/g and sample WRM2 (80:20) had the highest value and sample WRM3 (70:30) had the lowest value. Total viable count

**Table 7.** Proximate composition (%) of instant noodles from wheat, rice and mushroom flour blends

Sample	Protein (%)	Ash (%)	Fat (%)	Fiber (%)	Moisture (%)	Carbohydrate (%)
CTRL	10.84 <sup>a</sup> ± 0.01	1.39 <sup>a</sup> ± 0.04	0.90 <sup>b</sup> ± 0.01	1.50 <sup>a</sup> ± 0.05	7.92 <sup>a</sup> ± 0.02	77.45 <sup>e</sup> ± 0.02
WR0	9.49 <sup>b</sup> ± 0.01	1.66 <sup>b</sup> ± 0.02	1.50 <sup>f</sup> ± 0.01	1.79 <sup>b</sup> ± 0.28	12.21 <sup>e</sup> ± 0.02	73.35 <sup>f</sup> ± 0.00
WRM1	10.96 <sup>c</sup> ± 0.01	2.19 <sup>c</sup> ± 0.01	1.40 <sup>ef</sup> ± 0.01	2.30 <sup>c</sup> ± 0.14	8.28 <sup>b</sup> ± 0.02	74.87 <sup>e</sup> ± 0.00
WRM2	12.77 <sup>d</sup> ± 0.02	3.34 <sup>d</sup> ± 0.02	1.30 <sup>de</sup> ± 0.00	3.38 <sup>d</sup> ± 0.28	9.73 <sup>c</sup> ± 0.00	69.48 <sup>d</sup> ± 0.02
WRM3	13.21 <sup>e</sup> ± 0.01	4.47 <sup>e</sup> ± 0.01	1.20 <sup>cd</sup> ± 0.01	4.57 <sup>e</sup> ± 0.28	11.45 <sup>d</sup> ± 0.00	65.09 <sup>c</sup> ± 0.01
WRM4	13.59 <sup>f</sup> ± 0.01	5.30 <sup>f</sup> ± 0.01	1.10 <sup>c</sup> ± 0.15	5.39 <sup>f</sup> ± 0.01	12.18 <sup>c</sup> ± 0.05	62.49 <sup>b</sup> ± 0.01
WRM5	15.39 <sup>g</sup> ± 0.01	5.31 <sup>f</sup> ± 0.01	0.5 <sup>a</sup> ± 0.28	5.40 <sup>f</sup> ± 0.22	14.48 <sup>f</sup> ± 0.05	58.42 <sup>a</sup> ± 0.05

**Key:** CTRL=Control commercial sample; WR0 = 90 % wheat flour +10 % rice flour; WRM1= 90 % wheat/rice flour +10 % mushroom flour; WRM2 = 80 % wheat/rice flour +20 % mushroom flour; WRM3 = 70 % wheat/rice flour +30 % mushroom flour; WRM4 = 60 % wheat/rice flour + 40 % mushroom flour; WRM5 = 50 % wheat/rice flour + 50 % mushroom flour. Values are means ± standard deviation of duplicate replications. Means within a column with the same superscript were not significantly ( $p < 0.05$ ) different.

for the third week ranged from  $1.0 \times 10^5$  cfu/g to  $8.1 \times 10^4$  cfu/g, sample WRM2 (80:20) had the highest value and sample WRM1 (90:10) had the least value. For the fourth week of storage, the TVC ranged from  $1.7 \times 10^5$  cfu/g to  $2.95 \times 10^5$  cfu/g. Sample WRM2 (80:20) had the highest value and CTRL (commercial product) with the least value. The total viable count of the instant noodles produced increased exponentially with storage time. From the results, it can be deduced that as the storage week progressed, there were visible growth of microorganisms found on the product. The products were produced in a hygienic and conducive environment and stored at room temperature. The favorable condition of storage and pH of the products as well as high nutrient contents of the products must have stimulated the initial increases since microorganisms are ubiquitous. Likewise, the variation in the microbial load of the instant noodles from the various blends might be attributed to the way the samples were handled differently before and during analysis. According to the International Commission on Microbiological Specification of Foods (ICMSF, 1996), total plate counts between zero to  $10^3$  is acceptable, between  $10^4$  to  $10^5$  is tolerable and  $10^6$  and above is unacceptable. From the storage studies of this research, the developed noodle samples maintained an acceptable microbial count during the storage period. Total mould count is a microbial test carried out to evaluate the presence of spoilage fungi on food samples which makes it unfit for consumption since it can lead to severe health complications. Mold count was done to detect and quantify mold in the samples. For the first week mold count was not detected expect in sample CTRL (commercial product)  $1.0 \times 10$ . For the second week mold count was detected at  $1.0 \times 10$  level for samples CTRL (commercial product), WR0 (100:0) and WRM3 (70:30). On the third week, CTRL (commercial product) has the highest mold count  $4.0 \times 10$  cfu/g, WR0 (100:0) and WRM3 (70:30)  $2.0 \times 10$  cfu/g. Sample CTRL (commercial product) has the highest mold count on the fourth week ( $9.0 \times 10$  cfu/g) and sample WRM3 (70:30) with the lowest value ( $3.0 \times 10$  cfu/g). The presence of low numbers from the 3<sup>rd</sup> week of storage suggests post production contamination, especially as the products were

loosely packaged in polyethylene bags. The mould load of the products was therefore, within the acceptable level for mould in instant noodles which is 10 cfu/g.

### **Cooking characteristics of instant noodles made from wheat, rice and mushroom flour blends**

Table 9 represents the cooking characteristics of instant noodles formulated from wheat, rice and mushroom flour blends. Cooking yield of the blend samples ranged from 200 to 140%, with sample WRM5 (50:50) having the highest value and WRM1 (90:10) the lowest value. There was no significant ( $p > 0.05$ ) difference in all the samples. Cooking yield is dependent on the ability of noodles to absorb water during cooking. With increased mushroom flour, the cooking yield increased. This trend could probably be because sample WRM5 (50:50) has high water absorption capacity compared to other samples. The cooking yield of the CTRL (commercial sample) was higher than the noodles from the blends which could be probably due to other additives used in production.

Cooking loss is indicated by the loss of solid materials contained in noodle during cooking. Mushroom-supplemented noodles showed a significant increase in cooking loss compared with the control noodle as the mushroom flour increased. The cooking loss of the noodles with 50 % mushroom flour (4.58 per 100 g) was higher than those observed for the control sample (3.80 g per 100 g). All the cooking loss samples were below 8 g per 100 g, the value above which pasta quality was considered unacceptable (Foschia et al., 2015). The higher cooking loss in noodles with added mushroom could be attributed to a loss of continuity of the noodles protein-starch matrix, as a consequence of the competitive hydration tendency of the fibre which leads to uneven distribution of water within the matrix (Tudoric et al., 2002). Similarly, Kaur et al. (2013) studied the effect of button mushrooms on the leaching of solids from pasta and reported that the solids that leached into the cooking water increased as the level of mushroom powder was increased in the blend.



Cooking time is very important to characterize a product as instant. Optimum cooking time of mushroom fortified noodles is given in Table 9. Optimum cooking time of the noodle samples varied from 5.20 to 6.50 min. The addition of mushroom powder in noodles progressively increased the cooking time, although there was no significant ( $p < 0.05$ ) difference between

samples even up to 20% level of fortification as wheat flour replacement. This could be attributed to increase in protein content of noodles with addition of mushroom powder, resulting in firmer product. Kaur et al. (2013) also observed increase in cooking time in mushroom supplemented pasta.

**Table 8.** Total viable and mold counts of instant noodles from wheat, rice and mushroom flour blends

Period of storage	Sample	Total viable count (cfu/g)	Mould count (cfu/g)
Week 1	CTRL	$1.78 \times 10^3$	ND
	WR0	$1.42 \times 10^3$	ND
	WRM1	$8.3 \times 10^2$	ND
	WRM2	$1.0 \times 10^3$	ND
	WRM3	$1.2 \times 10^3$	ND
	WRM4	$4.3 \times 10^2$	ND
	WRM5	$6.1 \times 10^2$	ND
Week 2	CTRL	$2.8 \times 10^4$	$1.0 \times 10$
	WR0	$1.3 \times 10^4$	$1.0 \times 10$
	WRM1	$9.2 \times 10^3$	ND
	WRM2	$7.2 \times 10^3$	ND
	WRM3	$1.2 \times 10^4$	$1.0 \times 10$
	WRM4	$3.3 \times 10^3$	ND
	WRM5	$7.7 \times 10^3$	ND
Week 3	CTRL	$1.9 \times 10^3$	$4.0 \times 10$
	WR0	$1.6 \times 10^5$	$2.0 \times 10$
	WRM1	$1.0 \times 10^5$	ND
	WRM2	$8.1 \times 10^4$	ND
	WRM3	$1.4 \times 10^5$	$2.0 \times 10$
	WRM4	$7.2 \times 10^4$	ND
	WRM5	$6.1 \times 10^4$	ND
Week 4	CTRL	$2.9 \times 10^5$	$9.0 \times 10$
	WR0	$2.7 \times 10^5$	$4.0 \times 10$
	WRM1	$2.1 \times 10^5$	ND
	WRM2	$1.7 \times 10^5$	ND
	WRM3	$2.5 \times 10^5$	$3.0 \times 10$
	WRM4	$1.5 \times 10^5$	ND
	WRM5	$2.4 \times 10^5$	ND

**Key:** CTRL = Control commercial sample; WR0 = 90 % wheat flour +10 % rice flour; WRM1 = 90 % wheat/rice flour +10 % mushroom flour; WRM2 = 80 % wheat/rice flour +20 % mushroom flour; WRM3 = 70 % wheat/rice flour +30 % mushroom flour; WRM4 = 60 % wheat/rice flour + 40 % mushroom flour; WRM5 = 50 % wheat/rice flour + 50 % mushroom flour

ND = Not detected



### Sensory properties of instant noodles formulated from wheat, rice and mushroom flour blends

Table 10 shows the average mean sensory scores of noodles samples from blends of wheat, rice and mushroom flour. The result in Table 10 shows that there were significant ( $p < 0.05$ ) difference in all the sensory attributes (colour, appearance, taste, aroma, texture, mouth feel, aftertaste and overall acceptability) of the instant noodles from wheat, rice and mushroom flour blends as well as the sample with 100 % wheat-rice flour blend and commercial noodles. The sample CTRL (commercial sample) and sample with 100 % wheat-rice flour blend (WR0) had the highest level of acceptance for the sensory attributes. However, the sensory acceptability of the noodles from blends of wheat, rice and mushroom flour blends are as follows.

Sample WRM1 (90:10) was rated high while noodle sample WRM5 (50:50) had the lowest level of preference. According to Hou (2001), flour colour, protein content, ash content, yellow pigment and polyphenol oxidase activity are important factors responsible for noodle colour. The preference for colour decreased with increasing amount of mushroom flour and decreasing amount of wheat flour. This could probably be due to the decrease in the brightness of the noodles colour of the mushroom flour. Noodle sample WRM5 (50:50)

did not differ significantly ( $p > 0.05$ ) from WRM4 (60:40).

The average mean scores for mouth feel ranged from 2.00-4.95. Sample WRM1 had the highest mouth feel while WRM5 had the lowest value. There is no significant ( $p > 0.05$ ) difference between the mean scores of the noodles WRM2 (80:20) and WRM3 (70:30) and between WRM5 (50:50) and WRM4 (60:40). However samples CTRL (commercial sample), WR0 (100:0) and WRM1 (90:10) differed significantly ( $p < 0.05$ ).

The aroma of the noodles from the blends ranged from 4.90-7.15 with sample WRM5 (50:50) having the highest value and sample WRM1 (90:10) the lowest value. Characteristic flavour substances of wild-grown mushrooms could be classified into nonvolatile (taste) and volatile components (smell). Various volatile compounds such as terpenes, aromatic alcohols, aldehydes, ketones, eight carbon compounds and their derivatives, are the major aroma compounds in mushrooms. Eight-carbon volatiles are produced by oxidation of free linoleic acid catalyzed by lipoxygenase (Cheng et al., 2012). With increase in the mushroom flour, there was increase in the aroma of the samples. The preference for aroma for samples WRM4 (60:40) and WRM5 (50:50) did not differ significantly ( $p > 0.05$ ) from WR0 (100:0). CTRL (commercial sample) differed significantly ( $p < 0.05$ ) from all the samples.

**Table 9.** Cooking characteristics of instant noodles formulated from wheat, rice and mushroom flour blends

Sample	Cooking yield (%)	Cooking loss (%)	Optimum cooking time (mins)
CTRL	220 <sup>a</sup> ± 0.14	3.80 <sup>a</sup> ± 0.02	5.20 <sup>a</sup> ± 0.02
WR0	100 <sup>b</sup> ± 0.28	4.20 <sup>b</sup> ± 0.01	6.50 <sup>b</sup> ± 0.02
WRM1	140 <sup>c</sup> ± 0.42	4.35 <sup>c</sup> ± 0.02	7.40 <sup>c</sup> ± 0.28
WRM2	150 <sup>d</sup> ± 1.13	4.40 <sup>cd</sup> ± 0.04	7.80 <sup>d</sup> ± 0.14
WRM3	160 <sup>e</sup> ± 0.14	4.48 <sup>de</sup> ± 0.05	8.04 <sup>de</sup> ± 0.01
WRM4	190 <sup>f</sup> ± 0.40	4.52 <sup>ef</sup> ± 0.05	8.20 <sup>e</sup> ± 0.02
WRM5	200 <sup>g</sup> ± 0.28	4.58 <sup>f</sup> ± 0.02	8.55 <sup>f</sup> ± 0.02

Values are means ± standard deviation of duplicate replications. Means within a column with the same superscript were not significantly ( $p > 0.05$ ) different. **Key:** CTRL=Control, commercial sample; WR0 = 90 % wheat flour +10 % rice flour; WRM1 = 90 % wheat/rice flour +10 % mushroom flour; WRM2 = 80 % wheat/rice flour +20 % mushroom flour; WRM3 = 70 % wheat/rice flour +30 % mushroom flour; WRM4 = 60 % wheat/rice flour + 40 % mushroom flour; WRM5 = 50 % wheat/rice flour + 50 % mushroom flour

The taste of the formulated noodle blends ranged from 1.65-5.60 with sample WRM1 (90:10) having the highest value and sample WRM5 (50:50) having the lowest value. This could probably be because of the increasing proportion of mushroom flour in the samples and consumers did not find it very appealing. The preference for taste of CTRL (commercial sample) and WR0 (100:0) differed significantly ( $p < 0.05$ ) from the preference of the other samples.

The appearance of the formulated noodles from blends of wheat, rice and mushroom flour ranged from 4.80-1.55 with sample WRM1 (90:10) having the highest value and sample WRM5 (50:50) the least value. This is probably because, the noodles from 100% wheat-rice flour had longer noodle strands, a relatively brighter colour and less stickiness unlike the samples with increasing proportion of mushroom flour.

The texture of the formulated noodles from the blends ranged from 3.95-1.30. Sample WRM1 (90:10) ranked highest while sample WRM5 (50:50) was ranked the lowest. Starch characteristics, protein content and quality play major roles in governing the texture of cooked noodles (Hou, 2001). The preference for texture dropped with increase in mushroom flour incorporation. There was no significant ( $p > 0.05$ ) difference between the mean scores of

the samples but they differed significantly ( $p < 0.05$ ) from CTRL (commercial sample) and WR0 (100:0).

#### Micronutrient content of formulated instant noodles from wheat, rice and mushroom flour blends

#### Vitamin content of formulated instant noodles from wheat, rice and mushroom flour blends

The vitamin content of formulated instant noodles from wheat, rice and mushroom flour blends are presented in Table 11. As vitamins are very unstable, a lower value in the sample was expected, since heat processing was used which could contribute to the losses. Cooking and industrial processing of mushroom was found to have pronounced effects on the amount of vitamins in the product. Vitamin B<sub>1</sub> and B<sub>2</sub> are lost during industrial processing (canning) of Boletus at a rate of 21–57% and 8–74%, respectively (Zhou and Yin, 2008). Thiamin stability affected by formulation, processing, and storage has been reported by Bui and Small (2007a, 2007b, 2008) in noodles, and it was inferred that the potential to use thiamin in noodles where alkaline salts are used is limited due to its instability at higher pH. But with addition of mushroom flour there was an increase in the vitamin content. The values varied significantly ( $p < 0.05$ ) ranging

**Table 10.** Sensory evaluation of instant noodles from wheat, rice and mushroom flour blends

Sample	Colour	Mouthfeel	Aroma	Taste	Appearance	Texture	Overall acceptability
CTRL	8.80 <sup>f</sup> ±0.41	8.45 <sup>e</sup> ±0.60	8.30 <sup>c</sup> ±0.80	8.60 <sup>f</sup> ±0.50	8.65 <sup>f</sup> ±0.48	8.45 <sup>c</sup> ±0.51	8.60 <sup>f</sup> ±0.59
WRO	7.15 <sup>e</sup> ±0.74	6.85 <sup>d</sup> ±0.74	6.70 <sup>b</sup> ±0.73	6.70 <sup>c</sup> ±0.86	7.30 <sup>c</sup> ±0.47	6.75 <sup>d</sup> ±0.71	6.95 <sup>e</sup> ±0.60
WRM1	4.50 <sup>d</sup> ±1.53	4.95 <sup>e</sup> ±1.23	4.90 <sup>a</sup> ±1.16	5.60 <sup>d</sup> ±1.04	4.80 <sup>d</sup> ±1.50	3.95 <sup>c</sup> ±1.31	4.40 <sup>d</sup> ±1.35
WRM2	1.23 <sup>c</sup> ±0.27	3.65 <sup>b</sup> ±1.38	4.70 <sup>a</sup> ±1.17	3.90 <sup>c</sup> ±0.91	2.90 <sup>c</sup> ±1.5	2.85 <sup>b</sup> ±1.34	3.15 <sup>c</sup> ±1.30
WRM3	0.93 <sup>b</sup> ±0.21	3.20 <sup>b</sup> ±1.24	4.75 <sup>b</sup> ±1.58	3.20 <sup>b</sup> ±0.76	2.30 <sup>bc</sup> ±1.17	2.40 <sup>b</sup> ±1.14	2.50 <sup>bc</sup> ±1.14
WRM4	0.68 <sup>a</sup> ±0.15	2.35 <sup>a</sup> ±1.03	6.40 <sup>b</sup> ±1.14	2.15 <sup>a</sup> ±0.81	1.65 <sup>ab</sup> ±0.98	1.55 <sup>a</sup> ±0.75	1.95 <sup>ab</sup> ±1.05
WRM5	0.36 <sup>a</sup> ±0.08	2.00 <sup>a</sup> ±1.07	7.15 <sup>b</sup> ±1.30	1.65 <sup>a</sup> ±0.93	1.55 <sup>a</sup> ±0.82	1.30 <sup>a</sup> ±0.57	1.75 <sup>a</sup> ±1.02

Values are means ± standard deviation of duplicate replications. Means within a column with the same superscript were not significantly ( $p < 0.05$ ) different. **Key:** CTRL= Control, commercial sample; WR0 = 90 % wheat flour +10 % rice flour; WRM1 = 90 % wheat/rice flour +10 % mushroom flour; WRM2 = 80 % wheat/rice flour +20 % mushroom flour; WRM3 = 70 % wheat/rice flour +30 % mushroom flour; WRM4 = 60 % wheat/rice flour + 40 % mushroom flour; WRM5 = 50 % wheat/rice flour + 50 % mushroom flour

from 0.34 to 0.20 (mg/100 g) for vitamin B<sub>1</sub>, 0.65 to 0.06 (mg/100 g) for vitamin B<sub>2</sub> and (3.68-1.20 mg/100 g) for vitamin B<sub>3</sub>. Noodle sample WRM3 (70:30) had the highest content of vitamin B<sub>1</sub>, while sample WRM2 (80:20) had the highest for vitamin B<sub>2</sub> and WRM5 (50:50) had the highest for vitamin B<sub>3</sub> and CTRL (commercial sample) had the lowest value in all the vitamins.

Thiamin functions as the co-enzyme thiamin pyrophosphate (TPP) in the metabolism of carbohydrates and branched-chain amino acids. Hence, when there is insufficient thiamin, the overall decrease in carbohydrate metabolism and its inter-connection with amino acid metabolism (via  $\alpha$ -keto acids) have severe consequences, such as a decrease in the formation of acetylcholine for neural function. According to FAO (2001), the required daily intake of thiamin for the adult male and female is 1.2 mg/100g and 1.1 mg/100g, respectively. The sample WRM5 (50:50) containing 0.32 mg/10 g provides about 27% of the required daily intake for the adult male and 29% for the adult female. The major cause of hypo-riboflavinosis is inadequate dietary intake as a result of limited food supply, which is sometimes exacerbated by poor food storage or processing. According to FAO, (2001) the required daily intake of riboflavin for the adult male and female is 1.3 mg/100 g and 1.1 mg/100 g respectively. Sample WRM5 (50:50) containing 0.10 mg/100g noodles from wheat, rice and mushroom flour blends provides about 7 % of the required daily intake for adult male and 9 % for adult female. The contents of vitamins B<sub>1</sub> and B<sub>2</sub> found in the noodles indicate that mushrooms could not be considered as sources of vitamins B<sub>1</sub> and B<sub>2</sub>, since their contribution in terms of these vitamins to the diet is not significant although they might have contributed to the sums of these nutrients in the diet.

Niacin (nicotinic acid) deficiency classically results in pellagra, which is a chronic wasting disease associated with a characteristic erythematous dermatitis, that is bilateral and symmetrical, a dementia after mental changes including insomnia and apathy preceding an overt encephalopathy, and diarrhea resulting from inflammation of the intestinal mucous

surfaces (Tannenbaum et al., 1991). The required daily intake of niacin for the adult male and female is 16 mg/100g and 14 mg/100g respectively (FAO, 2001), and sample WRM5 (50:50) had the highest value (3.68 mg/100g) providing about 23% and 26% for both male and female adult, respectively.

#### **Mineral content of instant noodles formulated from wheat, rice and mushroom flour blends**

The mineral content of the instant noodles formulated from wheat, rice and mushroom blends is presented in Table 12. The iron content in this study ranged from 0.49 – 3.42 mg/100 g. Sample WR0 (100:0) had the lowest value and sample WRM5 (50:50) had the highest value and it was lower than the recommended daily allowance (RDA) - 10 mg of iron per day (Sandstead, 1995). Thus, it could be deduced from the Table that both the control sample and the WR0 (100:0) were not a good source of iron. Iron content increased significantly ( $p < 0.05$ ) with addition of mushroom powder. All the values varied significantly ( $p < 0.05$ ). The values obtained in this study was similar to those reported by Bello et al. (2017) who reported 1.68 – 2.89 mg/100g in biscuit made from wheat and mushroom flour blends. Iron is a major component of hemoglobin that carries oxygen to all parts of the body. Iron also has a critical role within cells assisting in oxygen utilization, enzymatic systems, especially for neural development, and overall cell function.

The potassium content of the samples ranged from 1.86 to 16.00 mg/100g. With CTRL (commercial product) having the lowest and WRM5 (50:50) having the highest value. The potassium content of the instant noodles increased with increase in the mushroom flour addition indicating that mushroom is a good source of minerals. The values obtained in this study were lower than the values obtained by other researchers; 40.63 - 154.07 mg/100g (Bello et al., 2017). The result could be attributed to variation in the different species of mushroom. All the values varied significantly ( $p < 0.05$ ).

Phosphorous content of the samples ranged from 0.16 to 0.070 mg/100g with sample WRM5 (50:50)

having the highest and sample CTRL (commercial product) with the lowest value. These values were lower than 12.5 - 54.62 mg/100g reported by Bello et al. (2017). There was no significant ( $p > 0.05$ ) difference between sample CTRL (commercial product) and samples WRO (100:0), WRM1 (90:10) and WRM2 (80:20). The mineral composition obtained in this study showed that there was an increase in the phosphorous, iron and potassium content of the noodles with an increase in the level of mushroom flour addition. This is an indication that mushroom is a good source of minerals as shown in Table 12.

## CONCLUSION

The study has shown that acceptable noodles could be produced from blends of wheat, rice and mushroom flour. Noodles from the composite flours were richer in nutrients and may confer nutritional advantages to consumers compared to the control. The incorporation of mushroom flour in noodles formulation affected the chemical, cooking and sensory properties. Mushroom flour therefore could be incorporated into instant noodles to obtain a product rich in dietary fiber, protein, vitamin B<sub>3</sub> and potassium and low in fat. Based on the study, the use of 10% mushroom and 50 % mushroom flour

**Table 11.** Vitamin content of instant noodles formulated from wheat, rice and mushroom flour blends

Sample	Vitamin B <sub>1</sub> (mg/100g)	Vitamin B <sub>2</sub> (mg/100g)	Vitamin B <sub>3</sub> (mg/100g)
CTRL	0.20 <sup>a</sup> ± 0.01	0.06 <sup>a</sup> ± 0.02	1.20 <sup>a</sup> ±0.28
WRO	0.22 <sup>ab</sup> ±0.02	0.07 <sup>a</sup> ±0.01	2.90 <sup>b</sup> ±0.14
WRM1	0.24 <sup>abc</sup> ±0.01	0.07 <sup>a</sup> ±0.02	3.00 <sup>b</sup> ± 0.14
WRM2	0.26 <sup>abc</sup> ±0.02	0.65 <sup>b</sup> ±0.21	3.20 <sup>bc</sup> ±0.02
WRM3	0.34 <sup>d</sup> ±0.01	0.04 <sup>a</sup> ±0.03	3.25 <sup>bc</sup> ±0.07
WRM4	0.30 <sup>bcd</sup> ±0.04	0.09 <sup>a</sup> ±0.02	3.60 <sup>cd</sup> ±0.28
WRM5	0.32 <sup>cd</sup> ±0.04	0.10 <sup>a</sup> ±0.02	3.68 <sup>d</sup> ±0.02

Values are means ± standard deviation of duplicate replications. Means within a column with the same superscript were not significantly ( $p < 0.05$ ) different. **Key:** CTRL: Control commercial sample; WRO = 90 % wheat flour +10 % rice flour; WRM1 = 90 % wheat/rice flour +10 % mushroom flour; WRM2 = 80 % wheat/rice flour +20 % mushroom flour; WRM3 = 70 % wheat/rice flour +30 % mushroom flour; WRM4 = 60 % wheat/rice flour + 40 % mushroom flour; WRM5 = 50 % wheat/rice flour + 50 % mushroom flour

**Table 12.** Mineral content of instant noodles from wheat, rice and mushroom flour blends

Sample	Iron (mg/100g)	Potassium (mg/100g)	Phosphorus(mg/100g)
CTRL	0.73 <sup>b</sup> ± 0.02	1.86 <sup>a</sup> ± 0.02	0.07 <sup>a</sup> ± 0.01
WRO	0.49 <sup>a</sup> ± 0.02	2.48 <sup>b</sup> ± 0.02	0.08 <sup>ab</sup> ± 0.01
WRM1	2.02 <sup>c</sup> ± 0.14	12.48 <sup>c</sup> ± 0.56	0.09 <sup>abc</sup> ± 0.01
WRM2	2.21 <sup>d</sup> ± 0.28	13.40 <sup>d</sup> ± 0.01	0.10 <sup>abc</sup> ± 0.01
WRM3	2.32 <sup>e</sup> ±0.14	14.00 <sup>e</sup> ± 0.04	0.13 <sup>bcd</sup> ± 0.02
WRM4	3.18 <sup>f</sup> ± 0.28	15.20 <sup>f</sup> ± 0.02	0.14 <sup>cd</sup> ± 0.02
WRM5	3.42 <sup>g</sup> ± 0.28	16.00 <sup>g</sup> ± 0.02	0.16 <sup>d</sup> ± 0.02

Values are means ± standard deviation of duplicate replications. Means within a column with the same superscript were not significantly ( $p < 0.05$ ) different. **Key:** CTRL: Control, WRO=90 % wheat flour +10 % rice flour; WRM1 = 90 %wheat/rice flour +10 % mushroom flour; WRM2 = 80 %wheat/rice flour +20 % mushroom flour; WRM3 = 70 %wheat/rice flour +30 % mushroom flour; WRM4 = 60 %wheat/rice flour + 40 % mushroom flour; WRM5 = 50 %wheat/rice flour + 50 % mushroom flour



showed great potential in improving the quality of noodles in terms of overall acceptability and nutrient contents respectively, and in turn beneficial to the consumers. Substitution of wheat flour with rice flour up to 10% is suggested as it compares well with the control in acceptability. It was also observed that decrease in the protein content of noodles was improved by the addition of mushroom flour to the blend up to 50%.

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Review Article/Derleme Makale

# Meyve ve sebzelerde bulunan biyoaktif bileşenlerin sağlık üzerine etkileri

## Health effects of bioactive components in fruits and vegetables

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### ÖZET

Doğal besin kaynağının yanı sıra biyoaktif bileşenler bakımından zenginleştirilmiş besinler fonksiyonel besinler (fermente gıdalar, meyve sebzeler, hayvansal gıdalar, sert kabuklu sebzeler vb.) olarak tanımlanmaktadır. Fonksiyonel besinlerin içerdiği biyoaktif bileşenler (karotenoidler, fenolik bileşikler, glikosinolatlar, lignanlar, organosülfür bileşikler ve bitki steroller vb.) kalp damar hastalıkları, kanser, hipertansiyon, ateroskleroz, diyabet gibi hastalıkların önlenmesinde ve tedavisinde etkili olan biyoaktif bileşenleri içeren fonksiyonel besinlerin günlük olarak tüketilmesi büyük bir önem kazanmaktadır. Bu çalışmada kullanılan literatür; 2000-2021 yılları arasında yapılan İngilizce ve Türkçe çalışmalar Pubmed, Science Direct, Google Akademik veri tabanları taranmıştır. Yapılan taramalarda “fonksiyonel besin (functional food), biyoaktif bileşenler (bioactive compounds), meyve ve sebze (fruit and vegetable), sağlık (health)” anahtar kelimeleri kullanılmıştır. Son zamanlarda fonksiyonel besin olan meyve ve sebzelerde bulunan biyoaktif bileşenler (askorbik asit, tokoferoller, karotenoidler, flavonoidler, fenolik asitler ve tiyoller gibi) üzerine gerçekleştirilen çalışmalardan elde edilen sonuçlar



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dikkate alınarak meyve ve sebzelerin sağlığımız üzerine etkisi literatür ışığında detaylı şekilde ele alınarak derlenmiştir. Bu bağlamda günlük meyve ve sebze tüketiminin kanser, kardiyovasküler hastalık, osteoporoz ve diyabet gibi hastalıkların etkilerini azalttığı, meyve ve sebzelerin sağlıklı yaşam için vazgeçilmez fonksiyonel besinler olduğu ortaya konulmaktadır. Bu nedenle meyve ve sebzelerde bulunan biyoaktif bileşenleri derinlemesine incelemek ve etki mekanizmalarını belirlemek gereklidir. Çalışmalar, meyve ve sebzelerde bulunan biyoaktif bileşenlerin gıda güvenliği açısından değerlendirilmesi ve meyve ve sebzelerin beslenmemize yer verilmesi sürecinde etkili olacaktır.

## ABSTRACT

Foods enriched with bioactive compounds beside the natural food source are defined as functional foods (fermented foods, fruit and vegetables, animal foods, hard-shelled vegetables, etc.). Bioactive compounds (carotenoids, phenolic compounds, glucosinolates, lignans, organosulphur compounds, plant sterols etc.) contained in functional foods are effective in the prevention and treatment of diseases such as cardiovascular diseases, cancer, hypertension, atherosclerosis, diabetes. Daily consumption of functional foods which are important for health, gains value. Pubmed, Science Direct, Google Academic databases were used in the research strategy in the review. The databases were searched for studies in English and Turkish from 2000 to 2021. Keywords searched in the screenings were: functional food, bioactive compounds, fruit and vegetable, health. Lately the effect of fruits and vegetables on health has been compiled in detail in the light of the literature, taking into account the results obtained from studies on bioactive compounds (such as ascorbic acid, tocopherols, carotenoids, flavonoids, phenolic acids and thiols) found in fruits and vegetables which are functional foods. In this context, it has been revealed that daily consumption of fruits and vegetables reduces the effects of diseases such as cancer, cardiovascular disease, osteoporosis and diabetes and fruits and vegetables are indispensable functional nutrients for a healthy life. Therefore, it is necessary to deeply study the bioactive components in fruits and vegetables and to determine their mechanism of action. Studies will be effective in the process of evaluating the bioactive components in terms of food safety and the inclusion of fruits and vegetables in our diet.

## GİRİŞ

Sağlıklı, uzun yaşam ve hastalıklardan korunma isteği insanları beslenme konusunda daha bilinçli ve duyarlı olmaya teşvik etmektedir (Sevilmiş ve ark., 2017). Gıda ve gıda bileşenlerinin sağlık üzerine olumlu ve olumsuz etkileri insanlar için her zaman ilgi odağı olmuştur (Hasler ve Brown, 2009). Son yıllarda, tüketiciler besin değeri yüksek ve gastrointestinal sistem, kanser, kalp-damar hastalıkları, diyabet, hipertansiyon, ateroskleroz gibi hastalıkların korunması ve önlenmesi amacıyla fonksiyonel besinleri tercih etmektedirler (Karaağaç, 2010; Coşkun, 2005). Yapılarında biyoaktif bileşenler bulunan veya bu bileşenler ile zenginleştirilen gıdalar fonksiyonel besinler olarak tanımlanabilmektedir (Coşkun, 2005). Fonksiyonel besinler, ilaç veya herhangi bir besin takviyesi olmayıp yeterli ve dengeli beslenme düzeninin önemli bir parçasıdır. Bu besinlerin içerdiği bileşenlerin, insan sağlığın korunmasında ve kronik hastalık riskinin azaltmasında etkili olduğu belirtilmiştir (Çakiroğlu ve ark., 2018; Badu-Gyan ve Owusu, 2017).

Fonksiyonel besinlerin potansiyel sağlık yararlarını araştıran ve fonksiyonel besinleri ayrı bir kategori olarak tanımlayan ilk ülke Japonya olarak bilinmektedir (Dayısoylu ve ark., 2014). Fonksiyonel besinler, sağlığımızı olumlu yönde etkilediği için diğer ülkeler de bu besinlere ilgi göstermiş ve bu besinler diyetlerinin vazgeçilmez bir parçası haline gelmiştir (Hasler ve Brown, 2009). Fonksiyonel gıda pazarları, özellikle Amerika Birleşik Devletleri, Japonya ve Avrupa'da gelişmektedir. Ülkemizde ise tüketicilerin, fonksiyonel besinlere karşı farkındalığını arttırmak, sağlıklı yaşamı benimsetmek amacıyla birçok gıda pazarı oluşmaya başlamıştır (Dayısoylu ve ark., 2014).

Fonksiyonel besinlerin yapısındaki biyoaktif bileşenler, sağlığın korunmasında ve astım, viral ve paraziter enfeksiyonlar, enflamatuar hastalıklar gibi birçok hastalığın önlenmesinde oldukça önemlidir (Castillo ve ark., 2018) ve bu bileşenleri içeren besinlerin tüketilmesi büyük önem kazanmaktadır (Uyar ve Sürücüoğlu, 2010). Karotenoidler, n-3 yağ asitleri, izoflavonlar, flavonoidler, izosiyanatlar, fenolik asitler, fitoöstrojenler, polifenoller, çözünür diyet lifleri, bitki stanoller ve



steroller, polioller, probiyotikler, prebiyotikler ve sinbiyotikler “biyoaktif bileşenler” olarak tanımlanmaktadır (Ashwell, 2002).

Bu çalışmada, meyve ve sebzelerin içerdiği biyoaktif bileşenler ve bu biyoaktif bileşenlerin sağlık açısından önemi hakkında kapsamlı literatür araştırması ışığında bilgiler verilecektir. Böylece günlük olarak tükettiğimiz meyve ve sebzelerin içerdiği biyoaktif bileşenleri ve bu biyoaktif bileşenlerin sağlığımız üzerine etkisi ve hangi hastalıkların önlenmesinde ve tedavisinde etkili olduğu değerlendirilecektir.

### 1. Besinlerde Bulunan Biyoaktif Bileşenler

Yeterli ve dengeli beslenme, bireyin fiziksel özellikler (yaş, cinsiyet), genetik, fiziksel aktivite, fizyolojik özellikler, hastalık durumu vb. farklı faktörlere göre enerji ve besin öğelerini yeterli

miktarda alması olarak tanımlanmaktadır. Yeterli ve dengeli beslenmenin sağlığını korunmasında ve yeniden kazandırılmasında etkili olduğu belirtilmiştir (Arlı ve Şanlıer, 2006). Yeterli ve dengeli beslenmek amacıyla tükettiğimiz sebze ve meyvelerde bulunan lif, vitamin ve polifenoller gibi besin öğeleri ile biyoaktif bileşenler sağlıklı yaşam, hastalıkların korunmasında ve tedavisinde oldukça önemlidir (Özdoğan ve ark., 2018). Besinlerde bulunan biyoaktif bileşenler yapılarına göre karotenoidler, fenolik bileşikler, glikosinolatlar, lignanlar, organosülfür bileşikleri ve bitki sterollerini olarak sınıflandırılmaktadır. Günlük olarak tüketilen meyve ve sebzelerde bulunan biyoaktif bileşenlerin ile biyolojik etkileri Tablo 1’de verilmiştir. (Meagher ve ark., 2000; Rao ve ark., 2007; Yılmaz ve Demirel, 2012; Shashirekha ve ark., 2015; Guasch-Ferré ve ark., 2017; Ruhee ve ark., 2020).

**Tablo 1.** Biyoaktif bileşenlerin sınıflandırılması

Biyoaktif bileşen	Örnekleri	Kaynakları	Biyolojik etkileri	Kaynak
Karotenoidler	$\alpha$ karoten, $\beta$ karoten, Lutein, Likopen, Zeaksantin, $\beta$ kriptoksantin	Havuç, domates, ıspanak, mısır, narenciye, patates, kabak, sarı ve kırmızı biber, havuç, kayısı, kavun, lahana,	Kanser, Koroner kalp hastalığı, Maküler dejenerasyon, Katarakt, Antioksidan, Serbest radikalleri hapsedici	Shashirekha ve ark., 2015; Rao ve ark., 2007
Fenolik Asitler	Hidroksibenzoik asitler, Hidroksisinnamik asitler, Hidroksifenilasetik asitler, Hidroksifenilpropanoik asitler, Hidroksifenilpentanoik asitler	Nar, yaban mersini, ahududu, erik, üzüm, çilek, elma, ceviz, çikolata, şarap, yeşil çay, kahve, tahıllar	Antioksidan, Anti-mutajenik, Antikanser, Antidiyabetik, Anti-enflamatuar	Shashirekha ve ark., 2015; Guasch-Ferré ve ark., 2017
Flavonoidler	Flavonoller, Flavanonlar, Flavanoller, Flavonlar, İzoflavonoidler, Antosiyaninler, Kalkonlar, Dihidroalkonlar	Çilek, üzüm, kiraz, erik, nar, elma, armut, maydanoz, kereviz, portakal, soğan, çay, bal, baharat, brokoli	Anti-enflamatuar, Anti-oksidatif etkiler, Antikanser, Antidiyabetik	Shashirekha ve ark., 2015; Guasch-Ferré ve ark., 2017
Glukosinolatlar	İzotiyosiyanatlar, Tiyosiyanatlar, Nitriller, Hidroksinitriller, Epitiyonitriller	Brokoli, karnabahar, lahana, kolza tohumu, hardal	Antioksidan, Antikanserojen, DNA koruyucu, Kalbi koruyucu, Anti-tümör, Anti-enflamatuar	Shashirekha ve ark., 2015; Yılmaz ve Demirel, 2012
Lignanlar	Asetoksinorezinol, Hidroksienterodiol, Sekoizolarisiresinol,	Keten tohumu, susam	Antidiyabetik, Kardiyovasküler hastalıklara karşı koruyucu	Meagher ve ark., 2000; Guasch-Ferré ve ark., 2017
Organosülfür Bileşikleri	İzotiyosiyanat, İndoller, Alilik kükürt bileşikleri, Sülforafan	Soğan, sarımsak, brokoli, lahana, karnabahar	Anti-enflamatuar, Antikanserojen, Kardiyovasküler hastalıklara karşı koruyucu	Ruhee ve ark., 2020
Bitki Sterollerini (Fitosteroller)	Sitosterol, Kampesterol, Stigmatrol, Sitostanol, Kampestanol, Stigmastenol	Fındık, tohumlar, tam tahıllar, baklagiller, sızma zeytinyağı	Koroner kalp hastalığı, Düşük yoğunluklu lipoprotein (LDL), kolesterolün bloke edilmesi	Shashirekha ve ark., 2015



### 1.1 Antioksidanlar

Radikal oluşumunun sınırlandırılması, radikal tepkimelerin sona erdirilmesi ve oluşan radikallerin etkisiz hale getirilmesinden “antioksidanların” etkili olduğu belirtilmiştir. Meyve ve sebzelerle doğal antioksidan etkili bileşikler (askorbik asit, tokoferoller, karotenoidler, flavonoidler, fenolik asitler ve tiyoller) vücudu hastalıklara karşı korumaktadır. Örneğin, flavonoid yapıları yüksek olan bazı bitkilerle beslenmek vücudumuzun birçok kronik hastalıklardan (kanser, kalp, akciğer, karaciğer) korunmasını sağlamaktadır (Arıdur, 2013).

### 1.2 Fenolik Bileşikler

Fenolik bileşikler, “fenolik asit” ve “flavonoidler” olarak sınıflandırılabilir. Flavonoidler grubunda flavonoller, flavanoller, antosiyaninler, kumarinler, tanninler ve lignin bulunmaktadır (Meral ve ark., 2012). Flavonoidler (polifenollerin bir alt sınıfı), bitki gıdalarında her yerde bulunan yapısal olarak ilişkili ikincil metabolitlerdir. Kakao ve çay gibi flavonoid bakımından zengin gıdaların tüketimi, daha düşük kardiyovasküler hastalık riski (KVH) ile ilişkilidir (Dower ve ark., 2015). Flavanonlar, flavanoller, flavonoller, izoflavonlar, flavonlar ve antosiyaninler gibi altı ana alt grubu olan flavonoidler diyabette apoptoz, inflamasyon ve nörodejenerasyon gibi retina dejeneratif faktörlerini iyileştirebilmektedir (Ola ve ark., 2018). Sebze, meyve, şarap, çay, kakao ve çilek gibi birçok bitki kaynaklı gıdada bulunan polifenolik bileşikler olan flavonoidlerin bilişsel bozulmayı azaltabileceği, nörodejeneratif bozukluklar üzerindeki koruyucu etkileri olduğu, oksidatif stresi azalttığı çalışmalarda gözlenmiştir (Devore ve ark., 2012; Hagan ve ark., 2016; Gildawie ve ark., 2018).

### 1.3 Karotenoidler

Yağda çözünebilir pigmentler olan karotenoidler, antioksidanlar ve A vitamini aktiviteleri fonksiyonlarına sahiptir. Karotenoidlerin tüketiminin, vücut içinde emilim, taşınım, metabolik reaksiyonlar ve depolanmalarına bağlı olarak çeşitli hastalıklara (görme, kanser ve kalp hastalığı) karşı etkili olduğu belirtilmiştir (Özçelik ve Davarcı, 2011). Karotenoidler, günlük beslenmemizde yaygın olarak bulunan çeşitli

sebze ve meyvelerin renklerinden sorumludur (Rao ve ark., 2007). Özellikle sarı-turuncu meyvelerde (havuç, domates, kabak, biber gibi) ve koyu yeşil yapraklı sebzelerde bol miktarda bulunur. Antioksidan kaynağı olan karotenoidler  $\alpha$  ve  $\beta$  karoten, likopen,  $\beta$ -kriptoksantin, zeaksantin ve lutein içermektedir (Xavier ve Gálvez, 2016).

### 1.4 Glukosinolatlar

Glukosinolatlar esas olarak Brassica familyasında yer alan brokoli, lahan, karnabahar gibi yaygın olarak yenilebilir bitkiler de dahil olmak üzere kolza tohumu, hardal, yaban turpu gibi turpgiller bitkilerinde bulunmaktadır (Prieto ve ark., 2019). Çiğneme, pişirme için yapılan kesme, doğrama gibi hazırlıklar, ısıtma ya da böcek saldırıları gibi dış etkenlerle hücrenin parçalanması sonucu aroma ve lezzet bileşikleri izotiyosiyanatlar, tiyosiyanatlar, nitriller, hidroksinitriller ve epitiyonitriller hidroliz ürünleri açığa çıkmaktadır. Bu ürünler antikanserojenik, antimikrobiyal, antioksidan, guatrojenik etki gibi birçok biyolojik aktivitelere sahiptir (Yılmaz ve Demirel, 2012).

### 1.5 Lignanlar

Keten, keten tohumu, küspesi ve unu benzeri ürünlerin içeriğinde fazla miktarda lignan bulunmaktadır. İlave olarak tahıllar, tahıl kepekleri, birkaç yağlı tohum, meyve ve sebzelerde de önemli miktarda lignan içermektedir (Meagher ve ark., 2000). Göğüs, kolon, prostat ve tiroid kanserinde lignanların etkisini araştırmak için yapılan çalışmalarda genel olarak faydalı olduğu belirtilmiştir. Lignanlar kalp hastalıkları ile ilgili risk faktörlerin azalmasında olumlu etki göstermektedir (Westcott ve Muir, 2003).

### 1.6 Organosülfür Bileşikleri

Organosülfür bileşikleri izotiyosiyanatlar, indoller, alilik kükürt bileşikleri ve sülfurafan içeren bir bileşiktir. Bu bileşikler kardiyovasküler hastalıklara karşı korumada, sistolik kan basıncının düşürülmesinde, kolesterol seviyelerinin düşürülmesinde etkilidir. Antienflamatuvar, antikanserojen etkileri bulunmaktadır. Allium ve

Brassica (Turpgiller) cinsinden sebzeler (soğan, sarımsak, brokoli, lahan, karnabahar gibi) organosülfür bileşiklerin iyi kaynağıdır (Ruhe ve ark., 2020).

### 1.7 Bitki Sterolleri

Bitki sterolleri (fitosteroller olarak da adlandırılır) meyvelerde, sebzelerde, kuruyemişlerde, tohumlarda, tahıllarda, baklagillerde, bitkisel yağlarda ve diğer bitkilerde doğal olarak bulunan, kolesterole benzer kimyasal bir yapıya sahip olan bitki bileşenleridir. Bitki sterollerin içerisinde fazla miktarda beta-sitosterol, kampesterol ve stigmasterol bulunmaktadır. Bitki sterolleri ile zenginleştirilmiş gıdaların kardiyovasküler hastalık riski, ateroskleroz, inflamasyon, antioksidan potansiyel, kanser, nörobilişsel fonksiyon ve göz hastalıkları üzerinde olumlu etkisi bilinmektedir (Rudkowska, 2009).

## 2. Meyve ve Sebzelerde Bulunan Biyoaktif Bileşenlerin Sağlık Üzerine Etkisi

“Dünya Sağlık Örgütü günde en az 400-500 g (günde en az beş porsiyon ilavesi) enerji içeriği düşük, mineral ve vitamin içerikleri yüksek olan meyve ve sebze tüketiminin kanser, hipertansiyon, kardiyovasküler, gastrointestinal ve kronik hastalıkların azalmasında ve yaşlanmanın gecikmesinde etkili olduğunu belirtmiştir.” Yapısında antioksidan ve fenolik bileşikler sayesinde antioksidatif ve antimikrobiyal etkilerinden dolayı sağlığımızı koruyan meyve ve sebzeler fonksiyonel besinler olarak tanımlanmaktadır (Giampieri ve ark., 2012). Bu bölümde, günlük olarak tükettiğimiz fonksiyonel besinlerden meyve ve sebzelerin içerdiği biyoaktif bileşenler ve bu bileşenlerin sağlığımız üzerine etkileri ele alınmıştır. İncelenen çalışmalar, fonksiyonel besinlerin içerdiği biyoaktif bileşenlerin hastalıkların önlenmesinde ve tedavisinde etkili olduğunu göstermektedir.

### 2.1 Çilek

Çilek, vitaminler, mineraller, folat ve lif olmak üzere birçok önemli diyet bileşeninin yanı sıra polifenoller bakımından zengin fitokimyasal bileşik kaynağı olarak tanımlanmaktadır (Giampieri ve ark., 2012). Bayram ve ark. (2013), doğal olarak fazla miktarda C vitamini, antioksidanlar,

biyoaktif bileşenler içermesinden dolayı meyve grubunda yer alan çileği değerlendirmişlerdir. Çileğin fonksiyonel özellikleri, yetiştirilmesi sırasında kullanılan yöntemlerle, antioksidan, fenolik madde ve C vitamini fazla olması ile fonksiyonel özelliklerinin artabileceğini ifade etmişlerdir. Çileğin içerdiği biyoaktif bileşenler iltihaplanma, oksidatif stres, kardiyovasküler hastalık, belirli kanser türleri, tip 2 diyabet, obezite ve nörodejenerasyon gibi hastalıkların korunmasında etkilidir (Giampieri ve ark., 2012).

### 2.2 Yaban Mersini

Kırmızı yaban mersini (cranberry), antioksidan, antimikrobiyal ve antikanserojen gibi sağlık üzerine olumlu etki gösteren flavonoller (flavonols), antosiyaninler (anthocyanins), tanenler (tannins) ve fenolik asit türevleri (ferulik asit, p-kumarik asit vb.) gibi biyoaktif bileşenleri içermektedir (Cote ve ark., 2010). Lifli yapıya sahip kırmızı yaban mersini bağırsak sisteminin düzenlenmesinde, kan şekeri ve kolesterolün düşürülmesinde, göz yorgunluğu ve şeker hastalığından kaynaklı görme bozukluklarının engellenmesinde etkilidir (Çağlar ve Demirci, 2017). Polifenoller, antosiyaninler, flavanoller ve tanenler bakımından zengin biyoaktif bileşen içeren kırmızı yaban mersini, kansere karşı etkili olan yüksek miktarda elajik asit içermektedir (Çağlar ve Demirci, 2017). Prediyabetli veya yeni tanı konmuş 160 diyabetli hastaya 12 hafta boyunca antosiyanin takviyesinin verilmesi ile hastaların serum adiponektininde bir artış olduğu ve yeni tanı konmuş diyabetli hastalarda açlık glikozunun azalmasında etkili olduğu gösterilmiştir (Yang ve ark., 2020).

### 2.3 Üzüm

Üzüm malvidin 3-O-(6-O-p-kumarolglukozido)-5-glukozit gibi antosiyaninler, flavonoller, ve prosiyanidin B2 3-O-gallat gibi antioksidan ve zengin fenolik bileşikler içermektedir (Gülcü ve ark., 2008; Sağlam ve ark., 2021). Vitamin ve mineral bakımından zengin besin kaynağı olan üzüm enerji kaynağı olarak kullanılmaktadır. Kansere, alerji ve kireçlenmelerde iltihap oluşumunu engellemektedir. Aminoasit ve antioksidan içeren üzümün sağlık üzerine olumlu etkiye sahip olduğu belirtilmiştir. Bağışıklık sisteminde, böbrek ve karaciğer fonksiyonlarında, kara-

ciğer hastalıklarının ve kalp damar sisteminin korunmasında ve kansızlığın önlenmesinde etkili olduğu bilinmektedir (Ateş, 2015). Resveratrol, üzümde, kırmızı şarapta bulunan ve birçok organizmada ömrünü uzattığı gösterilen doğal bir polifenoldür. Son birkaç yılda, resveratrol kardiyoprotektif, antiinflamatuvar ve antioksidan özelliklere sahip olarak karakterize edilmiştir (Baur ve Sinclair, 2006). Üzümde bulunan resveratrol takviyesinin alkolsüz yağlı karaciğer hastalığına karşı etkinliğini, resveratrolün herhangi bir dozajdaki etkilerini plasebo veya müdahale etmeyenlerle karşılaştıran bir çalışmada LDL ve toplam kolesterol seviyelerini önemli ölçüde azalttığı ve lipid metabolik parametrelerini iyileştirebileceği gözlenmiştir (Zhang ve ark., 2016).

#### 2.4 Domates

A, B1, B2, B3, C, K vitaminleri, protein, yağ, karbonhidrat, organik asitler, potasyum, demir içeren domates (*lycopersicum esculentum*), karoten grubundan likopen içermektedir. Domateste bulunan likopen bu sebzededen elde edilen ürünlerin kırmızı renkli olmasını sağlamaktadır (Konar, 2008). Norrish ve ark. (2000), prostat kanser riski ile havuç, yeşil yapraklı sebzeler ve domates gibi besinlerin içerdiği  $\beta$ -karoten ve likopen arasındaki ilişkiyi araştırmışlardır.  $\beta$ -karoten bakımından zengin besinlerin prostat kanserine karşı koruyucu olmadığı fakat domates gibi likopen içeren besinlerin prostat kanserinin azalmasında etkili olduğunu belirtmişlerdir. Karotenoid alımı ile prostat kanseri riski arasında ilişkiyi inceleyen bir çalışmada, diyetle  $\alpha$ -karoten alımı ve likopen tüketiminin (hem diyet alımı hem de kan seviyeleri) prostat kanser riskinin azalmasıyla ilişkili olduğu gözlenmiştir (Wang ve ark., 2015). Çift kör, randomize, kontrollü mekanik bir çalışmada ise 2 ay boyunca 7 mg/gün likopen ile tedavi edilen 36 kişide endotel fonksiyonunda iyileşme olduğu gözlenmiştir (Gajendragadkar ve ark., 2014).

#### 2.5 Turunçgiller

Naringin ve hesperidin flavonoidlerince zengin olan turunçgiller yapısında bulunan askorbik asit, folik asit, karotenoidler ve flavonoidler içermesi bakımından fonksiyonel besindir. Portakal, üzüm ve limon gibi turunçgillerde bulunan

hesperidin ve hesperetin, nörodejeneratif hastalıklar, hafıza, kanser, oksidatif stres, depresyon ve diğer kronik enflamatuvar morbiditeler dahil olmak üzere insan sağlığına birçok faydası olan portakal kabuğundan ekstrakte edilen flavonoidlerdir (Maldonado ve ark., 2020). Narenciye flavonoidleri serbest radikalleri temizler, glukoz toleransını ve insülin duyarlılığını artırır, lipid metabolizmasını ve adiposit farklılaşmasını modüle eder, inflamasyonu ve apoptozu engeller ve endotel disfonksiyonunu iyileştirir (Mahmoud ve ark., 2019). Turunçgillerin insanlarda görülen çeşitli kanser hastalıklarına karşı etkili olduğu belirtilmiştir. Ayrıca bu bileşenler hiperglisemi, hiperlipidemi, hipertansiyon, inflamasyon ve obezite üzerine olumlu etkileri de vardır (Cin ve Pezer, 2017; Güven ve Gülmez, 2006). Portakal suyunun kan basıncı üzerindeki etkisini inceleyen bir çalışmada, 11 kişilik 2 gruba ayrılan 22 sağlıklı bireye bir ay süresince günde iki kez 500 ml/gün portakal suyu tüketilmesiyle diastolik ve sistolik kan basıncında anlamlı olarak azalma olduğu gözlenmiştir (Asgary ve Keshvari, 2013). Polifenolik bileşiklerin nörobilişsel fonksiyonlardaki potansiyel rolünü ve oksidatif strese karşı önlemeyi değerlendiren başka bir çalışmada sıçanlar 16 gün boyunca sırasıyla 50 mg/kg, 200 mg/kg ve 50 mg/kg dozlarında oral olarak naringenin (NAR), kurkumin (CUR) ve quercetin (QUE) ile tedavi edildi. Çalışmanın sonucunda flavonoid bakımından zengin gıdaların tüketiminin, bilişsel işlevlerde iyileşme sağladığı, nörodejeneratif bozuklukların başlangıcını geciktirdiği ve yaşa bağlı bilişsel gerilemeyi önlemede etkili olduğu gözlenmiştir. Tedavi edilen sıçanlarda lipid peroksidasyonu açısından oksidatif stres önemli ölçüde önlendi (Liaquat ve ark., 2018).

Antioksidan, antiinflamatuvar ve antikanserojen etkilerinden dolayı hastalıkların önlenmesinde ve tedavisinde etkili olan portakal ve elma günlük beslenmede tercih edilmektedir. Meyve grubundan fonksiyonel besin olan portakal ve elmada flavanoid grubundan hesperidin, tangeretin ve phloretin biyoaktif bileşikler içermektedir. Bu biyoaktif bileşiklerin obezite, diyabet, kardiyovasküler hastalıklar, kanser üzerinde olumlu etkisinin olduğu belirtilmiştir (Arslan, 2021).

## 2.6 Turpgil Sebze

Lahana, brokoli, karnabahar ve brüksel lahanası gibi turpgil sebzelerde bulunan glukosinolatlar antikanserojenik etki göstermektedir. Bu sebzeler C vitamini, karoten, folik asit, kalsiyum ve demir bakımından zengin besin kaynağıdır (Güven ve Gülmez, 2006). Turpgil sebze ve izotiyosiyanat alımı ile çeşitli sağlık sonuçları arasındaki ilişkilerin kanıtlarını açıklığa kavuşturmak için, insanlarda meta-analizlerin ve sistematik incelemelerini yapan bir çalışmada turpgillerden elde edilen fitokimyasalların, kanserojen metabolizmasını modüle ederek kansere karşı koruma sağladığı gözlenmiştir. Ayrıca turpgil sebzeler mortalitenin azalmasıyla ilişkili karoten, tokoferol ve askorbat gibi antioksidan mikro besinler bakımından da zengindir (Li ve ark., 2022).

## 2.7 Sarımsak

Özdoğan ve ark. (2018), sarımsakta kükürtlü bileşikler (alliin, allisin, ajoen, diallil disülfid, diallil trisülfid vb.), saponin, fruktooligosakkaritler, inülin gibi biyoaktif bileşenler içerdiğini belirtmişlerdir. Sarımsakta bulunan kükürtlü bileşikler, obezite, diyabet, kardiyovasküler hastalıklar, kanser gibi hastaların tedavisinde etkili olduğunu ifade etmişlerdir (Özdoğan ve ark., 2018). İnsanlardaki kanser riskinin azaltılmasında sarımsağın etkili olduğu belirtilmiştir. Örneğin, menopoz sonrası dönemde oluşan kolon kanserlerinin %50 azalmasında etkilidir (Güven ve Gülmez, 2006). Son on yıldır, sarımsak hipertansiyonlu hastaların kullandığı kan basıncının kontrolü amacıyla en çok tercih edilen takviyelerden biridir (Xiong ve ark., 2013). Sarımsak takviyelerinin kan basıncını azalttığı düşünülmektedir. Wang ve ark. (2015), tarafından gerçekleştirilen meta analizi sonucunda, sarımsak takviyesinin özellikle hipertansiyon hastalarında kan basıncının azalmasında etkili olduğu belirtilmiştir. Şangay, Çin'de yapılan vaka kontrol çalışmasında, sarımsak ve soğan çeşitleri olmak üzere allium sebzelerinin tüketilmesi durumunda prostat kanseri üzerine etkisini araştıran bir çalışmada ise 10.0 g/gün'den fazla allium sebze tüketen erkeklerin, 2.2 g/gün'den az tüketenlere kıyasla prostat kanseri riskinin azaldığı tespit edilmiştir. Allium sebzelerinin tüketimi ile ilişkili prostat kanseri riskinin azalması, diğer bireysel sebzelerin veya sebze gruplarının

etkilerinden çok daha belirgin olduğu gözlenmiştir (Hsing ve ark., 2002).

## 3. Fonksiyonel Meyve ve Sebzeler

Bilimsel ve teknolojik alanlardaki gelişmeler ile tüketiciler, besin değeri yüksek, faydalı ve hastalıklardan koruyan fonksiyonel gıdalara yönelmiştir. Tahıl, baklagiller, fermente gıdalar, hayvansal gıdalar, sert kabuklu meyve-sebzeler ve meyve-sebzelerin biyoyararlanımının artması için bu gıdaların üretim teknolojisinde fonksiyonel bileşenler eklenmektedir (Şengün ve Yahşi, 2021). Geleneksel olarak probiyotikler, yoğurt ve diğer fermente süt ürünlerinde kullanılmaktadır. Son zamanlarda, süt ürünü olmayan probiyotiklerin üretimi için uygun substratlar olup olmadığını belirlemek için alternatif hammaddeler üzerinde çalışılmıştır. Meyve sularının içerisinde mineraller, vitaminler, lif ve antioksidanlar gibi faydalı besinleri içerdikleri için probiyotik bakteri yetiştiriciliği için uygun substratlar olduğu gösterilmiştir. Ayrıca meyve sularının hoş bir tadı olduğu için her yaş grubundaki insanlar için tercih edilen bir içecektir. Ananas, kavun, kaju elması, elma, portakal, siyah frenk üzümü, muz ve yaban mersini, probiyotik bakteri iletimi için gıda matrisi olarak kullanılan meyve sularından bazılarıdır (Pereira ve Rodrigues, 2018).

Meyve suyunu probiyotik gıdaya dönüştürmenin iki yolu vardır: meyve suyuna mikroorganizma ilavesi ve probiyotik mikroorganizmalarla fermantasyon işlemidir. Probiyotik meyve sularının üretimi için ilk olarak eklenen mikroorganizma Laktobacillus'tur. Fermantasyon işleminde probiyotikler sonucunda oluşan ve besin değeri yüksek olan birçok metabolitler (siringik asit, ferulik asit, gallik asit ve laktik asit gibi) sağlığımızı olumlu yönde etkilemektedir (Wu ve ark., 2021).

Laktik asit bakterileri, probiyotik gıdaların depolanması sırasında askorbik asit (C vitamini) kaybını engellemesi, antioksidan kapasitesinde artışı sağlaması gibi birçok fayda sağladığı gözlenmiştir. Meyve suları probiyotik taşıyıcı olarak kullanıma uygun olduğu için dünya çapındaki bazı şirketler probiyotik meyve suyu içecekleri satışa sunmaya başlamıştır. Bu nedenle fonksiyonel meyve bazlı içecekler gelecek vaat eden



bir pazar haline gelecektir (Pereira ve Rodrigues, 2018).

Şeker miktarı az olan farklı meyvelerden oluşan (%22 armut, %16 elma, %16 ayva, %12 kivi, %12 çilek, %8 havuç, %8 siyah havuç, %4 yaban mersini ve %2 portakal kabuğu) marmelatların üretimi gerçekleştirilmektedir (Acoğlu ve Ömeroğlu, 2020). Marmelatın fonksiyonelliğini artırmak amacıyla yapılan bir çalışmada, su yerine biyoaktif bileşenler ve mineral bakımından zengin olan nar suyu ve yeşil çay ilave edilmiştir. Kalori miktarını azaltmak amacıyla bal ve stevia şekeri ilave edilerek oluşturulan farklı ürünler elde edildiğinde, stevia şekeri ile kullanılan marmelatta en yüksek fenolik madde miktarı olduğu belirlenmiştir. Ayrıca bal ilaveli marmelatın en yüksek antioksidan kapasitesine sahip olması hammaddedeki nar suyu ve yeşil çay sayesinde olduğu düşünülmektedir. Ürünün fonksiyonel özelliğini artıranlar düşük şekerli, ballı ve stevia formunda bulunmasıdır. Ticari stevia şekerinin, endüstriyel alandaki üretimlerde gıda katkı maddesi olarak kullanılan steviol glikozitleri yerine kullanılması önerilmektedir. Ayrıca ısı işlem sonrasında hidrosimetilfurfural (HMF) oluşumunu tespit etmek için HMF oluşumunu etki eden faktörlerin detaylı olarak incelenmesi önerilmektedir. Çalışmadan elde edilen bulgular sonucunda; yapay olmayan tatlandırıcılar ve su yerine nar suyu ve demlenmiş yeşil çay ilave edilerek üretilen marmelatların tüketici tarafından tercih edilen, besin değeri yüksek, fonksiyonel bileşenleri fazla olan, düşük kalorili marmelat çeşitlerinin üretilebileceği önerisi yapılmıştır (Acoğlu ve Ömeroğlu, 2020).

Siyah havuçlar yüksek fenoliklere, antosiyaninlere ve antioksidan kapasiteye sahip olma özelliği sayesinde koroner kalp hastalığı ve inme riskindeki azalma, antitümör özellikler, antiinflamatuvar etkiler ve gelişmiş bilişsel davranış gibi bazı sağlık yararlarına katkıda bulunmaktadır. Taze hazırlanmış siyah havuç suyunun genel nitelikler üzerindeki etkilerini inceleyen bir çalışmada ultraviyole (UV-C ve UV-B) radyasyon tedavilerinin zamana bağlı olarak (0, 5, 15, 30 ve 60 dk) etkileri değerlendirilmiştir. Değerlendirilen kalite kriterleri; pH, toplam çözünür katı, titre edilebilir asitlik, su aktivitesi, viskozite ve

renk, esmerleşme indeksi, berraklık, antioksidan aktivite, toplam fenolik içerik, toplam flavonoid içeriği, toplam monomerik antosiyanin içeriği, askorbik asit içeriği ve mikrobiyal özellikler gibi fizikokimyasal özelliklerdir. UV-B ve UV-C işlemleri sonucunda antioksidan değerlerinde artış gözlenmiştir. Sonuç olarak UV işlemlerinin, genellikle belirli biyoaktif bileşikler ve aktiviteyi (toplam fenolik içeriği, flavonoid içeriği) artırabileceği gözlenmiştir. Ultraviyole (UV-C ve UV-B) radyasyon tedavilerinin çok geniş olduğu ve meyve suyunda kalite parametrelerini korumak için başarıyla kullanılabilirliği söylenebilir. Kombine tedavi geliştirmek için daha fazla araştırma çalışması gerekebilir ve karşılaştırılabilir ısı işlem olabileceği önerilmektedir. Bununla birlikte çalışmanın sonuçları, ultraviyole radyasyon tedavilerinin, bir pilot ölçekte termal olmayan bir koruma modu olarak endüstriyel olarak etkili bir şekilde araştırılabileceğini de göstermektedir (Türkmen ve Takci, 2018).

Başka bir çalışmada kabak yapraklarının doğal fermentasyonunun, hasat sonrası kayıpları azaltmak, ürünün raf ömrünü uzatmak ve pişirme işlemi yapmadan beslenme özelliklerini arttırmak için yararlı bir araç olup olmadığını ortaya çıkarmak amacıyla yapılmıştır. Kurutulmuş kabak yapraklarının %3 NaCl ve %3 sükröz ile fermente işleminden önce besin bileşimi ve daha sonra 168 saat sonra hava ile kurutulmuş veya termal olarak işlenmiş kabak yapraklarının besin bileşimi karşılaştırılmıştır. %3 NaCl ve %3 sükröz işleminde B3 (niasin) konsantrasyonu anlamlı olarak azalırken, B6 (pridoksin) ve B9 (folat) anlamlı olarak daha yüksek olduğu gözlenmiştir. Fermentasyon işlemi sonrasında karotenoidler içerisinde olan  $\beta$ -karoten ve ürünün besin değerinde artış gözlenmiştir (Misci ve ark., 2021).

Meyve ve sebzelerin içerdiği biyoaktif bileşenler, başka besinlere eklendiği zaman o besinlerin fonksiyonelliği artırdığını gösteren çalışmalar da mevcuttur (Tonyali ve ark., 2020).

Ekstrüde ürünlerin fonksiyonel özelliklerini, meyve ve sebze yan ürünlerinin dahil edilmesiy-le geliştirmek mümkündür. Zengin bir quercetin ve lif kaynağı olan soğan kabuğu, endüstride



atık olarak kabul edilir ve ekstrüde ürünlerin besin değerini artırmak için alternatif bir bileşen olarak kullanılabilir. Ekstrüzyon işleminin quercetin, toplam fenolik içerik ve numunelerin antioksidan aktivitesi üzerindeki etkisini araştıran çalışmada, buğday unu üç seviye (%3, %6 ve %9) soğan kabuğu tozu (OSP) ilave edilerek kontrol (%0 OSP) ile karşılaştırılmıştır. Soğan kabuğu tozu seviyesinin arttırılmasının quercetin içeriğini arttırdığı göstermiştir. Soğan kabuğu tozu seviyesinin arttırılması, numunelerin antioksidan aktivitesini ve toplam fenolik içeriğini arttırmıştır. Bu çalışma, ekstrüzyon işlemi ile ürünlerin biyoaktif bileşenlerin seviyesini artırabileceğini ve biyoaktif bileşenler bakımından zenginleştirilmiş ürünlerin elde edilmesinde etkili olduğunu göstermiştir (Tonyali ve ark., 2020).

Başka bir çalışmada, geleneksel ekmeğe kıyasla, glutensiz ekmeğin pişirme sonrası birçok kusur, daha düşük bir besin ve fonksiyonel değer göstermektedir (Krupa-Kozak ve ark., 2021). Brokoli yaprakları atık ürünler olarak algılansa da, yüksek besin içeriği ve biyoaktif bileşikler açısından önemli bir besindir. Bu çalışma, brokoli yaprağı tozu (BLP) ile zenginleştirilmiş glutensiz ekmeğin gelişmiş glikasyon son ürünlerinin oluşumuna karşı besin değeri, teknolojik kalite, antioksidan özellikler ve inhibitör aktivitenin analizine dayanarak brokoli yaprağı tozunun (BLP) glutensiz ekmeğin bileşeni olarak uygunluğunu ve işlevselliğini değerlendirmiştir. Brokoli yaprağı tozu ile oluşan glutensiz ekmeğin, daha yüksek besin içeriği (proteinler ve mineraller) ve antioksidan özelliği göstermiştir. Sonuç olarak araştırmacılar bu çalışmada, geliştirilmiş teknolojik ve fonksiyonel özelliklere sahip yeni geliştirilen brokoli yaprağı tozu ile zenginleştirilmiş glutensiz ekmeğin, glutensiz diyet yapan denekler için antioksidan kapasitede ve oksidatif stabilitede önemli bir artış sayesinde kronik hastalıkların önlenmesinde ve tedavisinde faydalar sağlayabileceği gözlemlenmiş ancak olumlu sağlık etkilerini doğrulamak için insan müdahalesi çalışmalarına ihtiyaç olduğunu vurgulamışlardır (Krupa-Kozak ve ark., 2021). Meyve ve sebzelerin üretim teknolojilerinde fonksiyonel bileşenlerin eklenmesi ile ilgili yapılan çalışmalar Tablo 2' de verilmiştir. Tablo 2'de verilen farklı fonksiyonel besinlere sahip meyve ve sebzelerin fonksi-

yonelliğini artırmak için farklı yöntemlerin kullanıldığı görülmektedir.

#### 4. Meyve ve Sebzelerde Bulunan Fonksiyonel Besinlerin Sürdürülebilirlik Açısından Değerlendirilmesi

Farklı alanlarda oluşan atık malzemelerin tekrar geri dönüştürülerek kullanılması, hem ülke ekonomisine destek sağlaması hem de atık malzemelerin çevre üzerindeki olumsuz etkilerini azaltması bakımından oldukça önemlidir. Günümüzde özellikle nüfus artışına bağlı olarak gıda üretim fabrikalarının artması sonucunda oluşan gıda atıklarının geri dönüştürülmesine büyük önem verilmektedir. Özellikle fonksiyonel bileşenler (antioksidanlar, polifenoller, fenolik maddeler vb.) bakımından zengin meyve ve sebze atıklarının tekrar geri dönüştürülmesi ile hem besinsel bakımdan zenginleştirilmiş hem de sağlığımız açısından faydalı gıdalar elde edilmektedir (Yağcı ve ark., 2006). Örneğin meyve ve sebze atıkları bağırsak sağlığına, kilo yönetimine, düşük kan kolesterol seviyelerine, glisemik ve insülin yanıtına katkı sağladığı belirtilmiştir (Gomez ve Martinez, 2018). Atık meyve ve sebzeler, karotenoidler, polifenoller, diyet lifleri, vitaminler, enzimler ve yağlar gibi potansiyel olarak değerli biyoaktif bileşenleri içeren tohum, meyve kabuğu ve posadan oluşmaktadır. Bu fitokimyasallar, fonksiyonel ya da zenginleştirilmiş gıdaların geliştirilmesinde, sağlık ve tekstil endüstrilerinde kullanılabilir. Atık malzemelerden elde edilen biyoaktif bileşenlerinin farklı alanlarda kullanılması sürdürülebilirlik kalkınmayı desteklemektedir (Sagar ve ark., 2018). Bu amaçla zengin antioksidan bileşimi içeren domatesin kabuğu ve çekirdekleri değerlendirilmiştir. Stajcic ve ark. (2015), tarafından gerçekleştirilen bir çalışmada beş farklı domates genotipinden elde edilen domates atığı ekstraktlarının karotenoid içeriği, antioksidan ve hücre büyüme aktiviteleri araştırılmıştır. Elde edilen sonuçlar domates atıklarının potansiyel nutrasötik kaynak olarak görülmesi gerektiğini ve fonksiyonel bir gıda maddesi olarak kullanılabilirliğini göstermektedir. Araştırmacılar, antioksidan ve anti-proliferasyon aktiviteleri gösteren domates atığının, insan besin arzını iyileştirmek ve kanser gibi oksidatif hasarın neden olduğu hastalık risklerini

azaltmak için iyi bir karotenoid kaynağı olarak kabul edilmesi gerektiğini vurgulamışlardır. Bir başka çalışmada, zengin bir lif ve polifenol kaynağı olarak bilinen ve elma endüstrisinin bir yan ürünü olan elma prinanın içerisindeki lifin kimyasal yöntemlerle analiz edilerek, kek yapımı üzerindeki etkisi incelenmiştir. Yüksek miktarda diyet lifi içeren elma posasının, kek yapımında diyet lifi olarak kullanılabilmesi tespit edilmiştir (Sudha ve ark., 2007). Gomez ve Martinez (2018), meyve ve sebze atıklarının unlu mamullere ilave edilmesi durumunda çözünmeyen ve çözünür diyet lifinin arttığını gözlemlemişlerdir.

## SONUÇ

İnsanların sağlığı ve zindeliği, büyük ölçüde besleyici gıdaların tüketimine bağlıdır. Çeşitli araştırmalar, gıdaları dejeneratif hastalıkla mücadelede yardımcı olarak ilişkilendirmiştir. Bu nedenle, farklı fonksiyonel özellikler taşıyan besinlerin sağlık üzerine olumlu etkilere sahip olduğu tespit edilmiştir. Kansere, kalp ve damar hastalıkları, obezite (şişmanlık) ve sindirim sistemi hastalıklarının korunmasında meyve ve sebzelerde bulunan biyoaktif bileşenlerin etkili olduğu gözlemlenmiştir. Sağlıklı beslenmede vitamin, mineral gibi besin öğelerini içeren mey-

**Tablo 2.** Meyve ve sebzelerin üretim teknolojilerinde fonksiyonel bileşenlerin eklenmesi ile elde edilen ürünler ve özellikleri

Elde Edilen Ürün	Ürünün Fonksiyonelliğini Artırmak İçin Uygulanan İşlemler	Sonuçlar	Referans
Probiyotik meyve suları	Meyve suyuna mikroorganizma ilavesi ve probiyotik mikroorganizmalarla fermantasyon işlemi	Meyve suları probiyotik taşıyıcı olarak kullanıma uygun olduğu görülmüştür.	Pereira ve Rodrigues (2018)
Fonksiyonel besin açısından zengin siyah havuç	Ultraviyole (UV-C ve UV-B) radyasyon uygulamaları	UV-B tedavisi, kontrol numunelerine göre toplam fenolik içerikte önemli bir artış olduğu, UV-B ve UV-C işlemleri sonucunda antioksidan değerlerinde artış gözlemlenmiştir.	Türkmen ve Takci (2018)
Antioksidan aktivitesi ve toplam fenolik içeriğince zengin olan buğday unu	Zengin bir quercetin ve lif kaynağı olan soğan kabuğunun buğday ununa ilave edilmesi	Soğan kabuğu tozu seviyesinin artırılmasının quercetin içeriğini arttırdığını göstermiştir. Soğan kabuğu tozu seviyesinin artırılması, numunelerin antioksidan aktivitesini ve toplam fenolik içeriğini arttırmıştır.	Tonyai ve ark. (2020)
Besin değeri yüksek, fonksiyonel bileşenler açısından zengin, düşük kalorili marmelat çeşitleri	Biyoaktif bileşenler ve mineral bakımından zengin olan nar suyu ve yeşil çay eklenmesi, şeker yerine bal ve stevia şekeri kullanılması	Stevia şekeri ile kullanılan marmelatta en yüksek fenolik madde miktarı olduğu belirlenmiştir, bal ilaveli marmelatının en yüksek antioksidan kapasitesine sahip olması hammaddeye nar suyu ve yeşil çay sayesinde olduğu düşünülmektedir.	Acoğlu ve Ömeroğlu (2020)
$\beta$ -karoten açısından zengin kabak yaprakları	%3 NaCl ve %3 sükröz ile fermentasyon işlemi	Fermantasyon işlemi sonrasında karotenoidler içerisinde olan $\beta$ -karoten ve ürünün besin değerinde artış gözlemlenmiştir.	Misci ve ark. (2021)
Yüksek besin içeriğine sahip glutensiz ekmekek	Yüksek besin içeriği ve biyoaktif bileşikler açısından önemli bir besin olan brokoli yaprağı tozunun glutensiz ekmeğe ilave edilmesi	Brokoli yaprağı tozu ile oluşan glutensiz ekmekek, daha yüksek besin içeriği (proteinler ve mineraller) ve antioksidan özelliği içermektedir.	Krupa-Kozak ve ark. (2021)

ve ve sebzelerin yer alması oldukça önemlidir. Bu çalışmada fermente gıdalar, meyve sebzeler, hayvansal gıdalar, sert kabuklu sebzeler gibi sınıflandırabildiğimiz fonksiyonel besinlerden meyve ve sebzelerin içerdiği biyoaktif bileşenler incelenmiştir. Çilek, portakal, elma, turunçgiller, domates, sarımsak, kırmızı yaban mersini, üzüm, brokoli, lahana gibi meyve ve sebzelerin biyoaktif bileşenleri ve bu bileşenlerin sağlığımız açısından önemi değerlendirilmiştir. Günümüzde sağlığımızın korunmasında ve hastalıkların önlenmesinde ve tedavisinde etkili olan fonksiyonel besinlerden meyve ve sebzelerin günlük beslenmemizde yeterli miktarda alınması gerektiği vurgulanmıştır. Ancak bilimsel verilerin yetersiz olmasından dolayı beslenme ve sağlık arasındaki bağın detaylı şekilde ele alınması için daha fazla çalışmalara ihtiyaç vardır. Fonksiyonel besin olan meyve ve sebze; kardiyovasküler, kanser, kan basıncı, kan yağlarının fazla olması, diyabet gibi hastalıkların tedavisinde yararlı etkileri oldukça fazladır. Mevcut klinik ve prelinik çalışmalar meyve ve sebzelerin alınmasının bu hastalıkların önlenmesinde olumlu etki gösterdiğini belirlemiş fakat bu konuda daha fazla çalışmalara ihtiyaç duyulmaktadır. Sebze ve meyve yan ürünleri gibi gıda atıklarının verimli bir şekilde kullanılması ile yeni fonksiyonel gıdaların geliştirilmesinde alternatif bir besin kaynağı olabilmektedir. Fonksiyonel gıdaların besin değerini artırmak için kullanılabilir yan ürünler ve yeterince kullanılmayan tarım yan ürünleri üzerinde daha fazla araştırma yapılabilir. Son olarak, sağlık yararları yeterli bilimsel literatürle desteklenen gıdalar, sağlıklı bir yaşam için vazgeçilmez bir besin olma ve halka ve gıda endüstrisine faydalı olma konusunda büyük bir potansiyele sahiptir.

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Review Article/Derleme Makale

# Kardiyovasküler hastalıklarda bağırsak mikrobiyotasının rolü

## *The role of microbiota in cardiovascular diseases*

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### ÖZET

Dünya genelinde kardiyovasküler hastalıkların morbiditesi ve mortalitesi oldukça yüksektir. Bu durum hastalığın patogenezi ile tedavi yöntemlerinin sürekli araştırılmasına neden olmaktadır. Son yıllarda yapılan çalışmalar, kardiyovasküler hastalıkların pek çok kronik hastalıkla ilişkisi olduğu gibi bağırsak mikrobiyotasıyla da ilişkili olduğunu göstermektedir. Bağırsak mikrobiyotası vücudun en büyük endokrin organı olarak kabul edilmektedir. Mikrobiyotanın çeşitliliği genetik ve pek çok çevresel faktörden etkilendiği için bireyler arasında farklılık göstermektedir. Bağırsak mikrobiyotasının, diyet tedavisi, probiyotik tedavi, antibiyotik tedavisi, fekal mikrobiyota transplantasyonu, mikrobiyal enzim inhibitörleri gibi çeşitli uygulamalarla düzenlenmesinin kardiyovasküler hastalıklar için de potansiyel bir tedavi olabileceği düşünülmektedir. Bu nedenle, bağırsak mikrobiyotasının kardiyovasküler hastalıkların oluşumunda ve gelişimindeki rolünü anlamak, hastalığın patogenezinin daha iyi anlaşılmasına ve tedavi için yeni fikirler sunulmasına yardımcı olacaktır. Bu derlemede, bağırsak mikrobiyotası ile metabolitlerinin kardiyovasküler hastalıklarla ilişkisi anlatılmıştır.

**ABSTRACT**

The morbidity and mortality of cardiovascular diseases are quite high worldwide. This situation causes the pathogenesis of the disease and the treatment methods to be constantly investigated. Studies conducted in recent years show that cardiovascular diseases are associated with many chronic diseases as well as with intestinal microbiota. Intestinal microbiota is accepted as the largest endocrine organ of the body. The diversity of the microbiota differs between individuals as it is affected by genetic and many environmental factors. It is thought that the regulation of the intestinal microbiota with various applications such as diet therapy, probiotic therapy, antibiotic therapy, fecal microbiota transplantation, microbial enzyme inhibitors may also be a potential treatment for cardiovascular diseases. Therefore, understanding the role of the gut microbiota in the occurrence and development of cardiovascular diseases will help to better understand the pathogenesis of the disease and provide new ideas for treatment. In this review, the relationship between intestinal microbiota and its metabolites and cardiovascular diseases is explained.

**GİRİŞ**

Kardiyovasküler hastalıklar (KVH), gelişmiş ülkelerde önde gelen mortalite ve morbidite nedenlerinden birisidir. Dünya Sağlık Örgütü; 2018 yılında tahmini 17.9 milyon insanın KVH sebebiyle hayatını kaybettiğini ve bunun bütün küresel ölümlerin %31'si olduğunu bildirmektedir (World Health Organization, 2018). Dünya genelinde obezite, Tip 2 Diabetes Mellitus (T2DM) ve metabolik sendrom gibi KVH risk faktörlerinde gözlenen artış da, kardiyometabolik bozuklukların seyrini önlemek ve değiştirmek için daha etkili stratejiler geliştirilmesini gerektirmektedir (Tang et al., 2017).

Kardiyovasküler hastalık riskinin azaltılmasında beslenme ve fiziksel aktivite önerilerinin yer aldığı yaşam tarzı değişiklikleri etkin rol oynamaktadır (Yeşil and Altıok, 2012, Ravera et al., 2016). Bunun yanı sıra son yıllarda yapılan çalışmalarda KVH gelişiminde bağırsak mikrobiyotasının da etkisinin olduğu, mikrobiyotaya yapılan müdahalelerin konak fizyolojisini etkilediği gös-

terilmiştir (Emoto et al., 2016, Tang et al., 2017, Witkowski et al., 2020). Beslenme alışkanlıkları, intestinal enfeksiyon veya çeşitli çevresel faktörler yetişkin bireylerin bağırsağında yer alan mikroorganizmaların türünde ve miktarında değişikliklere yol açmaktadır. Bu durumun sonucunda gelişen bağırsak disbiyozunun da KVH gelişme riskini etkilediği tahmin edilmektedir (Jin et al., 2019). Bu derlemede, kardiyovasküler hastalıklarla bağırsak mikrobiyotası arasındaki ilişki ve bu ilişkide etkili olan mekanizmalar tartışılmıştır.

**1. KARDİYOVASKÜLER HASTALIKLARIN GELİŞİMİNDE BAĞIRSAK MİKROBİYOTASININ ROLÜ**

İnsan vücudunda çok sayıda bakteri, arke, virüs ve tek hücreli ökaryot bulunmaktadır. Konak ile birlikte yaşayan mikroorganizmalar topluluğu olarak adlandırılan mikrobiyota başta kolon olmak üzere özellikle gastrointestinal kanalda kolonize olmuştur (Tang et al., 2017). Sağlıklı insan bağırsağında *Firmicutes*, *Bacteroidetes*, *Actinobacteria*, *Proteobacteria* ve *Verrucomicrobia* filumlarına ait mikroorganizmalar çoğunlukta olup mikrobiyota ile konakçı arasında simbiyotik bir ilişki bulunmaktadır (Zabell and Tang, 2017).

Bağırsak mikrobiyotası konağın sindirim sürecine, sakkarolitik veya proteolitik olarak sınıflandırılan 2 ana katabolik yolla katılmaktadır. Sakkarolitik yol kısa zincirli yağ asidi (KZYA) üretimi ile sonuçlanırken ikinci katabolik yol protein fermentasyonu ile amonyak, çeşitli aminler, tiyoller, fenoller ve indoller gibi diğer kometabolitlerin üretimine yol açmaktadır (Tang et al., 2017). Fermentasyon sonucu ortaya çıkan metabolitler, bağırsak epitelinden emilerek distal organlara ulaşmakta ve vücut işlevini etkileyebilmektedir (Schiattarella et al., 2019). Mikrobiyotanın sindirim sürecine olan etkisinin dışında vitamin metabolizmasını düzenleme, patojenlere karşı koruma, bağışıklık hücrelerini uyararak bağışıklığı güçlendirme gibi farklı mekanizmalarla konakçı sağlığını etkilediği bilinmektedir (Katsi et al., 2019, Suganya et al., 2021). Bağırsak mikrobiyotası genetik faktörlerin yanı sıra diyet, yaşam tarzı, antibiyotik kullanımı, enfeksiyon hastalıkları gibi çevresel faktörlerden etkilenmekte (Tang et al., 2017), bunun sonucunda da mikrobiyotadaki

mikroorganizmaların miktarı ve çeşitliliği değişmektedir. Özellikle bağırsakta en fazla bulunan *Firmicutes* ve *Bacteroidetes* arasındaki oranın bozulması kronik hastalıkların gelişimine sebep olmaktadır (Schiattarella et al., 2019).

Aterosklerotik plaklarda bakteri DNA'sına rastlanması ve bu plaklarda gözlenen bakteriyel taksonların aynı bireylerin bağırsaklarında da gözlenmesi bağırsak mikrobiyotası ile KVH arasındaki ilişkiyi işaret etmektedir (Koren et al., 2011). Bu konuda yapılan bir çalışmada koroner arter hastalığı olan bireylerin bağırsak mikrobiyota profillerini tanımlamak için terminal kısıtlama fragman uzunluğu polimorfizm yöntemi kullanılmış, *Lactobacillales* ve *Clostridium alt kümesi XIV'de* bir artış ile dışkı örneklerinde yer alan *Bacteroides'te* bir azalma görülmüştür (Emoto et al., 2017). Başka bir çalışmada konjenital kalp hastalığı olan bebeklerin bağırsaklarındaki hem toplam bakteri sayısının hem de *Bacteriodes* ve *Bifidobacter* sayısının sağlıklı bebeklerden daha az olduğu belirlenmiştir (Ellis et al., 2013). Aterosklerotik kalp hastalığı olan bireylerde *Enterobacteriaceae* ve *Streptococcus spp* içeriğinin sağlıklı kontrollerden daha fazla bulunması da KVH olan bireylerde mikrobiyota dengesinin değiştiğini göstermektedir (Jie et al., 2017).

Bağırsak mikrobiyotası konakçı ile, lipopolisakarit (LPS) ve peptidoglikan gibi metabolizmadan bağımsız yollar ile KZYA, trimetilamin (TMA), trimetilamin N-oksit (TMAO) ve safra asitleri aracılığıyla etkileşime girmektedir (Brown and Hazen, 2015). Bağırsak bakterileri, konağın kardiyovasküler durumunu etkileyebilen metabolik ürünler üretebilmektedir. Buna örnek olarak, dolaşımdaki dalı zincirli amino asit metabolitleri, triptofan ve histidin seviyeleri, insülin direnci ve vasküler bozukluk arasındaki ilişkiyi vermek mümkündür.

### 1.1. Trimetilamin ve Trimetilamin N-Oksit

Bağırsak mikrobiyotası, fosfatidilkolin, kolin veya l-karnitin gibi diyet bileşenlerini TMA'ya dönüştürür. Oluşan TMA, konakçı tarafından atılır veya emilerek hepatik flavin içeren monoksijenazlar (FMO'lar) yoluyla TMAO'ya çevrilmektedir (Albenberg and Wu, 2014).

Fosfatidilkolin kırmızı et, kabuklu deniz ürünle-

ri ve yumurtada bulunan bir fosfolipittir. Fosfatidilkolin ile ilişkili olan metabolitlerden l-karnitin ve  $\gamma$ -bütirobetain plazma seviyelerinin artmış kardiyovasküler hastalıklar ile ilişkili olduğu belirlenmiştir (Wang et al., 2011, Koeth et al., 2013, Koeth et al., 2014, Skagen et al., 2016). Yapılan bir çalışmada ateroskleroza olan hastaların serum  $\gamma$ -bütirobetain düzeylerinde artış olduğu ve yüksek  $\gamma$ -bütirobetain ve öncüsü trimetilizin seviyeleri ile kardiyovasküler mortalitenin ilişkili olduğu bulunmuştur (Skagen et al., 2016).

Fosfatidilkolinin bir bileşeni olan kolin alımı da plazma TMAO seviyesini değiştirmektedir (Lawson-Yuen and Levy, 2006). Çalışmalarda plazma TMAO düzeyinin aterosklerotik stenoz ile pozitif kolerasyon gösterdiği ve yüksek plazma düzeyinin kardiyovasküler olay riskindeki artışla ilişkili olduğu belirlenmiştir (Senthong et al., 2016, Tang et al., 2013). TMAO ters kolesterol taşınımını inhibe ederek ateroskleroz gelişimine katkıda bulunmaktadır (Koeth et al., 2013). Ayrıca TMAO'nun vasküler hücrelerde prostaglandin G/H sentaz2 (siklooksijenaz 2), interlökin-6 (IL-6), E-selektin ve nükleer faktör- $\kappa$ B'nin aktivasyonu yoluyla hücre içi adezyon molekülü 1 dahil olmak üzere ateroskleroza neden olan inflamatuvar proteinleri indüklediği (Seldin et al., 2016) ve artan TMAO seviyesinin trombosit aktivasyonunu arttırdığı gösterilmiştir (Zhu et al., 2016). Flavon içeren monoksijenaz 3 (FMO3) ekspresyonu kadınlarda ve dişi farelerde daha yüksek seviyededir (Bennett et al., 2013). Yapılan bir çalışmada, TMA üreten bakteri suşları ile kolonize edilen gnotobiotik dişi farelerin erkek farelere göre daha yüksek TMAO seviyelerine ve hepatik FMO3 aktivitesine sahip olduğu bulunmuştur (Romano et al., 2015). Farelerde FMO3'ün yıkımı ise, plazma TMAO seviyesini ve aterosklerotik lezyonları azaltmaktadır (Shih et al., 2015, Miao et al., 2015).

### 1.2. Kısa Zincirli Yağ Asitleri

Kısa zincirli yağ asitleri (KZYA) bağırsak mikrobiyotası tarafından diyet lifi, dirençli nişasta, prebiyotik gibi besinlerin sindirilemeyen kısımlarının fermantasyonu sonucunda üretilmektedir (Topping and Clifton, 2001, Turnbaugh et al., 2006). Kolondaki KZYA'lerinin büyük çoğunluğu, basit alifatik iki, üç veya dört karbonlu olan

asetat, propiyonat ve bütirattan oluşmaktadır (Topping and Clifton, 2001). KZYA'ları metabolizmasındaki değişiklik, bağırsak bariyerinin bozulması ve bağırsak mikrobiyotasının disbiyozu ile ilişkilendirilmektedir (De la Cuesta-Zuluaga et al., 2018). Eksojen KZYA'ları kısmen splanknik yaktan geçerek endojen dolaşımdaki KZYA ile karışıp periferik kan dolaşımına ulaşmaktadır (Pouteau et al., 2003). Hayvan çalışmaları, KZYA'ların spesifik G proteinine bağlı reseptörlere bağlanabildiğini, böylece kan basıncını ve renin-anjiyotensin-aldosteron sistemini (RAAS) düzenlediğini belirlemiştir (Pluznick et al., 2013, Natarajan et al., 2016). Bazı çalışmalar KZYA'ların konakçının enerji metabolizmasına katıldığını (De la Cuesta-Zuluaga et al., 2018) ve ateroskleroz ile yakından ilişkili olduğunu göstermektedir (Aguilar et al., 2016).

Yüksek lifli diyet ile asetat takviyesi alan hipertansif farelerin bağırsak mikrobiyota profilinin düzeldiği ve kan basıncı, inflamasyon, kardiyak hipertrofi ve kardiyorenal fibrozda iyileşme olduğu belirlenmiştir (Marques et al., 2017). Benzer şekilde, sistolik kalp yetersizliği (KY) olan 84 hasta üzerinde yapılan bir çalışmada, düşük lif alımının disbiyozis ile ilişkili olduğu ve bunun da önemli düzeyde artmış kalp nakli gereksinimine ve yüksek mortaliteye yol açtığı bulunmuştur (Pasini et al., 2016). Bunun aksine genetik dilate kardiyomyopati bir fare modelinde lif ve asetat takviyesinin kardiyomyositleri apoptozdan veya olumsuz yeniden yapılanmadan koruyamadığı gösterilmiştir (Jama et al., 2020). Literatürde yer alan çelişkili çalışma sonuçları nedeni ile KY tedavisinde yüksek lifli diyet ve KZYA takviyesinin kullanımı araştırılmaya devam etmektedir (Marques et al., 2017, Jama et al., 2020, Pasini et al., 2016).

### 1.3. Safra Asiti

Safra asitleri bağırsak mikrobiyota kompozisyonunu etkilemekte ve bağırsaktaki mikrobiyal büyümeyi inhibe etmektedir (Hofmann and Eckmann, 2006). Kolona az bir kısmı ulaşan primer safra asitlerini mikrobiyota dekonjugasyon, 7 $\alpha$ -dehidroksilasyon ve 7 $\alpha$ -hidrojenasyon dahil olmak üzere çeşitli modifikasyonlarla sekonder safra asitlerine dönüştürmektedir (Begley et al., 2005). Safra asitleri, nükleer bir reseptör

olan, glikoz ve lipit metabolizması gibi çeşitli fizyolojik süreçleri düzenleyen farnesoid x reseptör (FXR)'üne bağlanmaktadır (Lefebvre et al., 2009). FXR agonistlerini kullanan çalışmalar, FXR'nin ateroskleroza eğilimli farelerde koruyucu bir rolü olduğunu göstermiştir (Miyazaki-Anzai et al., 2014, Hartman et al., 2009, Mencarelli et al., 2009). Fareler üzerinde yapılan bir çalışmada, FXR fonksiyon kaybı ile ateroskleroz ve lipit metabolizmasındaki defektlerin artması arasında pozitif ilişki olduğu bulunmuştur (Hanniman et al., 2005). Buna karşılık FXR eksikliğinin, aort lezyonlarının ve plazma LDL kolesterolünün azalmasına yardımcı olduğunu bulan çalışma sonuçları da literatürde yer almaktadır (Guo et al., 2006, Zhang et al., 2006). Bu nedenle safra asitleri ile FXR'nin aterogenezdaki rolünün tam olarak anlaşılabilmesi için daha fazla araştırma yapılması gerekmektedir.

Primer safra asitlerinin miyokard üzerinde negatif inotropik ve kronotropik etkilere neden olduğu hayvan çalışmalarında gösterilmiştir (Binah et al., 1987, Joubert, 1978). Yapılan bir çalışmada, safra asidi reseptörü türü olan Takeda G proteinine bağlı reseptör 5 (TGR5) aktivasyonunun, miyokard sağ kalımını ve stres yanıtını iyileştirdiği bulunmuştur (Eblimit et al., 2018). Başka bir çalışmada, KY olan hastalarda kontrol grubuna göre primer safra asit miktarının daha düşük, sekonder safra asiti ile primer safra asiti oranının ise daha yüksek olduğu gösterilmiştir (Mayerhofer et al., 2017). Von Haehling ve ark. (Von Haehling et al., 2012) sekonder safra asidi olan ursodeoksikolik asit verdikleri KY hastalarında, periferik kan akışının iyileştiğini ve inflamasyonun azaldığını saptamıştır. Literatürde yer alan bu sonuçlar göz önüne alındığında, safra asitleri arasındaki dengesizliğin, artış veya azalmanın, KY'nin gelişimi ve progresyonu ile ilişkili olduğu düşünülmektedir.

## 2. KARDİOVASKÜLER HASTALIKLARIN TEDAVİSİNDE BAĞIRSAK MİKROBİYOTA MÜDAHALELERİ

Bağırsak mikrobiyotasının iyileştirilmesi amacıyla uygulanan prebiyotik ve probiyotik tedavi, fekal mikrobiyota transplantasyonu, antibiyotik tedavisi, TMA-liyaz inhibitörlerinin kullanımı gibi yöntemlerin kardiyovasküler hastalıkların



önlenmesinde ve tedavi sürecinde kullanılmasına yönelik çalışmalar bulunmaktadır (Gan et al., 2014, Kumar et al., 2016, Hu et al., 2019, Lam et al., 2012, Roberts et al., 2018). Tüm bunların yanında KVH'ları önleme ve tedavi etmede kullanılan beslenme tedavisinin gösterdiği olumlu etkilerin bağırsak mikrobiyotasının düzenlenmesi ile sağlandığı düşünülmektedir (Anselmi et al., 2021).

Kardiyovasküler hastalıklardan koruyucu olan Akdeniz veya DASH (Dietary Approaches to Stop Hypertension)" diyetleri aynı zamanda KVH mortalitesini de azaltmaktadır (Lopez-Garcia et al., 2014, Xu et al., 2020). Her iki diyet tedavisinin de temelinde saf karbonhidrat, doymuş yağ, kolesterol ve tuz alımının azaltılması, sebze, meyve, posa ve tekli doymamış yağ asitlerinin alımının artırılması yer almaktadır. Bu beslenme değişikliği barsak mikrobiyotasını da iyileştirerek KVH, çeşitli kanser türleri gibi bulaşıcı olmayan kronik hastalıklardan korunmada fayda sağlanmaktadır (Duttaroy, 2021).

### 2.1. Prebiyotik ve Probiyotik Tedavi

Probiyotikler esas olarak bifidobakterileri, mayaları, laktik asit bakterilerini içermektedir. İnflamasyonu inhibe etme, bağırsak mukozal bariyerini koruma ve onarma, bağırsak fonksiyonunu iyileştirme fizyolojik etkilerinden bazılarıdır (Kurtaran, 2021). Gan ve ark. yaptıkları çalışmada, akut miyokard enfarktüsü geçiren sıçanlarda probiyotiklerin (*Lactobacillus rhamnosus* GR-1) sol ventrikül hipertrofisini ve ejeksiyon fraksiyonunu önemli ölçüde iyileştirdiğini belirlemiştir (Gan et al., 2014). Kronik KY olan hastalarda *Saccharomyces boulardii* takviyesinin sol atriyal çap ve ventrikül ejeksiyon fraksiyonu üzerinde olumlu etkisi olduğu bulunmuştur (Costanza et al., 2015). Prebiyotik olan oligofruktozun sıçanlarda inflamatuvar hücrelerin infiltrasyonunu azalttığı gösterilmiştir (Kumar et al., 2016). Koroner arter tıkanıklığına sahip farelerde yapılan bir çalışmada probiyotik olarak *Lactobacillus rhamnosus* GR-1 kullanımının KY gelişimini geciktirebileceği bulunmuştur (Gan et al., 2014). Ayrıca *Methanomassiliococcus luminyensis* B10'un TMA'yı hidrojen ile indirgeyip TMA seviyesini dolayısıyla plazma TMAO seviyesini de azaltarak KVH'yi tedavi etmek için kullanabileceği

düşünülmektedir (Brugère et al., 2014).

### 2.2. Fekal Mikrobiyota Transplantasyonu

Fekal mikrobiyota transplantasyonu (FMT), yararlı mikroorganizmaların sağlıklı konaklardan hastaların gastrointestinal sistemine aktarılması yoluyla bağırsak patojenlerini inhibe etmek böylece bağırsak mikrobiyotasının normal işlevlerini yeniden oluşturmak amacıyla tasarlanmış olası bir terapötik müdahaledir. FMT çoğunlukla *Clostridium difficile* enfeksiyonunu tedavi etmek için kullanılmaktadır. Tedavi edilen vakalarda yan etki görülme sıklığı düşük olmasına rağmen FMT'nin diğer hastalıklara uygulanabilirliği hala tartışılmaktadır (Cammarota et al., 2014). Araştırmalar otolog dışkı transplantasyonunun, antibiyotik kullanımından sonra sağlıklı insanların bağırsak mikrobiyota çeşitliliğini hızla geri kazanabileceğini göstermiştir (Taur et al., 2018). Otoimmün miyokardit geliştiren hastalarda fareler üzerinde yapılan bir çalışmada, FMT bağırsak mikrobiyota ağını yeniden dengelemekte ve miyokarditle ilişkili miyokardiyal hasarı azaltabilmektedir (Hu et al., 2019).

Tüm bu umut vadeden sonuçların yanı sıra FMT ile konakta bulunan endotoksinlerin veya bulaşıcı ajanların da transfer edilme riski bulunmaktadır. FMT'nin KVH tedavisinde kullanılabilmesi için, yan etki ve diğer risklerin olasılığını azaltmak ve uygulamanın verimliliğini artırmak için araştırılması gerekmektedir (Xu et al., 2020). Obstrüktif uyku apneli (OSA) sıçanlarla yapılan bir çalışmada; yüksek yağlı diyetle beslenen hipertansif OSA sıçanlardan, normal diyetle beslenen normotansif sıçanlara FMT yapılmıştır. Çalışmanın sonunda alıcı hayvanlarda da yüksek kan basıncı geliştiği gözlemlenmiştir (Durgan et al., 2016). Kalp yetersizliği risk faktörleri veya mevcut KY olan hastalarda, düşük TMAO içeren bağırsak mikrobiyotası transferinin yüksek TMAO düzeyini azaltabileceği düşünülse de henüz böyle bir klinik çalışma bulunmamaktadır.

### 2.3. Antibiyotikler

Kardiyovasküler hastalıklara yönelik yapılan antibiyotik müdahalelerinin farklı klinik sonuçları mevcuttur. Lam ve ark (Lam et al., 2012). ratlarda vankomisin kullanımının miyokard infarktüsü riskini %27 azalttığını göstermiştir.

İlacın bu etkiyi; bağırsak mikrobiyotasında yol açtığı değişiklik ile leptinin plazma seviyesini azaltarak sağladığı düşünülmüştür. Yapılan ikinci çalışmada leptin baskılayıcı probiyotik olan *Laktobasillus plantarum* 299v uygulamasının miyokart enfarktüsünde %29'luk azalma sağlaması da bu düşünceyi doğrulamıştır (Lam et al., 2012). Ancak yapılan bir çalışmada koroner arter hastalığı olan 4012 kişiye 1 yıl boyunca 600 mg azitromisin ve plasebo verilmiş ve çalışmanın sonunda antibiyotik tedavisinin kardiyak vaka gelişme riskine herhangi bir etkisinin olmadığı belirlenmiştir (Grayston et al., 2005). Andraws ve arkadaşlarının yaptığı meta-analiz çalışmasında da antibiyotik tedavisinin kardiyovasküler olay veya mortalite için önemli bir etkisinin olmadığı sonucuna varmıştır (Andraws et al., 2005). Kardiyovasküler hastalıklarda antibiyotik tedavisinin kullanımı ve bu tedavinin mikrobiyota ile olan bağlantısı konusunda daha fazla klinik araştırma yapılması gerekmektedir.

#### 2.4. Mikrobiyal TMA-Liyaz İnhibitörleri

Yapılan bir çalışmada resveratrol, bağırsak mikrobiyotasını yeniden şekillendirerek bağırsak mikrobiyal TMA oluşumunu inhibe ettiğini, TMAO seviyesini azalttığını ve böylece TMAO kaynaklı gelişen ateroskleroz etkisinin azaldığını bulmuştur. (Chen et al., 2016) Ratlarda yapılan bir çalışmada, kolin analogları kullanılarak TMA sentezinde anahtar enzim olan CutC/D'yi inhibe edilmiş ve bu sayede plazma TMAO seviyesi düşürülerek kardiyovasküler hastalık riski azaltılmıştır (Roberts et al., 2018). Başka bir çalışmada TMA üretimini ve dolayısıyla TMA'nın TMAO'ya dönüşümü azaltmak için batı diyetiyle beslenen farelere, bir TMA-liyaz inhibitörü olan 3,3-dimetil-1-butanol (DMB) verilmiştir. Çalışmanın sonunda TMA ve TMAO üretiminin inhibisyonunun hemodinamik parametrelerin iyileşmesini sağladığı belirlenmiştir (Chen et al., 2017). Ayrıca kolinin TMA'ya dönüşümünde etkili olan kolin trimetilamin-liyaz enziminin inhibisyonu da plazma TMAO seviyesini ve aort lezyonlarını azaltmaktadır (Wang et al., 2015).

#### SONUÇ

Sonuç olarak; bağırsak mikrobiyotasının kalp hastalıkları ile ilişkisi olduğuna dair kanıtlar

giderek artmaktadır. Ancak mikrobiyota bazlı tedavilerin kardiyovasküler hastalıkları başarılı bir şekilde tedavi edip edemeyeceği hala net değildir. Mikrobiyotayı değiştirmek amacı ile antibiyotik ve FMT ile tedavi ciddi yan etkilere neden olabileceği için mümkün görülmemektedir. Mikrobiyota bileşiminin diyet müdahaleleri ile değiştirilmesi veya TMAO gibi proaterojenik metabolit üreten yolların düzenlenmesi ile değiştirilmesi yeni terapötik stratejiler arasındadır. Daha pragmatik bir düzeyde, bağırsak-kardiyovasküler etkileşimlerden sorumlu kesin hücresel ve moleküler mekanizmaları belirlemek için daha fazla çalışmaya ihtiyaç vardır.

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Review Article/Derleme Makale

# Alternatif ve sürdürülebilir bir gıda kaynağı olarak algler

*“Algae as an alternative and sustainable food source”*

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## ÖZET

Algler; güneş ışığı, su ve karbondioksiti biyokütleyle dönüştürebilen hücre fabrikaları olarak bilinirler. Yaygın olarak büyüklüklerine göre sınıflandırılan algler (mikroalgler ve makroalgler), çok fazla çeşitlilik gösterebilen heterojen organizma gruplarıdır. Algler türe, yetiştiği bölge, mevsim, hasat şekli, depolama koşulları ve gıda işleme tekniklerine bağlı olarak değişiklik göstermek ile birlikte, yapılarında yüksek miktarda lipit (%20-80), protein (%39-71) ve diyet lifi içermektedir. Ayrıca sterol, vitamin, pigment,  $\alpha$ -tokoferol,  $\beta$ -karoten, glutatyon, askorbik asit, flavonoidler, hidrokinonlar, fikosiyeninler, prolin, fenolik bileşikler, poliaminler ve özellikle çoklu doymamış yağ asitleri ( $\omega$ -3 yağ asitleri) içerikleri nedeniyle iyi bir besin kaynağı olarak kabul edilmekte ve fonksiyonel gıda üretiminde kullanılmaktadır. Alglerin barındırdıkları bu değerli biyoaktif bileşenler sayesinde antioksidan, antimikrobiyal, antiinflamatuvar ve antikarsinojen etkiye sahip oldukları düşünülmektedir. Uzun yıllardır insan diyetinin bir parçası olarak olan alglerin tüketiminin en fazla görüldüğü ülke Japonya'dır. Alg üretimi konusunda ise Çin ve Endonezya önderlik etmektedir. Algler, gıda olarak kullanımının yanı sıra, gıda takviyesi üretiminde, hayvan yemi olarak, kozmetik ve ilaç endüstrisinde, biyoenerji ve biyoyakıt üretimi sırasında hammadde olarak tercih edilmektedir. Algler azot sabitleyici biyogübreler olarak kullanımlarının

yanı sıra, aynı zamanda sera gazı emisyonunun azaltılması ve biyolojik iyileştirme uygulamalarında kullanılmaktadır. Bu çalışmada alglerin bileşimi, özellikleri, sınıflandırılmaları, üretimi ve hasatı, ayrıca alg yağı hakkında bilgi verilmiştir. Çalışmanın amacı sürdürülebilir, alternatif, yenilikçi ve daha iyi değerlendirilme potansiyeli oldukça yüksek olan bir kaynağa dikkat çekmek, özellikle bir  $\omega$ -3 kaynağı olarak alglerin tanıtılması ve alg kullanımı ile zenginleştirilmiş gıdaların takviye edici olarak insan diyetinde yer alması konusunda bilgi sunmaktır.

## ABSTRACT

Algae are known as cell factories that can convert sunlight, water and carbon dioxide into biomass. Algae, commonly classified by their size (microalgae and macroalgae), are heterogeneous groups of organisms that can vary greatly. Algae contain high amounts of lipid (20-80%), protein (39-71%) and dietary fiber depending on species, the region where it grows, the season, the way of harvesting, storage conditions and food processing techniques. Moreover, due to their there are sterol, vitamin, pigment,  $\alpha$ -tocopherol,  $\beta$ -carotene, glutathione, ascorbic acid, flavonoids, hydroquinones, phycocyanins, proline, phenolic compounds, polyamines and polyunsaturated fatty acids ( $\omega$ -3 fatty acids) contents, they are considered as good food sources and are used in the production of functional food. Thanks to these valuable bioactive components, algae are thought to have antioxidant, antimicrobial, anti-inflammatory and anticarcinogenic effects. The country with the highest consumption of algae, which is a part of the human diet for many years, is Japan. China and Indonesia lead the way in algae production. In addition to its use as food, algae is preferred as a raw material in the production of food supplements, animal feed, in the cosmetics and pharmaceutical industries, and in the production of bioenergy and biofuels. Algae are also used in greenhouse gas emission reduction and biological remediation applications, as well as their usage as nitrogen-fixing biofertilizers. In this study; information about the composition, properties, classification, production and harvesting of algae as well as algal oil is given. The aim of the study is to draw attention to a resource that is sustainable, alternative, innovative and has a high potential for better evaluation and to provide information about the introduction of algae as a source of  $\omega$ -3 and the inclusion of foods enriched with the use of algae in the human diet as supplements.

## GİRİŞ

Algler, güneş ışığını da kullanarak karbondioksiti potansiyel biyoyakıtlara, gıdalara, yemlere ve yüksek değerli biyoaktif maddelere dönüştüren hücre fabrikalarıdır (Paul Abishek vd., 2014). Mikroalgler, yapılarında kuru maddede yüksek miktarda lipit (%20-80), protein (%39-71) ve diyet lifi (bazı türlerde %74.6'ya kadar ulaşmış) içerirler. Sahip oldukları karotenoid, karbonhidrat, sterol, vitamin, pigment, çoklu doymamış yağ asitleri (ÇDYA), özellikle  $\omega$ -3 nedeniyle ek bir besin kaynağı olarak kabul edilirler (Balasubramanian vd., 2011; Akyıl vd., 2016). Ayrıca mikroalgler, içermiş oldukları  $\alpha$ -tokoferol,  $\beta$ -karoten, glutatyon, askorbik asit, flavonoidler, hidrokinonlar, fikosiyeninler, prolin, fenolik bileşikler ve poliaminler gibi bileşikler sayesinde yüksek antioksidan etkiye sahiptirler (Yılmaz, 2019).

Alglerin, arkeolojik kanıtlara dayanarak binlerce yıldır insan diyetinin bir parçası olduğu söylenebilir. Makroalgelere ek olarak, bazı mikroalglerin de gıdalar ve gıda katkı maddeleri için yetiştirildiği belirtilmiştir. Sadece Çin ve Endonezya, alg üretiminin %95'ini karşılamaktadırlar. Bir gıda maddesi olarak alglerin tüketimi en fazla Japonya'da görülmektedir. 2010 ile 2014 yılları arasında alg tüketimi "9.6-11 g/günlük tüketilen makroalg" aralığında değişim göstermiştir. Alg türü, yetiştirildiği bölge, mevsim, hasat, depolama ve gıda işleme tekniklerine göre değişiklik göstermektedir (Wells vd., 2017).

Balık yağları ÇDYA'nın ana kaynağı olarak bilinse de, aslında balıklar bu yağ asitlerini üretmezler, dışarıdan mikroalg tüketerek bünyelerine alırlar. Ayrıca balık yağı gıda takviyesi olarak arzu edilmeyen kokusu ve tadı nedeniyle sınırlı tüketildiğinden, alg yağları alternatif  $\omega$  yağ asitleri kaynağı (özellikle  $\omega$ -3 yağ asitleri) olarak fonksiyonel gıdaların üretiminde tercih edilmektedir (Akyıl vd., 2016). ÇDYA, koroner kalp hastalığını önleme, iltihabı hafifletme, hiperlipidemi ve hipertansiyonu iyileştirme gibi beslenme ve sağlığı geliştirici etkilere sahip olduğundan, bu yağlar ticari olarak giderek daha fazla önem kazanmaktadır (Cebi vd., 2017). Fakat, gıdalardaki lipitlerin yüksek doymamışlık derecesi, daha hızlı oksidasyon reaksiyonları anlamına gelmek-

tedir. Özellikle, alg yağı ile zenginleştirilmiş gıdalarda, lipit oksidasyonuna karşı korunma ve hoş olmayan balık kokularının maskelenmesi büyük bir zorluktur. ÇDYA'nın oksidatif stabilitesinin düşük olması, bu yağ asitlerinin gıdalarda kullanımını sınırlandırır ve bu yağları oksidasyondan korumak için farklı strateji ve teknikler geliştirilmesini gerekli kılmaktadır (De Ciriano vd., 2010).

Algler, gıda (hem insan hem de hayvan beslenmesi), kozmetik ve ilaç endüstrilerini destekleyen çok sayıda ürünün değerli bir kaynağı niteliğindedir (Mercer ve Armenta, 2011). Mikroalgler, verimli güneş enerjisi dönüştürücüleri olduğundan çok çeşitli metabolitler üretebilirler. Biyoenerji üretimi (biyometan, biyohidrojen, biyoetanol) veya biyoyakıt üretimi (biyodizel) ve CO<sub>2</sub> konsantrasyonunu düşürmek için kombine uygulamalarda kullanılabilirler (Demirbas ve Demirbas, 2011). Ayrıca bu fotosentetik mikroorganizmalardan biyolojik iyileştirme uygulamalarında ve azot sabitleyici biyogübreler olarak faydalanılmaktadır (Paul Abishek vd., 2014).

Mikroalglerin bir yakıt kaynağı olarak kullanılması fikri yeni değildir, ancak petrolün artan fiyatı ve daha da önemlisi, fosil yakıtlarla ilişkili küresel ısınma ve sera etkisi ile ilgili ortaya çıkan sorunlar sebebiyle ciddi bir şekilde gündeme alınmaktadır (Paul Abishek vd., 2014). Algler sayesinde, kömür yakıtlı enerji santrallerinden ve diğer karbon yoğun endüstriyel işlemlerden kaynaklanan sera gazı emisyonunun azaltılması amaçlanmaktadır (Demirbas ve Demirbas, 2011).

Alg türevi gıdaların tüketimi, bileşimleri ve potansiyel besin değerleri nedeniyle sağlık açısından faydalı olduğu düşünülmektedir, ancak bu faydaların ve olası yan etkilerin ölçülmesi devam etmektedir. Alg kaynaklı gıdaların, insan metabolizmasında nasıl etkiler yarattığı ayrıntılı şekilde araştırılmalıdır (Wells vd., 2017).

Bu çalışma kapsamında, alglerin bileşimi ve özellikleri, sınıflandırılmaları, alg kültürlerinin üretimi ve hasatı, toplanan biyokütlelerden yağ eldesi, ayrıca alg yağının özellikleri ve kullanım alanları hakkında bilgi verilmiştir.

## 1. ALGLER

Algler; güneş ışığı, su ve karbondioksiti biyokütleye dönüştürebilen, tuzlu veya tatlı su ortamlarında yaşayan fotosentetik mikroorganizmalardır. Gerçek birer minyatür biyokimyasal fabrika gibi çalışırlar (Demirbas ve Demirbas, 2011) ve birçok gıda ağının hayati parçalarıdır (Abubakar vd., 2012).

### 1.1. Alglerin Sınıflandırılması

Algler, yaygın olarak büyüklüklerine göre mikroalg (fitoplankton) ve makroalg (deniz yosunu-ipliksi algler) olarak sınıflandırılmış heterojen bir organizma grubudur (Abubakar vd., 2012; Leal vd., 2013). Özellikle fitoplanktonlar, yüksek üretkenlik gösterirler ve hızla büyüyebilirler. Daha yüksek yapılı bitkiler gibi davranarak, triaçilgiserol formunda lipit depolarlar. Kuru kütlelerinin %20-80'i kadar lipit içerebilirler. Bu oran çoğu zaman kuru biyokütlelerinin %60'ından daha fazla miktarlarda gözlenmektedir (Demirbas ve Demirbas, 2011; Schlagermann vd., 2012).

Ayrıca alglerin; diatomlar, yeşil algler, mavi-yeşil algler ve altın algler olmak üzere dört ana gruba ayrılarak incelendiği kaynaklarda mevcuttur (Demirbas ve Demirbas, 2011; Schlagermann vd., 2012). Algler, diğer deniz ve tatlı su bitkilerine göre çok daha fazla çeşitlilik göstermektedirler (Abubakar vd., 2012).

### Mikroalgler

Mikroalgler, tek hücreli ve basit çok hücreli mikroorganizmaları kapsar (Paul Abishek vd., 2014). En önemli üç mikroalg sınıfı; diatomlar (*Bacillariophyceae*), yeşil algler (*Chlorophyceae*) ve altın algler (*Chrysophyceae*) olarak belirtilebilir. Özellikle diatomlar, fitoplanktondaki baskın yaşam biçimidir ve muhtemelen dünyadaki en büyük biyokütle grubunu temsil ederler. Sadece diatomlar için bile 100 000'den fazla türün olduğu tahmin edilmektedir (Demirbas ve Demirbas, 2011).

Ayrıca mikroalgler, prokaryotik ve ökaryotik mikroalgler olarak da sınıflandırılabilir. Prokaryotik mikroalgere, siyanobakteriler (*Chloroxybacteria*); ökaryotik mikroalgere ise yeşil algler (*Chlorophyta*) ve diatomlar (*Bacillariophuta*) örnek olarak verilebilir (Paul Abishek vd., 2014).



Mikroalgler, özellikle deniz kültüründe yaşayan canlılar için besin kaynağı olarak önemli bir role sahiptirler (Volkman vd., 1989).

Çoğunlukla mikroskopik boyutlarda olan bu organizmalar, karbondioksit varlığında hızla büyüebilirler (Demirbas ve Demirbas, 2011; Paul Abishek vd., 2014). Biyokütlenin kuru ağırlığının %20-50'si kadar yağ içerebilirler. Ekili olmayan arazide, tuzlu su varlığında çoğalabildikleri gibi, atık sularda da görülebilirler ve tüm yıl boyunca bu alglerin üretiminin yapılması mümkündür (Paul Abishek vd., 2014).

Mikroalglerden elde edilen hektar başına yağ verimi; soya fasulyesi, palm, hindistancevizi veya ayçiçeği gibi hammaddelerden elde edilen yağ verimini büyük ölçüde aşmaktadır (Paul Abishek vd., 2014). Mikroalglerin bitkilere göre diğer bir avantajları ise, metabolik esneklikleridir. Bu durum, biyokütlenin biyokimyasal bileşimindeki (lipit, karbonhidrat veya protein) bir varyasyonun, yetiştirme koşullarının değiştirilmesiyle düzenlenebileceği anlamına gelir. Ayrıca, tüm yıl boyunca gerçekleştirilen aşılama, bakım, hasat ve benzeri işlemler tarım uygulamalarına göre çok daha fazla mümkün olmaktadır (Abubakar vd., 2012; Schlagermann vd., 2012; Paul Abishek vd., 2014).

### Makroalgler

Makroalgler, tuzlu veya tatlı suda büyüyen çok hücreli bitkilerdir (Demirbas ve Demirbas, 2011). Yosun olarak da bilinen makroalgler, karmaşık ve dinamik bir sınıflandırma sergilemektedir. Makroalgler pigmentasyonlarına göre, kahverengi algler (*Phaeophyceae*), yeşil algler (*Chlorophyceae*) ve kırmızı algler (*Rhodophyceae*) olarak sınıflara ayrılırlar (Demirbas ve Demirbas, 2011; Leal vd., 2013). Özellikle kahverengi algler, ılıman bölgelerdeki denizlerde önemli miktarda bulunurlar. Bu alglerin gıda, ilaç ve kozmetik endüstrilerinde uygulama alanları mevcuttur (Pereira vd., 2017).

### 1.2. Alglerin Bileşimi

Algler glikoz, nişasta, selüloz/hemiselüloz ve çeşitli polisakkarit türlerini içermektedirler. Kırmızı alglerin ana polisakkarit bileşeni karragenan ve agar iken, kahverengi makroalglerde bulunan

başlıca polisakkaritler gluklan, mannitol ve alginatdır. Alg bileşiminde yer alan bu karbonhidratlar çeşitli gıda ürünlerinde emülgatör, jelleştirici ajan ve stabilizatör olarak kullanılmaktadırlar (Draget vd., 2005).

Protein içeriği, alg grupları arasında büyük farklılıklar sergilemektedir. Protein içeriğindeki bu farklılıklar, mevsimsel ve çevresel koşullardan ziyade, daha çok alglerin türünden etkilenmektedir. Bir ipliksi siyanobakteri olan *Arthrospira platensis* ve tek hücreli yeşil alg *Chlorella*'nın çeşitli ticari türleri kuru ağırlıklarının %70'i kadar protein içerebilirler. Ayrıca bu mikroalgler tüm elzem aminoasitleri de içeren bir profile sahiptirler (Akyıl vd., 2016). Yüksek protein içeriği dışında; vitamin, mineral ve birçok aktif biyolojik maddeyi bünyesinde barındıran *Arthrospira platensis* hücre duvarının %86 sindirilebilirliğe sahip olması ve insan vücudu tarafından kolayca absorbe edilebilen polisakaritlerden oluşması nedeni ile gıdalarda diyet takviyesi olarak kullanılmaktadır. Ayrıca akvaryum balıkları, kümes hayvanları endüstrisi ve su ürünleri yetiştiriciliği sırasında da tercih edilmektedir (Akyıl vd., 2016).

Makroalgler arasında yer alan kırmızı ve yeşil algler: *Porphyra* türleri (laver), *Pyropia* türleri (nori), *Palmaria palmata* (dulce), *Ulva* türleri (deniz marulu) de proteince zengindirler. Her tür için farklılık göstermek ile birlikte, alglerde en fazla yer alan aminoasitler arasında taurin, glutamik asit ve aspartik asit sayılabilir (Wells vd., 2017). Ayrıca *Anabeana* türleri, *Chlorella vulgaris*, *Chlorella pyrenoidosa*, *Dunaliella* ve *Euglena* türleri ve *Scenedesmus obliquus* yüksek protein içeriğine sahip olarak bilinen diğer alg türleri arasında sayılmaktadır (Becker, 2007).

Mikroalglerin bileşiminde yer alan ÇDYA, sağlık üzerine faydası olan önemli biyoaktif bileşenlerdendir. ÇDYA içerisinde bulunan özellikle  $\omega$ -3 ve  $\omega$ -6, elzem yağ asitleri olup insan vücudunda sentezlenemediklerinden mutlaka diyetten dışarıdan alınmaları gerekmektedir (Akyıl vd., 2016). Özellikle  $\omega$ -3 tüketiminin nöral ve görsel gelişim, kalp hastalığı, hipertansiyon, kanser, diyabet, kistik fibroz, astım, artrit, depresyon, şizofreni ve dikkat eksikliği gibi hastalıkların önlenmesi üzerinde yararlı etkileri olduğu düşünülmekte-



dir (Takahata vd., 1998; Kris-Etherton vd., 2002; Chee vd., 2005).

Algler, beslenmede bir hayli öneme sahip  $\omega$ -3 yağ asitleri bakımından oldukça zengindirler (Pereira vd., 2017). Genellikle, öncelikli  $\omega$ -3 kaynağı olarak balıklar akla gelmektedir. Fakat balıkların uzun zincirli ÇDYA'yı sentezleyememeleri, zamanla biyolojik çeşitliliklerinin azalması, sık tüketimlerinin karaciğerde civa birikimi riski oluşturması ve balık yağındaki  $\omega$ -3 yağ asitlerinin istenmeyen lezzet, koku, stabilite sorunları yaratması besin takviyesi ve gıda katkı maddesi olarak balık yağı kullanımını sınırlamaktadır. Bu nedenle, algler iyi bir alternatif  $\omega$ -3 yağ asitleri kaynağıdır (Lenihan-Geels vd., 2013; Cebi vd., 2017). Alg yağları, yüksek  $\omega$ -3 yağ asidi içeriğinin yanı sıra, doğaya zarar vermemesi nedeni ile de özellikle fonksiyonel gıdaların üretimi sırasında tercih edilmektedir. Fakat bu kadar yüksek ÇDYA içeriğine sahip bir yağ kolayca oksidasyona uğrayabilir, bu durum da yağın stabilitesinin ve kalitesinin kolayca bozulması anlamına gelmektedir (Hu vd., 2004).

Makroalglerde baskın yağ asitlerini  $\alpha$ -linolenik asit (ALA) ve dokosaheksaenoik asit (DHA) oluştururken, özellikle kırmızı alglerde baskın yağ asidi eikosapentaenoik asit (EPA)'tir. Mikroalgler de; zooplanktonlar, balık ve diğer çok hücreli organizmalar için DHA ve EPA'nın birincil kaynakları olarak sayılabilirler (Wells vd., 2017).  $\gamma$ -linoleik asit (GLA) ve araşidonik asit (ARA) gibi özel yağ asitleri de alg yağlarının bileşiminde yer almaktadır. *Amphidinium* türleri, *Cryptocodinium cohnii*, *Prorocentrum triestinum*, *Schizochytrium* türleri yapılarında DHA sentezleyebilirken, *Porphyridium cruentum* yapısında yüksek miktarda EPA yer almaktadır (Bellou ve Aggelis, 2013; Akyıl vd., 2016). Ayrıca *Chrysophyceae* (yeşil alg), *Crypthecodinium* ve *Gyrodinium* türleri de iyi birer EPA ve DHA kaynaklarıdır (Makri vd., 2011). *Arthrospira* türleri ve *Porphyridium cruentum* de sırasıyla GLA ve ARA kaynağı olarak belirtilmiştir (Akyıl vd., 2016). Arterburn vd. (2008) tarafından yapılan ve gıda takviyelerinin doğal gıda kaynakları ile eşdeğer olup olmadığı sorgulandığı çalışmada, 20 ile 65 yaş arası 32 sağlıklı erkek ve kadına alg yağı kapsülleri ve pişmiş somon (600 mg DHA/gün olacak şekilde) verilmiş-

tir. 2 haftalık çalışma süresi sonunda, kişilerin plazma fosfolipitleri ve eritrosit DHA seviyeleri belirlenmiştir. DHA seviyeleri her iki grupta da plazma fosfolipitlerinde %80, eritrositlerde %25 artmıştır. Plazma fosfolipitleri ve eritrositlerde meydana gelen DHA düzeylerindeki değişiklikler gruplar arasında benzerlik göstermiştir. Bu sonuçlar, alg yağı DHA kapsüllerinin ve pişmiş somonun plazmaya DHA sağlamada biyoeşdeğer olduğunu göstermiştir. Graeve vd. (2002) tarafından yapılan çalışmada, Antarktika Yarımadası'nda 6 Arktik ve 14 Antarktik makroalg türünün (*Rhodophyta*, *Phaeophyta* ve *Chlorophyta*) yağ asidi bileşimleri incelenmiştir. Makroalglerde, en fazla bulunan yağ asitleri EPA ve palmitik asit olarak belirlenmiştir. Arktik *Palmaria palmata* ve Antarktik *Audouinella purpurea*, çok yüksek oranlarda EPA (sırasıyla %67.3 ve %60.3) ile karakterize edilirken, bu türlerde ayrıca palmitoleik ve araşidonik asit varlığı da saptanmıştır. *Phycodrys rubens* ve *Delesseria lancifolia*'da sırasıyla %35.3 ve %31.1 oranlarında baskın yağ asidi olarak araşidonik asit tespit edilmiştir. *Ptilota gunneri* ve *Rhodymenia subantarktika*'da palmitoleik asit içeriği sırasıyla %39.9 ve %32.7'dir. *Phaeophyta*'da baskın yağ asidi olarak ise palmitik asit bildirilmiştir. *Desmarestia muelleri*'de nadir görülen tekli doymamış yağ asidi palmitoleik asit (%11.1) yüksek miktarda bulunmuştur. Arktik *Prasiola crispa* ve Antarktika *Lambiya antarktika*'sında, baskın ÇDYA linolenik ve linoleik asittir. *Rhodophyta*'da yüksek miktarda EPA tespit edilirken, *Chlorophyta*'da yüksek yapılı bitkilerde görülen stearik asit varlığı gözlenmiştir. Bu çalışma makroalg türleri arasında yağ asidi bileşimi açısından farklılıkları ortaya koymuştur. Van Ginneken vd. (2011) tarafından yapılan çalışmada; iltihaplanma, kardiyovasküler hastalıklar ve zihinsel bozuklukların önlenmesi ile ilişkili olarak makroalgler (*Ulva lactuca*, *Chondrus crispus*, *Laminaria hyperborea*, *Fucus serratus*, *Undaria pinnatifida*, *Palmaria palmata*, *Ascophyllum nodosum*, *Caulerpa taxifolia*, *Sargassum*) araştırılmıştır. Örneklerde  $\omega$ -3 ve  $\omega$ -6, 2-14 mg/g kuru madde (KM) aralığında hesaplanırken, toplam lipid içeriği 7-45 mg/g KM arasında değişim göstermiştir. n-6/n-3 oranı 0.05-2.75 aralığında (çoğu durumda 1'in altında) hesaplanmıştır. Özellikle *P. palmata* ve *S. natans* sırasıyla yüksek miktarda EPA ve DHA içermektedir. Çalışma sonucunda,

deniz makroalgleri ÇDYA için iyi, dayanıklı ve neredeyse tükenmez bir kaynak olarak bildirilmiştir. Schmid vd. (2014) tarafından yapılan çalışmada, farklı mevsimlerde İrlanda Batı Sahili'nden hasat edilen 16 farklı makroalg türünün (dokuz *Phaeophyceae*, beş *Rhodophyta* ve iki *Chlorophyta*) yağ asidi bileşimi araştırılmıştır. Baskın yağ asitleri; palmitik, oleik,  $\alpha$ -linolenik, araşidonik asit ve EPA olarak belirlenmiştir. Yağ asidi profilleri alg grupları arasında ve içinde oldukça değişkenlik göstermiştir. İncelenen çoğu  $\omega$ -3 açısından zengin tür 1'e yakın n-6/n-3 oranına sahiptir, bu da insan sağlığı açısından faydalı oldukları şeklinde yorumlanmıştır. Ayrıca bu çalışmada, farklı mevsimlerde hasat edilen algler arasında, toplam yağ asidi ve EPA içeriğinde, önemli farklılıklar gözlenmiştir. Yılın her iki zamanında da *Palmaria palmata*, araştırılan tüm türler arasında kuru ağırlığın %0.44-0.58'i oranında değişen seviyelerle EPA içerdiğinden, umut verici bir kaynak olarak tanımlanmıştır. Gubelit vd. (2015) tarafından yapılan çalışmada, *Cladophora glomerata* ve *Ulva intestinalis* incelenmiştir. Bu iki makroalgden elde edilen yağın, yağ asidi bileşimleri incelendiğinde, esansiyel ÇDYA seviyeleri dahil önemli ölçüde farklılıklar saptanmıştır. *C. glomerata*'nın biyokütlesinde nispeten yüksek (4.14 mg/g KM) bir EPA içeriği tespit edilmiştir. *U. intestinalis*'in EPA içeriği (0.45 mg/g KM) daha düşüktür. McCauley vd. (2015) tarafından yapılan çalışmada ise, altı farklı Avustralya deniz makroalginin ÇDYA'ları incelenmiştir. Tüm örneklerde baskın yağ asidi, palmitik asit olarak saptanmıştır. Diğer doymuş yağ asitleri kaprik asit, miristik asit, stearik asit ve araşidik asit olarak bildirilmiştir. Çalışma sırasında 31 farklı yağ asidi tanımlanmıştır. Altı türün hepsinde çok farklı n-6/n-3 oranları bulgulanmış ve yeşil deniz yosunu *Ulva* türleri, 2 kat daha yüksek linolenik asit içerdiğinden, en düşük n-6/n-3 oranına sahiptirler. Bu çalışmada *Ulva* türleri, DHA içeren tek tür olmuştur. Altı alg türü de, inflammatuar aracı nitrik oksidin üretimini güçlü bir şekilde inhibe ettiğinden, antiinflammatuar etki göstermiştir.

Alglerin toplam sterol içeriği ve sterol bileşimi de zamanla incelenmiştir ve türlerine göre farklılık gösterdiği belirtilmiştir (Holdt ve Kraan, 2011). Geçmiş yıllarda yapılan çalışmalarda, ya-

paları benzer olduğundan algal steroller, genellikle kolesterol ile karıştırılmıştır. Fakat özellikle kırmızı ve kahverengi makroalglerde baskın sterol, fukosterol olarak belirlenmiştir (Pereira vd., 2017; Wells vd., 2017).  $\beta$ -sitosterol ise alg yağında önemli miktarda bulunan bir diğer sterol olarak bildirilmiştir (Fahy vd., 2005). Fukosteroller, diyabet ve hipertansiyon komplikasyonlarının yanı sıra diğer sağlık sorunlarının da tedavisinde önemli bir değere sahiptirler (Abdul vd., 2016). Ayrıca algler, hücre zarlarının düzenlenmesinde rol oynayan zengin bir sterol kaynağı olarak kabul edilirler (Grattan, 2013). Terapötik uygulamalar açısından düşünüldüğünde, sterollerin antiinflammatuar, antioksidan ve antikarsinogen gibi farklı biyolojik aktivitelerinden de bahsedilmiştir (Pereira vd., 2017). Fitosterollerin, vitaminler gibi bazı biyoaktif moleküllerin öncüleri olduğu, nutrasötik ve farmasötik endüstrilerde de önemli bir rol oynadığı bilinmektedir. Ayrıca algal sterollerin bağırsaktan kolesterol emilimini engelleyerek toplam ve düşük yoğunluklu lipoprotein (LDL) kolesterol seviyelerini düşürdüğü de bildirilmiştir (Francavilla vd., 2010). Pereira vd. (2017) tarafından yapılan çalışmada, Antartika'da görülen altı farklı kahverengi makroalg türünün (*Adenocystis utricularis*, *Ascoseira mirabilis*, *Cystosphaera jacquinotii*, *Desmarestia anceps*, *Desmarestia antarctica* ve *Himantothallus grandifolius*) sterol içeriği ve bileşimi incelenmiştir. Örneklerde ergosterol, brassikasterol, fukosterol,  $\beta$ -sitosterol, kampesterol, kolesterol ve stigmasterol varlığı tespit edilirken, baskın sterol fukosterol olarak bildirilmiş ve miktarı 6.60 ile 48.13 mg/kg aralığında değişim göstermiştir. Ardından en yüksek miktarda belirlenen sterol  $\beta$ -sitosterol olmuştur ve 5.29 ile 16.49 mg/kg aralığında tespit edilmiştir. Ayrıca örneklerde stigmasterol miktarı 2.69-14.84 mg/kg aralığında belirlenirken, kampesterol ise incelenen tüm örneklerde daha düşük konsantrasyonlarda tespit edilmiştir (0.07-0.15 mg/kg).

Alglerin klorofil içeriği de oldukça yüksektir. Ayrıca klorofil dışında karotenoidler ve fikobiliproteinler gibi pigmentleri de sentezlemektedirler. Karotenoidler, fotosentez sırasında ışığın absorbe edilmesinde, oksijenin toksik etkilerine karşı koruyucu olarak ve fototaksid görev yaparken, biliproteinler *Porphyridium* ve *Sp-*

*ulina* türlerinden ticari olarak elde edilmekte ve doğal renk maddesi olarak gıdalarda kullanılabilirler. Özellikle *Spirulina platensis*'den elde edilen mavi doğal pigmentin (fikosiyenin) pazarı oldukça geniştir (Demiriz, 2008).

Yapılan çalışmalar ile mikroalglerin antibakteriyel, antifungal, antiviral etkileri de doğrulanmıştır. Özellikle antimikrobiyal aktivite algin türü ve algin ekstraksiyonunda kullanılan çözücü ile ilgilidir (Özçimen, 2018). Demiriz (2008) tarafından yapılan çalışmada, uygun kültür koşullarında üretilmiş olan alg türlerinin (*Chlorella vulgaris*, *Oscillatoria limosa*, *Oscillatoria limnetica*, *Phormidium tenue*, *Spirulina major*) farklı çözümler (aseton, etanol, hekzan, metanol, n-bütanol, 0.5 M Tris-HCL pH:8.00 ) kullanarak elde edilen ekstraktlarının antibakteriyel aktiviteleri araştırılmıştır. Elde edilen ekstraktların antibakteriyel etkileri disk difüzyon yöntemi kullanılarak *Bacillus subtilis*, *Escherichia coli*, *Staphylococcus aureus*, *Salmonella enteritidis* bakterileri üzerinde denenmiştir. *Spirulina major*'ün incelenen algler arasında en yüksek antibakteriyel aktiviteye sahip olduğu saptanmıştır. Özçimen (2018) tarafından yapılan çalışmada, farklı mikroalg türlerinden elde edilen yağların, çeşitli mikroorganizmalara karşı *in vitro* antimikrobiyal ve/veya antifungal aktivitesi araştırılmıştır. Çalışma sırasında, dimetil sülfoksit, etanol ve metanolde, 50 ve 100 mg/mL olacak şekilde çözülen *Chlorella protothecoides* mikroalg yağının, *Botrytis cinerea* ve *Aspergillus niger* gibi fungal mikroorganizmalara karşı, antifungal etkisi incelenmiştir. Çalışmanın sonucunda, mikroalg yağının farklı çözücülerde ve farklı oranlarda hazırlanmış ekstraktlarının çalışılan patojenlere karşı antifungal aktiviteye sahip olduğu saptanmıştır. Benzer şekilde Yılmaz (2019) tarafından yapılan çalışmada da, mikroalg türlerinden elde edilen yağların fungal mikroorganizmalara karşı antimikrobiyal etkisi ve gıdalarda koruyucu olarak kullanımı araştırılmıştır. Gıdalarda kayıplara yol açan *Penicillium chrysogenum* ve *Aspergillus parasiticus* funguslarına karşı, kimyasal gıda koruyucularına alternatif olabilecek *Chlorella protothecoides* mikroalg yağının antifungal etkinliği incelenmiştir. Yağın, çalışılan *P. chrysogenum* ve *A. parasiticus*'a karşı antifungal aktiviteye sahip olduğu görülmüştür.

Bu sonuçlar doğrultusunda *C. protothecoides* yağının gıda endüstrisinde gıda koruyucu olarak kullanılabileceği belirtilmiştir.

Eleren ve Öner (2019) tarafından yapılan çalışmada, bazı alg türlerinin karbonhidrat, protein ve lipit içeriklerine yer verilmiştir. Çalışma kapsamında farklı alg türlerine ait karbonhidrat içeriği %15-77, protein içeriği %44-76 ve lipit içeriği ise %23-62 aralığında belirtilmiştir. İçeriği oldukça zengin böyle bir kaynağın gıda denemelerinde kullanıldığı çalışmalar da literatürde yer almaktadır. Chee vd. (2005) tarafından yapılan çalışmada, yoğurt oksidatif olarak stabil ve  $\omega$ -3 bakımından zengin alg yağı emülsiyonu ile desteklenmiştir ve ürünlerin oksidatif stabilitesi, peroksit sayısı tayini ve duyu analizler sonucu belirlenmiştir. Takviye edilmiş yoğurtların peroksit içeriği, emülsiyonun ilave edildiği aşamadan etkilenmeksizin depolama işlemi boyunca artış göstermiştir. Eğitimli panelistler tarafından yapılan duyu analizler sonucunda, örneklerde 22 gün depolama süresi sonrası belirgin bir balık tadı algılanmıştır. Hem kontrol grubu hem de takviye edilmiş örnekler, panelistler tarafından orta derecede beğenilmiştir. Blouin vd. (2006) tarafından yapılan çalışmada, *Porphyra yezoensis*, *P. umbilicalis* ve *P. amplissima* üzerinde araştırmalar yapılmış ve tüketici tarafından kabul edilebilirlik seviyeleri duyu analizler ile değerlendirilmiştir. Bu amaçla 67 çocuk ve 84 yetişkin tarafından duyu analizler gerçekleştirilmiştir. *Porphyra* türleri kraker formülasyonuna dahil edilmiştir ve patlamış mısır kaplama materyali olarak kullanılmıştır. Her iki ürünün de kabul edilebilirliği aynı olarak bulgulanmıştır. Ayrıca yeni hasat edilmiş bu alglerin yağ asidi bileşimlerinin de incelendiği çalışmada, EPA ve palmitik asit en baskın yağ asitleri olarak tespit edilmiştir. EPA içeriği taze alglerde 3.2 mg/g kuru madde olarak bulgulanmıştır. Çalışılan bu türlerin  $\omega$ -3/ $\omega$ -6 oranının (2-3:1) gıdaların besin değerine katkıda bulunabilecek seviyelerde olduğu bildirilmiştir. Chee vd. (2007) tarafından yapılan çalışmada, dondurma örnekleri alg yağı ile zenginleştirilmiş ve duyu özelliklerini iyileştirmek amacıyla vanilya ve çilek aromaları kullanılmıştır. Ürünlerin imalatından itibaren 2 hafta içerisinde eğitimli panelistler tarafından gerçekleştirilen duyu analizleri



sonucunda takviyeli dondurmalarda güçlü bir balık lezzeti algılanırken, vanilya ve çilek aromalı örnekler daha çok tercih edilmiştir. Cofrades vd. (2008) tarafından yapılan çalışmada, üç farklı alg türü *Himanthalia elongata*, *Undaria pinnatifida* ve *Porphyra umbilicalis*' den elde edilen emülsiyonlar, farklı konsantrasyonlarda et ürünlerine dahil edilmiştir. Alg ilavesi, et ürününün su ve yağ bağlama özelliklerini geliştirmiştir. Pişirilen takviyeli ürünlerin, sertliği ve çiğnenebilirliği kontrol örneklerine göre daha iyidir ve yapışkanlık daha düşüktür. Et sistemlerindeki renk değişiklikleri algin türünden etkilenmiştir. Kaynakçı (2012) tarafından yapılan çalışmada, insan sağlığına yararlı olduğu düşünülen farklı yağ kaynaklarının et üretiminde kullanımı ve bu yağların ürünlerin kalite kriterleri üzerine etkileri incelenmiştir. Bu amaçla çeşitli oranlarda (%25, 50, 75, 100) hayvansal yağ ile bitkisel yağ (ALG: alg yağı, AF: antepfıstığı yağı, AY: aspir yağı, ÇY: çörekotu yağı, ÜY: üzüm çekirdeği yağı) yer değiştirilmiş ve emülsiyon ürünü olan sosis denemeleri yapılmış ve sosis örneklerinde pişirme kaybı, nem, tekstür, yağ asidi profili, kolesterol, protein, kül, duyu analizler gerçekleştirilmiştir. Ayrıca sosisler 30 gün boyunca depolanmış ve aralıklarla pH, renk, TBARS ölçümlerine bakılmıştır. Çalışma sonucunda ÜY grubu hariç diğer tüm sosis gruplarında nem değerleri istatistiksel olarak azalma göstermiştir. Tekstür özelliklerine bakıldığında ise ALG, AF, ÜY gruplarının sertlik değerleri kontrol grubuna göre zamanla azalma göstermiştir. ALG grubu hariç diğer tüm bitkisel yağ kullanımı ile kolesterol değerleri önemli düzeyde azalma göstermiştir. Tüm sosis deneme gruplarında doymuş yağ asitleri ve tekli doymamış yağ asitleri değerleri azalmış buna karşın ÇDYA değerleri dikkat çekici şekilde artmıştır. Depolama sonucunda, TBARS değerleri ÜY grubu hariç diğer tüm gruplarda kontrole göre daha yüksek olarak tespit edilmiştir. ALG ve ÜY grupları hariç tüm sosis grupları kontrol grupları ile benzer genel kabul edilebilir olarak değerlendirilmiştir. Chen vd. (2016) tarafından yapılan çalışmada ise, suda alg yağı nanoemülsiyonu elde edilmiştir. Püskürtmeli kurutucuda kurutulduktan sonra elde edilen toz ürün, 30 günlük depolama süresinden sonra bile mükemmel bir yapısal davranış sergilemiştir. Doğal antioksidanlar olan  $\beta$ -sitos-

terol ve  $\gamma$ -orizanol ile hazırlanmış formülasyon yoluyla hem birincil hem de ikincil oksidasyon ürünlerinin azaltılması sağlanmış ve böylece oksidatif stabilite de artırılmıştır. Hazırlanmış olan alg yağı yüklü nanoemülsiyon, istenmeyen balık lezzeti açısından daha iyi bir performans sergilediğinden, hazırlanan emülsiyonun fonksiyonel yiyecek ve içecekler için uygun olduğu sonucu bildirilmiştir. Alglerin fonksiyonel gıda üretiminde kullanıldığı daha birçok çalışma literatürde yer almaktadır. Bu çalışmalarda tercih edilmiş olan alg türü, gıdaya ilave edilen formu ve üretilen ürün ile ilgili bilgiler Tablo 1'de özetlenmiştir.

### 1.3. Alg Kültürlerinin Üretimi

Alg kültürü üretimi; tübüler, düz plaka veya diğer tasarımlara dayalı olarak açık ya da kapalı havuzlarda veya kapalı fotobiyoreaktörlerde gerçekleştirilebilir (Patil vd., 2005; Demirbas ve Demirbas, 2011).

Açık havuz sistemleri, alglerin yetiştirildiği sığ havuzlardır. Bu sistemlerde algler için gerekli besin maddeleri yakındaki kara alanlarından gelen akıntı sularından veya kanalizasyon/su arıtma tesislerinden gelen sularından sağlanmaktadır. Özellikle güvenilir sonuçlar elde edilebilen birkaç açık sistem mevcuttur (Demirbas ve Demirbas, 2011). Bazı mikroalgler, koşulların çok spesifik olduğu durumlarda (yüksek tuz ya da alkali) dahi açık üretim için çok uygundur (Aydın, 2014). Açık havuz kültürleri kapalı sistemlere göre ekonomik olarak daha elverişlidir ve kolay düzenlenebilirler, ancak arazi kullanım maliyeti ve su gerekliliğinin fazla olması, uygun iklim koşulları gerektirmesi ve istenmeyen türlerin oluşması olumsuz yönlerini oluşturmaktadır (Demirbas ve Demirbas, 2011; Aydın, 2014; Eleren ve Öner, 2019). *Spirulina* ve *Chlorella*, açık sistemlerde en çok yetiştirilen mikroalg türlerini oluşturmaktadır (Eleren ve Öner, 2019). Bu sistemlerin teknik ve biyolojik sınırlamaları, kapalı fotobiyoreaktörlerin geliştirilmesine yol açmıştır (Demirbas ve Demirbas, 2011).

Fotobiyoreaktörler, alglerin yetiştirildiği farklı tipte tanklar veya kapalı sistemlerdir. Korunaklı, istilacı mikroorganizmalara karşı nispeten güvenli olan bir kültür ortamı sunarlar (Demirbas

ve Demirbas, 2011). Fotobiyoreaktörler, enerji santrali baca gazlarını temizleme veya atık sudaki besin maddelerini kullanma gibi yararlı görevleri yerine getirirken, aynı zamanda alg üretme yeteneğine sahiptirler (Chisti, 2007; Mwangi vd., 2015). Bu sistemler, fotosentetik verimliliği arttırdıklarından, daha yüksek biyokütle üretimini sağlamaktadırlar (Jez vd., 2017). Kapalı sistemler içinde en fazla boru ve plaka şeklinde fotobiyoreaktörler kullanılmaktadır. Bunun dışında torba şeklinde biyoreaktörler de mevcuttur. Fotobiyoreaktörler kurulurken çeşitli şeffaf plastikler veya konteynırlar kullanılabilir. Uzun yıllar süren çalışmalar sonucu; düz panel, tübüler ve dikey-kolon fotobiyoreaktörler gibi yüksek verimli özelliklere sahip birbirinden farklı fotobiyoreaktörler tasarlanmıştır (Eleren ve Öner, 2019).

Bu teknoloji, altyapı maliyetleri nedeniyle açık havuzlara kıyasla nispeten daha da pahalıdır fakat algleri büyütme için çok daha az ışık ve tarım arazisi gerektirmektedir (Demirbas ve Demirbas, 2011). Fakat üretilen suşların özgülüğü de bu sistem açısından dezavantaj olarak sayılabilir (Jez vd., 2017). Fotobiyoreaktör, büyümeyi optimize ederek yüksek yağlı mikroalg türlerinin üretimine imkan sağlamıştır. Bu sistemlerde bir yılda dönüm başına 19 000-57 000 litre mikroalg yağ üretilebilmektedir. Bu değerler, en yüksek verim ile elde edilen bitkisel yağ miktarının dahi 200 katından fazladır (Demirbas ve Demirbas, 2011).

İdeal bir biyokütle üretim sistemi, güneş ışığını rahatça kullanabilmelidir (Patil vd., 2005). Alglerin yetiştirilmesi ortamın aldığı ışık dışında, sıcaklık (çevresel faktörler) ve besin durumuna göre de değişiklik göstermektedir (Volkman vd., 1989). Jiang ve Chen (2000) tarafından yapılan çalışmada, sıcaklığın deniz mikroalgi *Cryptocodinium cohnii*'nin yağ asidi bileşimi, DHA içeriği ve verimliliği üzerine etkileri araştırılmıştır. Mikroalg, çalışılan tüm sıcaklık aralığında (15-30°C) iyi bir gelişim göstermiştir. 30°C'de en yüksek spesifik büyüme oranına saptanırken, düşük sıcaklık ÇDYA'nın oluşumunu desteklemiştir. En yüksek DHA içeriği 15°C'de erken durağan fazda (72 saat) tespit edilmiştir. 25°C'den (48 saat) 15°C'ye (24 saat) geçiş, hücrel DHA içeriğinde %19.9 ve verimlilikte %6.5 artış ile sonuçlanmıştır.

Ayrıca algler; kültür yetiştirme koşulları dışında, reaktör konfigürasyonları ve toplama seçeneklerine göre çevresel yararlılık ve enerji üretimi açısından farklı performans sergilemektedirler (Jez vd., 2017). Algler üretimleri sırasında başlıca besin maddeleri olarak azot ve fosfora da ihtiyaç duyarlar. Büyüme için gerekli diğer elementler arasında Na, Mg, Ca, K (makro besinler) ve Mo, Mn, B, Co, Fe, Zn (eser miktarda) sayılabilir (Aydoğdu, 2019). Tüm bu faktörler iyileştikçe, alglerin üreme düzeyi artar, fakat üretimin belli bir düzeyi aşması halinde alglerin ve diğer bakterilerin faaliyetleri sonucu bulanıklık oluşur (ötrofikasyon), dolayısıyla algler ışıktan tam olarak faydalanamaz hale gelebilirler (Demiriz, 2008).

Yetiştirilen mikroalgler; mikro basınçlı elekler, sedimentasyon, santrifüj, flokülasyon ya da membran filtrasyonu gibi ayırma prosesleriyle fermentasyon ortamından hasat edilebilirler (Aydın, 2014). Uygun hasat yöntemi, seçilmiş olan mikroalgin özelliklerine, yoğunluğuna, boyutuna ve ayrıca istenen ürünün özelliklerine bağlıdır. Seçilen yöntem türden bağımsız olmalı, daha az kimyasal madde ve enerji gerektirmeli ve mümkünse hücre içi materyalleri serbest bırakmalıdır (Aydoğdu, 2019). Toplanan (hasat edilen) biyokütle, vakum altında su miktarı sabit bir değere gelene kadar kurutulur ve yağ ekstraksiyonu için hazır bir hale getirilmiş olur (Aydın, 2014).

#### 1.4. Alg Yağlarının Elde Edilmesi

Ekstraksiyon, yağ elde etmek için yaygın olarak kullanılan bir yöntemdir. Mikroalglerden yağın geri kazanılması sırasında kullanılan ve iyi bilinen ekstraksiyon yöntemleri arasında mekanik presleme (basınç), çözgen ekstraksiyonu (sokshlet ekstraksiyonu), enzimatik, süperkritik ve ultrasonik ekstraksiyonlar sayılabilir (Szentmihályi vd., 2002; Shah vd., 2005; Mercer ve Armenta, 2011). Bu ekstraksiyon yöntemlerinin kimi avantaj ve sınırlamaları mevcuttur. Mekanik presleme, kullanımı oldukça kolay ve çözgen gerektirmezken, işlem sırasında çok miktarda numune gerekir ve işlem yavaş ilerler. Çözgen ekstraksiyonu sırasında kullanılan çözücüler nispeten ucuzdur, sonuçlar tekrarlanabilir fakat organik çözücülerin çoğu yüksek derecede yanıcı ve/veya toksiktir, çözücü geri kazanımı pahalı



ve yoğun enerji gerektirdiği gibi yüksek miktarda çözgen kullanımı vardır. Süperkritik sıvı ekstraksiyonu sırasında, toksik ya da yanıcı bir çözgen kullanılmaz ve işlem oldukça kolaydır, fakat bu yöntemin de yüksek güç tüketimi ve yatırım maliyeti olumsuz yönlerini oluşturmaktadır. Ultrasonik destekli ekstraksiyon da ise, ekstraksiyon süresi oldukça kısa ve çözücü tüketimi azdır, çözücü hücresel materyallere daha fazla nüfuz etmektedir. Bu işlem sırasında da yüksek güç tüketimi söz konusudur (Mercer ve Armenta, 2011). Bu yöntemler arasında özellikle çözgen ekstraksiyonu, alglerden yağ elde edilmesi sırasında sıklıkla tercih edilmektedir. Ekstraksiyon sırasında hücre biyokütlesinden yağı başarılı, verimli ve zarar görmeyecek bir şekilde çıkarabilmek gerekmektedir. Ek olarak, yağın çevresel açıdan en sürdürülebilir şekilde ekstrakte edilmesi de çok önemlidir, bu nedenle çözgen ekstraksiyonu, yağı alg biyokütlesinden almak için her zaman en iyi çözüm olmayabilir (Mercer ve Armenta, 2011; Pragma vd., 2013). Hangi ekstraksiyon yöntemi uygulanırsa uygulansın işlem sırasında ışık, sıcaklık, pH, hava (karbondioksit) ve besin derişimi gibi kültür şartları kontrol altında tutulmalıdır (Aydın, 2014). Hammaddenin türüne bağlı olarak da, bazen yağ ekstraksiyonundan önce biyokütlenin ön işlemi gerekebilmektedir. Hücre parçalamanın amaçlandığı bu yöntemler; mikrodalga uygulaması, sonikasyon, otoklavlama, öğütme, ozmotik şok, homojenizasyon, dondurarak kurutma, NaCl ilavesi vb. olarak sıralanabilirler (Pragma vd., 2013).

Wiyarno vd. (2011) tarafından yapılan çalışmada, *Nannochloropsis* türlerinden yağ ekstraksiyonu gerçekleştirilmiş ve elde edilen yağ analiz edilmiştir. Ekstraksiyon sırasında sokshlet (SE) ve ultrasonik (US) yöntemler kullanılmıştır. Araştırılan iki yöntem sırasında da etanol çözücü olarak seçilmiştir. SE'de çeşitli etanol konsantrasyonu ve süresi denenmiştir. UE'de ise çeşitli etanol hacmi, süresi ve sıcaklığı araştırılmış, tüm denemeler sırasında frekans 40 kHz olarak tercih edilmiştir. SE'de, en iyi kombinasyon etanol konsantrasyonu %70 ve süre 200 dakika olarak tespit edilmiştir. Bu parametreler ile elde edilen örnekte serbest yağ asitliği %9.4 ve sabunlaşma sayısı 286.8 olarak hesaplanmıştır. UE'de ise, 51.6 dakika, %98 etanol konsantrasyonu ve 69.62°C, yağ

veriminin maksimum olduğu parametrelerdir. İki yöntem karşılaştırıldığında, SE'de daha yüksek çözücü konsantrasyonu, daha yüksek serbest yağ asitliği ve sabunlaşma sayısı saptanmıştır. UE ise, daha kısa sürede gerçekleştirilmiştir. Sonuç olarak, her iki ekstraksiyon yönteminin de güçlü ve zayıf yönleri mevcuttur. Ultrasonik ekstraksiyon, geleneksel ekstraksiyona kıyasla nispeten daha kısa bir operasyon süresinde tamamlanırken, ayrıca ultrasonik ortamda çalışılırken daha düşük sıcaklığa ihtiyaç duyulmuştur. Balasubramanian vd. (2011) tarafından yapılan çalışmada da, yeşil alglerden (*Scenedesmus obliquus*) yağ ekstraksiyonu işlemi tasarlanmış ve optimize edilmiştir. Bu işlem için 1.2 kW, 2450 MHz rezonant sürekli mikrodalga sistemi kullanılmıştır. Alg-su süspansiyonu (ağırlıkça 1:1) 80 ve 95°C'ye kadar ısıtılmış ve 30 dakikaya kadar ekstraksiyon işlemine tabi tutulmuştur. Maksimum yağ verimi (%76-77), 95 °C'de 30 dakika boyunca uygulanan işlem sırasında elde edilmiştir. Mikrodalga uygulamasının doymamış ve esansiyel yağ asitlerine zarar vermediği, yüksek kalitede yağ elde edildiği saptanmıştır.

### 1.5. Alternatif Enerji Kaynağı Olarak Algler

Dünya üzerinde ihtiyaç duyulan enerji her geçen gün artmaktadır. Enerji ihtiyaçlarını karşılamak için ilk sırada kullanılan fosil yakıtların sınırlı olması ve gelecekte daha fazla enerji ihtiyacı söz konusu olacağı göz önüne alındığında, fosil yakıtların da bir süre sonra tükeneceği düşünülmelidir (Eleren ve Öner, 2019).

Ayrıca fosil yakıtların kullanımı sonucu CO<sub>2</sub> miktarı önemli ölçüde artmaktadır. Bu durum dünya üzerinde sera etkisi yaratmakta ve küresel ısınmaya sebep olmaktadır. Tüm bunlar göz önüne alındığında, çevre dostu alternatif enerji kaynakları arayışına girilmiştir. Günümüzde güneş, rüzgâr ve okyanus enerjisi, jeotermal, biyoyakıt gibi birçok enerji kaynağı fosil yakıtların yerini almaya başlamıştır (Eleren ve Öner, 2019). Biyoyakıt olarak alglerin kullanımı çevreye karşı nispeten daha zararsızdır (Paul Abishek vd., 2014).

Biyoyakıt üretimi için yenilenebilir bir kaynak olarak alglerin potansiyeli, yüksek büyüme oranları nedeni ile umut vericidir (Schlager-

mann vd., 2012). Biyodizel üretiminde tür ve boyut aralıklarına göre (birkaç mikrometre-birkaç yüz mikrometre) makroalgler veya mikroalgler kullanılabilir (Paul Abishek vd., 2014). Biyodizel üretiminde genellikle tercih edilen mikroalglerle *Pleurochrysis carterae*, *Botryococcus braunii*, *Dunaliella* ve *Chlorella* türleri örnek verilebilirken, makroalgler arasında ise *Gracilaria*, *Sargassum*, *Ulva* türleri örnek verilebilir (Paul Abishek vd., 2014). Yağlı bitkilerden 1 litre biyoyakıt üretimi için yaklaşık 3000 litre suya ihtiyaç duyulurken, %50 lipit içeriğine sahip mikroalglerden 1 litre biyoyakıt elde etmek için gerekli su miktarı 10 ile 20 litre arasında değişmektedir (Schlagermann vd., 2012).

Ayrıca dönüm başına yüksek verime sahip olmalarının yanı sıra, pratik olarak her yerde büyüyebilmeleri, belirli türlerin günlük olarak hasat edilebilmesi, kükürt içermemeleri, toksik olmamaları, daha az atık oluşması, daha az çevre kirliliğine sebep olmaları, biyolojik olarak yüksek oranda parçalanabilmeleri, küspelerinin hayvan yemi olarak kullanılabilmesi ve hatta etanole işlenebilmesi, yüksek miktarda ÇDYA içermeleri nedeniyle soğuk iklimler için dahi uygun olmaları, üretimi diğer fosil yakıtlara göre çok daha ekonomik olması, karbon emisyonunu azaltmaları ve sürdürülebilir bir kaynak oluşu alg yağından biyodizel üretiminin avantajlı yönlerini oluşturmaktadır (Demirbas ve Demirbas, 2011; Paul Abishek vd., 2014).

## SONUÇ

Besin zincirinin önemli bir parçasını oluşturan ve su ortamındaki primer üretici olan alglere, küresel talep gün geçtikçe artmaktadır. Yüksek değere sahip bileşiklerin üretiminde sürdürülebilir bir kaynak olarak algler, geleneksel beslenme ve biyoyakıt üretiminin yanı sıra, fonksiyonel özellikleri nedeni ile sağlık faydaları düşünülerek giderek daha fazla tüketilmektedir. Ülkemiz alg üretimi için gerekli olan güneş enerjisi açısından son derece elverişli olduğundan, kesinlikle alg kültürü üretimi gerçekleştirilmeli ve alglerin sahip olduğu bu üstün özelliklerinden yararlanılmalıdır.

Mikroalg ve makroalg gıdaların ve takviyelerin içeriği hakkında geniş bir literatür mevcut

olsa da, insan sağlığına niceliksel katkılarını değerlendiren çalışma sayısı oldukça azdır. Son yıllarda ticarileşme potansiyeli giderek artan alglerin, hala keşfedilmeyi bekleyen bileşikleri mevcuttur. Çevre dostu bir hammadde olarak tercih edilen algler, daha fazla araştırılmalı ve insan metabolizmasında nasıl etkiler yarattığı üzerine ayrıntılı çalışmalar yapılmalıdır. Özellikle alg yağının oksidatif stabilitesini arttırmaya ve oluşabilecek balık kokusunu maskeleymeye yönelik çalışmalara ağırlık verilmelidir. Alg yağının, yüksek verimi ve özellikle ÇDYA ( $\omega$ -3 ve  $\omega$ -6 yağ asitleri) açısından iyi bir kaynak olması nedeniyle, kullanım alanları daha da genişletilmelidir.

## Teşekkür

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## EK

Tablo 1. Alglerin fonksiyonel gıda üretiminde kullanıldığı çalışmalar

Alg Türü	Yapı (Form)	Ürün	Kaynak
<i>Chlorella vulgaris</i> , <i>Spirulina maxima</i> (mikroalg)	Biyokütle	Makarna	Fradique vd. (2010)
<i>Lamina japonica</i> (makroalg)	Bütün (toz)	Sosis	Kim vd. (2010)
<i>Undaria pinnatifida</i> (makroalg-wakame)	Bütün (toz)	Dana köftesi	López-López vd. (2010)
<i>Himanthalia elongata</i> (makroalg-kahverengi alg)	Bütün (toz)	Tavuk bifteği	Cofrades vd. (2011)
<i>Ascophyllum nodosum</i> (makroalg-kahverengi alg)	Bütün	Ekmek	Hall vd. (2012)
<i>Saccharina latissima</i> , <i>Palmaria palmata</i> , <i>Gracilaria verrucosa</i> , <i>Saccharina japonica</i> (makroalgler)	Bütün	Dondurma, taze peynir, ekmek	Mouritsen vd. (2012)
Kırmızı alg	Bütün (toz)	Yenilebilir film (peynir ve pastırma için)	Shin vd. (2012)
<i>Gelidium corneum</i> (Makroalg-kırmızı alg)	Bütün	Yenilebilir film (füme ördek eti)	Song vd. (2013)
<i>Arthrospira fusiformis</i> (Spirulina)	Biyokütle (toz)	Ekmek	Achour vd. (2014)
<i>Spirulina platensis</i>	Biyokütle	Makarna	De Marco vd. (2014)
Alg (belirtilmemiş)	Alg yağı	UHT süt	Huimin vd. (2014)
<i>Spirulina</i>	Toz	Atıştırmalık	Joshi vd. (2014)
<i>Schizochytrium</i> türleri (mikroalg)	Alg yağı	Çilek aromalı yoğurt	Lane vd. (2014)
<i>Spirulina</i> (mikroalg, mavi-yeşil alg)	Biyokütle	Ekstrüde gıda	Morsy vd. (2014)
<i>Spirulina platensis</i>	Biyokütle (toz)	Makarna	Özyurt vd. (2015)
Kahverengi alg	Bütün	Sosis	Salagean vd. (2015)
<i>Spirulina platensis</i> (mikroalg)	Biyokütle (toz)	Beyaz peynir ve dondurma	Agustini vd. (2016)
<i>Arthrospira platensis</i>	Biyokütle	Ekmek	Ak vd. (2016)
<i>Fucus vesiculosus</i> (makroalg-kahverengi alg)	Ekstrakt (su, aseton, etanol)	Mayonez	Honold vd. (2016)
<i>Spirulina</i> türleri	Biyokütle (toz)	Çikolata aromalı takviye edici toz gıda	Santos vd. (2016)
<i>Dunaliella salina</i> (mikroalg)	Toz	Makarna	El-Baz vd. (2017)
<i>Pyropia yezoensis</i> (makroalg-kırmızı alg)	Enzim ve kültür varlığında fermente edildi	Yüksek tuz içerikli sos (toz karışım)	Uchida vd. (2017)
<i>Nannochloropsis</i> türleri (mikroalg)	Biyokütle	Makarna	Rodriguez De Marco vd. (2018)
<i>Pyropia yezoensis</i> (makroalg-kırmızı alg)	Kültür varlığında fermente edildi	Sos	Uchida vd. (2018)
<i>Spirulina</i> türleri	Biyokütle (toz)	Atıştırmalık	Lucas vd. (2018)
<i>Spirulina platensis</i> (mavi-yeşil alg)	Biyokütle (toz)	Kurabiye	Onacik-Gür vd. (2018)

## EK

**Tablo 1. (devamı)** Alglerin fonksiyonel gıda üretiminde kullanıldığı çalışmalar

<i>Arthrospira platensis</i> , <i>Tetraselmis suecica</i> , <i>Phaeodactylum tricornutum</i> , <i>Chlorella vulgaris</i> (mikroalg)	Biyokütle	Kraker	Batista vd. (2019)
<i>Spirulina platensis</i>	Biyokütle (toz)	Ayran	Çelekli vd. (2019)
<i>Spirulina platensis</i>	Biyokütle	Yoğurt	Da Silva vd. (2019)
<i>Spirulina</i> türleri	Biyokütle (toz)	Atıştırmalık bar	Lucas vd. (2019)
<i>Spirulina platensis</i> (mavi-yeşil alg)	Biyokütle (toz)	Ekmek	Rajmohan ve Bellmer (2019)
<i>Arthrospira platensis</i> (Spirulina)	Biyokütle (toz)	Makarna	Grahl vd. (2020)
<i>Spirulina platensis</i>	Biyokütle (toz)	Buğday ekmeği	İlhan vd. (2020)
<i>Arthrospira platensis</i> (Spirulina)	Toz	Soya fasulyesi içeceği	Niccolai vd. (2020)
<i>Gracilaria gracilis</i> (makroalg-kırmızı alg)	Ekstrakte edilen fikobiliprotein (pigment)	Pankek ve yoğurt	Pereira vd. (2020)
<i>Spirulina</i> türleri	Biyokütle	Makarna	Zen vd. (2020)
<i>Spirulina</i> türleri	Biyokütle (toz)	Sos (toz karışım)	Almeida vd. (2021)
<i>Spirulina platensis</i>	Biyokütle (toz)	Kefir	Atik vd. (2021)
<i>Sargassum boveanum</i> (makroalg-kahverengi alg)	Bütün	Mayonez	Savaghebi vd. (2021)

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Review Article

# Dietary analysis of traditional food cultures in India: An overview of 2600 BCE to the 21<sup>st</sup> century

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## ABSTRACT

Around 415 BC, Hippocrates proclaimed, “let food be thy medicine, and let medicine be thy food.” A report published in Elsevier’s Journal of Archaeological Science: Reports in 2021 by Archeological Survey of India (ASI), New Delhi, that ‘seven similar big-size brown ‘laddoos’ high protein, multigrain ‘laddoos’ (food balls)’ was found at the Harrapan site in Rajasthan India. Excavation results were surprisingly shocking to the world as many civilizations, sects & religions were not born then. Although India has capacity, if one Jute Sack of food grains is staked over another, they will reach moon and can come back and can still cover more than 1/3rd journey of the moon (2.347 times moon distance capacity). But even in 21st century scientific community are still talking about global hunger. Unfortunately, in the 2021 Global Hunger Index, India ranked 101st out of the 116 countries in 2021 GHI scores and fell under the serious group with a score of 27.5. Poshan Abhiyaan’s convergence mission to address malnutrition must be implemented through ensured community mobilization and participation. Multigrain Ladoos were used in ancient times need to be in the mainstream again to alleviate the hidden hunger. The research concludes that India in the past may have more food security than present India. Hence, ayurvedic foods are recommended for different health disorders as diet plays the most critical role in maintaining health parameters.

## INTRODUCTION

The number of people suffering from severe food insecurity worldwide is more than 2.5 times (precisely 2.56 times) from the earlier which is 135 million to 345 million since 2019 as the onset of COVID-19 (United Nations 2021; World Food Programme 2022). Several studies showed that lockdown hardly hit the farming community vis-à-vis their production, livelihoods, and food security (Acharya, 2020; Azim Premji University, 2020; Cebellos et al., 2020; Harris et al., 2020; Jaacks et al., 2021; Totapally et al., 2020; Vikas Anvesh Foundation 2020; Lal et al., 2021; Lindsay et al., 2021). The scores are improving while comparing the earlier Global Hunger Index (GHI) data. India has a Buffer stock of Wheat (602.91 lakh MT) & Rice (299.25 lakh MT), making the pooled value of food grains (902.16 lakh MT) as of June 2021 which is enough to meet the operational need for food grains and exigencies at any moment (Food Corporation of India, 2022). With this capacity, if India stake one Jute Sack of food grains over another, they will reach the moon and can come back and still cover more than 1/3rd journey of the moon (2,347 times moon distance capacity). But the irony is that recently, India ranked 101st out of 116 nations in the Global Hunger Index (GHI) 2021 (IFPRI, 2022). It is estimated that 537 million people are currently suffering from diabetes and this number is estimated to increase globally as time passes with current projections showing this number may rise up to 783 million globally by 2045 (Statista, 2022). India is often referred as the “Diabetic capital of the world” and it accounts for 17% of total diabetic patients globally currently close to 80 million people in India are living with diabetes (Times of India, 2022). Due to this fast lifestyle in the 21<sup>st</sup> century diabetes is increasing and ultra-processed food being in trend in this day and age that has increased obesity (NIH, 2019). The use of palm oil in developing countries in fast food and fried snacks has drastically increased the risk of cardiovascular disease (Zuckerman, 2018). But, fact of the matter is India has the traditional wisdom of Ayurveda. Ayurveda is a union of 2 words from the Sanskrit language, ‘ayus’, meaning ‘life’ and ‘veda’, which denotes ‘science’; thus, Ayurveda precisely means the

‘science of life. There are different food habits of the Indian population. Different food habits have their own pros & cons. Every Ayurvedic food is traditional, but every traditional food cannot be called Ayurvedic in nature. Mostly, traditional & Ayurvedic foods are considered the same, but there is a pinpoint difference between these, which has been discussed in detail in this manuscript. Ayurvedic foods are very useful and are found to be balanced & best for consumption for a healthy lifestyle. Ayurvedic foods can prevent various diseases. This study focuses on the traditional food culture prevalent in our country.

## History

Urbanization began in the Indus plains in the Indian subcontinent around 2600 BC. The world’s largest metropolises at that era were Harappa and Mohenjo-Daro, which rose to prominence around 2600 BCE (Ahamed et al., 2019). The Harappan folks consumed protein-rich, multigrain “laddoos” (edible balls) more than four thousand six hundred years ago (4600 years ago), according to a scientific investigation of the discovered material in the course of excavation in Rajasthan, which reveals the residents of Harappa practiced agriculture under favorable climatic circumstances. In 2017, while excavating a Harappan archaeological site at a settlement, seven “laddoos” were found in village 4 MSR (4 MSR is the name of a village and has no specified full form; 29°12’ 24.48” N, 73°09’ 20.16” E) (formerly called Binjor) in Anupgarh Tehsil in Sri Ganganagar district of western Rajasthan near the Pakistani border amid 2014 & 2017 (Agnihorti et al., 2021; Tewari, 2021). At the Harappan site in the Anupgarh Tehsil of Rajasthan, seven large brown “laddoos,” two bull sculptures, and a hand-held copper adze (implement similar to an axe for shaping & cutting wood) were discovered. These well-preserved food balls, which date to roughly 2600 BCE, were discovered. (Agnihorti et al., 2021; TFPJ, 2021). Moong dal, millet, cereals and other legumes were the main ingredient in these laddoos. Finger millet was the richest source of calcium and iron. Calcium deficiency leads to bone and teeth issues; while iron deficiency

leads to anaemia and these anomalies can be reduced by incorporating finger millet in one's daily diet (Pragya & Rita, 2012). Later, complete organic-geochemistry of laddoos was carried out at BSIP (Birbal Sahni Institute of Palaeosciences) & NBRI (National Botanical Research Institute), Lucknow, India. Multinutritious compact "laddoos" were consumed by the people of Harappa as a meal supplement for immediate nutrition. Multigrain Laddoos used in ancient times, need to be in the mainstream again to alleviate the hidden hunger.

## MATERIALS AND METHODS

Analysis of data that has already been acquired by others is referred to as secondary data analysis (Srivastava & Lal, 2021). In this research, secondary data were used to show the changing scenarios of the country's nutritional status with various parameters from the primary data collected by the Ministry of Health and Family Welfare (MoHFW) and the International Institute for Population Sciences (IIPS & ICF, 2021). National Family Health Survey (NFHS-5), 2019-21; and the hunger status of various countries from the primary data collected by the International Food Policy Research Institute

(IFPRI) in the Global Hunger Index Report (IFPRI, 2022) have also been investigated.

## RESULTS AND DISCUSSION

The world hunger is increasing and people are suffering from malnutrition and scarcity of food. Due to this people die and become vulnerable to many health problems. Situations like COVID-19 global pandemic, and the Russia-Ukraine war drastically escalated the problem of hunger and food insecurity globally.

### World Hunger Index 2021

The Global Hunger Index computes hunger globally, regionally, and by nation. As shown in Table 1, India ranked 101 among 116 countries in the World Hunger Index of 2021, scoring 27.5, thus falling under the "serious" category.

In Fig. 1, different categories have been shown vis-à-vis the condition of the countries based on their ranking in the World Hunger Index 2021, as shown in Table 1 (IFPRI, 2022).

### Breastfeeding Status in India

Breastfeeding is the most effective way to ensure children's health & survival right from the birth

**Table 1.** World Hunger Index Ranking in the 21<sup>st</sup> century.

2021 Rank	Countries	2000	2006	2012	2021
101	Republic of India	38.80	37.40	28.80	27.50
102	Papua New Guinea	33.60	30.30	33.70	27.80
103	Islamic Emirate of Afghanistan	50.90	42.70	34.30	28.30
103	Nigeria	39.50	32.50	30.40	28.30
105	Republic of Congo	34.90	34.60	28.50	30.30
106	Mozambique	48.00	38.20	31.50	31.30
106	Sierra Leone	57.70	52.70	34.70	31.30
108	Timor-Leste	-	46.10	36.20	32.40
109	Haiti	42.00	43.60	35.20	32.80
110	Liberia	48.10	40.00	35.00	33.30
111	Republic of Madagascar	42.80	41.60	34.30	36.30
112	The Democratic Republic of the Congo	50.60	45.30	42.30	39.00
113	Republic of Chad	50.80	51.20	45.70	39.60
114	Republic of Central Africa	48.90	48.00	40.50	43.00
115	Republic of Yemen	41.00	38.80	38.40	45.10
116	Federal Republic of Somalia	58.10	57.90	65.10	50.80

Low	Moderate	Serious	Alarming	Extremely Alarming
(≤ 9.9)	(10.0–19.9)	(20.0–34.9)	(35.0–49.9)	(≥ 50.0)

**Fig.1.** Colour-coded World Hunger Index Continuum (Low to Extreme)

to up to 6 months. As in this time span child is solely dependent on the mother and breast milk for their proper physical & mental development. Breastfed children perform better on IQ test (WHO 2022).

The percentage of new-borns who were breastfed within 1 day of birth has been shown in Fig. 2. India has been divided into 6 zones viz., North, Central, East, West, South, and North-East, as per breastfed nature of the new-born. The breastfeeding status of the states having the maximum and the minimum percentage per zone is given. In the North zone, Ladakh had the highest breastfeeding percentage (92.5%) while Chandigarh had the lowest breastfeeding percentage (73.4%). For the Central zone, Chhattisgarh has the highest (92.1%), and Uttar Pradesh has the lowest (81.0%) percentage. In the East zone, Odisha accounts for the highest (93.3%) & Bihar for the lowest (84.5%). In the Northeast zone, Meghalaya has the highest (94.6%) & Arunachal Pradesh has the Lowest (81.7%) percentage. Goa has the highest (92.6%) in the West zone, whereas Gujarat has the lowest (85.9%) percentage. For South zone, Lakshadweep has the highest (99.1%), and

Telangana has the lowest (87.8%) value. The overall breastfeeding status country stands at 86.8% (NFHS- 5, 2021).

At Fig. 3 unveiled the different reasons for which growing children require nutrition, viz., for development, physical activities, learning, and better immunity. In the COVID-19 pandemic, immunity played a vital role which is directly related to good nutrition.

### Comparison of Nutritional Status of Children (NFHS-4 vs. NFHS-5)

The frequency of stunting and under-weight has dropped since 2015-16. As shown in Fig. 4, stunting reduced from 38% in 2015-16 to 36% in 2019-21 during the last 5 years. The prevalence of wasting has decreased during the same time frame, falling from 21% in 2015-16 to 19% in 2019-21.

Now it's time to think a way out to overcome from this vicious cycle of hunger. For this problem, awareness programme like National Nutrition Week will come handy.

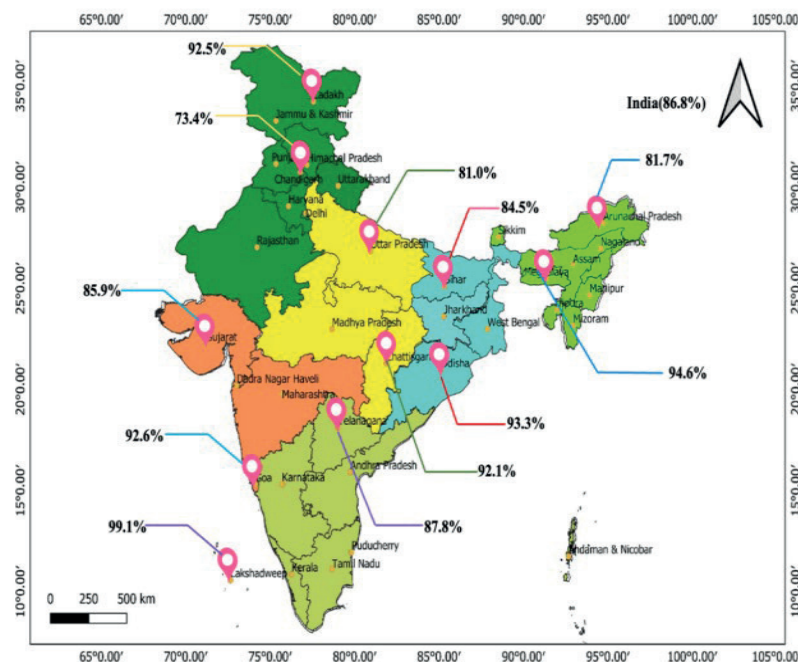


Fig. 2. Percentage of the new-born who was breastfed within 1 day of birth (Source: NFHS- 5, 2021)

### Themes of National Nutrition Week

National Nutrition Week is celebrated annually from 1<sup>st</sup> to 7<sup>th</sup> September in India, and since 2018 the Government of India has been celebrating National Nutrition Month (Lal and Kumari, 2022). The week is celebrated to raise awareness among people regarding proper nutrition and diet. Table 2 exhibits the themes of National

Nutrition Week for the past 10 years.

Five Keys to a healthy diet are recommended by WHO (2020), as shown in Fig. 5. If one follows these 5 keys, then the propensity for healthy life and body increases.

Every Ayurvedic food is traditional, but not every traditional food can be Ayurvedic.



Fig. 3. Nutritional requirements of growing children

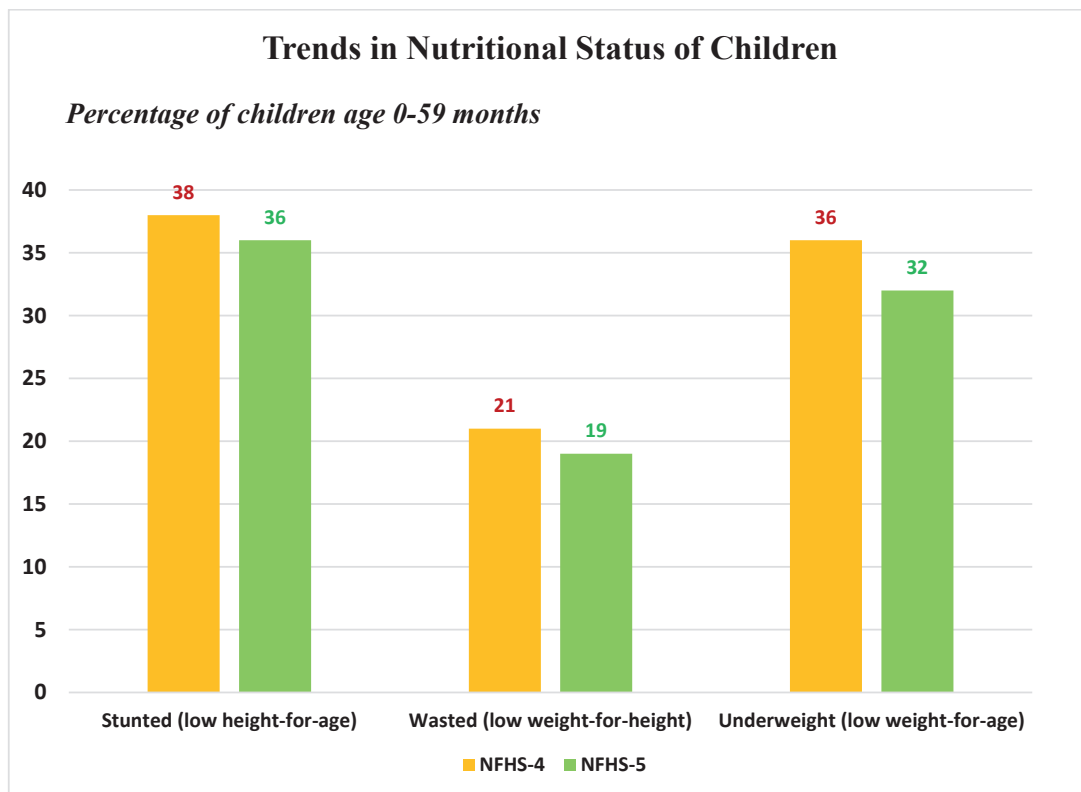


Fig. 4. Trends in nutritional status of children (Source: NFHS- 5, 2021)



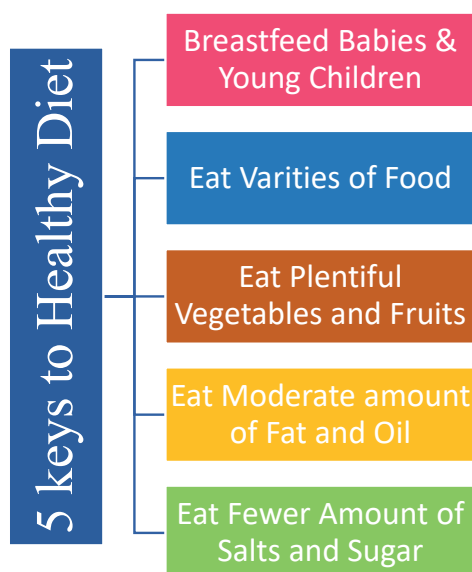
Ayurveda is one of the oldest healthcare systems that evolved in the Indian Subcontinent. The word Ayurveda encompasses 2 words- ayu (life) and veda (knowledge), it deals with various aspects related to health and wellbeing in their diverse aspects, such as happy life, healthy body, and longevity (Payyappallimana & Venkatasubramanian, 2016). Ayurvedic foods consists of satvika food and are thought to be pure and balanced food, offering feelings of calmness, happiness, and mental clarity. The word "sattvika" means "pure essence". Table 3 divulged the difference between Ayurvedic and Traditional Foods in India.

## CONCLUSION

After spreading awareness through National Nutrition Week, various government-sponsored schemes, and other awareness programmes, their impact must be accessed. For this proper questionnaire should be framed and by testing its reliability and validity by using various tools and techniques viz., Kappa coefficient, quartile method, intraclass correlation by Fisher. In comparing chief cereals, the finger millet grains have a much higher level of calcium (>300 mg/100 g). Finger Millet is treated as an orphan crop that may help in mitigating the calcium deficiency in the semi-arid tropics of Asia and Africa. So, nutritionally the finger millet is one the richest

**Table 2.** Theme of National Nutrition Week for the past 10 years

S.No.	Year	Theme
1	2013	Dinnertime Project – Eat, Cook, and Enjoy
2	2014	Nutritional Diet: Country's Foundation
3	2015	Better Nutrition: Key to Development
4	2016	Life cycle approach for better Nutrition
5	2017	Optimal Infant & Young Child Feeding Practices: Better Child Health
6	2018	Go Further with Food
7	2019	Nutrition Behaviour, Every home
8	2020	Eat Right, Bite by Bite
9	2021	Feed smart right from the start
10	2022	Celebrate a World of Flavors



**Fig. 5.** Five keys to a healthy diet (WHO, 2020)

source of Fe and Ca among all crops. In present India, millet festivals are still observed (Niyogi, 2018). The research concludes that a healthy, nutritious diet is the need of the mitigation both ends of the continuum i.e., from wasting (acute malnutrition) to obesity.

**Table 3.** Difference between Ayurvedic and Traditional Foods

S.No.	Particulars	Ayurvedic Food	Traditional Food
1.	Food Categories	Consists of only Satvika food	Consists of Satvika, Tamsika & Rajsika foods
2.	Therapeutic Effect	More	Comparatively less
3.	Immunity	Enhances immunity through Rasayana therapy	No therapy is used but can induce immunity
4.	Focus Area	More on health	More on taste & flavors
5.	Knowledge	Gained through ancient texts like Charaka Samhita & the Sushruta Samhita	Gained from ancestors and passed on from generation to generation
6.	Food Diversity	Less	More
7.	Principles	5 viz., Air, Water, Space, Earth, and Fire	No such principles
8.	Balance	Balancing diet depending on prakriti (Nature) of food	Balancing diet based on food ingredients
9.	Nutritional Element	Panchamahabhuta (5 elements)	Calories, protein, fats, carbohydrates, vitamins, minerals
10.	Variation according to region	Do not varies	Varies according to different regions
11.	Why we eat	To take in prana to live	Personal preference, habit, flavors etc.
12.	Dietary Recommendations	Depends on the nature of the food	Depends on the food groups
13.	Effect on	Body, mind and soul	Body specifically
14.	Importance	Deals with Tridosha namely vata, pitta, and kapha	Calorific value of food
15.	Documentation	More	Less
16.	Examples	Herbal rice drink, Bamboo shoot curry, Jaljira powder juice	Saag, Rabdi, Ginna

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