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Provide a context or background for the study (i.e.,the nature of the problem and its significance). State the specific purpose or research objective of, or hypothesis tested by, the study or observation; the research objective is often more sharply focused when stated as a question. Both the main and secondary objectives should be made clear, and any pre-specified subgroup analyses should be described. Give only strictly pertinent references and do not include data or conclusions from the work being reported.

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Present your results in logical sequence in the text, tables and illustrations, giving the main or most important findings first.

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Cook NR, Rosner BA, Hankinson SE, Colditz GA. Mammographic screening and risk factors for breast cancer. American Journal of Epidemiology. 2009 Dec;170(11):1422-32. doi: 10.1093/aje/kwp304.

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Conclusion should elucidate how the results communicate to the theory presented as the basis of the study and provide a concise explanation of the allegation of the findings.

ACKNOWLEDGEMENT

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Synergistic Role of Probiotic Based Diet in Managing IBD

OPEN ACCESS

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Inflammatory bowel disease (IBD) is characterized by repetitive episodes of inflammation of the gastrointestinal tract caused by an abnormal immune response to gut microflora. Gut flora and diet act as vital role for cause and management of Inflammatory bowel disease as the gut microbiota variety and richness is determined by food we eat on daily basis. High fiber diet and fermented products increase the amount of beneficial bacteria Lactobacillus and Bifidobacterium in the gut while high fats, sugar or processed food increase the amount of pathogenic bacteria and they are responsible for gut inflammation. In Pakistan rising cases of inflammatory bowel disease are due to unhealthy diets, sedentary lifestyles, stress, and misuse of widespread use of antibiotics for self-medication disrupts gut balance, leading to adverse side effects, increased costs, and complications in managing disease.

An imbalance between the types of organism present in a body's natural microflora, especially that of the gut, contribute to a range of conditions of ill health or dysbiosis Many cases of inflammatory bowel disease in Pakistan go undiagnosed, which may be linked to the rising number of colorectal cancer. Studies show that ulcerative colitis is more common among South Asians, including Pakistanis, than in Europeans. It is expected that in 2030 the prevalence of IBD, will be significantly increasing with some of the highest rates of increase seen within pediatrics and the elderly populations due to consumption of junk foods. IBD growing issue demands sustainable and targeted solutions to ease the healthcare burden and improve quality of life. In current years, probiotic-based therapies have emerged as promising adjuncts in IBD management. Probiotics are live beneficial microorganisms that work by restoring microbial balance, enhancing mucosal barrier function, and modulating the immune response. Several clinical and preclinical studies have demonstrated that certain probiotic strains can reduce intestinal inflammation, alleviate symptoms, and even promote mucosal healing in IBD patients. Human health and disease are significantly influenced by the gut microbiome. Human gastrointestinal tract is home to trillions of microorganisms that form a dynamic and essential ecosystem. In healthy individuals, these microbes maintain immune homeostasis, protect the mucosal barrier, and inhibit pathogenic organisms. In IBD, however, this balance is disrupted a phenomenon known as gut dysbiosis. This microbial imbalance leads to an overactive immune response and sustained inflammation

Probiotics have ability to alter gut composition and rejuvenate the growth of useful gut microbes, therefore there is a need to develop probiotic based products improve the gut-microbiota. This method can be regarded as a safe alternative for an over-reliance on antibiotics and may strengthens immunity, may cultivate a healthy gut flora, and will help in fighting infections. Mutually dependent approaches for managing IBD are probiotic and diet. As diet While diet shapes gut environment while restore microbial balance by probiotics and together they deal a harmless, natural, and holistic attitude to regulate inflammation and recover patient health state in low resources.

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Review Article

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A Review on Nutritional Composition and Bioactive Compounds of Black Wheat (*Triticum aestivum* L)

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ABSTRACT

Black wheat (*Triticum aestivum* L) is a pigmented wheat type that is nutritionally superior. It is packed with bioactive chemicals, such as anthocyanins. Black wheat has different nutritional properties and medicinal potential, and more iron (60%), zinc (35% more), vitamin B3, vitamin B5, vitamin E, vitamin K, dietary fiber, and antioxidants than the average food. The high concentrations of phenolic acids, carotenoids, flavonoids and essential amino acids in black wheat provide it with powerful antioxidant, anti-inflammatory and antibacterial capabilities. With anthocyanin levels up to 28 times greater than standard white wheat, this kind of wheat is effective against oxidative stress-related metabolic syndrome and chronic medical issues like diabetes, cancer, obesity and heart disease. Additional compounds like tricin derivatives, ferulic acid, quercetin and luteolin present in black wheat help neuroprotection, manage blood sugar and boost immunity. Due to its diverse composition, black wheat shows promise as a nutritional supplement, therapeutic diet, and long-term answer to today's food problems.

INTRODUCTION

The hybridization of purple and blue wheat cultivars produces black wheat, a colored variety of wheat. Black wheat gets its colour from anthocyanins in the pericarp and aleurone layers. The Shanxi College of Agricultural Science's Institute of Crop Genetics was the first to cultivate black wheat 20 years later. In 1970, the long-term endeavor began. His name was "Black 76." It was the original black wheat type ever developed. The gradual creation of this variety was achieved by uniting "blue-purple 114" with "purple 12-1" in a cross-pollination that occurred as a result of a pericarp mutation. Black wheat, instead of a whole grain, is a type of seed that is consumed as food. Their speciality is that they do not grow on grass like other cereals. These are included in the group of other common pseudo-cellular grains, quinoa and amaranth [1]. Because of its high nutrient density, pleasant flavour, and beneficial effects on health, black wheat has become the most talked-about pigmented wheat type. It is also used to

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manufacture functional meals and colorants, with the main goal of creating items that are much better than conventional wheat [2]. Nevertheless, studies are being carried out using the given data to enhance its nutritional and functional properties, economic value, and outcomes to the fullest extent possible (Figure 1).

Figure 1: Black Wheat Plant and Seed

This study aims to compile data on the bioactive components and nutritional profile of black wheat. In addition to highlighting the health advantages of black wheat, this study will compare it to conventional white wheat.

Nutritional Composition of Black Wheat

Black wheat contains high nutritional compounds like protein, carbohydrates, dietary fiber and minerals (Figure 2).

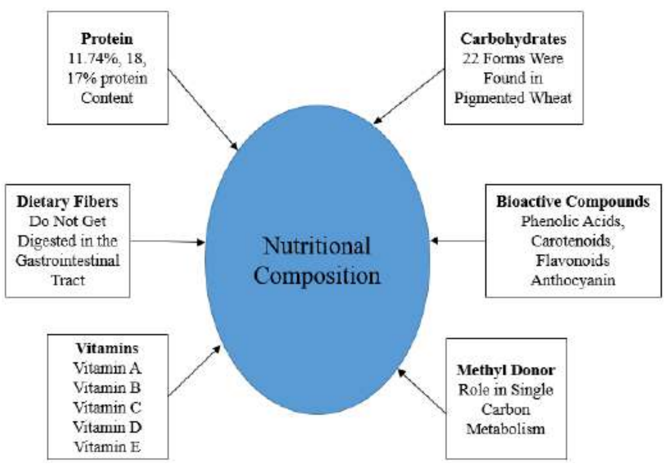

Figure 2: Nutritional Composition of Black Wheat, Source: [Ref: 25]

Protein

A good source of all ninety-six amino acids and their byproducts is black wheat. The bulk of the storage proteins found in black wheat are glutamine and gliadin, which are prolamins that are soluble in alcohol. The calculated total amino acid content is 15.74%, but the required concentration is around 4.45%. A lower gelatin index of 69.74 is used to evaluate the ultimate quality of black wheat flour, as compared to regular wheat, which has a higher value of 98-99. However, black wheat flour may still be used to make bread as its antigen index is still within the optimal range of 60 to 90. Because of this, black wheat may now be processed into low-gluten food products that those suffering from celiac disease can consume instead [3].

Carbohydrates

One of the most important components that provide energy is starch, the primary carbohydrate found in wheat. In the kernel, it is found in the endosperm, also called the flour. A total of 22 different carbohydrate types were discovered in wheat cultivars with red, blue, purple, and black coloring. Black wheat has a higher polysaccharide content compared to white wheat and 75% of the carbohydrates in black wheat are found in the endosperm [4]. It was found that moved lines and black donor lines had a greater amount of sugars and carbohydrates compared to blue and white wheat lines, as well as lines generated through crossing white with pigmented types of wheat (purple, black and blue) [5].

Dietary Fibers

The gastrointestinal tract's enzymes are unable to break down several polysaccharides, including cellulose, hemicellulose, pectin and others that are part of dietary fiber, which increases the food's bulk and, as a result, makes starch granules less accessible to the digestive tracts enzymes. Total dietary fibers (TDF) make up approximately 1.15 % of the weight of black wheat, and insoluble fiber, which makes up a larger portion [6]. Constipation can be avoided and intestinal transit can be stimulated by insoluble fiber, which gives the body and faces volume.

Vitamins

Black wheat contains 19 different vitamins. Bone metabolism and blood coagulation are mediated by vitamin K. A particularly good source of vitamin K is black wheat. Energy metabolism, DNA synthesis, and the preservation of healthy skin are all impacted by vitamin B3 [7]. The main focus of vitamin E is preventing hemolysis of the red blood cell. Vitamin B5 is involved in gland function, hormone release, blood cell development, and the transformation of food into energy. The BGW 76 variety has a vitamin K content of 11.47 mg/. Significant levels of B3, B5, and E have been found in black wheat.

Minerals

A person's normal development depends on the creation of bones, which in turn requires calcium. It was found that compared to Jinchun 9 (a conventional wheat variety), black-grained grains 76 (BGW-76) have four times the amount of calcium [8]. A person's teeth and bones would not exist without the mineral phosphorus. The phosphorus levels in Jinchun 9 were 2.41 g/kg, but in BGW 76 they were 4.10 g/kg. The immune system, inflammation and oxidative stress may all benefit from selenium (Se). Black wheat has 1.04 mg/kg of Se, which is much more than normal wheat's 0.26 mg/kg. Colored wheat, as opposed to white wheat, has a greater concentration of zinc and iron elements and benefits from double-fortification [9].

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Functional Compounds

Several bioactive chemical compounds are found in black wheat, which is higher than in normal wheat (Table 1). Wheat is primarily composed of carbohydrates, protein, and fat, but wheat also contains phenolic acids, flavonoids, anthocyanins, and carotenoids, among other useful compounds. Both flavonoids and PAs include hydroxyl groups and one or more aromatic rings. One kind of flavonoid is anthocyanin, which is a naturally colored flavonol. The anthocyanin, antioxidants, folic acid, the elements selenium, magnesium, calcium, and the metals manganese, zinc, iron, calcium, magnesium, copper, potassium, a mineral fiber, and amino acids found in black wheat are some of its fascinating health benefits. The anthocyanine content of black wheat is twenty-eight times higher than that of white wheat [10].

Phenolic Acids

The bioactive chemicals known as phenolic acids protect plants from both biotic and abiotic stresses. Furthermore, phenolic acids govern gastronomic properties like astringency, colour, flavour, aroma, bitterness, and resistance to lipid oxidation. Phenolic acids have several beneficial effects, including reducing inflammation, killing germs, slowing cell growth, preventing cancer and reducing free radicals [11]. Phytochemicals known as phenolic acids may be present in wheat in two forms: free (soluble reactive) and bound (insoluble). Phenolic chemicals are more typically found on the outside of wheat grains compared to the endosperm. This means that the bran extracts consistently had greater TPC levels [12]. The accumulation of amethyst in the outermost portion of the grain is responsible for the formation of colour on the grain [13]. The first category of PAs consists of compounds derived from hydroxybenzoic acid, whereas the second category includes compounds derived from hydroxycinnamic acid. The extraction of soluble phenolic acids is simpler than that of bound ones. Antioxidant activity is shown by both of such substances when attached to or dissolved in phenolic acid [14]. The phenolic acids found in black wheat in the highest concentrations were protocatechuic acid and gentisic acid. There are several phenolic acids found in black wheat. These include o-coumaric, p-hydroxybenzoic, ferulic, gentisic, syringic, and Gallic acids. The percentage of phenolic acid in the bran is greater than that in the whole meal. The majority of black wheat's phenolic acid content is ferulic acid. The soluble phenolic content of black wheat is greater than that of white wheat. It was found that, in contrast to purple and yellow cultivars, black wheat has 1.6 times more linked total phenolic chemicals (TPC) and 6 times greater quantities of free TPC. To manage innate immune processes, dividing cells, apoptosis, and cell survival, the PAs found in wheat modify a transcriptional factor called nuclear factor kappa B (NF-kB) [15].

Flavonoids

Fruits, vegetables, bark, stems, roots, grains, flowers, tea, and wine are all good sources of flavonoids, which are naturally occurring compounds with varying phenolic structures. Plants produce flavonoids as a byproduct of aerobic metabolism. C6-C3-C6 is an abbreviation for the skeleton of a flavonoid, which consists of a three-carbon heterocyclic ring containing two rings of phenol rings with six carbon atoms each. Many subgroups of flavonoids have positive impacts on health. These subgroups include isoflavones, flavones, flavan-3-ols, flavanones, flavanonols, flavonols, anthocyanidins, and chalcones. Flavonoids assist in strengthening the immune system and have an anti-inflammatory impact [16]. Flavonoids and anthocyanins have several positive impacts on health, including preventing cardiovascular disease, reducing the risk of cancer, lowering blood pressure, preventing diabetes, protecting neurons, and improving eye health [17]. Wheat contains the following flavonoids: apigenin, naringenin, tricin, luteolin, a substance known as quercetin, and vitexin. Black wheat contains around 174 flavonoids. In the investigation, found that at 876 μg/g dry weight, BW had the greatest total flavonoid concentration. Thanks to the presence of apigenin, tricin, anthocyanins, and luteolin, black wheat has a high concentration of flavonoids. Several black wheat types have antioxidant-rich tricin derivatives, such as O-glucuronic acid [2].

Antioxidants

Some species contribute to the development of several illnesses via inducing oxidative stress; they include peroxide anions, hydroxyl radicals, superoxide anion radicals, and singlet oxygen [18, 19]. Because of their ferric reducing capabilities and ability to scavenge free radicals, phenolic and flavonoid chemicals mitigate oxidative stress-induced cell damage [20]. In one research, [9] showed that, in comparison to white wheat, colored wheat flour and anthocyanin-rich wheat grass juice had a significant antibacterial activity and a good antioxidant value against free radicals. Black wheat has a greater Trolox equivalent (TE) content (450.92 μmol/100 g DW) compared to lavender grains (273.40 μmol/100 g DW), turquoise wheat (159.66 μmol/100 g DW), plus yellow wheat (89.93 μmol/100 g DW). Another research found that compared to purple and white grain wheat, complete meal black-grained wheat had a DPPH radical scavenging activity of 33.51%. This is 1.31 times greater and 1.41 times when compared to blue-grained wheat [8].

Carotenoids

The bioactivity and colour of the majority of fruits and vegetables are caused by carotenoids, which are fat-soluble plant pigments. Xanthophylls are hydrocarbon carotenoids, whereas carotene is an oxygen derivative.

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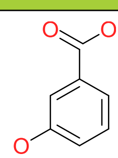
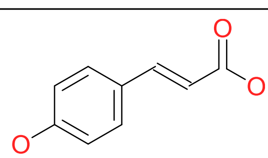
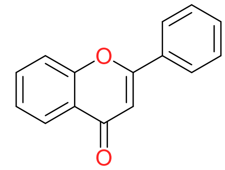
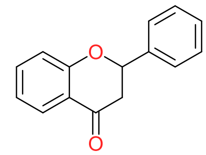
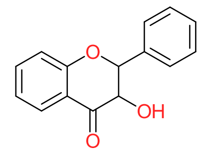
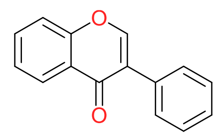
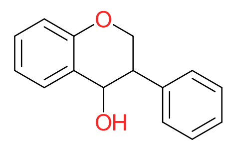
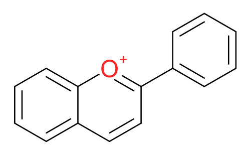
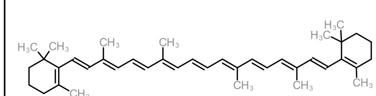
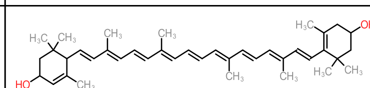
Carotenoids are isoprenoids with 40 carbon atoms and a lengthy conjugated polyene chain. Like ordinary wheat and other tinted wheat varieties (blue, red, purple), BW contains carotenoids. Compared to white and red wheat, BW had a higher net carotenoid content value [21]. The process of converting alpha-carotene, beta-carotene, gamma-carotene, and nor beta-cryptoxanthin into retinol (vitamin A) utilizes four different types of carotenoids. Although they do not convert to tretinoin (vitamin A), other carotenoids, including lutein, lycopene, and zeaxanthin, do have antioxidant properties. Durum wheat is valued for its high carotenoid content, which is responsible for the colour of wheat products. Carotenoids shield cells from harmful oxidative stress by acting as antioxidants. Carotenoids promote immunological activities, reduce LDL cholesterol levels via cholesterol synthesis, and increase iron absorption in humans by binding iron to its aromatic ring [22].

Anthocyanin

Anthocyanins, which are found in the outermost levels of the wheat kernel, are crucial for the formation of grain colour. Compared to other wheat varieties, BW had a significantly higher concentration of 13 of the 14 anthocyanins studied [7]. The researchers compared the antibacterial efficacy of white wheat extracts with coloured wheat amethyst extracts against human infections. For every pathogen tested, black wheat gluten had a minimum inhibitory concentration (MIC) between 50 and 150 mg/mL; the highest MIC value was 50 mg/mL against *S. aureus* and *P. aeruginosa*. Black wheat flour extract at concentrations of 100 and 150 mg/mL completely inhibited the formation of *Staphylococcus aureus* and *Pseudomonas aeruginosa*, respectively. The higher dose of 200 mg/mL was the only one that reduced Candida albicans and *Escherichia coli* [9]. Several bioactive chemical compounds are found in black wheat, which is higher than in normal wheat (Table 1) [2].

Table 1: Bioactive Compounds and Their Chemical Structures [2]

Compounds	Phytochemical Name	Structures
Phenolic acids	Hydroxybenzoic Acid	
	Hydroxycinnamic Acid	
Flavonoids	Flavones	
	Flavanones	
	Flavonols	
	Isoflavones	
	Flavan-3-ols	
	Flavylium salts (anthocyanins)	
Carotenoids	B-Carotene	
	Xanthophyl	

Comparison of Black Wheat and White Wheat

Black wheat is an improved variety of conventional yellow wheat that is richer in protein, fiber, calcium, vitamin K, antioxidant activity, total flavonoid content, and phenolic content. The quantity of anthocyanins is the primary differentiator between white and black wheat. Black wheat has 28 times more anthocyanins than white wheat. There are fewer than five parts per billion (ppm) of anthocyanin in a normal wheat grain. On the other hand, 100-200 ppm is found in black wheat grain. Increasing one's intake of antioxidants improves one's health in general by assisting the body in flushing out harmful pollutants and free radicals. Approximately 225 metabolites, mostly flavonoids, are responsible for the colour variation across black and yellow wheat. Zinc, which is present in black

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wheat at a 35% higher concentration than white wheat, aids in metabolic and immune system processes. In addition, most wheat cultivars contain around 25% iron. In contrast, the iron content of black wheat is 60% higher, leading to a significant elevation of hormone and hemoglobin levels in the body. The protein content of black wheat was 17.71% higher than that of the control wheat. Vitamin K levels in black wheat 76 were 1.6 times higher than in conventional wheat [8]. Comparative Analysis of Nutritional Composition of Different Colored Wheat [1] (Table 2).

Table 2: Comparative Analysis of Nutritional Composition of Different Colored Wheat [1]

Parameters	Black Wheat	White Wheat
Anthocyanin (ppm)	100-400	5
Energy (kcal)	318	322
Carbohydrate (g)	64.8	67.8
Protein (g)	12	10
Dietary fiber (g)	12	11
Fat (g)	1.2	1.2
Moisture (%)	10	10
Iron (mg)	45	38
Zinc (mg)	35	28

Health Benefits of Black Wheat

Heart disease, inflammation, cancer, diabetes, and obesity are all prevented by consuming whole black wheat. Black wheat's high anthocyanin content helps diabetes patients' blood sugar metabolism and cholesterol levels. Black wheat types' high anthocyanin content serves as a protective barrier against lifestyle diseases. Wheat flour's total anthocyanin content ranges from 6.61 to 95.04 mg k-1, with the following sequence: black > blue > purple > white. According to epidemiological studies of whole grains, black wheat has emerged and serves as a better choice in the form of dietary supplements for anyone suffering from breast, colon, liver, ovarian, or prostate cancer [23]. Whole grain consumption and the risk of obesity are inversely correlated, according to numerous observational studies. Naturally occurring antioxidants that form in the grain fill the void and enhance the nutritional value of colored wheat, particularly black wheat. Eye-related issues among adults can be resolved with black wheat. Furthermore, by reducing age-related neurodegeneration and psychological deterioration, it aids in the cerebrum's proper function. Compared to conventional wheat, black wheat has more anthocyanins and antioxidants, which help our bodies regulate free radicals and antibodies while also enhancing immunity. Malnutrition can also be alleviated with black wheat. Additionally, black wheat limits body weight gains and dramatically lowers fat pads [24]. Health benefits of black wheat are analyzed [25] (Figure 3).

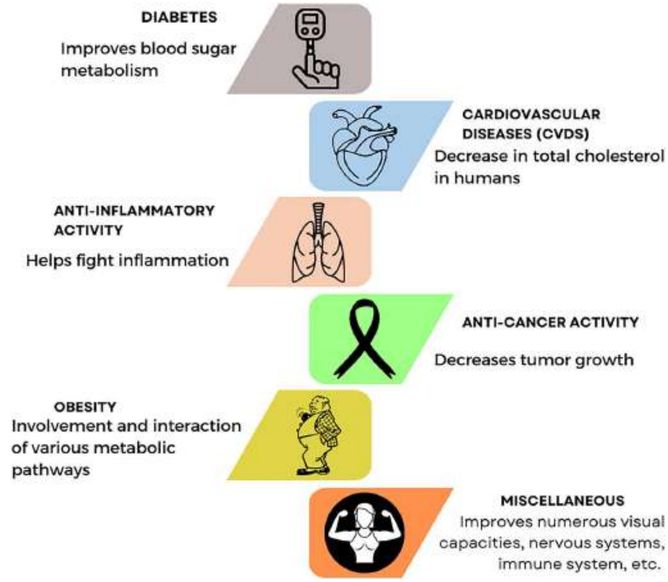

Figure 3: Health Benefits of Black Wheat [25]

CONCLUSIONS

Black wheat has emerged as a nutritionally superior alternative to conventional white wheat due to its rich composition of proteins, minerals, vitamins, and bioactive compounds like anthocyanins, flavonoids, phenolic acids, and carotenoids not only improve metabolic and cardiovascular health but also offer antimicrobial and anti-inflammatory. Prevent obesity, diabetes, cancer, neurodegeneration effects, while also alleviating malnutrition and eye-related disorders. Its health-promoting attributes make it an ideal candidate for inclusion in dietary supplements, functional foods, and specialized therapeutic diets. In short, black wheat offers enhanced nutritional value and represents a promising avenue for health-focused agricultural innovation.

Authors Contribution

Conceptualization: MKS
Methodology: MKS, NZ, SA
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Writing review and editing: QS, SHIA

All authors have read and agreed to the published version of the manuscript.

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Review Article

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A Diminutive Review on "Quinoa Seed's Nutrient Content, Biological Active Compounds and Potential Food Applications

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ABSTRACT

Cereals are regarded as a vital component of the human diet. They supply roughly half of the world's energy. Quinoa (*Chenopodium quinoa* Wild) is a very nutritious plant that has been dubbed "one of the grains of the 21st century" due to its nutritional and functional qualities as well as its ability to withstand harsh weather conditions. It has gained widespread recognition as a superfood due to its significant nutritional and physiological benefits. The current review offers a brief introduction to the nutrients, mineral makeup, amino acids, polyphenols, and bioactive ingredients of quinoa. It is revealed that quinoa contains a wealth of essential components, including vitamins, minerals and amino acids, which directly contribute to its remarkable nutritional value. Its antioxidant properties and unique composition of physiologically active chemicals, such as polyphenols and other compounds, increase its potential as a functional meal. The conclusion of this review is to provide a useful understanding of the potential nutritional and health benefits of quinoa as well as its potential culinary usage. Furthermore, fermented quinoa has a high biological and nutritional value that may be utilized to create a variety of functional food products that are highly palatable to consumers and have positive health effects.

INTRODUCTION

Countries with rapidly expanding populations must discover new sources of nourishment. Despite being readily available to the local population and having a high nutritional value, indigenous crops in the Andes are sometimes overlooked. While some of the crops are wild and regarded as weeds, others are grown [1]. Quinoa (*Chenopodium quinoa* Wild), a very nutritious plant with 250 variants worldwide, is a member of the Amaranthaceae family. It is categorized according to the form of the plant or the colour of the fruits and plants. In Europe, North America, Asia and Africa, this grain may be planted. In Europe, North America, Asia, and Africa, this grain may be planted. Due to its remarkable nutritional profile and adaptability as a food source, this pseudo-cereal has garnered a lot of interest lately. It became an essential part of many indigenous societies' diets. It is a vital staple that can improve food security and nutrition, especially in areas experiencing agricultural difficulties, due to its remarkable

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resilience and ability to grow in a range of environmental obstacles, such as high altitude and nutrient-poor soil. In both cold (-5°C) and hot (up to 35°C) regions, quinoa has shown a strong resistance to salty, acidic, or alkaline soils [2]. Quinoa was brought to South Asia recently, and because of its adaptability to a variety of climates, its production there is becoming more and more popular. Its ability to thrive in both dry and semi-arid environments is a result of Pakistan's shifting agro-ecological zones. Farmers must choose a quinoa variety that is appropriate for their environment in order to cultivate it successfully. To promote healthy growth and a suitable supply of nutrients, plants should be spaced appropriately apart. Quinoa is a drought-resistant crop that requires little irrigation, making it a sustainable agricultural option in arid regions of South Asia. Due to its high nutritional value and environmental adaptability, it has been cultivated as a new alternative crop in many places [3]. The method of cultivation includes preparing soil that drains properly and planting seeds 1-2 cm deep (Figure 1).

Figure 1: Quinoa Plant and Its Seeds

Its low glycaemic index makes it an excellent meal choice for diabetics because it helps control blood sugar levels. Because it includes antioxidants, which also improve general health, consuming it greatly reduces the risk of acquiring chronic diseases. Quinoa serves as an intriguing crop with a long history and a lot of possibilities for modern diets. Its numerous health advantages make it a wonderful addition to a variety of culinary techniques, and its extensive cultivation can help to improve food security and nutrition in the South Asian region [4]. Quinoa is a gluten-free food, and studies have demonstrated that regular consumption of quinoa improves small intestine health in people with celiac disease, allowing intestinal villi to heal more quickly than with a simple gluten-free diet [5].

Varieties Found in Pakistan

Quinoa is a nutritious crop that has great adaptability to a variety of environmental situations. In Pakistan, it has a great chance of expanding. Its remarkable resistance and durability imply that it might flourish in Pakistan's many climates, even though the majority of research has concentrated on its growth in the Andes. It can withstand harsh weather conditions as salt, drought, and frost. Because of this, quinoa is a great choice for Pakistan's underserved regions [6]. According to research, quinoa agriculture is more affected by soil sodicity a high percentage of sodium ions, than by salinity. Genotypes such as A1 and A7, on the other hand, exhibit improved nutritional quality and stress resistance, making them suitable for regions impacted by salt. Quinoa genotypes grown in Pakistan have been the subject of recent research. C. quinoa V7, V2, and V1 are among them. In Pakistan's food industry, quinoa's potent nutritional profile, which includes high levels of protein and fatty acids, can provide enormous advantages for human consumption. It may find application in the pharmaceutical, food, and animal feed sectors [7]. Because of its broad leaves and starchy, dicotyledonous seeds, quinoa is categorized as a pseudo-cereal crop, which means it is not a cereal. The FAO views quinoa production as a way to fight famines because of its natural resistance to dry soils [8]. It is extremely suited to the various soil types and climate zones, having been domesticated in the Andes. Quinoa is a potential substitute for increasing food security in Pakistan and provides a path towards sustainable agriculture.

Nutrients of Quinoa

Quinoa is well-known for its resilience, but it also maintains its position as a superior nutritional and health food. For vegans and vegetarians, it is a wonderful source of nutrients. the chemical makeup of quinoa seeds was as follows: 62.07% carbohydrates, 14.40% protein, 6.88% fat, 5.12% crude fibre, 2.63% ash and 8.90%moisture [9]. Quinoa is regarded as one of the best sources of vegetable protein (12–23%); its protein level is higher than that of actual cereals like rice, wheat, and maize and comparable to that of milk. Additionally, quinoa contains a high percentage of starch (52–60%) and a low percentage of amylose (7–11%). Additionally, quinoa has a greater dietary fibre content than other cereals [10]. Protein, dietary fibre, vitamins (particularly B vitamins and vitamin E), and vital minerals including calcium, iron, potassium, and magnesium are also abundant in it [11] (Table 1).

Table 1: Nutritional, Mineral and Vitamin Composition of Quinoa Seed

Variables	Parameters	Values
Nutritional Composition (g/100g)	Moisture	8.9
	Ash	2.63
	Fat	6.88
	Fiber	5.12
	Protein	14.4
	Carbohydrates	62.07
	Energy Kcal/100g)	464.6
Minerals (mg/100g)	Calcium (Ca)	70
	Potassium (K)	855
	Sodium (Na)	2.7
	Phosphorus (P)	462
	Zinc (Zn)	3.2

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	Iron (Fe)	6.3
	Magnesium (Mg)	161
	Manganese (Mn)	3.5
Vitamins (mg/100g)	Riboflavin (B2)	0.60
	Folic Acid (B9)	6.80
	Niacin (B6)	1.24
	Alpha Tocopherol (E)	2.01

Amino acids

A significant source of dietary proteins is the grain quinoa [12]. The proteins found in quinoa grains are globulins (37%) and albumins (35%), with a lesser proportion of prolamins. Quinoa's protein content is on par with that of casein, a protein found in milk. Tryptophan, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tyrosine, and valine are among the necessary amino acids that are found in quinoa proteins [13]. For this reason, it is considered a complete food. Protein consumption is a challenge for communities that rarely eat animal proteins; consequently, plant-based proteins should be an integral part of their diet. In this sense, quinoa-based products are a good choice because, in contrast to other traditional cereals, quinoa does not significantly lose protein content during industrial processing. Furthermore, quinoa's organic value surpasses that of other conventional cereals due to the amino acid makeup of its proteins. Furthermore, lysine is regarded as the first limiting amino acid and is a vital nutrient for children, as it can boost immunological function and support human development [14] (Table 2).

Table 2: Amino Acids Composition of Quinoa Seed

Amino Acids Profile	Concentration (g/100g Protein)
Glutamic acid	17.30
Aspartic acid	10.54
Arginine	9.71
Leucine	6.88
Lysine	6.30
Glycine	6.26
Serine	5.62
Alanine	5.53
Phenylalanine	4.52
Threonine	4.41
Histidine	4.09
Valine	3.67
Tyrosine	3.66
Proline	3.54
Isoleucine	3.02
Methionine	2.27
Half cystine	1.39

Polyphenolic profile

The phenolic profile of quinoa seeds powder by HPLC which are Protocatechuic, kaempferol, caffeic, syringic, vanillic, ferulic, chrysin, sinapic, p-coumaric apigenin-7-glucoside, rosmarinic and cinnamic equal to 21.2, 13.6, 49.0, 22.8, 285.9, 3059.2, 9.3, 244.9, 65.8, 52.1, 14.6, 342.1, 107.7 and 44.1(µg/100g), respectively (Table 3).

Table 3: Polyphenolic Profile of Quinoa Seed Powder

Phenolic Profile	Concentration (µg/100g)
Ferulic	3059.2
Rosmarinic	342.1
Vanillic	285.9
Sinapic	244.9
Cinnamic	107.7
p-coumaric	65.8
Rutin	52.1
Caffeic	49
Apigenin	44.1
Syringic	22.8
Protocatechuic	21.2
Apigenin-7-glucoside	14.6
Kaempferol	13.6
Chrysin	9.3

When compared to other phenolic compounds that are present in moderate concentrations, like vanillic and sinapic, it is evident that rosmarinic and ferulic acid are found in the highest proportions. However, data in the same table showed that the lowest levels of kaempferol and chrysin are present. Phenolic is a broad and varied class of chemicals that have at least one aromatic hydrocarbon ring joined to a hydroxyl group or groups. Because of their high structural stability, phenolics have substantial antioxidant properties [15]. Quinoa has a lot of polyphenols, which are significant byproducts of biologically active secondary metabolism in plants. They have a structure that includes one or more aromatic rings and hydroxyl groups. Quinoa's functional qualities and flavour are influenced by the amount of these phenolic chemicals it contains. Touil *et al.*, (20124) used the HPLC-DAD analysis to identify the phenolic compounds of the various extracts of quinoa genotypes [16]. They found seven phenolic compounds, which may be categorized into four families: coumarins, stilbenes, flavonoids and phenolic acid. Higher phenolic contents and antioxidant activity are found in darker quinoa seeds. Through their regulation of the gut's microbial balance, dietary phenolics contribute to gut health maintenance. According to Melini and Melini, (2022), individual phenolic acids have important anticancer, anti-inflammatory, anti-obesity, antidiabetic and cardioprotective properties because they enhance metabolism and cell signalling [17]. The environmental conditions at the planting location also have an impact on the polyphenol content of quinoa. Phenolic compounds have attracted a lot of attention in the past 20 years because of their potential to prevent chronic diseases and

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provide other health advantages [18].

Biologically Active Compound

Carbohydrates that are glycosidic linkages bind with high molecular weight, called polysaccharides, which have a range of biological functions. Starch and non-starch polysaccharides are the two main types of quinoa polysaccharides. About 60% of it is crude starch, and of those starches, amylopectin has a larger proportion than amylose. Microorganisms in the large intestine can partially digest and metabolize dietary fibre, a form of carbohydrate with nutritional functional activity that cannot be broken down by the human small intestine. Quinoa contains between 7.7% and 9.7% total dietary fibre and between 1.3% and 6.1% soluble dietary fibre. People with celiac disease symptoms can effectively address gluten-free fibre shortages in their diet by substituting fibre-rich, gluten-free whole grains like quinoa for refined grains [19]. Several other bioactive substances, each with unique health advantages, include saponins, polyamines, polyphenols, phytosterols, and bioactive peptides. Saponins are prevalent in the quinoa seed husk. Because saponins are bitter, they are generally considered to be anti-nutritional. They may also lower cholesterol, facilitate nutrition absorption, and increase the permeability of the cell membrane. Additionally, saponins are used as fungicides, insecticides, and detergents [20]. Quinoa also contains phytohormones, including genistein and daidzein, which have antioxidant properties, boost bone density, and preserve vascular health [21]. Polyamines like spermidine and spermine give quinoa its "earthy" flavour. On the other hand, bioactive peptides, phytosterols, and polyphenols have anti-inflammatory, antibacterial, and antioxidant qualities. According to Hernández-Ledesma (2019), it is also a good source of fatty acids, such as palmitic, oleic, linoleic, and α-linoleic acids [22].

Potential Food Applications of Quinoa

Quinoa and other ancient foods have been saved by the growing demand for healthier and more functional diets. A pseudo cereal that was crucial to many ancient societies was quinoa. It is becoming more and more important in the markets because of its superior nutritional value compared to many of the cereals that are a staple of modern diets. Ascorbic acid, protein, and minerals like P and K are all abundant in it [23]. The production of synbiotic foods, lactose-free fermented foods, gluten-free fermented foods, and bioactive peptides, as well as their use as food additives in fermented dairy and non-dairy foods, was all made possible by quinoa seeds because of their functional ingredients [24, 25] (Figure 2).

Figure 2: Functional Food Products by Fermented Quinoa

CONCLUSIONS

Quinoa is a high-nutritional, historically significant pseudo cereal that can withstand environmental stress. A substantial portion of the daily requirements for key nutrients, primarily amino acids, vitamins, minerals, carbohydrate, fibre and antioxidants, are present in quinoa. Its capacity to nourish millions of undernourished individuals globally has been acknowledged. Additionally, quinoa is gluten-free, and it is generally safe for those who have celiac disease. Fermenting quinoa enhances its nutritional and sensory qualities and yields useful functional foods with increased biological activity. In order to effectively improve public nutrition and health, new functional foods that incorporate quinoa into contemporary diets should be developed.

Authors Contribution

Conceptualization: MKS
Methodology: MKS, SA
Formal analysis: NZ, KF, AK
Writing review and editing: HMJ

All authors have read and agreed to the published version of the manuscript.

Conflicts of Interest

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Original Article

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Low Calcium Intake of Pre-Adolescent Girls from Customary Diets in A Semi-Rural Setting in Khyber Pakhtunkhwa, Pakistan

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ABSTRACT

Calcium is a critical nutrient for adolescent bone development and overall health, yet intake often remains below recommended levels in many populations. **Objectives:** To assess the calcium intake of pre-adolescent girls, their calcium fortified preferences, and perceived barriers to calcium-rich foods to inform targeted nutritional interventions. To assess the calcium intake of adolescent girls, their calcium fortified preferences, and perceived barriers to calcium-rich foods. **Methods:** A descriptive cross-sectional study was conducted, where 78 school girls aged 9-14 years completed FFQ with a focus on foods rich in calcium. The questionnaire also contained questions inquiring about the perceived barriers which could result in low intake/no intake of calcium-dense foods. **Results:** A total of 174 calcium-rich foods and products were available in Charsadda city. Mean intake of calcium (275 ± 52) was well below the RDI. Notable gaps in students' awareness of calcium sources and their importance existed. The top 8 Key barriers that potentially hinder the intake of nutritious foods were identified, including cost, low availability in the school environment, low awareness, inappropriate storage facilities at school, food habits, low nutrition education, food stigma, and junk food pressure. The data highlights a clear preference among students for certain calcium-fortified food types over others. **Conclusions:** Mean calcium intake from foods available to pre-adolescent girls was low, with numerous barriers identified that hinder calcium intake in this age group. Further, pre-adolescent girls preferred a variety of calcium-fortified foods, biscuits and milk on top of the list.

INTRODUCTION

Calcium is a critical micronutrient important for skeletal system development, especially during pre-adolescence, a time characterized by bone growth acceleration and hormonal development [1]. Proper calcium consumption during this window of development is critical in the attainment of peak bone mass and the prevention of osteoporosis, fractures, and other bone diseases in adulthood [2]. Global guidelines, like those of the World Health Organization (WHO) and Institute of Medicine (IOM), recommend that children 9–12 years of age consume 1000 to 1300 mg of calcium each day to maximize growth and physiological functioning [3]. Nonetheless, there is evidence that calcium consumption in children living in South Asian nations, such as Pakistan, continues to be woefully inadequate, especially in rural and resource-poor populations [4]. This is further compounded by socio-economic limitations [5], restricted dietary variety [6], minimal exposure to dairy or fortified foods [7], and widespread ignorance about nutritional requirements [8]. The outcomes of this research are anticipated to guide specific nutrition interventions and assist with the design of culturally sensitive approaches to enhancing calcium consumption among school-age girls in comparable contexts. In Pakistan, where the dietary habits are

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predominantly cereal-based and calcium-containing foods are missing from typical diets, the risk of calcium deficiency is particularly high among young girls. Pre-adolescent females are a group of special concern because poor calcium consumption during this age can result in impaired bone structure, growth delay, and increased risk of persistent nutritional deficiency. Although the seriousness of this problem is so considerable, little evidence supports the availability of calcium-rich foods in the semi-rural regions of Pakistan, especially KP, where socio-cultural and economic obstacles are unique. Information regarding the calcium sources in foods is critical to any nutrition policy development in addressing the common calcium deficiencies[9, 10].

This study aims to know pre-adolescent girls' daily dietary intake of calcium in a semi-rural area in KP and to ascertain the primary sources of calcium in their traditional diets. Also, to know what foods these girls prefer to eat as a source of calcium and what perceived barriers they have while trying to achieve calcium-rich foods.

METHODS

This descriptive cross-sectional study was carried out in a semi-urban school in Charsadda, targeting students in grades 5 through 9 who were between the ages of 9 and 14 during two survey phases (Jan–Feb and Jun–Jul 2022). 79 of the 120 enrolled students took part with permission from their guardians. Sample size was estimated using the standard formula for a single population proportion with a 70% expected response rate, 5% margin of error, and 95% confidence level [11]. Data were collected in four sections using a self-administered, validated questionnaire that was created with expert input: (1) demographics and familiarity with foods high in calcium; (2) food frequency and 24-hour recall with illustrations; (3) perceived barriers to calcium intake using a 5-point Likert scale; and (4) preferences for fortified foods that are available on the market. BMI-for-age percentiles were determined using CDC growth charts for girls aged 2–19 years. The questionnaire underwent a pilot test for clarity and relevance, and its content validity was established through expert review before data collection. The researcher explained the survey procedure one day before administration. A full census of eligible girls (ages 9–14, grades 5-9) was conducted at the selected semi-urban school. As the entire target group was surveyed, randomization or stratification was not applicable, ensuring representativeness and reducing selection bias. SPSS version 20.0 was used to analyze the data; descriptive statistics were used, and calcium intake was calculated from reported intakes using national food composition tables that were cross-checked with USDA data. RDAs and mean daily calcium intake were compared; a daily intake of less than 300 mg was deemed critically deficient [3]. Means and SDs on the Likert scale were also computed. No comparison between the season-based data was done as it was beyond the scope of the study.

RESULTS

Mean age was 13.5 (4.56) years. More than half of the girls (56.4%) were from grades 6-8 (Middle school grades). Only 53 of the 174 calcium-rich foods that had previously been identified [12] were recognized by the students, and only 26 of them had been consumed in the previous three months. Although fortified dairy products, such as high-calcium milks and cheeses, were widely known, consumption was found to be modest. Standard UHT and pasteurized milk, yoghurt, and malted beverages were among the frequently consumed dairy products; each serving included 150–200 mg of calcium. Although students were aware of meats and legumes as good sources of calcium, they rarely actually consumed them, particularly when it came to soy milk and red beans. On the other hand, subjects ate more turnip, okra, mint, and radish—vegetables that contain appreciable calcium (Table 1).

Table 1: Number of Food Items and Products Known to and Consumed by Students

Variables	Number of Items Known to the Responders	Number of Items Reports have been Consumed in the Last 3 Months
Meat and Meat Products	13	3
Vegetables	12	8
Dairy and Dairy Products	5	2
Legumes, Nuts, and Seed Products	6	3
Starchy Foods	15	10
Calcium Supplement Tablets	2	0
Total	53	26

The amount of calcium consumed and the calcium sources were recorded. The sample of 78 pre-adolescent girl's students surveyed, no student met the Pakistani Recommended Daily Intake (RDI) for calcium (≥800 mg/day). The median calcium intake was 275 ± 52 mg/day, indicating all students had inadequate calcium intake. The main calcium sources were dairy and meat products. Approximately 84% of participants consumed less than 200 mg/day of calcium, indicating a critical deficiency (Table 2).

Table 2: Summary of Daily Calcium Intake

Variables	Value
Mean Calcium Intake (mg/day)	275 ± 52
% Below 200 mg/day	84%
% Below RDA (1000 mg/day)	100%
Most Common Food Sources	Naan, Chapati, Tea, Spinach, Lassi, Mint, Mustard, Rice, Barsnada (Lotus Root), Okra, Turnip, Cabbage, Radish, Garlic

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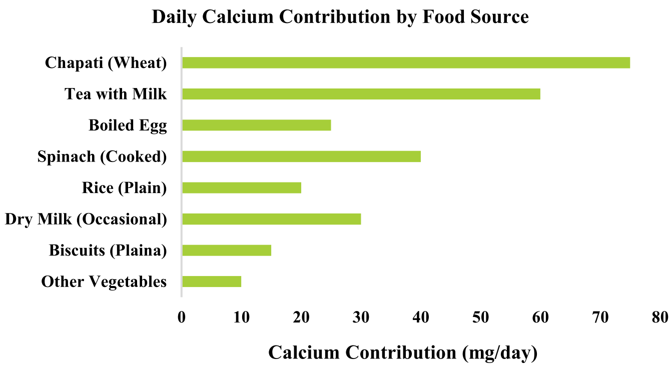

Figure 1: Daily Calcium Contribution by Food Source

Key barriers to the intake of calcium-containing foods were recorded. Using a 5-point Likert scale, the results list the main barriers to schoolgirls consuming enough foods high in calcium. The primary barrier to affordability was the high cost of nutrient-dense foods, which received the highest rating (Mean=4.5). Poor cafeteria diversity, low nutrition knowledge, inadequate food storage, cultural dietary habits, lack of nutrition education, junk food marketing, poor comprehension of food labels, and seasonal produce availability are some of the hurdles that received 4.0. Peer pressure, low staff participation, inadequate outside assistance, and the lack of school nutrition programs were all assessed as moderate hurdles (Mean=3.5). These results demonstrate how access to nutritious foods in semi-urban environments is restricted by both systemic and sociocultural issues (Table 3).

Table 3: Perceived Barriers hindering Calcium Rich Foods Intake

Barriers	Descriptions	Likert Scale Mean ± SD	Median (IQR)
High Cost	Healthy options like fruits, dairy, lean meats, and fortified products are often more expensive.	4.5 ± 1.5	5 (4–5)
Limited Variety in School Canteens	Canteens mostly offer affordable, less nutritious snacks rather than balanced meals.	4 ± 1.0	4 (4–5)
Low Parental Nutrition Awareness	Many parents lack knowledge about balanced diets and the importance of specific nutrients.	4 ± 1.5	4 (3–5)
Inadequate Food Storage Facilities	Lack of refrigeration limits the availability of perishable, nutritious foods like milk and yoghurt.	4.0 ± 1.5	4 (4–5)
Cultural and Traditional Food Habits	Local food preferences may exclude key sources of nutrients like cheese or fortified products.	4.0 ± 1.5	4 (3–5)
Insufficient Nutrition Education	Students are not regularly taught about healthy eating or the importance of specific nutrients.	4.0 ± 1.0	4 (3–5)
Peer Pressure and Food Stigma	Students may avoid healthy food to conform with peers or avoid being labelled "different."	3.5 ± 1.0	4 (3–5)
Marketing of Junk Food	Unhealthy snacks are aggressively advertised to children, influencing their choices.	4.0 ± 1.5	4 (3–5)
Poor Nutrition Label Understanding	Many students cannot interpret labels to assess calcium, iron, or vitamin content in foods.	4.0 ± 1.5	4 (3–5)
Seasonal Availability of Produce	Fruits and vegetables rich in nutrients may not be available year-round.	4.0 ± 1.3	4 (3–4)
Limited Staff Involvement	Teachers and staff often do not monitor or encourage healthy eating during school hours.	3.5 ± 1.4	3 (3–4)
Lack of External Support	Government or NGO-supported nutrition initiatives may be absent or irregular.	3.5 ± 1.3	3 (3–4)
Absence of School Nutrition Programs	Few schools offer consistent programs like fortified meals, milk breaks, or health talks.	3.5 ± 1.3	3 (3–4)

Using a 5-point Likert scale, the study displays the preferences of students for meals supplemented with calcium. Dry milk powder and calcium-fortified biscuits were the most popular products (Mean=4.5), probably because of their familiarity, taste, and ease of use. Breakfast cereals (Mean=2), soy milk, and UHT fortified milk (Mean=2.5) were moderately desired items, perhaps as a result of their lesser cultural acceptance. Malt drinks, juices, snack bars, yoghurt, cheese slices, tofu, chocolate, and noodles were the least popular (Mean=1–1.5) and frequently regarded as strange, expensive, or less appetizing. These findings demonstrate how student preferences are influenced by flavor, cost, and cultural familiarity (Table 4).

Table 4: Students' Preferences for Calcium-Fortified Foods

Calcium-Fortified Food Type	Examples / Notes	Likert Scale Mean ± SD	Median (IQR)
Calcium-Fortified Biscuits	Sweet or savory; often marketed as nutritious snacks	4.5 ± 1.0	5 (4–5)
Calcium-Fortified Dry Milk Powder	Used in drinks or added to recipes; long shelf life	4.5 ± 1.0	5 (4–5)
UHT Calcium-Fortified Milk	Regular, low-fat, flavored options available	2.5 ± 1.4	3 (2–4)
Calcium-Fortified Soy Milk	Dairy alternative; often flavored and sweetened	2 ± 1.5	2 (1–3)
Calcium-Fortified Malt Drinks	Popular among students; often advertised for energy	1.5 ± 1.2	1 (1–2)
Calcium-Fortified Fruit Juices	Orange juice and other fortified foods with calcium	1.5 ± 1.3	1 (1–2)
Calcium-Fortified Breakfast Cereals	Usually eaten with milk; convenient and palatable	2 ± 1.4	2 (1–3)
Calcium-Fortified Snack Bars	Cereal bars or granola with added nutrients	1.5 ± 0.5	1 (1–2)
Calcium-Fortified Cheese Slices	Often expensive, but palatable; easy to use in sandwiches	1 ± 0.5	1 (1–1)
Fortified Yogurt and Yogurt Drinks	May not be labelled clearly as calcium-rich; needs awareness	1.5 ± 0.5	1 (1–2)

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Hard Tofu (Calcium-Set)	Traditional and inexpensive, though not always recognized by students	1 ± 0.5	1(1–1)
Calcium-Fortified Chocolate or Sweets	Sometimes marketed as health-enhanced treats	1 ± 0.5	1(1–1)
Fortified Instant Noodles or Soups	Rare but emerging in some markets	1 ± 0.5	1(1–1)

To find out if there were statistically significant variations in students' preference scores for the various kinds of calcium-fortified foods, a one-way ANOVA was performed. At least some groups' mean preference scores differed significantly, according to the study $F(12, 1677) = 24.56$, $p<0.001$). Calcium-enriched biscuits and dry milk powder were much more popular ($p<0.01$) than other products such as fortified cheese slices, tofu, snack bars, and instant noodles, according to post hoc comparisons using the Tukey HSD test. These findings imply that students' acceptance of fortified foods may be influenced by factors such as product form, convenience, and flavor familiarity.

DISCUSSION

The present research identified that despite the fact that numerous calcium-dense foods are present in Charsadda, fewer than one-third of them were identified by students as dietary sources of calcium. Milk, largely used in tea, was the predominant calcium source, presumably for reasons of publicity associating it with bone health and not through nutritional awareness [13, 14]. The students had no idea how much milk one should use to provide daily calcium needs. Cost was the greatest limiting factor: families had to incur 30–60 PKR/day/child on good sources, which is considerable in proportion to the average income (mean: 29,500 PKR/month; range: 24,000–45,000 PKR/month). There were universal misconceptions, e.g., many thought drinking milk with fruits (especially watermelon) was harmful. Intakes of calcium predominantly comprised chapati, tea and milk, and spinach, which form part of rural diets [15]. But their bioavailability of calcium is low owing to inhibitors such as phytates, oxalates, and tannins [16, 17]. Milk was seldom taken as a single beverage, further restricting proper calcium absorption. Students showed inadequate knowledge of non-dairy sources of calcium, including nuts, seeds, legumes, and fortified foods. This is consistent with other South Asian research estimating poor awareness and consumption levels in adolescents [18, 19]. Such evidence highlights the imperative for school-based nutrition education that is culturally appropriate, access to low-cost fortified foods, and public awareness campaigns [20, 21]. The present research also reports that students preferred fortified biscuits and milk as their food of choice. This preference must be taken into consideration in future clinical trials, policies and programs for school feeding programs. The present research emphasizes the imperative to develop focused interventions to enhance calcium consumption in school-aged girls, suggesting school-level supplementation, fortification of staple foods, increased promotion of affordable calcium foods such as sesame seeds and moringa, and education of parents as main strategies. It is, however, subject to the limitations of being based on self-reported data, having no biochemical validation, being geographically limited, and having no inclusion of contextual influences such as vitamin D status and socioeconomic factors that can influence the generalizability and validity of results.

CONCLUSIONS

Mean calcium intake from foods available to pre-adolescent girls was low, with numerous barriers identified that hinder calcium intake in this age group. Further, pre-adolescent girls preferred a variety of calcium-fortified foods - biscuits and milk on top of the list.

Authors Contribution

Conceptualization: SS
Methodology: SS, IA
Formal analysis: SS, AA, IA
Writing review and editing: SS, AA, IA

All authors have read and agreed to the published version of the manuscript.

Conflicts of Interest

All the authors declare no conflict of interest.

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Relationship Between Bone Mass Density and Nutritional Status of Women Diagnosed with Type 2 Diabetes Mellitus in Charsadda

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ABSTRACT

The prevalence of low bone density among women is common but remains under-documented. **Objectives:** To assess bone mass density (BMD) levels and their relationship with nutritional status in type 2 diabetes mellitus (T2DM) women visiting the NEAT Clinic in Charsadda, Khyber Pakhtunkhwa (KPK). **Methods:** Using an observational, cross-sectional design, data were collected from 408 women through a structured self-reported questionnaire covering osteoporosis risk factors, sociodemographic information, anthropometrics, general health, BMD, and dietary intake. BMD was measured via Quantitative Ultrasound Scan (QUS) at the calcaneus using the Sahara clinical bone sonometer. Initially, 605 women expressed interest, but data were collected for 450 participants due to drop-out and incomplete questionnaires. **Results:** The mean T-score was –1.76 ± 0.81 (median: –1.72), ranging from –3.5 to 1.4. Women classified as normal, osteopenic, and osteoporotic had mean T-scores of –0.42 ± 0.43, –1.66 ± 0.34, and –2.66 ± 0.25, respectively. The prevalence of osteopenia was 59.8% and that of osteoporosis was 22.2%, while only 18% had normal BMD. QUS scores showed significant correlations ($p<0.05$) with age, menopausal duration, BMI, waist-hip ratio (WHR), and education level. Nutritional factors such as energy, protein, calcium, and phosphorus intake were also positively associated with QUS scores. **Conclusions:** The study reveals a high prevalence of osteopenia and osteoporosis among women in this population, with bone health strongly linked to both demographic and dietary factors.

INTRODUCTION

Bone is a very active tissue that remodels continuously throughout life. It also serves as a storehouse of calcium (Ca), magnesium (Mg), phosphorus (P), and other elements that are required for homeostasis [1]. Optimal bone health can be achieved with a balanced diet and adequate dietary energy [2]. These include essential nutrients such as Ca, P, protein [4], potassium (K), vitamins A and K [3], fluorine, zinc, copper, and boron [4, 5]. Though adolescence is a key time for reaching optimal peak bone mass, scientific inquiry into the determinants of bone remodeling over this window of development is fairly underdeveloped. Of the many environmental determinants, nutritional status is especially crucial for ensuring healthy bone growth and mineralization. Sufficient calcium, vitamin D, protein, and other micronutrient intake is necessary to maintain the high rate of skeletal growth that characterizes this stage of life [6]. Yet, a recent and shocking finding from adolescent health surveys has been the following: around 25% of adolescents fall into the underweight category, with the other 75% sitting at the edge of normal body weight. This low-weight prevalence across most of the population is troubling in terms of potential suboptimal bone accrual since less body mass has been linked to impaired BMD and delayed skeletal maturation [7]. Although it is generally agreed across the literature that body weight and bone health are positively associated, less is known in terms of

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the effect of thinness or undernutrition on adolescent bone outcomes. Few empirical studies have explicitly tested this association, so more intensive research efforts are warranted to clarify the late-emerging implications of thinness in adolescents for osteoporosis risk and susceptibility to fracture. The specific objectives were to determine BMD levels in female of different age groups and to determine the relationship between BMD and nutritional status.

This study aims to determine the relationship between nutritional status and BMD of women who have been diagnosed with type 2 diabetes mellitus (T2DM) in Charsadda.

METHODS

The cross-sectional observational study was carried out in Charsadda in collaboration with NEAT Clinics at Charsadda between 2021 and 2022. The ethical approval was taken from the institute with reference no: 1835. Convenient sampling was used to enroll women aged 18 years and above who were literate and did not have speech or hearing impairment. Calculating using Cochran's formula at a 99% level of confidence and 5% precision, the sample size worked out to 408, with a malnutrition prevalence of about14.9% as indicated in the National Nutrition Survey Pakistan, 2018. To prepare for possible dropouts, the sample was boosted by 10%, targeting 450 participants. Information was gathered using standardized questionnaires on age, socioeconomic status (urban or rural), education, income, occupation, and history of bone fractures and osteoporosis in the family. The main anthropometrics (body weight, height, waist, and hip circumferences) were taken as previously reported [8]. Body Mass Index (BMI) was calculated (weight (kg) divided by the square of height (m^2). Waist-to-hip ratio was computed simply by dividing the waist circumference by the hip circumference. Dietary consumption data were obtained via the 24-hour dietary recall method and the Food Frequency Questionnaire (FFQ). Face-to-face interviews were conducted, and data were recorded in the questionnaires [8]. Nutrient consumption was estimated utilizing the Pakistan Dietary Composition Tables. Quantitative Ultrasound Scanning (QUS) at the right calcaneus was employed. Speed of Sound (SOS), Broadband Ultrasound Attenuation (BUA), Stiffness Index (SI), and estimated BMD (eBMD) were determined. WHO criteria were used to categorize participants as normal, osteopenic, or osteoporotic based on T-scores [9]. The ethical committee of Bacha Khan University approved the research. The informed consent of all respondents was taken, maintaining the confidentiality of individual data.

RESULTS

A total of 450 women participated in this study. Initially, 605 women expressed interest in participating. Of these, 90 did not fulfil the inclusion criteria. Out of the remaining 515, data could be completed for 450 women due to drop-out and incomplete questionnaires. This number was greater than the required sample size (408) and was considered for analysis in order to achieve more statistical power. The mean T-score was –1.76 ± 0.81 (median: –1.72) (range: –3.5 to 1.4). Normal BMD women had a mean T-score of –0.42 ± 0.43, followed by osteopenic women (mean T-score, –1.66 ± 0.34) and osteoporotic women (mean T-score, –2.66 ± 0.25). There was a high percentage of osteopenic (59.8%) and osteoporotic women (22.2%). Only 18% had normal BMD. Table 1 displays the osteoporotic and sociodemographic conditions. QUS-score (normal BMD, osteopenia, and osteoporosis) was significantly correlated with some independent variables (age, anthropometric indicators, e.g. BMI and WHR), and educational level ($p<0.05$) (Table 1).

Table 1: Characteristics of Subjects (n=450)

Variables	Total Sample (Mean ±SD) or n (%)	QUS Score, n (%)			p-Value
		Normal BMD	Low BMD		
		Normal	Osteopenia	Osteoporosis	
		81 (18.0%)	269 (59.8%)	100 (22.2%)	
Age (Years)	50.7 ± 8.23	46.7 ± 8.61	51.7 ± 10.6	54.2 ± 12.62	0.040^{a*}
Menopausal Age (Years)	44.1 ± 1.83	45.6 ± 2.21	48.9 ± 5.72	48.3 ± 1.98	0.015^{b*}
Menopausal Duration (Years)	6.8 ± 7.81	1.0 ± 2.45	6.79 ± 8.71	9.9 ± 5.56	0.002^{b*}
BMI, kg/m^2	24.9 ± 4.41	23.4 ± 4.89	25.6 ± 3.26	25.9 ± 5.9	0.000^{b*}
Waist to Hip Ratio	0.9 ± 0.07	0.9 ± 0.11	0.9 ± 0.16	0.9 ± 0.11	0.003^{a*}
Educational levels					
No Formal Education	312 (69.3%)	80 (30.7%)	110 (35.3%)	122 (39.1%)	0.013c*
Formal Education	138 (30.7%)	40 (28.9%)	70 (50.7%)	28 (20.3%)	
Monthly Income					
>5000 PKR	119 (26.4%)	39 (32.8%)	60 (50.4%)	20 (16.8%)	0.002^{c}
< 5000 PKR	331 (73.6%)	81 (24.5%)	120 (36.3%)	130 (39.2%)	

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Menopausal Status					
Postmenopausal	210 (46.7%)	30 (14.3%)	76 (36.2%)	104 (49.5%)	0.502[c]
Premenopausal	240 (53.3%)	90 (37.5%)	70 (32.0%)	80 (33.5%)	
Employment Status					
Not Working	360 (80.0%)	90 (25.0%)	140 (38.9%)	130 (36.1%)	0.003[c]
Working	90 (20.0%)	30 (33.3%)	40 (44.4%)	20 (22.2%)	
Living Place					
Urban	280 (62.3%)	35 (12.5%)	125 (44.6%)	120 (42.9%)	0.004[c]
Rural	170 (37.7%)	85 (50.0%)	55 (32.4%)	30 (17.6%)	
Family History of Osteoporosis					
Yes	232 (51.6%)	55 (23.7%)	82 (34.5%)	95 (40.8%)	0.462[c]
No	218 (48.4%)	65 (29.8%)	98 (45.0%)	55 (25.2%)	
Family History of Fracture					
Yes	223 (49.6%)	56 (25.1%)	83 (37.2%)	84 (37.7%)	0.208[c]
No	217 (48.2%)	64 (29.5%)	97 (44.7%)	56 (25.8%)	
Weight Status					
Overweight	300 (66.4%)	20 (6.7%)	138 (46.0%)	142 (47.3%)	0.003[c]
Normal Weight	160 (35.6%)	100 (62.5%)	42 (26.3%)	18 (11.3%)	

Continuous data are presented as mean, followed by Mean ± standard deviation (Mean ± SD). The p-values were derived from one-way ANOVA (for continuous variables normally distributed); b Continuous data are presented as mean ±standard deviation (M±SD). The p-values were derived from the Kruskal-Wallis test (for continuous variables not normally distributed); Categorical variables, expressed as frequency (percentage, %) of sample and p-values were derived from the Chi-square test; BMD, bone mineral density; LBMD, low bone mineral density; PKR, Pakistani Rupees; QUS, quantitative ultrasound; *p<0.05.

Findings compare the mean daily intake of various nutrients (± standard deviation) across three bone health categories: Normal BMD, Osteopenia, and Osteoporosis and show statistically significant differences for nearly all variables (Table 2).

Table 2: Nutrient Intake and Bone Health Status with Nutrition-Related Data

Parameters	Normal	Osteopenia	Osteopenia	p-Values
Energy Intake (Kcal/dy)	2851 (144.3%)	2448 (714.6%)	2609 (827.2%)	<0.001*
Proteins ((g)	90.8 (64.4%)	60.5(27.9%)	75.6 (51.7%)	<0.001*
Proteins (Animal) (g)	67.9 (42.4%)	40.2 (23.8%)	54.0 (45.1%)	<0.001*
Proteins (Vegetable) (g)	21.9 (41.5%)	19.7 (9.3%)	20.7 (15.6%)	<0.001*
Total Fat (g)	101.6 (60.4%)	74.8 (42.5%)	87.5 (55.5%)	<0.001*
Saturated Fats (g)	45.4 (27.3%)	41.41 (23.6%)	8.5 (26.7%)	<0.001*
Polyunsaturated Fatty Acids (g)	20.9 (14.1%)	16.9 (9.1%)	18.8 (12.5%)	<0.001*
Monounsaturated Fatty Acids (g)	44.5 (26.5%)	26.5 (19.1%)	40.5 (23.2%)	<0.001*
Carbohydrates (g)	180 (21.6%)	152 (80.2%)	166 (165.8%)	<0.001*
Vegetable Fibers (g)	26.7 (23.2%)	14.3 (12.5%)	20.6 (19.5%)	<0.001*
Vitamin C (mg)	141.5 (121.2%)	118.5 (102.0%)	124.7 (101.8%)	<0.001*
Vitamin D (µg)	9.9 (5.2)	6.4 (7.5%)	7.78 (6.8%)	<0.001*
Phosphorus (mg)	1012.3 (425.3%)	1142.3 (340.5%)	1071.3 (221.2%)	<0.001*
Calcium (mg)	964.9 (516.3%)	918.8 (211.0%)	942.8 (222.7%)	0.366
Potassium (mg)	6442 (3241.4%)	4433 (1562.0%)	5243 (1482%)	<0.001*
Magnesium (mg)	534 (235%)	441 (156.2%)	451 (189.7%)	<0.001*
Zinc (mg)	17.8 (9.0%)	14.2 (5.2%)	14.8 (7.3%)	<0.001*

In addition, the intake of different nutrients as % of RDA for these nutrients differed among the three groups. The intake of nutrients as % of RDA was ≥RDA for the majority of nutrients except vitamin D, Calcium and phosphorus (Figure 1).

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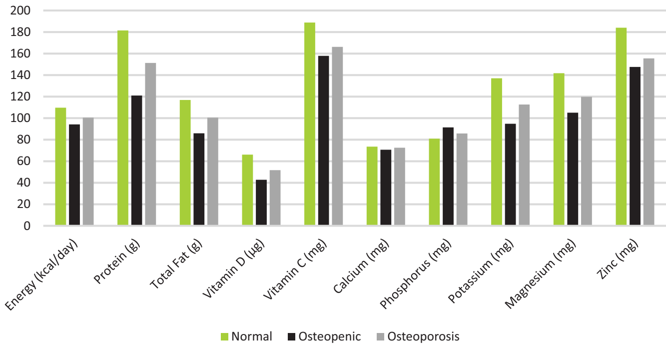

Figure 1: Intake of Nutrients as % of RDA

DISCUSSION

The current research evaluated bone mineral density (BMD) in a group of 450 women through the use of Quantitative Ultrasound (QUS) technology, generating significant information regarding the prevalence and severity of osteopenia and osteoporosis among them. The average T-score of the overall sample (-1.76 ± 0.81) suggests that the typical respondent lies within the osteopenic range, revealing an alarming trend of at-risk bone health in a significant percentage of the study sample. The grouping of mean T-scores also corroborates this observation, with osteopenic and osteoporotic subjects having much lower mean values (-1.66 ± 0.34 and -2.66 ± 0.25, respectively) than subjects with normal BMD (-0.42 ± 0.43). The application of QUS provides depth to the analysis. Mean values for BUA (48.9 ± 8.13 dB/MHz), SOS (1514.3 ± 7.14 m/s), SI (67.09 ± 15.40), and eBMD (0.41 ± 0.07 g/cm^2) reflect a range of bone health, the lower values of which in these parameters are usually related to weakened bone strength and fracture risk [10, 11]. The fact that QUS can measure both structural and elastic properties of bone qualifies it as a highly valuable, radiation-free substitute for DXA in community practice, particularly where DXA facilities are scarce. Dual-energy X-ray absorptiometry (DXA) has been deemed the gold standard in measuring bone mineral density (BMD) because of its accuracy and reproducibility. For this particular study, quantitative ultrasound (QUS) was chosen over DXA for several practical and ethical reasons. First, QUS is a radiation-free method, which makes it more suitable for adolescent or repeat-assessment populations. This is with ethical considerations for limiting radiation exposure, particularly among younger subjects. Second, QUS is non-invasive, portable, and affordable, making it suitable to be employed in field or community-based studies where access to sophisticated imaging equipment such as DXA is not available. The logistical convenience of QUS proved vital for this study involving semi-rural dwellers. Thirdly, while QUS is not a direct measurement of BMD like DXA, it gives clinically useful information pertaining to bone quality (e.g., stiffness index, speed of sound), which has predictive value for fracture risk as well. Various studies have established QUS as a valid screening tool for evaluating bone health, especially in population-based epidemiological research. The present research found a high prevalence of osteopenia (59.8%) and osteoporosis (22.2%) among the participants, with only 18% exhibiting normal BMD. These results concur with various population-based research studies showing an increasing low bone mass prevalence among females, especially menopausal women, as a result of hormonal alterations, inadequate calcium and vitamin D nutrition, and physical inactivity [12, 13]. The high percentage of women with suboptimal BMD in this study indicates that a high proportion of the population is at higher risk for fragility fractures, especially hip and vertebral fractures, that may cause considerable morbidity, mortality, and economic strain [14]. The current results underscore important nutritional distinctions among women with normal BMD and osteopenia or osteoporosis. In particular, women with normal BMD had greater energy and protein intake, specifically from animal sources, indicating that sufficient caloric and protein intake might prevent bone loss [15]. Protein not only contributes to the formation of bone matrix but also increases the absorption of calcium and the production of insulin-like growth factor 1 (IGF-1), both essential for bone formation [16]. The greater consumption of unsaturated fats, particularly monounsaturated and polyunsaturated fatty acids, by the normal BMD group is consistent with research that states they have an anti-inflammatory function in preventing bone resorption [17]. In addition, higher intakes of dietary fiber, vitamin D, vitamin C, magnesium, potassium, and zinc were positively linked with normal BMD status. These micronutrients are implicated in bone metabolism by providing cofactors and nutrients for collagen synthesis, osteoblast function, and mineralization [18, 19]. Even though the values in Table 2 suggest that calcium intake among the osteopenic and osteoporotic groups did not differ significantly, it is of interest that both groups had mean intakes that were below the RDA, specifically 91.88% for the osteopenic group and 94.27% for the osteoporotic group. This minimal inadequacy could perhaps remain of clinical importance, considering the cumulative impact of prolonged suboptimal calcium consumption on bone turnover. Notably, calcium's function to preserve bone mineral density is not singular. Its efficacy is affected by the availability and sufficiency of cofactors like vitamin D, magnesium, and phosphorus, which help assimilate and utilize calcium. In our population, although levels of calcium intake were generally similar, significant differences were seen in magnesium and vitamin D intakes, both of which were greater in the osteoporotic group,

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perhaps as a compensatory dietary response following diagnosis or through supplementation. This could partly account for why calcium on its own did not exhibit a distinguishing influence between groups. Thus, these results highlight the intricacy of nutrient interactions in bone metabolism and indicate that correcting calcium intake without securing the sufficiency of its synergistic nutrients would confine its protective action against bone loss [20]. Accordingly, our findings support the multifactorial etiology of dietary impact on bone, highlighting the value placed upon a well-balanced, nutrient-rich diet in osteoporosis prevention in women. As shown in Table 1, notable correlations were found between QUS scores and a number of demographic and anthropometric variables such as age, menopausal years, BMI, WHR, and educational level. These parameters can be potential confounders in the interaction between diet and BMD. For example, age and menopause duration are directly related to hormonal alterations, i.e., decreased levels of estrogen, which stimulate bone resorption and contribute to osteoporosis risk regardless of dietary consumption. In the same way, BMI and WHR can represent body composition and fat patterning, respectively, both of which can impact bone loading and estrogen synthesis from fat tissue, thereby altering outcomes for bone health. Increased BMI tends to be linked with increased bone density from mechanical loading, whereas central body fat (as indicated by WHR) has been shown to impair bone quality via inflammatory mechanisms. In addition, education level, as it relates to health literacy and socioeconomics, can shape food choices, consumption of nutrient-dense foods, and participation in bone-protective activities. Each of these factors can independently impact both patterns of nutritional intake and outcomes in bone health. Hence, in interpreting the relationships between diet and BMD, it should be noted that these demographic and anthropometric factors can confound or mediate the reported associations. Future studies utilizing multivariate modeling or stratified analyses are advised to adjust for these confounding influences and further remove the independent effect of diet on bone. This research demonstrates that low bone status in Charsadda women is associated with inadequate consumption of these important nutrients, such as vitamin D, protein, magnesium, and potassium, although calcium intake was similar between groups. Strong correlations with age, menopausal status, BMI, and WHR indicate that both nutritional and non-nutritional variables are related to bone status. To correct these, there is a requirement for specific public health interventions such as dietary education programs in the community, promotion of dietary intakes rich in nutrients, and exercise. Local dietary fortification and supplementation programs should be investigated as well. Future studies must test such interventions as well as investigate more general determinants influencing bone health among this population. Malnutrition is common in this part of the world [21-24], which needs appropriate strategic actions on local and national levels.

CONCLUSIONS

It was concluded that poor bone health among women in Charsadda is linked to suboptimal intake of key nutrients like vitamin D, protein, magnesium, and potassium, despite comparable calcium intake across groups. Significant associations with age, menopausal duration, BMI, and WHR suggest that both nutritional and non-nutritional factors influence bone status.

Authors Contribution

Conceptualization: AZ
Methodology: AZ, IA
Formal analysis: AZ, IA
Writing review and editing: AZ, IA

All authors have read and agreed to the published version of the manuscript.

Conflicts of Interest

All the authors declare no conflict of interest.

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Impact of Breakfast Skipping on Functional Capacity and Productivity among Office Workers with Standard Working Hours (9 AM–5 PM)

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ABSTRACT

Breakfast skipping is a prevalent behavior among working adults, yet its impact on workplace productivity in 9–5 office workers remains underexplored. **Objectives:** To assess the prevalence of breakfast skipping and its association with productivity, cognitive function, and well-being in this population. **Methods:** A cross-sectional study was conducted among 540 full-time office workers. Data were collected via an online questionnaire assessing breakfast habits, self-rated productivity (World Health Organization Health and Work Performance Questionnaire (WHO-HPQ)), presenteeism, and energy levels. A subset (n=312) completed an objective psychomotor vigilance task (PVT). Habitual breakfast skipping was defined as skipping breakfast ≥3 workdays/week. Multivariable regression analyses adjusted for age, sex, sleep duration, and caffeine intake. **Results:** The prevalence of habitual breakfast skipping was 39%. The most common reasons were lack of time (63%) and absence of morning hunger (41%). Habitual skippers reported significantly lower morning productivity (mean score 6.2 vs. 7.4, $p<0.001$) and higher rates of presenteeism (28% vs. 14%, $p=0.002$) compared to regular consumers. They were also more likely to experience low energy before lunch (52% vs. 29%, odds ratio (OR)=2.45). Objectively, skippers demonstrated slower reaction times on the PVT (median 345 ms vs. 310 ms, $p=0.01$). These associations remained significant after adjusting for confounders. **Conclusions:** Breakfast skipping is common among office workers and is significantly associated with reduced productivity, increased fatigue, and poorer cognitive performance. Workplace wellness initiatives should prioritize promoting regular breakfast consumption to enhance employee health and organizational efficiency.

INTRODUCTION

Breakfast is often described as the most important meal of the day, providing essential nutrients that help maintain energy balance and cognitive function throughout the morning hours [1]. Despite this, breakfast skipping has become increasingly common among working adults, particularly those engaged in 9–5 office jobs, due to time constraints, long commutes, or weight-control motives [2]. This habit is of particular concern in populations with high mental and physical demands during the workday, where sustained concentration and energy are critical for productivity. A growing body of evidence suggests that breakfast consumption is positively associated with better cognitive performance, mood stability, and work efficiency [3]. For instance, studies show that adults who regularly consume breakfast demonstrate improved memory, attention, and problem-solving abilities compared to those who skip it [4, 5]. Conversely, breakfast omission has been linked to mid-morning fatigue, irritability, reduced physical activity, and increased caloric intake later in the day [6, 7]. Over the long term, habitual breakfast skipping is

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associated with higher risks of obesity, insulin resistance, hypertension, and cardiovascular disease [8, 9]. Although numerous studies have explored breakfast habits among adolescents and students, fewer have examined their impact on working adults who spend prolonged hours in sedentary, mentally demanding environments [10]. This gap is critical because workplace performance and long-term health outcomes directly affect both individual well-being and organizational productivity. Furthermore, the study intends to provide evidence-based recommendations for workplace health interventions that promote regular breakfast consumption to improve both individual health outcomes and organizational performance.

This study aimed to assess the prevalence and underlying reasons for breakfast skipping among 9–5 workers, while also examining its immediate and long-term consequences. Specifically, it seeks to explore the short-term effects of breakfast omission on energy levels, concentration, and workplace productivity, alongside the potential long-term health risks, such as metabolic disorders and reduced overall well-being.

METHODS

This cross-sectional study collected original data from full-time office workers aged 18-60 in Lahore, Pakistan, from March to May 2024. A non-probability, convenience sampling method was employed. The sample size was calculated using the formula for estimating a single population proportion: $n = (Z^2 * P(1-P)) / d^2$. Assuming a 50% prevalence (P) of breakfast skipping (to maximize sample size), a 95% confidence level (Z = 1.96), and a 5% margin of error (d = 0.05), the minimum required sample size was 385. This study recruited 540 participants to account for potential non-response and to ensure sufficient power for subgroup analyses. A written informed consent was taken. Breakfast habits were categorized as daily consumption (0 days skipped), occasional skipping (1-2 days/week), or habitual skipping (≥3 days/week), defined as consuming ≥50 kcal within two hours of waking. A breakfast episode was objectively defined as consuming ≥50 kcal within two hours of waking to distinguish it from the consumption of very low-calorie beverages such as black coffee or tea. Data were collected via an anonymous online questionnaire and optional objective cognitive tests. The questionnaire gathered sociodemographic data and used validated instruments, including the WHO-HPQ for productivity [11], the Cognitive Failures Questionnaire for subjective cognitive function [12], and the PSQI for sleep quality [13]. All self-report instruments were forward-translated to Urdu and back-translated to English by bilingual experts to ensure conceptual equivalence, and were piloted for clarity and cultural relevance. A 5-minute psychomotor vigilance task (PVT) provided an objective measure of attention [14]. Primary outcomes were self-rated productivity, objective attention, and morning alertness, analyzed using bivariate tests and multivariable regression models. Adjusted model controls for age, sex, sleep duration, and caffeine intake. Data were analyzed using (R 4.5.1). The normality of continuous data was formally assessed using the Shapiro-Wilk test. Based on the results of this test, which indicated a significant deviation from normality for key variables ($p<0.05$), independent samples t-tests were used for normally distributed data, and the Mann-Whitney U test was used for non-normally distributed data. Chi-square tests were used for categorical variables. To complement null hypothesis significance testing, effect sizes were calculated, including Cohen's d for continuous outcomes and risk differences for proportional outcomes, along with their 95% confidence intervals. A p-value of <0.05 was considered statistically significant.

RESULTS

The sample consisted of 540 working adults with a mean age of 31.4 years (SD = 7.8), indicating a relatively young to middle-aged cohort. The gender distribution was nearly balanced, with a slight majority of male participants (54%). The workforce was predominantly drawn from the Information Technology sector (28%), followed by Finance (21%) and Education (17%), reflecting a sample engaged in primarily sedentary, cognitively demanding professions (Table 1).

Table 1: Demographic Characteristics of Participants (n=540)

Characteristic	Category	Mean ± SD / (%)
Age	(Years), Mean (SD)	31.4 ± 7.8
Gender	Male	54%
	Female	46%
Industry	IT	28%
	Finance	21%
	Education	17%
	Government	15%
	Other	19%

The data reveal that breakfast skipping is a common practice within the studied workforce, with a notable finding that 73% of office workers skip breakfast at least once weekly. Furthermore, habitual skipping (≥3 days/week) was the most prevalent pattern, reported by 39% of the cohort, indicating a substantial proportion at potential risk for associated negative outcomes (Table 2).

Table 2: Breakfast Habits among Participants

Breakfast Habits	n (%)
Habitual Skipper (≥3 Days/Week)	211 (39%)
Occasional Skipper (1–2 Days/Week)	183 (34%)

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Daily Consumer	146 (27%)

The reported effect sizes indicate that the associations are not only statistically significant but also substantively meaningful. The moderate-to-large Cohen's d values for productivity and sleep duration (d = 0.71 and d = 0.58, respectively), coupled with substantial risk differences for presenteeism and fatigue (14% and 23%), demonstrate that breakfast skipping has a clinically relevant impact on key functional outcomes. These findings confirm that the observed differences are practically significant beyond their statistical probability (Table 3).

Table 3: Key Outcomes by Breakfast Habit

Outcomes	Skippers (n=211)	Consumers (n=146)	p-Value	Effect Size (95% CI)
Morning Productivity (WHO-HPQ), Mean ± SD	6.2 ± 1.8	7.4 ± 1.6	<0.001	Cohen's d = 0.71 (0.51 to 0.91)
Presenteeism (>50% Day Lost), n (%)	59 (28%)	20 (14%)	0.002	Risk Difference = 14% (6% to 22%)
Low Energy Before Lunch, n (%)	110 (52%)	42 (29%)	<0.001	Risk Difference = 23% (14% to 32%)
Sleep Duration (Hours), Mean ± SD[1]	6.2 ± 1.1	6.8 ± 1.0	0.04	Cohen

The objective PVT data reveal that habitual breakfast skippers exhibited significantly slower median reaction times (345 ms vs. 310 ms, p=0.01) and a three-fold higher rate of attention lapses (median of 3 vs. 1, p<0.001) compared to daily consumers. These findings provide robust evidence that skipping breakfast impairs fundamental cognitive processes like processing speed and sustained attention. This cognitive deficit objectively substantiates the subjects' reports of reduced productivity and increased presenteeism (Table 4).

Table 4: Psychomotor Vigilance Task (PVT) Results by Breakfast Habit Group (n=312)

PVT Metric	Daily Consumers (n=146)	Habitual Skippers (n=135)	p-Value
Reaction Time (ms), Median (IQR)	345 (300-410)	310 (280-350)	0.01
Number of Lapses (≥500 ms), Median (IQR)	3 (1-6)	1 (0-3)	<0.001

The regression analysis confirms that while factors like shorter sleep explain part of the negative association, skipping breakfast itself has a strong and independent detrimental effect. After adjustment, habitual skippers still showed a clinically meaningful drop in productivity and were over twice as likely to experience morning fatigue. This underscores that promoting breakfast consumption can directly enhance employee well-being and performance, beyond other lifestyle factors (Table 5).

Table 5: Unadjusted and Adjusted Associations of Breakfast Skipping with Key Outcomes

Outcome	Model	Effect Estimate (95% CI)	p-Value
Morning Productivity (WHO-HPQ Score)	Unadjusted	β = -1.2 (95% CI: -1.5 to -0.9)	<0.001
	Adjusted*	β = -0.8 (95% CI: -1.1 to -0.5)	<0.001
Morning Fatigue (Low Energy before Lunch)	Unadjusted	OR = 2.45 (95% CI: 1.65 to 3.65)	<0.001
	Adjusted*	OR = 2.1 (95% CI: 1.3 to 3.4)	<0.01

DISCUSSION

Habitual breakfast skipping was prevalent among office workers and demonstrated a significant, dose-response association with lower self-reported productivity, greater presenteeism, and increased morning fatigue. These findings are consistent with international evidence and controlled trials showing breakfast improves cognitive performance [15, 16]. The objective measure of poorer psychomotor vigilance [17] and systematic review evidence [18] strongly corroborate these results. Our findings are strongly supported by a growing body of recent literature. A meta-analysis by Gwin and Leidy concluded that breakfast consumption significantly improves subjective alertness and aspects of cognitive performance compared to skipping, directly aligning with our observations on energy and productivity. Furthermore, a study demonstrated that changes in breakfast habits were associated with changes in self-rated health and depressive symptoms in Korean adults, providing temporal support for a potential causal link that our cross-sectional design can infer but not confirm [19]. The objective cognitive deficits study observed via the PVT is particularly telling. Research by Yoshizaki *et al.* similarly found that breakfast skipping was associated with slower reaction times and higher mental fatigue in Japanese workers, especially those with poor sleep quality, underscoring the cross-cultural validity of this effect and its interaction with other lifestyle factors [20]. The physiological mechanisms underlying these effects are likely multifaceted. Breakfast consumption helps restore glycogen stores and maintain stable blood glucose levels after an overnight fast, which is critical for optimal brain function [21]. Skipping breakfast can lead to metabolic stress and an increase in cortisol, potentially exacerbating feelings of stress and impairing higher-order cognitive processes like attention and executive function [22]. This metabolic disruption provides a plausible biological pathway for the increased irritability, fatigue, and reduced concentration reported by habitual skippers in our study. The study also found breakfast skipping was linked to shorter sleep duration, a correlation supported by research on meal timing and circadian rhythms [23]. Emerging evidence suggests that meal timing acts as a zeitgeber (synchronizer) for peripheral clocks, influencing

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sleep-wake cycles. A study by McHill *et al.* found that caloric intake timing was associated with circadian phase and sleep duration, suggesting that the habit of skipping breakfast may be part of a broader pattern of circadian misalignment that negatively impacts both sleep and daytime functioning [24]. Importantly, caffeine intake did not compensate for the deficits, echoing findings that breakfast offers unique cognitive benefits over caffeine alone [25]. This underscores that the nutritional components of a meal provide sustained energy and neurochemical precursors that a stimulant cannot replicate.

CONCLUSIONS

This study demonstrates that breakfast skipping is a common behavior among 9–5 workers and is significantly associated with reduced self-reported productivity, increased fatigue, and poorer cognitive performance, as objectively measured by slower psychomotor vigilance reaction times. These findings emphasize that breakfast consumption plays a crucial role in sustaining cognitive efficiency and energy levels during work hours. Workplace wellness initiatives should therefore prioritize promoting regular breakfast consumption to enhance employee well-being and organizational performance.

Authors Contribution

Conceptualization: MI, IF
Methodology: MI
Formal analysis: MI
Writing review and editing: IF

All authors have read and agreed to the published version of the manuscript.

Conflicts of Interest

All the authors declare no conflict of interest.

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Plant-Based Diet Adherence and Type 2 Diabetes Risk Factors

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ABSTRACT

Type 2 diabetes (T2D) prevalence is increasing worldwide. Dietary patterns, particularly plant-based diets, have been linked to improved glycemic control and lower T2D risk. **Objectives:** To analyze the relationship between plant-based dietary patterns and risk factors for T2D using publicly available secondary data. **Methods:** This cross-sectional secondary data analysis utilized data from the 2019 National Health and Nutrition Examination Survey (NHANES). Adults aged 20–65 years were categorized into four dietary patterns based on 24-hour dietary recalls: vegan, vegetarian, semi-vegetarian, and omnivore. Risk factors analyzed included body mass index (BMI), fasting blood glucose, and HbA1c. Statistical analyses were performed using SPSS version 27.0. ANOVA and post-hoc tests assessed differences among groups, while Spearman correlation evaluated associations between plant-based diet adherence and T2D risk factors. The p-values <0.05 were considered statistically significant. **Results:** The study included 5,200 adults (52% female; mean age 42.3 ± 12.5 years Mean BMI differed significantly among dietary groups, with vegans showing the lowest (23.1 ± 2.9 kg/m^2; 95% CI: 22.9–23.3) and omnivores the highest (27.2 ± 3.7 kg/m^2; 95% CI: 27.0–27.4). Fasting glucose and HbA1c were also lowest among vegans and highest in omnivores, indicating better glycemic control with higher adherence to plant-based diets. Higher plant-based diet adherence was associated with lower BMI, fasting glucose, and HbA1c, suggesting reduced metabolic risk. **Conclusions:** Plant-based diets are associated with reduced T2D risk factors. Public health strategies should promote plant-based eating patterns to mitigate the growing burden of T2D.

INTRODUCTION

The chronic illness known as type 2 diabetes (T2D) is characterized by elevated blood sugar levels brought on by either inadequate or resistant insulin production. Globally, the prevalence of T2D has reached pandemic levels, affecting over 537 million adults in 2021 and projected to rise further in the next decades [1]. Diet and other lifestyle variables are important in preventing type 2 diabetes. Plant-based diets that prioritize fruits, vegetables, whole grains, legumes, and nuts while reducing animal products may be beneficial for metabolism. Evidence suggests that these diets improve insulin sensitivity, reduce body weight, and lower chronic inflammation [2-4]. Observational studies indicate that vegetarian and vegan diets are linked to reduced T2D incidence, but cross-population analyses are limited [5]. Type 2 diabetes (T2D) represents a major public health challenge worldwide, with escalating prevalence driven by aging populations, urbanization, and lifestyle changes [6]. In recent decades, dietary shifts toward higher consumption of processed foods, refined carbohydrates, animal products, and saturated fats have paralleled rising T2D rates [7]. In contrast, plant-based diets—rich in fiber, antioxidants, micronutrients, and phytochemicals—are thought to confer metabolic benefits through multiple pathways, including improved insulin sensitivity, modulation of gut microbiota, reduction of oxidative stress, and mitigation of low-grade inflammation

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[8]. Interventional trials have also demonstrated that transitioning to vegetarian or vegan diets yields improvements in glycemic control, weight loss, and lipid profiles among individuals with T2D [9]. However, many existing studies focus on specific populations or clinical settings, and there is still limited evidence from large, representative datasets that classify varying degrees of plant-based adherence (e.g., semi-vegetarian, vegetarian, vegan) in relation to multiple T2D risk markers [10]. Therefore, by leveraging the NHANES 2019 dataset, this study aims to fill this gap by exploring how gradations of plant-based dietary patterns are associated with BMI, fasting glucose, and HbA1c in a broad adult population, thereby contributing to the evidence base for diet-based preventive strategies in diabetes.

The aim of this study was to evaluate the association between plant-based dietary patterns and T2D risk factors (BMI, fasting glucose, HbA1c).

METHODS

This cross-sectional secondary data analysis utilized data from the 2019 National Health and Nutrition Examination Survey (NHANES). The dataset was accessed and extracted in 2024, while the statistical analysis was performed between January and March 2025 conducted at the Centers for Disease Control and Prevention (CDC). Written informed consent was taken. As this design captures associations at a single point in time, causal relationships between dietary patterns and type 2 diabetes risk factors cannot be inferred conducted by the Centers for Disease Control and Prevention (CDC). NHANES collects demographic, dietary, and health-related data using standardized methods, and publicly available datasets were obtained from the NHANES website. The survey employs a complex, multistage probability sampling design to obtain a nationally representative sample of the U.S. population. This design was considered in data collection, and the weighted nature of NHANES enhances generalizability of the findings. Participants aged 20–65 years with complete dietary recall, BMI, fasting glucose, and HbA1c data were included, while those with missing data or diagnosed type 1 diabetes were excluded. Dietary patterns were classified based on 24-hour dietary recall into four groups: vegan (no animal products), vegetarian (no meat or fish but dairy/eggs allowed), semi-vegetarian (occasional meat or fish consumption less than three times per week), and omnivore (regular meat or fish consumption). Key risk factors were assessed as follows: BMI was calculated as weight (kg)/height^2 (m^2), fasting glucose (mg/dL) was measured using standard laboratory methods, and HbA1c (%) was determined by high-performance liquid chromatography (HPLC). Statistical analyses were conducted using SPSS version 27.0, with continuous variables reported as mean ± standard deviation (SD). Before applying ANOVA, assumptions of normality (Shapiro–Wilk test and inspection of histograms) and homogeneity of variance (Levene's test) were assessed and met. Group differences were then examined using one-way ANOVA followed by Tukey's post-hoc test, and correlations between plant-based diet adherence (coded 1–4, vegan to omnivore) and type 2 diabetes (T2D) risk factors were evaluated using Spearman correlation coefficients, with significance set at $p<0.05$. As this analysis used publicly available secondary data, no direct human subject interaction occurred, and institutional review board (IRB) approval was not required.

RESULTS

The study included data of 5,200 adults aged 20–65, with 52% female and a mean age of 42.3 ± 12.5 years. Although no a priori power calculation was performed due to the secondary nature of this analysis, the relatively large NHANES sample size (n=5,200) provides adequate statistical power to detect small-to-moderate effect sizes in comparisons across dietary groups. Prior studies using NHANES data with comparable sample sizes have successfully identified associations between dietary patterns and metabolic outcomes, supporting the adequacy of the sample for this analysis. Table 1 presents that participants were categorized according to dietary habits as vegan (n=312), vegetarian (n = 598), semi-vegetarian (n = 1,024), and omnivore (n = 3,266). Mean Body Mass Index (BMI) differed significantly among dietary groups ($p < 0.01$), with vegans showing the lowest mean BMI (23.1 ± 2.9 kg/m^2), followed by vegetarians (24.5 ± 3.2 kg/m^2), semi-vegetarians (25.8 ± 3.5 kg/m^2), and omnivores (27.2 ± 3.7 kg/m^2) (Table 1).

Table 1: Mean BMI Across Dietary Patterns

Dietary Pattern	Mean BMI (kg/m^2)
Vegan	23.1 ± 2.9
Vegetarian	24.5 ± 3.2
Semi-vegetarian	25.8 ± 3.5
Omnivore	27.2 ± 3.7

The study represents fasting glucose and HbA1c levels varied significantly across dietary groups ($p<0.01$), with the lowest values observed in vegan participants, followed by vegetarians, semi-vegetarians, and omnivores. Mean fasting glucose was 92 ± 8 mg/dL in vegans and 102 ± 12 mg/dL in omnivores, while mean HbA1c was 5.4 ± 0.3% and 5.9 ± 0.5%, respectively, indicating better glycemic control among individuals adhering to plant-based diets (Table 2).

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Table 2: Comparison of Fasting Glucose and Hba1c Across Dietary Patterns

Dietary Pattern	Fasting Glucose (mg/dL)	HbA1c (%)
Vegan	92 ± 8	5.4 ± 0.3
Vegetarian	95 ± 10	5.6 ± 0.4
Semi-vegetarian	98 ± 11	5.7 ± 0.4
Omnivore	102 ± 12	5.9 ± 0.5

The study presents higher adherence to plant-based diets was significantly associated with lower BMI ($r = -0.31$, $p<0.01$; 95% CI: −0.34 to −0.28), fasting glucose ($r = -0.28$, $p<0.01$; 95% CI: −0.31 to −0.25), and HbA1c ($r = -0.29$, $p<0.01$; 95% CI: −0.32 to −0.26), indicating reduced metabolic risk (Table 3).

Table 3: Correlation Between Plant-Based Diet Adherence and Diabetes Risk Indicators

Variables	Correlation coefficient (r)	p-Value
BMI	-0.31	<0.01
Fasting Glucose	-0.28	<0.01
HbA1c	-0.29	<0.01

DISCUSSION

This secondary analysis demonstrates that greater adherence to plant-based diets is associated with lower BMI, fasting glucose, and HbA1c markers strongly linked to T2D risk. These results are consistent with global findings that dietary composition exerts a pivotal influence on metabolic health [11, 12]. A large-scale meta-analysis by Clemente-Suárez *et al.* reported a 23 % lower relative risk of developing T2D among individuals following plant-based patterns [13]. Similarly, Lv *et al.* found that vegetarian and vegan diets significantly reduced HbA1c and BMI in patients with T2D [14]. These outcomes closely mirror the gradients observed in our NHANES data, suggesting a dose-response relationship between plant-based adherence and glycemic outcomes. Earlier studies reported that plant-based diets enhance insulin sensitivity by improving gut-microbiota diversity, lowering endotoxemia, and reducing low-grade inflammation [15, 16]. Additionally, replacing animal fats with plant-derived unsaturated fats and fibers improves hepatic glucose metabolism and lipid profiles [17]. From a public-health standpoint, promoting semi-vegetarian or predominantly plant-based eating could yield measurable reductions in diabetes burden [18, 19]. However, the quality of plant-based diets remains crucial unrefined, fiber-rich foods confer benefits, whereas refined starches or sweetened plant products may not [8, 20]. The cross-sectional design limits causal inference, and the 24-hour dietary recall may not capture habitual intake. Additionally, reliance on self-reported 24-hour dietary recall introduces the possibility of recall bias and under- or over-reporting of specific foods, which may affect classification accuracy of dietary patterns. Nonetheless, using nationally representative NHANES data enhances generalizability and aligns with emerging evidence linking plant-forward diets to improved cardiometabolic health.

CONCLUSIONS

Plant-based dietary patterns are significantly associated with lower BMI, fasting glucose, and HbA1c, indicating reduced T2D risk. Promoting plant-based diets at the population level may be a feasible strategy for T2D prevention

Authors Contribution

Conceptualization: TB, AM, RN
Methodology: TB, AM, RN
Formal analysis: TB, AM, RN
Writing, review and editing: TB

All authors have read and agreed to the published version of the manuscript.

Conflicts of Interest

All the authors declare no conflict of interest.

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Experimental Trial on Ginger Plus Lemon Shots

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ABSTRACT

Acne vulgaris is a widespread skin disease that is affected by several factors, such as inflammation, hormonal status, and dietary factors. Natural products like ginger and lemon, which contain anti-inflammatory and antioxidant phytochemicals, have been potential in controlling symptoms of acne. **Objectives:** To examine the impact of the ginger plus lemon shot intervention on the severity of acne and associated hematological parameters in young adult women. **Methods:** This interventional study included 100 female volunteers between 21–24 years of age with mild to moderate acne who were included in a 10-day interventional trial. All volunteers took 100 mL each of fresh ginger and lemon shots once a day. Clinical evaluation by the Global Acne Grading System (GAGS), complete blood count (CBC), and sensory questionnaires were performed before and after the intervention. Dietary consumption and compliance were observed during the study. **Results:** The intervention significantly decreased acne lesions and redness of the skin, backed by hematological improvement. Reduction in white blood cell (WBC) count, erythrocyte sedimentation rate (ESR), and C-reactive protein (CRP) levels reflected a reduction in systemic inflammation. Sensory evaluation revealed excellent acceptability with scores higher than 4 on a 5-point hedonic scale for taste, aroma, color, and overall acceptability. **Conclusions:** Intake of ginger along with lemon shots on a daily basis for 10 days showed favorable results in alleviating acne symptoms and systemic inflammation markers without any side effects.

INTRODUCTION

Acne vulgaris is a pervasive dermatological condition affecting over 80% of individuals at some point in their lives, posing significant psychological and social challenges, particularly among young adults [1]. Multiple pathogenic mechanisms underlie the pathogenesis of acne, which includes the overproduction of sebum, hyperkeratinization of follicles, the colonization of Cutibacterium acnes, and inflammatory cascades [2]. Although there are traditional medications like retinoids, antibiotics, and hormonal therapy, they are accompanied by various side effects like skin inflammation, systemic complications, and so forth, which prompts the emergence of the desire to find more natural methods of treatment [3]. The last few years have seen much attention being given to the use of herbal and functional foods in dermatology. Around 80 percent of acnes sufferers have stated that they have been using natural or plant-based products in the search of a solution that does not only deal with the underlying causative factors but also has few side effects [4]. Ginger (Zingiber officinale) and lemon (Citrus limon) are among them, and these two have been promising because of broad-spectrum pharmacological activities. Ginger contains bioactive compounds that have a high level of anti-inflammatory, antioxidant, and antimicrobial properties including gingerols and shogaols [5]. The compounds will suppress the inflammatory actions that

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are the primary cause of acne development and suppress the proliferation of the pathogens associated with acne [6]. At the same time, lemon is rich in citric acid, vitamin C, and flavonoid and possesses antibacterial, antioxidant, and astringent properties, that help to clean pores, decrease excess sebum, and resolve post-acne hyperpigmentation [7]. Preliminary studies have indicated the potential of the synergistic effect of ginger and lemon. Previous literature has shown that the combined intake of ginger and lemon may have great effect on the skin radiance and acne lesions by increasing the anti-inflammatory and antibacterial effect [8]. Also, a recent study has pointed out the effectiveness of ginger extracts in Staphylococcus aureus and Propionibacterium acnes and lemon juice had strong bactericidal effect, which was better than in using some traditional cleansers [9, 10]. Despite these promising findings, there remains a scarcity of clinical studies evaluating the combined effect of ginger and lemon as a dietary intervention for acne management. This study, therefore, aims to investigate the effects of a 10-day intervention with ginger and lemon shots on acne severity and associated hematological markers in young adult women, providing preliminary evidence for its use as a natural, complementary approach to acne care.

METHODS

This interventional study employed a pre-post-trial design conducted over 10 days, from February 10, 2025 to February 19, 2025 to evaluate the effects of ginger-lemon shots on acne severity in young adults, with each participant serving as their own control. The study received approval from the institutional supervisor, and informed consent was obtained from all participants before enrollment. A total of 100 female volunteers aged 21–24 years with mild to moderate acne vulgaris were recruited via purposive sampling. Exclusion criteria encompassed individuals using antibiotics or other disease-based medications, those with chronic skin conditions, and persons with known allergies to ginger or citrus fruits. Participants were instructed to maintain their existing skincare regimens while refraining from initiating any new topical or oral treatments for the duration of the trial. The ginger-lemon shot was prepared daily using 50 g of fresh ginger root (*Zingiber officinale*), 50 mL of fresh lemon juice (*Citrus limon*), and 100 mL of filtered water. The preparation process involved washing, peeling, and chopping the ginger, blending it with lemon juice and water for 60 seconds using a high-speed blender, and then straining the mixture through a fine mesh to remove pulp and fibers. Each participant consumed a 100 mL shot daily for 10 consecutive days, typically in the evening. Before the intervention, a 3-day observation period was implemented to establish baseline dietary habits, skin condition, and protocol compliance, during which no shots were administered. Assessments included anthropometric measurements (height, weight, and BMI using a calibrated digital scale), biochemical analysis via complete blood count (CBC) to evaluate inflammatory markers, clinical evaluation of acne severity using the Global Acne Grading System (GAGS) with standardized photographic documentation on days 0 and 10, and dietary assessment through 3-day dietary recalls and food diaries to monitor nutrient intake and control for confounding factors such as dairy, sugar, and omega-3 consumption. All statistical analyses were performed using IBM SPSS Statistics version 27.0.

RESULTS

The high mean scores (≥4.1) across all sensory attributes indicate excellent acceptability of the ginger-lemon shots among participants. This strong palatability is crucial for adherence in nutritional interventions, suggesting that the formulation is both appealing and feasible for regular consumption. Such positive sensory reception supports its potential as a sustainable, consumer-friendly functional beverage (Table 1).

Table 1: Sensory Evaluation (N=100)

Sensory Attribute	Excellent (5)	Good (4)	Fair (3)	Poor (2)	Very Poor (1)	Mean Score
Taste	40	30	30	0	0	4.1
Aroma	60	20	20	0	0	4.4
Color	50	40	10	0	0	4.3
Overall Acceptability	40	40	20	0	0	4.2

The post-intervention results indicate a notable reduction in systemic inflammation, as evidenced by the decreases in WBC count, ESR, and CRP levels. The shift in differential counts, specifically a decrease in neutrophils and an increase in lymphocytes, suggests a positive immunomodulatory effect. These hematological improvements align with the observed clinical reduction in acne severity, supporting the anti-inflammatory properties of the ginger-lemon intervention (Table 2).

Table 2: Clinical and Hematological Outcomes Before and After a 10-Day Ginger-Lemon Shot Intervention (N=100)

Parameters	Reference Range	Baseline (Mean ± SD)	After 10 Days (Mean ± SD)	p-Value
WBC ($\times10^9$/L)	4.0 – 11.0	9.8 ± 1.5	7.3 ± 1.3	<0.001
Neutrophils (%)	40 – 75	68 ± 6	62 ± 5	<0.001
Lymphocytes (%)	20 – 45	26 ± 5	31 ± 4	<0.001
Hemoglobin (g/dL)	12.0–16.0 (female)	13.4 ± 0.8	13.5 ± 0.7	0.301
Hematocrit (%)	36 – 46	40.1 ± 2.1	40.4 ± 1.9	0.215
Platelet Count ($\times10^9$/L)	150 – 400	268 ± 40	270 ± 38	0.685
ESR (mm/hr)	0 – 20 (female)	18 ± 4	11 ± 3	<0.001
CRP (mg/L)	< 5	4.8 ± 0.9	2.1 ± 0.8	<0.001

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DISCUSSION

The results of this 10-day interventional trial indicate that Ginger plus Lemon Shots formulation is tolerable and has potential efficacy in skin health, mainly in alleviating symptoms of acne vulgaris. The high acceptability ratings of the sensory analysis are a decisive result, because palatability is one of the major determinants of long term adherence during functional food and nutraceutical interventions [11]. Clinically, the hematological parameters provide preliminary evidence of the formulation's systemic anti-inflammatory and immunomodulatory effects. The observed reductions in total leukocyte (WBC) count, erythrocyte sedimentation rate (ESR), and C-reactive protein (CRP) levels are indicative of a decrease in systemic inflammation [12]. This is highly relevant to acne pathogenesis, which is characterized by chronic low-grade inflammation [13]. The modest shift in leukocyte differentials, a decrease in neutrophils and an increase in lymphocytes, further suggests a positive modulation of the immune response, a property attributed to the key bioactive compounds in ginger, such as gingerols and shogaols [14]. The safety profile of the intervention is supported by the stable levels of hemoglobin, hematocrit, and platelet counts. The significant reduction in CRP provides further validation of the anti-inflammatory properties of the ginger and lemon mixture [15, 16]. These systemic improvements correlate with the participants' self-reported dermatological outcomes, including a reduction in acne lesions and skin redness. The concordance between improved inflammatory markers and clinical skin manifestations suggests that the mechanism of action likely involves anti-inflammatory and immunomodulatory pathways, justifying its exploration as a natural complementary approach for acne management [17, 18]. Moreover, the increasing interest in plant-derived bioactives for cosmetic and dermatological applications aligns with the growing trend of green cosmetics, where natural food ingredients like ginger and lemon are being incorporated for their antioxidant and skin-healing properties [19]. Additionally, emerging research highlights the psychodermatological benefits of nutraceuticals and psychobiotics in improving skin conditions through gut-skin and stress modulation pathways, which may further complement the effects observed in this trial [20, 21]. The interpretation of these promising results must consider the study's limitations. The uncontrolled, single-arm design, absence of a placebo group, small sample size (n=10), and short intervention period (10 days) limit generalizability and the assessment of long-term effects or side effects. The use of a single daily dose without pharmacokinetic data leaves the optimal regimen undefined. Despite these limitations, the study provides a clinical rationale for using a simple, natural formulation as an adjunct therapy for acne, aligning with the growing demand for plant-based health solutions. The improvement in hematological markers offers objective support for ginger and lemon's traditional benefits. Future research should focus on double-blind, placebo-controlled trials with larger samples, longer follow-up, standardized dermatological assessments, and exploration of mechanisms such as the gut-skin axis.

CONCLUSIONS

In conclusion, this preliminary investigation indicates that daily consumption of fresh ginger and lemon shots over 10 days may be a viable, natural, and well-accepted complementary strategy for improving skin condition and reducing acne symptoms in young adult women. The intervention was associated with significant reductions in key systemic inflammation markers (WBC, ESR, CRP) and was highly rated for its sensory properties, which is crucial for adherence.

Authors Contribution

Conceptualization: AF, MA
Methodology: AF, NW
Formal analysis: HI,
Writing review and editing: SI, EA, Q, IGH, TA, M, MA

All authors have read and agreed to the published version of the manuscript.

Conflicts of Interest

All the authors declare no conflict of interest.

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