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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(S)

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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Artificial intelligence and revolutionization in the field of food science

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The trend of AI gradually becoming prominent in most disciplines of science and it was not long before it affected biology. It is revolutionizing every aspect of animals concerning health, nutritional status, and productivity in the course of animal agriculture. For instance, the requirements for food and animal products are increasing day by day which is increasing the demand for operational effectiveness. People are using Artificial Intelligence in feeding strategies to improve and also to advance the health of the animals within farms and to increase food production thus bringing visions and innovations for a sustainable agriculture sector.

Animal nutrition did not meet individual demands based on some of the standards that were provided. AI presents various solutions since, through the use of big data, it is possible to identify animal responses to various feeding regimens. This makes it possible to come up with diets depending on individual animals which are a healthier way to make the animals more productive.

It also helps in tracking other factors such as feed consumption, weight gain, feed-to-gain ratio, and general production proficiency. In such cases feeding adjusting strategies can be ensured in the best way possible to suppress the given condition in the shortest period. An efficient feeding system in our software makes sure that the animals get the right feed at the right time to maximize its/her growth and minimal wastage likely to pollute the environment. This precise feeding practice enhances feed usage by the animals, which is another factor that contributes to the economic feasibility of the animal farming business. One of the major innovations in refined livestock farming is precision farming which focuses on feeding processes whereby feeds are prescribed depending on the needs of particular animals. An automated feeding model was devised that was able to avoid either underfeeding or overfeeding, the nutrient intake, and the time at which the food was going to be fed to the body.

AI also improves knowledge of the animal's gut microbial ecosystem which is a mix of microorganisms that inhabit the animal gut. Animal microbiota plays an important role in participating in digestion and nutrient assimilation processes and the immune system of an animal. AI in turn assists in the determination of microbial population that enhances animal health and performance and synthesizes new microbial consortiums that can be added to animal feed. Amenable animal feeds that unsurpassed the microbiota of healthy human beings may help in digestion, health, and feed conversion. These interventions are well-known ways of increasing the general health and productivity of animals among farmers, and with the help of AI, these interventions can be improved. The application of AI to the genetics of animals also extends it more into nutrition. AI uses feeding regimens from a pet's DNA to develop unique feeding plans that best suit the genetics of the animal. Not only does this method enhance efficiency but also the health of the animals is enhanced as well.

Artificial intelligence is taking up the challenge of feeding, nutrition, and welfare, as well as the excellent performance of animals and making the process more efficient and specific. Artificial Intelligence remains to develop better approaches in the enhancement of animal well-being, mechanisms of farming, and general food production.

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Original Article

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Quality Assurance of Commercial Dog Food in Terms of Proximate and Aflatoxins Analysis

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ABSTRACT

The domestication of the dog has been extensive and continues to grow, predominantly in terms of their dietary habits. Primarily, wild dogs depended exclusively on hunting for their food, but with the passage of time and domestication, they started consuming meals provided by humans. **Objective:** To evaluate the quality of dry dog food, focusing particularly on evaluating its aflatoxin contamination and proximate analysis comparable to established nutritional standards for dogs. **Methods:** This experimental research was conducted at the Food Department within the PCSIR Laboratories Complex in Lahore, Pakistan. The sample was chosen purposively to check the quality of pet dog food. The analysis included determining the moisture content, ash, crude fiber, and crude protein levels of the dog food using standardized methods. **Results:** The purchased dog food exhibited a protein content of 20.64 ± 0.40% and a calculated energy value of 376.50 ± 0.25 Kcal/100g. The results were evaluated against the nutrient benchmarks established by AAFCO (Association of American Feed Control Officials). The minimum protein level in adult dog food, calculated on a dry matter basis, must be no less than 18% according to AAFCO guidelines. **Conclusion:** Conspicuously, the analysis depicted that aflatoxins (B1, B2, G1, G2) were not present in the dog food, confirming its safe consumption by dogs based on the aflatoxin assessment. So, it was revealed that according to nutritional and aflatoxin analysis of dog food obtained from the market, it was for the dog's health.

INTRODUCTION

The increasing prevalence of pets in households is driving dynamic growth in the pet food market. Today, pet food is commonly utilized by numerous pet owners due to its ease, affordability, and accessibility, providing a trustworthy means of feeding pets throughout their lives. With an improved understanding of dogs' nutritional requirements, diets can now be tailored based on breed, age, and activity level. As dogs are often seen as integral members of the family, owners are increasingly focusing on the dietary needs of both their pets and themselves [1]. It is assumed that the pet food industry will be increasing at an annual rate of 2.6% [2]. The contamination by biological, physical and chemical components in the food of animals can pose significant risks in that they may cause sickness or loss in animals; if appropriate controls in production are not taken [3]. The most common kind of pet food in the market is normally dry, hence quite convenient in storage and effectively satisfies the nutritional needs of pets. Under EU directives (EC 767/2009), a complete diet of whole pet food used as the only nutrient source over a prolonged time meets the complete dietary requirements of the given animals in terms of their species and their stage of life [4]. Consequently, it is imperative to assess the quality of pet food, and numerous studies have been conducted to

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examine dog food [5, 6]. Another serious potential threat related to animal feed security is the occurrence of fungi and mycotoxins [7]. Comprehending the link between food, feed, and fungi is important for assessing the threat of mycotoxin infectivity. The studies have shown that these toxins in pet foods can present significant dangers to the health of pets, resulting in both severe and chronic poisoning based on the degree of contamination and the duration of contact [8]. The dogs, especially, are particularly vulnerable to the severe hepatotoxic impacts of aflatoxins [9]. Aflatoxins represent a class of tremendously harmful mycotoxins that can spoil various feed products, including commercially accessible dog food. This contamination presents a grave health threat to dogs, as aflatoxins are linked to many health issues such as liver damage, weakened immune response, and cancer. The study underscores the requirement of regular inspection for aflatoxin contamination in commercial dog food, mainly in repackaged products. It is necessary to uphold high hygiene standards during the storage and repackaging processes to prevent the formation of aflatoxins [10].

This study aimed to assess the quality of dry food for adult dogs, focusing specifically on evaluating its proximate analysis and adherence to established nutritional guidelines for dogs.

METHODS

This experimental research was conducted at the Food Department within the PCSIR Laboratories Complex in Lahore, Pakistan. The sample was chosen purposively to check the quality of pet dog food. The dry dog food (1 kg) in pellet form was obtained from a supermarket in Lahore, Pakistan, to analyze the information regarding proximate aspects of packaging material (March to May 2025). The dog food sample was ground to a mesh size of 1mm and was stored in sealed plastic bags in a cool, dry location to conserve its nutritional quality. The sample was preserved till the experimental lab analysis. Analysis involved an evaluation of moisture content, ash, crude fiber and crude protein contents of the dog food on standard standards. Dietary assessment of the reviewed pet food was determined using a dry matter (DM), crude protein (CP), crude fiber (CF) and crude ash (CA). All tests were done using the standardized methods with ISO 17025 [11] accreditation with guidance according to AOAC-2023 [12]. An amount of 10 g of the sample was dried in a hot air oven at 105°C until a stable weight was recorded, and the difference in the weight was recorded as moisture content. In case of ash content, 1g of sample was put in a pre-weighed porcelain crucible and ashed in an ashing furnace at 600°C until white ash formed and a stable weight was recorded. It determined the crude protein content as a result of multiplying the nitrogen value by 6.25 after assessing the nitrogen content through the micro-Kjeldahl procedure. Moreover, the carbohydrate content was assessed according to the guidelines established by AOAC-2023. The carbohydrate content (expressed as a percentage on a wet basis) is calculated by deducting the total percentages of crude protein, crude fat, ash, and moisture content from 100. Carbohydrate % (wet basis) = 100-(% of crude protein + % of Crude fat + % ash content + % content of moisture). The incidence of aflatoxins in dog food was assessed using the thin-layer chromatographic method [13]. A 50g powdered sample was combined with 250ml of chloroform and shaken for 30 minutes. Subsequently, 50ml of the resulting solution was evaporated using a water bath. The volume was adjusted, and the sample was applied to a TLC plate as per the given procedure. Different concentrations of standards were utilized to compare against the sample extract for aflatoxin quantification (AOAC, 2023). The total aflatoxins (AFB1+AFB2+AFG1+AFG2) were calculated using the specific formula [14]. The formula for the detection of aflatoxin B1 is given below: Aflatoxin B1 (g/kg) = S x Y x V/ W x Z. Where, S=volume in L of aflatoxin standard which is equivalent intensity to Z = L of the sample, Y=Concentration of aflatoxin standard in g/mL, V=Volume in L of solvents requisite to dilute final sample extract, Z =Volume in L of extracted sample requisite to confer fluorescence intensity similar to that of S=L of the aflatoxin standard and W=Effective weight (in grams) of original sample contained in final extract.

RESULTS

According to the nutrient content guiding principle for dry dog food, the recommendations include a minimum of 26% crude protein, at least 15% crude fat, a maximum of 5% crude fiber, approximately 37% carbohydrates, around 7% minerals, and no more than 10% moisture. Nutrient profile for dog food intended for growth and reproduction, it is suggested that adult dogs get a minimum of 18% protein and 5.5% fat on a dry matter basis (DMB), with a caloric density of 300 to 400 kilocalories of metabolizable energy (ME) per 100 grams of dry matter (DM) (Table 1).

Table 1: Proximate Assessment of Food for Pet Dog

Sr. No	Parameter (%)	Values
1	Moisture	7.00 ± 0.30
2	Ash	4.40 ± 0.09
3	Protein	20.64 ± 0.40
4	Fat	8.10 ± 0.50
5	Fibre	4.60 ± 0.20
6	Carbohydrates	55.26 ± 0.10
7	Energy (Kcal/ 100g)	376.50 ± 0.25

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In this analysis, the product was checked for aflatoxin contamination and was found safe, as no contamination of aflatoxins was detected. This indicates that the dog food sample is safe for canine health (Table 2).

Table 2: Aflatoxin Assessment of Food for Dogs

Sr. No	Aflatoxins	Concentration (ppb)
1	B1	Not Detected
2	B2	Not Detected
3	G1	Not Detected
4	G2	Not Detected
5	Total Aflatoxins	Not Detected

DISCUSSION

The contemporary findings signify that the average levels of crude fat were found to be lacking, while crude protein, fiber and ash align closely with recommendations of AAFCO, 2014. Though, notably, dry dog food exhibited lower levels of crude protein according to the recommendation of Case *et al*. [15]. The moisture levels are less than 10% as per recommendations. The proximate analysis results showed that this dog food can be regarded as a high-calorie option, providing 376.50 ± 0.25 Kcal per 100g, to meet the dietary needs of dogs. The inadequate fat content in dry dog food may create a momentous threat, as fat serves as the main energy source and provides necessary fatty acids, while also being one of the most easily digestible nutrients. Additionally, fat enhances the palatability and quality of pet food. A considerable lack of fat in dry dog food may result in energy deficiencies, mostly for working dogs [16, 17]. Aflatoxins are harmful and cancer-causing fungal metabolites that can drastically impact food quality. The most prevalent aflatoxins include aflatoxin B1, B2, G1, and G2. The relation of aflatoxins with dietary lipids may lead to digestive issues, as it reduces the activity of digestive enzymes, resulting in a malabsorption syndrome [18]. The domestic dogs, similar to other household animals, are exposed to aflatoxin poisoning that may be found in their food, which poses a considerable threat to their health [19], while fatalities from aflatoxicosis are usually linked to extremely high concentrations of aflatoxin (ranging from 100 to 6700 ppb), there have also been instances of deaths occurring at lower levels of aflatoxin (between 13 and 91 ppb). In spite of the implemented protection protocols and protective measures designed to protect dog food components and ensure precise recipe preparation, incidents of food contamination by aflatoxins may lead to health issues and even fatalities among animals. Veterinarians play a crucial role in promptly identifying the negative health impacts linked to contaminated food products [20]. The well-timed communication with manufacturers and regulatory bodies, along with comprehensive documentation, sample collection, and analytical testing, will ease the validation or dismissal of potential toxic exposure to aflatoxins.

CONCLUSIONS

The growing reliance on dogs for camaraderie and consolation is believed to be a key factor driving the noteworthy growth of the pet food sector, which in turn has led to a consequent increase in demand for dog food among pet owners and breeders. To our knowledge, the claims regarding nutritional sufficiency made by pet food companies in Pakistan have not been carefully verified. Providing dog food without accurate information regarding nutrient concentrations cannot satisfactorily meet a dog's nutritional requirements. It is suggested that manufacturers should accurately declare the nutrient concentrations in their dog food products and ensure the quality of pet food in terms of toxins. The pet dog food free of any contamination may ensure the well-being of pet dogs.

Authors Contribution

Conceptualization: NZ
Methodology: SU, SN, AY
Formal analysis: MKS, AS
Writing review and editing: NZ

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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Original Article

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Nutritional, Antioxidative and Aflatoxin Safety Assessment of Ginger-Turmeric Tea: A Functional Herbal Beverage Tea

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ABSTRACT

Turmeric and ginger belong to the family *Zingiberaceae,* which contains bioactive chemicals with a variety of uses. Ginger *(Zingiber officinale)* has several bioactive components, such as gingerols, zingiberene, caffeic acid, salicylate and capsaicin. It is utilized in a wide range of food and beverages. Turmeric *(Curcuma longa)* contains numerous physiologically active substances, including Curcumin, dimethoxycurcumin and bis-demethoxycurcumin. Both ginger and turmeric have been used to treat nausea, indigestion, stomachaches, toothaches, insomnia, asthma, diabetes, infertility, neurological issues, rheumatism and memory loss. Apart from water, tea is the most popular beverage in the world. **Objectives:** To develop ginger-turmeric tea and assess of nutritional facts, aflatoxin and antioxidant potential. **Methods:** The nutritional facts and aflatoxin were determined by standard AOAC methods, and the antioxidant activity of the ginger-turmeric tea was evaluated by DPPH assay. **Results:** The results of this study revealed that ginger-turmeric tea has 7.5 ± 0.84% moisture, 5.12 ± 0.65 ash, 0.22 ± 0.01% fat, 14.75 ± 1.67% fiber, 8.10±0.82% protein, carbohydrates 64.25 ± 3.90, and energy 292 ± 5.30 kcal/100g, while the results of aflatoxin study showed that aflatoxins (B1, B2, G1 and G2) were absent. The significant antioxidant capacity DPPH (% inhibition) was found to range from 25.50 ± 1.20–74.80 ± 2.77% at concentrations of 0.2–1.0 mg/mL, significant differences in various concentrations $p<0.05$ were found. **Conclusions:** This study concluded that the developed ginger-turmeric tea is a good source of protein, fiber and has a high content of natural antioxidants, making it advantageous for treating oxidative stress.

INTRODUCTION

Herbs and spices have long been used to improve the flavor of food and are also used medicinally to cure a variety of illnesses in many civilizations worldwide. Polyphenols and flavonoids are abundant in many plants used in traditional medicine, and these compounds represent a significant source of phytochemicals for both human and animal health [1]. Most spices have a variety of possible health benefits, including positive effects on lipid metabolism, anti-diabetic effects, stomach-stimulating properties and probable anti-inflammatory and antioxidant properties [2]. Ginger, often known as ginger root, is the rhizome of the *Zingiber officinale* and is used as a spice, medicinal or

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delicacy. Ginger contains chemical compounds, especially gingerol and shogaol, which are thought to have functions as anti-cancer, anti-inflammatory, anti-hypertensive, anti-diabetic, anti-hyperlipidemic and antioxidant [3]. The *Zingiberacea* family includes the rhizomatous herbaceous perennial plant known as turmeric. Turmeric has been shown to contain around 250 phytochemicals, mostly terpenoids and phenolic compounds including curcumins, turmerone, α-zingiberene, carotene, syringic, p-coumaric, ascorbic acid, ferulic acid, nd vanillic acid [4]. Curcumin is its active element that has garnered attention due to its potential to treat a wide range of disorders, such as diabetes, cancer, Alzheimer's disease, allergies, arthritis, and other chronic conditions [5]. Following harvest, both ginger and turmeric are dried and kept in extremely unsanitary circumstances, which increases the risk of mycotoxins and aflatoxins. Aflatoxins are fungal metabolites mostly produced by the *Aspergillus flavus* and *Aspergillus parasiticus* species. AFB1 is the most poisonous of the four main aflatoxins, which are AFB1, AFB2, AFG1, and AFG2. Hepatocellular cancer, acute aflatoxicosis, immunological suppression, malnourishment and growth deficits are all associated with AFB1 [6]. Numerous pre-harvest and post-harvest factors, such as agronomic and agricultural practices, transportation, storage and processing techniques, can result in aflatoxin contamination along the supply chain. As a result, aflatoxins should not be present in the raw material. The medicinal qualities of plants have been thoroughly investigated in the current scientific environment due to their strong antioxidant qualities, lack of side effects and viability from an economic standpoint. With a large percentage of the world's population depending on herbal medicines, herbal therapy is essential to maintaining human health and prosperity [7]. Furthermore, the usage of natural elements produced from plants has been essential since the start of the global COVID-19 epidemic. As a result, the COVID-19 pandemic has brought attention to the necessity of natural antioxidants that improve immunity, improve human health and possibly even stop oxidative stress [8].

This study aims to develop the ginger-turmeric tea and assessment of its nutritional facts, aflatoxin and antioxidant activity.

METHODS

This experimental study was conducted in March-May 2025 at the Food and Biotechnology Research Centre, PCSIR, Labs complex, Lahore. Before being utilized, the raw materials green tea *(Camellia sinensis)*, ginger *(Zingiber officinale)*, and turmeric were collected from the local market in Lahore, Pakistan and kept at room temperature (25 ± 2°C). After washing, chopping and three hours of oven drying at 70°C, the three materials were blended until a consistent powder was obtained. After that tea was prepared by properly combining these ingredients with a small cardamom and filling it in a tea bag. The ginger-turmeric tea was prepared by immersing the tea bag in hot water. 50g of the ground raw samples and the developed tea were added to 500mL of the conical flask. Add 200 mL of dichloromethane and 10 mL of water, respectively and shake it a half-hour using a shaker. In a 300 mL glass flask containing 10g of Na_2SO_4, the mixture was filtered via filter paper. After letting the contents settle gently, the solution was re-filtered into a 100 mL beaker using Whatman filter paper. On TLC plates, samples were spotted. Before being used for analysis, the TLC plates, which included a coated gel of silica on a glass plate, were activated in a hot air oven for an hour at 80°C. After drying, this plate was placed in a diethyl ether-containing TLC tank (first mobile phase) and then, for 30 minutes, it was placed in a second mobile phase tank that contained acetone and chloroform in an 88:12 (v/v) ratio. Blue fluorescence under UV light after plate development verified the presence of aflatoxin, which was then compared to standard AFB1(2µg/ml), B2 (0.508µg/ml), G1 (2.01µg/ml) and G2 (0.515µg/ml) Biopure Romar Lab Austria (Product-10003656; Lot: 1000046444). The spray of aqueous sulphuric acid (50/50 v/v) was done on the TLC plate for the confirmation of aflatoxins presence [9]. The aflatoxins were determined by using a formula. Total aflatoxins(µg/Kg)= S × Y × V/ W × Z

Where:

S = volume of aflatoxins standard in µL of equivalent intensity to Z (µL of sample).

Y = aflatoxins standard concentration in µg/mL.

V = volume of solvents in µL required to dilute final extract.

W = weight of original sample in g contained in final extract.

Z = volume of sample extract (µL) require to give fluorescence intensity comparable to that of S. Fs total = concentration of AFB1 + AFB2 + AFG1 + AFG2.

Nutritional profile (moisture, ash, fat, crude fiber, crude protein and carbohydrate contents were determined for raw ginger, turmeric and tea samples developed by AOAC standard methods [10]. By immersing a 1.5g tea sachet sample in 100ml of boiling water, the tea sample's aqueous extract was produced. These were left for one to two minutes. Additionally, their level of antioxidant activity was evaluated as well. The approach adapted by Saeed *et al.* was used to test the extracts' scavenging capabilities on the stable free radical DPPH [11]. 2.9 mL of methanolic 2,2-diphenylpicrylhydrazyl radical scavenging capability (DPPH) solution was combined with 0.1 mL of tea water extract. Each sample was incubated in the dark for 30

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minutes, and its absorbance at 517 nm was compared to a blank. By measuring the sample's and standard BHT absorbance and using the following formula, the proportion of percentage inhibition (DPPH) was determined: Percentage inhibition (DPPH)=[1−(As/A0)]×100. Where A0 represents the absorbance of the DPPH solution, and as represents the absorbance of the samples. The results were statistically analyzed using SPSS Minitab software version 25. Data were represented as mean ± SD, and analysis of variance (ANOVA) was performed. Statistical significance was defined at $p<0.05$.

RESULTS

The moisture, ash, fat, fiber, protein, carbohydrate contents and energy of ginger, turmeric and ginger-turmeric tea were determined by AOAC standard methods. According to this study, turmeric had the highest moisture content (13.20%), followed by ginger (10.60%) and ginger-turmeric tea (7.50%). Turmeric had the lowest amount of ash (3.10%), followed by developed tea (5.12%) and ginger (5.80%). Turmeric had the highest lipid content (4.70%), followed by ginger (3.70%) and developed tea (0.22%). According to the fiber analysis, developed tea had the highest fiber content (14.75%), followed by ginger (13.50%), and turmeric had the lowest (2.78%). The highest protein level was found in ginger (8.90%), followed by developed tea (8.10%) and turmeric (5.85%). Ginger had carbohydrate content (57.50%), while developed tea (64.25%) and turmeric had the highest (68.37%). Turmeric has the maximum energy 343 ± 5.80Kcal/100g, followed by ginger 299 ± 5.38Kcal/100g and developed tea 292 ± 5.30 Kcal/100g (Table 1).

Table 1: Nutrition Information of Raw Material and Ginger-Turmeric Tea

Parameters	Ginger	Turmeric	Ginger-turmeric Tea
Moisture	10.60 ± 1.44	13.20 ± 1.56	7.50 ± 0.84
Ash	5.80 ± 0.69	3.10 ± 0.40	5.12 ± 0.65
Fat	3.70 ± 0.43	4.70 ± 0.50	0.22 ± 0.01
Fiber	13.50 ± 1.61	2.78 ± 0.67	14.75 ± 1.67
Protein	8.90 ± 0.84	5.85 ± 0.56	8.10 ± 0.82
Carbohydrates	57.50 ± 3.80	68.37 ± 3.98	64.25 ± 3.90
Energy (Kcal/100g)	299 ± 5.38	343 ± 5.80	292 ± 5.30

Aflatoxin was measured in this study using thin-layer chromatography (TLC), and the findings indicated that all samples of raw ginger, turmeric, and developed ginger-turmeric tea were free from aflatoxins. All the samples are fit for human consumption according to the EU limit (2023), i.e. 5-10 ppb for ginger and turmeric, while it is 0-20 ppb for developed tea (Table 2).

Table 2: Determination of Aflatoxin Contamination in Raw Ingredients and Ginger-Turmeric Tea

Samples	Aflatoxins (ppb) B1, B2, G1 and G2	Total Aflatoxins	Fit/ Unfit	EU Limit (2023)
Raw Material Ginger Powder	Not Detected	Not Detected	Fit	5-10 ppb
Raw Material Turmeric Powder	Not Detected	Not Detected	Fit	5-10 ppb
Raw Material Green Tea	Not Detected	Not Detected	Fit	0-20 ppb
Ginger-turmeric Tea	Not Detected	Not Detected	Fit	0-20 ppb

Five distinct concentrations, ranging from 0.2 to 1.0 mg/ml, were employed to assess the antioxidant activities of the developed ginger-turmeric tea. After 30 minutes, measurements were made. The findings show that the % inhibition (DPPH), ranging from 25.50 ± 1.20 to 74.80 ± 2.77, increased with increasing concentration based on the ANOVA test. it concluded that the antioxidant activity of the developed tea was significantly different at different concentrations ($p<0.05$). The highest free radical scavenging activity was seen at a concentration of 1 mg/ml; however, all tea extracts demonstrated a notable capacity to eliminate free radicals (Figure 1).

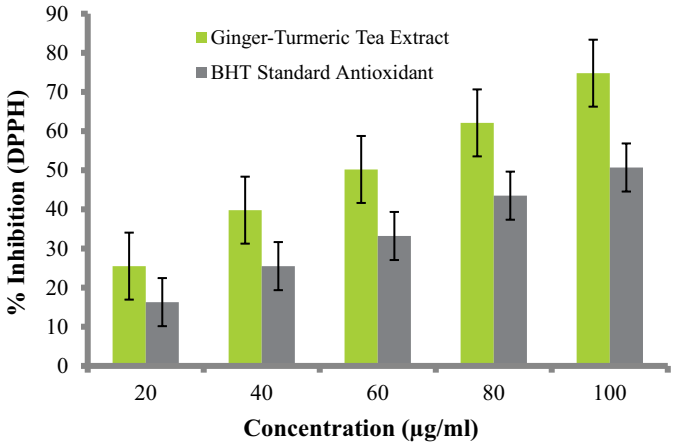

Figure 1: Antioxidant Activity % Inhibition (DPPH) Ginger-Turmeric Tea and BHT

DISCUSSION

According to reports, dried ginger has the following contents: 7.0–10.90% moisture, 8.50–12.4% protein, 70–72% carbohydrate, 5.70–6.64% ash, 7.17–14.1% fiber, and 1.8–4.0% fat. While the turmeric's overall composition is between 11 and 13% moisture, 6 to 9% protein, 5 to 10% fat, 60 to 70% total carbohydrates, 3 to 7% ash and 2 to 7% fiber [12]. The 8.85% crude protein content of ginger-turmeric tea suggests that it may be a useful source of protein. Turmeric's 5.12% ash percentage indicates that it has a respectable amount of minerals. By eliminating possible carcinogens from the body and preventing the absorption of excess cholesterol, the fiber (14.75%) in the developed product will aid in cleansing the consumer's digestive tract. Additionally, fiber gives meals volume and

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keeps people from consuming too many starchy foods, which may protect against metabolic diseases, including diabetes mellitus and hypercholesterolemia [13]. The developed product has 64.25% carbohydrate content, according to the current investigation. It has a high calorie value and was suggested by the high carbohydrate content [14]. The Food and Agriculture Organization (FAO) estimates that mycotoxins contaminate about 25% of the world's food crop supply each year, with aflatoxins being the most dangerous [15]. It is concerning to note that over 5 billion individuals globally are regularly exposed to elevated quantities of aflatoxin, surpassing 1000 parts per billion (ppb) [16]. More than 100 countries and regions have put in place unique aflatoxin threshold requirements for different food products in order to guarantee food safety. In this regard, the European Union (EU) has imposed comparatively strict regulations. They specifically state that the overall amounts of aflatoxins (AFB1, AFB2, AFG1 and AFG2) in all cereals should not be more than 4 ppb, while the values for spices should be between 5 and 10 ppb and for raw green tea powder, 20 ppb [17]. Aflatoxins are a major contributor to the formation of primary hepatocellular carcinoma, a form of malignant liver tumor and can seriously harm the liver. According to studies, aflatoxins are responsible for over half of the risk of liver cancer in Asia and Africa [18, 19]. Both our novel tea and its raw material are safe for human consumption and free of aflatoxins. The sample's ability to scavenge 2,2-diphenyl-1-picrylhydrazyl (DPPH) is a gauge of its antioxidant capacity. In the presence of oxidants, DPPH is reduced and loses its purple-blue blue when it takes an electron provided by an antioxidant component, which can be measured by changes in absorbance [20]. Because ginger, a raw ingredient in ginger-turmeric tea, includes non-nutritional chemicals with antioxidant qualities, it helps strengthen the body's immune system. Mahmudati *et al.* investigated the antioxidant activity of three different types of ginger and depicted that red ginger had the maximum antioxidant activity by the infusion method, 79.83%, followed by elephant 70.43% and emprit ginger 61.70%, respectively [3]. On the other hand, the decoction of elephant ginger showed the highest antioxidant activity, 78.76%, followed by red ginger, 70.56% and elephant ginger, 60.93%. According to Priyanka *et al.* who examined the antioxidant activities of different varieties of turmeric, other ingredients of this tea also have significant antioxidant activity [20]. They showed that the Prathibha variety's higher antioxidant activity might be caused by a higher percentage of phenolics, flavonoids, curcuminoids and natural antioxidants. Antioxidants found in ginger help prevent free radicals from harming the body's immune system cells by preventing their entry. Additionally, antioxidants help to boost immuno-stimulatory activity, and cells help to maximize the immune system, and turmeric is less immune-stimulating than ginger [20]. To preserve the body's immune system, ginger can have a therapeutic effect by boosting DNA repair, increasing antioxidants, and reducing lipid peroxidation [21]. Green tea *(Camellia sinensis),* a popular beverage high in polyphenols, was the third component of this innovative tea. Studies showing green tea's anti-carcinogenic, anti-microbial, antiviral, anti-hypercholesterolemic, anti-obesity, anti-inflammatory and antioxidant qualities have stoked interest in the beverage over time. Green tea's high catechin content, which makes up between 15 and 27% of the dry weight of the leaves, is thought to be responsible for its health benefits [22]. The inclusion of phytochemicals such as catechins, flavonoids, polyphenols, and tannins may be the cause of the developed novel tea's high level of antioxidant activity. The substantial antioxidant qualities of several plant-derived substances, including tannins, flavonoids, terpenoids, glycosides, alkaloids, and phenols, have also been demonstrated by earlier research [23]. As far as there isn't a tea like this in this combination, and it's a novel tea that was developed first time in Pakistan.

CONCLUSIONS

According to the findings of this study, a new ginger-turmeric tea has been developed that offers natural antioxidants in addition to two essential nutrients fiber and protein. Moreover, developed tea contains numerous bioactive compounds that can be utilized to prevent or cure a variety of human health conditions, including diabetes, inflammation, oxidative stress, immune system diseases and neurological disorders.

Authors Contribution

Conceptualization: MKS
Methodology: MKS, SA
Formal analysis: HKA, AK, SJ
Writing review and editing: NZ, AS

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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The Rising Trend of Energy Drink Consumption among University Students: A Hidden Health Risk

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ABSTRACT

Energy drinks, highly caffeinated beverages marketed to combat fatigue, are a global public health concern due to their association with adverse effects like insomnia and cardiovascular symptoms, particularly among young adults. **Objective:** To determine the prevalence, predictors, and self-reported health effects of energy drink consumption among university students in Pakistan. **Methods:** A cross-sectional online survey was conducted among 460 students at the University of the Punjab, Lahore. Data on consumption patterns, knowledge, and health effects were collected. First, bivariate analyses were performed to identify factors associated with frequent energy drink use. Subsequently, multivariable logistic regression was used to identify predictors of frequent energy drink use (≥3 times/week). The AOR from this test quantifies the strength of association between a predictor and the outcome, after controlling for the influence of other variables in the model. **Results:** The prevalence of current energy drink use was 175 (38.0%), with 22.9% of users classified as frequent consumers. Common consumption triggers were exam preparation (65.1%) and late-night studying (52.0%). Notably, 44.6% of users reported adverse effects, primarily insomnia (29.7%) and anxiety/jitters (18.2%). Knowledge about safe caffeine limits was low (28.5%). Significant predictors of frequent use included male gender (AOR=1.9; 95% CI: 1.1–3.3), residing in a university hostel (AOR=2.4; 95% CI: 1.4–4.2), and part-time employment (AOR=1.7; 95% CI: 1.0–2.9). **Conclusions:** The findings highlight an urgent need for targeted university-level health promotion interventions to educate students, particularly those in high-risk groups, about the potential dangers of energy drinks and to promote healthier alternatives for managing academic stress.

INTRODUCTION

The consumption of energy drinks has become increasingly popular among young adults, particularly university students, over the past two decades. Marketed as beverages that enhance alertness, concentration, and physical performance, these products are widely accessible and heavily promoted through targeted advertising campaigns. Their use is often linked to coping with academic stress, late-night study routines, and social pressures that characterize student life [1]. Globally, research has raised concerns about the health risks of frequent energy drink consumption. Studies have shown that the high caffeine and sugar content in these beverages is associated with adverse effects, including insomnia, elevated blood pressure, obesity, dental caries, and an increased risk of cardiovascular complications [2, 3]. Furthermore, mixing energy drinks with alcohol, a common practice among students, has been linked to impaired judgment and engagement in risky behaviors [4]. Despite

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these documented risks, awareness about safe consumption limits remains low among young populations [1]. In South Asian countries, including Pakistan, energy drink consumption among university students is an emerging trend, yet the body of scientific literature remains limited. Existing studies highlight that students often use these beverages to combat academic fatigue and enhance performance, but few investigations have explored the extent of this habit, its determinants, and its potential health implications in local contexts [5].

This study aims to assess the prevalence of energy drink consumption among university students in Pakistan, identify the sociodemographic and academic factors that influence their use, and examine students' knowledge and perceptions regarding the associated potential health risks.

METHODS

This cross-sectional study utilized a web-based survey conducted at the University of the Punjab, Lahore, from October 2023 to January 2024. A convenience sampling method was employed, with eligibility restricted to students aged 18 years or older enrolled in undergraduate or postgraduate programs. The minimum sample size was calculated to be 384, but a target of 500 was set to account for subgroup analyses. Participation was voluntary and confidential, and no personal identifiers were collected. The questionnaire, which was available in both English and Urdu after a rigorous translation process, was piloted with 30 students. The data from this pilot phase were used solely to refine the instrument and were not included in the final analysis. Frequent consumption was defined as an intake of four or more standard servings per week. This threshold was selected to identify the upper tier of consumers and was consistent with cut-offs used in prior public health research to classify high or habitual caffeine intake, which was associated with increased risk of adverse effects. It covered sociodemographic characteristics, consumption behaviors, knowledge, and self-reported health outcomes. Data collection involved measuring both the frequency and volume of energy drink consumption. "Frequent consumption" was defined as an intake of four or more standard servings per week, a threshold derived by multiplying the average volume per occasion by a habitual frequency cut-off. The minimum sample size was calculated using Cochran's formula for cross-sectional studies: $n = Z^2P(1-P)/d^2$. The assumptions were a 95% confidence level (Z = 1.96), an expected prevalence (P) of 50% (0.5) to maximize the sample size due to the lack of prior precise estimates in the local population, and a margin of error (d) of 5% (0.05). This calculation yielded a minimum sample size of 384. A target sample size of 500 was set to account for potential incomplete responses and to ensure sufficient power for subgroup analyses. Written informed consent was taken. The original English questionnaire underwent a rigorous process of forward and back translation to ensure conceptual equivalence in Urdu. First, two independent bilingual translators produced two Urdu versions (T1 and T2), which were synthesized into a single version. This version was then back-translated into English by two other translators, who were blinded to the original instrument. The research committee compared the back-translated versions with the original to identify and resolve any discrepancies in meaning. For cultural adaptation, a panel of experts (including a nutritionist, a public health specialist, and a linguist) assessed the content validity of the Urdu version, evaluating the relevance and clarity of each item. The Content Validity Index (CVI) was calculated, and items with a CVI below 0.78 were revised. Finally, the pre-final Urdu version was pilot-tested on 30 students to assess face validity, comprehensibility, and contextual appropriateness (e.g., ensuring local names for energy drinks were used). Minor adjustments to wording were made based on pilot feedback. The data from this pilot phase were used solely to refine the instrument and were not included in the final analysis. Data analysis was performed using IBM SPSS Statistics version 27. Descriptive statistics summarized the data, and bivariate analyses (Chi-square and t-tests) identified factors associated with frequent use. For the continuous variable (age), the use of parametric tests (t-test) was deemed appropriate given the large sample size (n=460) and the robustness of the t-test to minor deviations from normality, as supported by the Central Limit Theorem. Variables with a p-value<0.10 were included in a final multivariable logistic regression model to identify independent predictors, with a p-value<0.05 considered significant. Measures to mitigate bias included emphasizing anonymity and using validated survey items.

RESULTS

The study cohort comprised 460 university students with a mean age of 21.4 years, reflecting a typical undergraduate population. The sample was predominantly composed of undergraduates (84.8%) and was nearly evenly split by sex, with a slight majority of females (52.6%). A key characteristic of the sample is that nearly half of the participants (47.0%) resided in university hostels, a factor often associated with independent lifestyle choices and increased exposure to peer influences (Table 1).

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Table 1: Sociodemographic Characteristics of the Study Participants(n=460)

Characteristic	Category	Mean ± SD, n (%)
Age	Years	21.4 ± 2.1
Sex	Male	218 (47.4%)
	Female	242 (52.6%)
	–	460 (100.0%)
Academic Level	Undergraduate	390 (84.8%)
	Postgraduate	70 (15.2%)
	–	460 (100.0%)
Residential Status	Hostel	216 (47.0%)
	With Family	205 (44.6%)
	Other	39 (8.4%
	–	460 (100.0%
Part-Time Employment	Yes	98 (21.3%)
	No	362 (78.7%)
	-	460 (100.0%)

The bivariate analysis revealed that male students and those residing in university hostels had significantly higher odds of being frequent energy drink users, with crude odds ratios of 2.83 and 2.26, respectively. A positive, though not statistically significant, association was observed for part-time employment. No significant associations were found for age or academic level (Table 2).

Table 2: Bivariate Associations Between Sociodemographic Factors and Frequent Energy Drink Use (≥4 Servings/Week)

Variables	Category	Frequent Users (n=40)	Non-Frequent Users (n=420)	Crude Odds Ratio (COR)	95% CI	p-Value
Categorical, n (%)						
Sex	Male	28 (70.0%)	190 (45.2)	2.83	1.40 – 5.73	0.003
	Female	12 (30.0%)	230 (54.8%)			
Residential Status	Hostel	26 (65.0%)	190 (45.2%)	2.26	1.18 – 4.35	0.014
	With Family/Other	14 (35.0%)	230 (54.8%)			
Part-time Employment	Yes	12 (30.0%)	86 (20.5%)	1.67	0.83 – 3.36	0.152
	No	28 (70.0%)	334 (79.5%)			
Academic Level	Undergraduate	32 (80.0%)	358 (85.2%)	0.69	0.31 – 1.56	0.377
	Postgraduate	8 (20.0%)	62 (14.8%)			
Continuous (Mean ± SD)						
Age	Years	21.6 ± 2.2	21.4 ± 2.1	1.05*	–	–

The multivariable analysis, adjusting for all other factors, confirmed that male sex, hostel residence, and part-time employment were significant independent predictors of frequent energy drink consumption. Notably, students living in hostels had 2.4 times higher adjusted odds of being frequent consumers compared to those living with family or elsewhere. Age and academic level were not significant predictors in the adjusted model (Table 3).

Table 3: Multivariable Logistic Regression Analysis of Predictors of Frequent Energy Drink Use (≥4 Servings/Week)

Variables	Category/ Comparison	Adjusted Odds Ratio (AOR)	95% CI	p-Value
Sex	Male vs. Female	1.9	1.1 – 3.3	0.020
Residential Status	Hostel vs. Family/ Other	2.4	1.4 – 4.2	<0.001
Part-time Employment	Yes vs. No	1.7	1.0 – 2.9	0.040
Age	Per One-Year Increase	1.1	0.9 – 1.3	0.280
Academic Level	Postgraduate vs. Undergraduate	1.3	0.7 – 2.6	0.400

AOR: Adjusted Odds Ratio; CI: Confidence Interval.

DISCUSSION

This study contributes to the growing body of international literature confirming energy drink (ED) consumption as a significant public health concern among university students. The observed prevalence of 38.0% among Pakistani students is consistent with rates reported in recent studies from the UAE (35%), Italy (48.7%), and Saudi Arabia (up to 60%), underscoring the global nature of this trend [6-8]. This convergence suggests that common drivers, such as escalating academic pressure, the need for cognitive enhancement, and aggressive marketing targeting young adults, are influential across diverse cultural contexts [9, 10]. A central finding of this study is the robust association between ED consumption and self-reported adverse health effects, with insomnia being the most common (29.7%). This aligns perfectly with physiological evidence on high-dose caffeine's disruption of sleep architecture by antagonizing adenosine receptors [11]. Recent large-scale studies have further solidified this link; for instance, the Norwegian SHOT2022 study, which included over 50,000 students, found a strong, dose-dependent relationship between ED consumption and poor sleep quality, including longer sleep latency and reduced sleep duration [6]. Similarly, the reported anxiety and jitters (18.2%) are consistent with known sympathomimetic effects of caffeine and taurine, which can induce physiological arousal and heighten stress responses, as demonstrated in controlled laboratory studies [11, 12]. A 2023 systematic review further affirmed that these subjective reports are often correlated with measurable physiological changes, including transient elevations in

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blood pressure and heart rate variability, highlighting a tangible cardiovascular strain even in young, presumably healthy individuals [13]. A critical and concerning finding is the significant gap between consumption and knowledge. Only 28.5% of respondents were aware of safe caffeine intake guidelines. This "knowledge-practice" dissonance is a recurring theme in contemporary literature. A 2022 study among Malaysian medical students found that despite high awareness of general ED risks, detailed knowledge of caffeine content and safe limits was poor, and this knowledge was not a significant deterrent to consumption [14]. This pattern is echoed in studies from Jordan and the US, where perceived functional benefits like improved concentration and fatigue reduction often override factual knowledge, leading to continued use [15, 16]. The primary motivations identified in our sample, coping with academic stress and enhancing concentration for late-night studying, are echoed globally. This often creates a vicious cycle of dependency, where ED use leads to poor sleep, which in turn increases daytime fatigue and perpetuates the need for stimulants [9, 10]. The multivariable analysis identified key sociodemographic predictors of frequent use. The association with male gender (AOR=1.9) is well-documented and often attributed to higher risk-taking propensity and greater engagement in sensation-seeking behaviors among young men [17]. The strong independent predictor of hostel residence (AOR=2.4) is particularly insightful. It suggests that the environment itself—characterized by independence from parental supervision, irregular routines, peer influence, and heightened academic stress in a confined setting is a powerful driver of consumption. It is important to note, however, that some predictors, notably part-time employment (AOR=1.7; 95% CI: 1.0–2.9), had wide confidence intervals that crossed the null value of 1. This suggests a degree of statistical imprecision and indicates that the study may have had limited power to detect a more precise effect size for this variable, potentially due to the smaller subgroup of students engaged in employment. A 2023 study from the Philippines specifically highlighted that university dormitory residents had significantly higher odds of ED consumption compared to their family-dwelling peers, reinforcing the role of the residential environment in facilitating this health behavior [18]. The attenuation of the crude odds ratios for these variables in the multivariable model indicates shared variance; for example, the hostel environment may amplify consumption behaviors that are already more prevalent among males. This underscores that public health interventions should target high-risk settings like hostels, rather than demographics alone [19]. The findings collectively underscore an urgent need for evidence-based, multi-level interventions. University policies should consider restricting the sale and marketing of EDs on campus, particularly in and around student hostels. Furthermore, targeted educational campaigns are needed that move beyond generic warnings. These should incorporate behavioral science principles, explicitly deconstruct the "vicious cycle" of ED use and poor sleep, and promote evidence-based strategies for stress management and cognitive enhancement, such as mindfulness and time management workshops [20]. Finally, the cross-sectional design of this study precludes causal inference. Future longitudinal research is essential to establish the temporal sequence between ED consumption and health outcomes and to understand the long-term health trajectories of young, frequent consumers. Furthermore, the wide confidence intervals for some associations indicate limited statistical power for those specific predictors, suggesting the findings for variables like part-time employment should be interpreted with caution and require confirmation in larger studies

CONCLUSIONS

In conclusion, this study found a high prevalence of energy drink consumption (38.0%)among university students in Pakistan, with a significant proportion being frequent users. The consumption was primarily driven by academic pressures and was significantly associated with self-reported adverse health effects, notably insomnia and anxiety. A critical finding was the poor knowledge of safe caffeine limits among the students. Male gender, hostel residence, and part-time employment were identified as key predictors of frequent use. These findings highlight an urgent need for targeted educational interventions and health promotion strategies within university settings, particularly those reaching high-risk student groups, to mitigate the associated health risks.

Authors Contribution

Conceptualization: IF
Methodology: IF, 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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Association Between Plant-Based Dietary Intake and Anthropometric Measurements

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ABSTRACT

The benefits of plant-based diets in enhancing health and lowering the risk of chronic illnesses are becoming more widely acknowledged. **Objectives:** To establish a correlation between a plant-based dietary intake and anthropometric measurements among adults. **Methods:** This cross-sectional study was conducted on 200 adults (20–50 years old) from community centers in Lahore. The validated food frequency questionnaire was used to measure dietary intake of foods. Anthropometric Indicators were measured. Groups of participants were organized according to how much or how little they consume plant-based foods. SPSS version 26.0 was used to analyze the data, and independent t-tests were used for comparison. **Results:** Among 200 participants, 55% consumed legumes three or more times per week, 40% had three or more servings of whole grains daily, 70% consumed at least two servings of vegetables per day, and 65% had three or more servings of fruit daily. **Conclusions:** Individuals with higher intake of plant-based foods had significantly lower waist-to-hip ratio, body mass index, and lower waist circumference than those with lower intake.

INTRODUCTION

Plant diets have been noted to have the capacity to enhance health and prevent chronic illnesses [1, 2]. These diets contain large amounts of essential nutrients, fiber, and bioactive compounds, which help to achieve better nutritional status and decrease the risk of developing many health conditions [3, 4]. Diet is very important in preserving human health and in the prevention of chronic diseases. There is growing evidence that plant-based diets are a source of necessary nutrients, bioactive compounds, and antioxidants that promote metabolic health, weight control, and disease prevention [5, 6]. Studies have shown that dietary patterns that involve following vegan-based diets are linked to healthy results. To illustrate, it has been found that people who adhere to plant-based diets are likely to be less fat, experience lowered blood pressure, and have better lipid profiles [7-9]. Moreover, Plant-based diets have been linked to decreased levels of inflammation and oxidative stress, which has been attributed to the development of chronic diseases [10, 11]. Although these advantages exist, nutritional trends in most communities have been moving to high consumption of processed and animal-based food products, and many studies are needed

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regarding the impact of plant-based diets [12, 13]. To help guide dietary recommendations for the local population, this study examines the relationship between plant-based food intake and health indicators. There is currently little data on how plant-based diets affect nutritional and anthropometric indicators in Pakistani adults.

This study aims to establish a correlation between a plant-based dietary intake and anthropometric measurements among adults.

METHODS

This analytical cross-sectional study was conducted on 200 adults aged 20–50 years to analyze the association between plant-based dietary intake and anthropometric measurements. The study was conducted for 5 months from May 2024 to September 2024. Participants were recruited using a convenience sampling method from multiple community centers located in urban areas of Lahore, Pakistan. This non-probability sampling approach was chosen for practicality, though it may limit the generalizability of the findings. A sample size of 200 was used for this study. A post-hoc power analysis was conducted using G*Power software, which indicated that this sample size provided over 80% power to detect a medium effect size (d=0.4) in the independent t-test comparisons of anthropometric measures between the high and low plant-based diet groups, assuming a two-tailed alpha of 0.05. Participants were recruited from multiple community centers located in urban areas of Lahore, Pakistan. Participants were categorized into 'high' or 'low' plant-based diet intake groups based on a composite score derived from the food frequency questionnaire. The score was calculated from the sum of weekly servings of fruits, vegetables, whole grains, and legumes. A median split was used, with participants scoring above the median classified as 'high intake' and those at or below the median as 'low intake'. Participants included healthy individuals consuming either a mixed or plant-based diet who were willing to provide dietary information, while those with chronic diseases, pregnancy, or special dietary restrictions were excluded. Informed written consent was obtained from all participants. Dietary intake was assessed using a validated food frequency questionnaire with a 0–7 consumption scale [14]. The questionnaire was adapted for the local population by including region-specific foods and portion sizes. Its reliability was confirmed in a pilot study (n=30) with a two-week test-retest, showing good reproducibility (Cronbach's alpha = 0.78). Anthropometric measurements included BMI, waist circumference, hip circumference, and waist-to-hip ratio, measured using standard procedures [15]. Data were analyzed in SPSS version 26.0 using an independent t-test and descriptive statistics. For the independent t-tests used for inter-group comparisons, the assumption of homogeneity of variances was verified using Levene's Test. As the data met the assumptions of normality and homogeneity of variances, parametric tests were appropriately applied.

RESULTS

A total of 200 participants were recruited, with a mean age of 34.2 ± 8.5 years. The sample had a balanced gender distribution (51% male, 49% female). Dietary assessment showed that 40% of participants consumed three or more daily servings of whole grains, 55% consumed legumes at least three times per week, 70% consumed at least two daily servings of vegetables, and 65% reported three or more fruit servings daily (Table 1).

Table 1: Participant Characteristics and Dietary Intake by Gender

Variables	Total (n=200)	Male (n=102)	Female (n=98)
Age (Years)	34.2 ± 8.5	35.1 ± 8.2	33.3 ± 8.7
Fruit Intake ≥3 Servings/Day	130 (65%)	60 (59%)	70 (71%)
Vegetable Intake ≥2 Servings/Day	140 (70%)	68 (67%)	72 (73%)
Legume Intake ≥3 Times/Week	110 (55%)	56 (55%)	54 (55%)
Whole Grain Intake ≥3 Servings/Day	80 (40%)	38 (37%)	42 (43%)

Participants with higher plant-based diet intake exhibited significantly better anthropometric outcomes. Specifically, their mean BMI and waist-to-hip ratio were lower compared to those with lower plant-based intake (Table 2).

Table 2: Anthropometric Outcomes by Plant-Based Diet Score and Gender

Plant-Based Diet Score	Gender	BMI (kg/m²)	Waist Circumference (cm)	Waist-to-Hip Ratio
High Intake	Male	23.5 ± 2.4	80.2 ± 7.0	0.86 ± 0.05
	Female	22.7 ± 2.6	76.9 ± 7.3	0.84 ± 0.04
Low Intake	Male	26.8 ± 3.3	90.1 ± 8.4	0.93 ± 0.06
	Female	26.0 ± 3.1	86.4 ± 7.9	0.91 ± 0.05
p-Value	–	<0.01	<0.01	–

Although both groups had anthropometric measurements within WHO-recommended ranges, participants with higher plant-based diet scores exhibited significantly lower BMI, waist circumference, and waist-to-hip ratio compared to those with lower scores ($p<0.05$), indicating a trend toward improved body composition among plant-based diet consumers.

DISCUSSION

Plant-based diets are linked to better fat distribution, a lower risk of obesity, and an improved nutritional status [16, 17]. According to the current study, eating more plant-based meals is associated with having a healthier body composition. Although the anthropometric indicators of all participants were within the WHO-recommended normal

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ranges, individuals with higher plant-based diet scores exhibited significantly lower BMI, waist circumference, and waist-to-hip ratio compared to those with lower scores. These findings indicate an association between plant-based dietary intake and more favorable fat distribution, rather than a direct causal relationship. Our results align with the findings of Rahbar et al. who reported that individuals adhering to plant-based diets exhibited lower BMI and waist-to-hip ratios compared with those consuming predominantly animal-based foods [18]. Similarly, Ferguson et al. observed that female participants following plant-based dietary patterns demonstrated significantly reduced BMI and waist circumference compared to their counterparts [19]. These studies support the idea that high plant food intake contributes to reduced overall adiposity and less fat accumulation around the abdomen, which is especially important given the role of central obesity as a risk factor for cardiometabolic diseases. A strong association between treatment allocation and positive outcome, regardless of the technique used. This is evidenced by the consistent, strong within-group improvements (all $p<0.01$) and the absence of a statistically significant or clinically meaningful association between the specific technique (holding vs. pistoning) and the degree of improvement (all between-group $p > 0.05$, with small effect sizes). Mechanistically, plant-based diets are typically high in dietary fiber, lower in energy density, and often lead to greater satiety and lower overall caloric intake, which could help explain the anthropometric differences observed. The correlation with waist measures indicates effects not just on overall weight, but primarily on central fat deposition. Given our cross-sectional design, causality cannot be assumed; however, when taken together with longitudinal cohort studies (e.g., fruit intake being inversely associated with weight gain and waist circumference over time), the evidence suggests that promoting higher intake of fruits, vegetables, legumes, and whole grains might be a fruitful strategy to prevent obesity and central adiposity in our population [20]. While associations between plant-based diets and positive health metrics highlight their potential benefits, cross-sectional studies cannot prove cause and effect. The results suggest that promoting greater intake of plant-based foods could be an affordable and practical approach to improve body composition and reduce chronic disease risk. These findings can inform dietary guidelines, community health programs, and policy initiatives encouraging plant-based nutrition.

CONCLUSIONS

Dietary factors derived from plant-based foods significantly influence nutritional status and health outcomes. Promoting regular inclusion of fruits, vegetables, legumes, and whole grains in the daily diet offers an affordable way to strengthen community health and lower the likelihood of chronic disorders. Future longitudinal studies are recommended to establish causality and explore specific plant-based components.

Authors Contribution

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

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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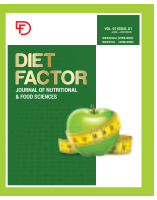

Original Article

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A Comprehensive Survey Report on Caffeine Effect on Academic Performance

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ABSTRACT

Caffeine is consumed by students and is very common in different forms worldwide. While it has been researched to increase alertness and focus, it also imparts negative effects, such as disturbing a person's sleep cycle. **Objective:** To assess how much people are aware of drinks containing caffeine and also their consumption of it. **Methods:** This was based on a report of 100 university-going students aged between 17 to 28 years who are the potential subjects to assess how the consumption of caffeine affects academic performance. The data were collected from The University of Lahore students. **Results:** Caffeine consumption was widespread among university students (71%), mainly through tea. While most saw no impact on academics, 31.6% noted improvements, and 6.5% reported declines. Nearly half experienced sleep issues, and over a third reported side effects like anxiety. Despite high awareness, many noted peer over-reliance, with 40.7% open to healthier alternatives. **Conclusion:** caffeine use among students reflects a balance between perceived cognitive benefits and potential health risks. Its mixed effects suggest the need for greater awareness and informed decision-making. Social influences and habitual use further highlight the importance of education on healthier alternatives.

INTRODUCTION

Caffeine consumption among university students is a widespread phenomenon, primarily driven by the belief that it enhances attention and productivity [1]. However, this practice is coupled with potential negative health outcomes, creating a complex public health consideration [2, 3]. The consumption of caffeinated beverages has grown dramatically among young adults, who often justify their use as a means to increase mental vigilance, improve productivity, and enhance brain function, all of which can subsequently impact academic outcomes [4]. This central nervous system stimulant is found in a diverse range of products, including coffee, tea, soft drinks, and over-the-counter medications [5]. While moderate intake is common, consumption between 100-400 mg has been associated with adverse effects such as anxiety, tremors, and agitation [6]. Despite this, students frequently report using caffeine to cope with demanding academic situations, claiming it boosts alertness, attention span, and long-term memory [7]. The popularity of caffeine stems from its ability to combat fatigue, though this benefit often comes at the cost of sleep quality. Research indicates that caffeine consumption can reduce total sleep time by approximately 45 minutes and decrease sleep efficiency by 7%, necessitating careful timing of intake relative to bedtime [8]. Behavioral studies suggest that moderate caffeine consumption can reduce fatigue and improve

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vigilance, particularly in monotonous situations or during tasks requiring sustained attention [9]. However, the effects on complex cognitive tasks are less predictable and can be influenced by individual factors such as personality and time of day [10]. The impact is evident in student populations, with one study noting that caffeine consumption led to reduced sleep duration, negatively affecting concentration, even as it helped students feel more awake and alert [11]. Furthermore, increased caffeine use has been associated with heightened depressive and anxiety symptoms among university students, highlighting the need for further investigation into this relationship and the promotion of healthier alternatives [12]. The health implications of caffeine are dose-dependent. While it possesses antioxidant and anti-inflammatory properties that may offer protection against various diseases, long-term or excessive use is linked to adverse effects, including insomnia, migraines, and specific risks for vulnerable groups like pregnant women and adolescents [13]. The environmental impact of caffeine consumption is also noteworthy, with significant levels detected in water sources globally, posing potential risks to aquatic ecosystems [14]. Among university students, those in demanding fields like medicine are particularly prone to high caffeine consumption. They face unique pressures from rigorous academic workloads and clinical training, often leading to disrupted sleep patterns and elevated stress levels [15]. This makes caffeine an attractive, readily available aid for maintaining energy and alertness. The habitual use of caffeine in this demographic is especially concerning, as these future healthcare professionals' personal health behaviors may later influence the advice and care they provide to patients [16].

This study aims to comprehensively explore the consumption patterns, perceived benefits, and associated risks of caffeine use among university students in relation to their academic performance.

METHODS

This cross-sectional study was conducted for three months from July to September 2024 to assess caffeine consumption patterns and their perceived effects among university students. Data were collected via a self-structured and pre-tested questionnaire, and ethical approval was obtained from the University Institute of Diet and Nutritional Sciences, The University of Lahore. Sample size calculation was done using Rao's online software. A total of 106 students were recruited using a non-probability convenience sampling method from multiple universities, including the University of Lahore (42%), University of Central Punjab (7%), University of Management and Technology (6%), Punjab University (4%), and Beaconhouse National University (1%). Inclusion criteria comprised university students aged 17–32 years who had a habit of caffeine consumption, and who voluntarily consented to participate, while individuals having caffeine allergy, from outside this age range, non-university students, and uncooperative respondents were excluded. The questionnaire consisted of four sections: demographic information, caffeine-related sleep disturbances, beliefs about caffeine-induced dehydration, and the perceived impact of caffeine on academic performance. All participants provided informed consent before completing the survey through online or offline modes. Data were entered and analyzed in SPSS version 24.0.

RESULTS

The survey reveals high caffeine prevalence (71%) among students, primarily consumed as tea. Despite most respondents (61.7%) perceiving no significant academic impact, a notable proportion reported caffeine-induced sleep disturbances (53.3%) and side effects like burnout (25.9%). This indicates a complex relationship where perceived short-term benefits for alertness coexist with significant health trade-offs, underscoring a need for targeted education on responsible consumption (Table 1).

Table 1: Caffeine Consumption and Perceived Effects Among University Students (n=106-108)

Variable	Category	Findings (%)
Age	17-20 Years	24.75%
	21-24 Years	70.4%
	25-28 Years	2.85%
	28+ Years	2%
Marital Status	Married	5.6%
	Unmarried	94.4%
Department	Medical and Allied Health	74.1%
	Other	25.9%
Residence	Day Scholar	81.5%
	Hostelite	18.5%

The caffeine consumption patterns of the participants are summarized, along with their preferred form, timing, and variations in use during stressful academic times. The majority of respondents (71%) said they drank caffeine, with tea accounting for the majority (69.2%), followed by coffee (21.2%) and other drinks including soda (9.6%). While a lower percentage reported drinking caffeine in the afternoon (13.9%), late at night (16.7%), or evening (28.7%), the majority (40.7%) preferred to use it in the morning. 35.2% of respondents said they used it more frequently during tests or deadlines, 16.7% said they used it occasionally, and 48.1% said they did not. Out of those studying at night, 24.1% always drank coffee, 29.6% occasionally, 12% infrequently, and 34.3% never. The

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majority (61.7%) reported no change, despite 31.6% of respondents believing caffeine enhances academic performance. Although 47.2% were ambivalent, about 35.8% agreed or strongly agreed that caffeine has academic benefits. While 45.4% were doubtful, one-fifth (20.4%) said it gives a competitive edge. Regarding classroom alertness, 28.3% of respondents said they always felt more aware after consuming caffeine, while 39.6% said they occasionally felt more alert. Furthermore, 40.7% of those surveyed said they had advised their peers to take caffeine. After consuming coffee in the evening, 46.7% of respondents reported having trouble falling asleep, and more than half (53.3%) reported having disturbed sleep. Caffeine has an impact on sleep patterns, according to half of the respondents (50%) who responded. The majority of individuals (52.8%) reported sleeping 6–8 hours on average, followed by 4–6 hours (27.3%), more than 8 hours (13.2%), and less than 4 hours (5.4%). Furthermore, after consuming large amounts of caffeine, 25.9% felt burnout or weariness, 25% occasionally reported it, and 49.1% reported no such effects (Table 2).

Table 2: Caffeine Consumption Habits among Study Participants

Variable	Category	Findings (%)
Consumption Habits	Caffeine Consumers	71%
Preferred Form:	Tea	69.2%
	Coffee	21.2%
	Other (Soda, etc.)	9.6%
Time of Day	Morning	40.7%
	Evening	28.7%
	Late Night	16.7%
	Afternoon	13.9%
Increased Use During Exams/Deadlines	Yes	35.2%
	Sometimes	16.7%
	No	48.1%
Use During Night Study Sessions	Always	24.1%
	Sometimes	29.6%
	Rarely	12%
	Never	34.3%
Perceived Effects on Academic Performance	Improves	31.6%
	No Change	61.7%
	Decreases	6.5%
Perceived Academic Benefits	Agree/Strongly Agree	35.8%
	Neutral	47.2%
	Disagree/Strongly Disagree	16.3%
Believe it Provides a Competitive Edge	Yes	20.4%
	No	34.3%
	Not Sure	45.4%
Alertness in Class	Always	28.3%
	Sometimes	39.6%
	Rarely	17%
	Never	15.1%
Have Recommended Caffeine to Peers	Yes	40.7%
	No	59.3%
Sleep and Health Impacts	Experience Disturbed Sleep	53.3%
	Difficulty Falling Asleep After Evening Intake	46.7%
	Agree Caffeine Intake Affects Sleep Patterns	50%
Average Sleep	<4 Hours	5%
	4-6 Hours	27.4%
	6-8 Hours	52.8%
	>8 Hours	13.2%
Experience Burnout/ Exhaustion After High Intake	Yes	25.9%
	Sometimes	25%
	No	49.1%
Awareness & Attitudes	Believe Caffeine Causes Dehydration	65.4%
Feel Guilt/Concern About Caffeine Use	Yes	19.4%
	No	80.6%
Actively Limit Caffeine Intake:	Yes	48.3%
	No	51.7%
Willing to Consider Healthier Alternatives	Yes	40.7%
	No	59.3%
Interested in Attending Seminar on Caffeine	Yes	66.7%
	No	33.3%

DISCUSSION

The findings of this study illuminate the complex and dualistic nature of caffeine consumption among university students in Pakistan. The high prevalence rate of 71% aligns closely with global trends reported in recent literature, reinforcing caffeine's status as a ubiquitous psychoactive substance in academic environments [17]. The overwhelming preference for tea (69.2%) as the primary vehicle for caffeine intake highlights a significant cultural and regional consumption pattern, distinguishing this cohort from Western populations where coffee is typically dominant [18]. A central finding of this research is the discrepancy between perceived academic benefit and measurable outcome. While a combined 35.8% of students agreed that caffeine offered academic benefits, the largest group (61.7%) reported no significant change in their actual academic performance. This perception-reality gap was supported by Yasmeen et al. which concluded that while caffeine reliably enhances subjective feelings of alertness and concentration, its effect on complex cognitive tasks and long-term academic grades is often negligible or inconsistent [19]. The small but notable percentage of students (6.5%) who reported a decline in performance underscores the risk of adverse effects like anxiety and burnout, which can ultimately undermine academic success. The significant sleep disturbances reported by our participants (53.3% experiencing disturbed sleep, 46.7% having difficulty falling asleep after evening intake) present a critical public health concern.

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These results are consistent with the mechanistic understanding that caffeine, as an adenosine receptor antagonist, directly disrupts sleep architecture [4]. A recent longitudinal study by Alqawasmi et al. specifically among medical students, found that nocturnal caffeine use was a stronger predictor of poor sleep quality than academic stress alone, creating a vicious cycle where students use more caffeine to combat fatigue induced by caffeine-related sleep loss [6]. This is further evidenced by our data showing that 24.1% of students always consume caffeine during all-night study sessions. Furthermore, an interesting awareness gap was observed. A majority of students (65.4%) correctly identified caffeine's dehydrating effect, demonstrating general health knowledge. However, this awareness does not appear to translate into proactive behavioral change, as 41.7% do not actively limit their intake, and a substantial portion continue to consume it in the late evening. This suggests that knowledge alone is insufficient to modify habits driven by academic pressure and peer influence, the latter being evident from the 40.7% who have recommended caffeine to peers. This social reinforcement of caffeine use has been identified as a key factor in normalizing its consumption among student populations [20]. The strong interest (66.7%) in attending a seminar on caffeine and academic health indicates a clear opportunity for university health services and student affairs departments to intervene. Educational campaigns should move beyond simply listing caffeine's effects and instead focus on practical strategies for responsible use. This includes promoting caffeine curfews, highlighting healthier alternatives for energy management (e.g., brief naps, physical activity), and integrating time-management and stress-reduction workshops into student orientation programs. Future research should employ longitudinal designs with objective measures of caffeine intake and academic records to better understand the causal pathways.

CONCLUSIONS

In conclusion, caffeine use among university students is a multifaceted issue characterized by a trade-off between perceived short-term cognitive benefits and tangible negative impacts on sleep and well-being. While students turn to caffeine as a tool to meet academic demands, its unregulated use can paradoxically hinder performance through sleep disruption and side effects. A proactive, educational approach that empowers students with strategies for sustainable energy management is crucial to fostering a healthier academic environment.

Authors Contribution

Conceptualization: MA, AD
Methodology: AD, LK
Formal analysis: MNL
Writing review and editing: NZ, MAR, AK

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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