Cureus

Review began 10/07/2023 Review ended 10/20/2023 Published 10/26/2023

Gupta et al. This is an open access article distributed under the terms of the Creative Commons Attribution License CC-BY 4.0., which permits unrestricted use, distribution

and reproduction in any medium, provided the original author and source are credited

© Copyright 2023

Millets: A Nutritional Powerhouse With Anticancer Potential

Mansha Gupta<sup>1</sup>, Dina Medhanie Asfaha<sup>2</sup>, Govintharaj Ponnaiah<sup>3</sup>

 Medicine, Kasturba Medical College, Manipal, Manipal, IND
 Medicine, Orotta School of Medicine, Orotta Hospital, Asmara, ERI 3. Molecular Biology/Plant Breeding and Genetics, International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Hyderabad, IND

Corresponding author: Mansha Gupta, mansha.g16@gmail.com

### Abstract

Millets are important food crops widely grown by smallholder farmers in the arid and semi-arid regions of the world. Millets are rich in protein, dietary fiber, micronutrients, and have a low glycemic index (GI) and desirable bioactive compounds. Due to their higher nutritional content, millets are popularly known as "nutricereals". Coinciding with the United Nations and the Food and Agriculture Organization's declaration of 2023 as the "International Year of Millets," this review underscores the nutritional value of these grains from the Poaceae family.

The consumption of nutricereals is associated with several health benefits including lowering of blood sugar levels (diabetes), controlling blood pressure, and providing protection against thyroid, cardiovascular, and cancer diseases. A review of the literature from PubMed and Google Scholar was done focusing on the health benefits and anti-cancer properties of different millets. Millets have a rich content of macronutrients like carbohydrates and proteins, as well as micronutrients and bioactive compounds, including dietary fibers, essential fatty acids, and phytochemicals. This article explores millets' nutritional elements, i.e., macronutrients, micronutrients, and bioactive compounds, and provides insights into the types of carbohydrates present, the prebiotic function of dietary fibers, and millets' low GI. The study identified the mechanisms by which millets may deter cancer growth, focusing on the roles of dietary fibers, plant protease inhibitors, and bioactive peptides. Additionally, it compared the mineral and vitamin content of millets to other common grains, such as rice and wheat, and explored the potential health advantages of millets over other cereal crops. This review systematically investigated the health advantages of millets, particularly, their anti-cancer capabilities. Dietary fibers, plant protease inhibitors, and bioactive peptides present in millets have the capacity to induce apoptosis, inhibit cell proliferation, and interact with gut microbiota leading to potential anti-cancer effects. This review also identified existing challenges in the bioavailability and effective delivery of millets' bioactive peptides, advocating for further research to maximize their health benefits.

Categories: Nutrition, Internal Medicine, Oncology Keywords: micronutrients, bioactive compounds, anti-cancer properties, nutrition, millets

# Introduction And Background

Food and Agriculture Organization and the United Nations have recognized 2023 as the "International Year of Millets" (IYM 2023) to spread awareness about the health and nutritional benefits of millets. Millets are a group of small-seeded nutrient-dense food crops that are widely grown on marginal lands in dry areas around the world. This group of crops belongs to the family Poaceae and consists of several species including pearl millet (Pennisetum glaucum), foxtail millet (Setaria italica), proso millet (Panicum miliaceum), finger millet (Eleusine coracana), kodo millet (Paspalum setaceum), little millet (Panicum sumatrense), and barnyard millet (Echinochloa utilis) [1]. Millets are designated as "nutricereals" as they are rich in macro- and micronutrients, carbohydrates, proteins, dietary fibers, important fatty acids, and a vast number of minerals and vitamins. Millet carbohydrates have less starch compared to the other staple cereal crops. Millet dietary fibers act as prebiotics that help in the development of healthy gut microbiota [2]. The dietary fiber content of millets also slows down the glucose absorption in the small intestine and can help in blood glucose regulation and reducing the glycemic index (GI) of food [3]. Millet proteins are rich in all essential amino acids and are particularly high in cysteine and methionine (sulfur-containing amino acids). Millets are gluten-free cereal grains, and hence can be consumed regularly by celiac patients [4]. The presence of higher levels of phytochemicals such as phenolics, tannins, carotenoids, and flavonoids exhibit strong antioxidant activity and reduce tumor growth [5]. Millets are also well recognized for their potential health benefits. including fighting against many diseases like diabetes, cardiovascular disease, high blood pressure, thyroid disorders, and celiac disease [6]. This review highlights the anti-cancer properties of millets to sensitize scientists to work in this direction.

### **Review**

#### Millets: the powerhouse of nutrients

Millets are nutritionally comparable to or superior to major cereal grains. They possess many additional benefits such as high dietary fiber content, gluten free, lower glycemic index, and the availability of abundant bioactive compounds, making them "ideal foods" for human health [7]. The proximate composition and calorific values, dietary fibers, essential amino acids, and micronutrients of millets are provided in Table 1. Millet carbohydrates can be categorized into two: (i) non-structural (sugars, starch, and fructosans) carbohydrates and (ii) structural (cellulose, hemicelluloses, and pectin substances) carbohydrates [8]. The carbohydrate content of millets varies from 56.88 to 72.97 g/100 g. The highest content of carbohydrates has been reported in finger millet, whereas the lowest carbohydrate content is reported in barnyard millet [9]. Millets' dietary and crude fiber content is much higher than that of the other staple cereals like rice, wheat, and maize [4]. The soluble dietary fiber offers many health benefits. preventing constipation, enhancing gut health, and reducing heart disease. Also, millets' dietary fiber reduces the GI of the food. The GI of millets varies from 33 to 65, which is significantly lower than the GI of rice and corn. Furthermore, dietary fiber directly interacts with gut microbiota and releases short-chain fatty acids (SCFAs) through fermentation, which can help prevent pathogen invasion and colonization by reducing the gut pH [10]. Such dietary fibers are known as "prebiotics". These suppress the growth of pathogens and remove carcinogens from the body and enhance nutrient absorption and immunity [11].

# Cureus

Crop Name	Proximate and calorific values				Dietary fiber contents				Micronutrient composition							
	Carbohydrates (%)	Protein (%)	Fat (%)	Calorific value (calories/100g)	Total dietary fiber (%)	Insoluble dietary fiber (%)	Soluble fiber (%)	Glycemic index <sup>†</sup> (numbers)	Calcium (mg/100 g)	Phosphorus (mg/100 g)	Zinc (mg/100 g)	Iron (mg/100 g)	Thiamine- B1 (mg/100 g)	Riboflavin- B2 (mg/100 g)	Niacin- B3 (mg/100 g)	Folic acid (µg/1 g)
Pearl millet	67-70.4	10.6- 12.6	4.8- 5.0	363-412	8.0- 13.5	5.0-8.0	2.0-3.0	55.0	27.0- 43.0	339	3.1	6.0-17.0	0.3-0.4	0.09-4.0	1.0-3.0	45.5
Finger millet	66.5-75.0	6.0-8.2	1.3- 1.5	332-376	15.0- 20.0	3.6-19.7	2.5-3.0	55.0-65.0	335.0- 352.0	250	32.0- 41.0	4.0-6.0	0.3-0.5	0.02-0.19	0.8-2.0	18.3
Proso millet	62.3-72.2	10.6- 12.5	1.8- 4.0	341-364	8.5- 14.2	7.0-12.0	1.5-2.0	50.2-64.7	20.0- 42.0	300	60.6	3.0-5.0	0.6	0.05-0.16	0.5-3.2	15.0
Foxtail millet	61.6-72.4	9.9- 12.3	2.5- 4.0	351-353	19.1	7.0-17.8	1.3	33.0	7.0-13.5	200-206	2.4-3.7	2.2-4.8	0.4	0.28	4.5	
Barnyard millet	49.0-65.5	8.9- 11.9	2.2- 4.5	300-310	13.6	4.0-14.7	4.2	41.7-50.0	12.0- 25.0	280	55.0- 60.0	15.0- 19.0	0.3	0.1	4.2	
Kodo millet	56.1-74.0	8.3- 11.6	1.3- 4.2	346-353	6.8- 37.8	4.0-6.0	2.0-3.0	49.5	27.0- 37.0	300	30.0- 35.0	1.8-4.0	0.2	0.09	0.1-1.2	23.1
Little millet	64.2-67.0	7.6- 10.0	2.4- 2.8	325-336	6.4- 12.2	5.0-7.0	2.0-3.0	41.5-61.8	15.0- 17.0	220	32.2- 35.1	1.2-1.7	0.3	0.05-0.09	1.3-3.2	9.0
Rice (brown rice)	72.8-75.8	8.5- 9.90	1.1- 1.3	325-340	0.7- 6.0	0.6-4.3	0.7-1.0	65.0-81.0								
Wheat	61.2-66.8	10.0- 12.0	1.4- 15	310-334	2.8- 12.1	2.1	1.0-2.0	44.0-60.0								
Maize	63.2-66.4	8.0-9.5	3.6- 4.1	328-338	3.9- 13.4	2.3-12.4	0.8-1.2	78.5-86.3								

# TABLE 1: Millets' nutrient composition

<sup>†</sup>Glycemic Index Scale: low, 55 or less; mid, 56-69; high, 70+ (source: Glycemic Index Foundation, https://www.gisymbol.com/)

Source: [4,8]

Millets are reservoirs of many minerals and vitamins. Among millets, finger millet is rich in calcium (348 mg/100 g) [4], which is about eight times higher than wheat. Furthermore, Singh and Raghuvanshi reported that the calcium content estimated on the 36 finger millet genotypes varied from 162 to 487 mg/100 g] [12]. Finger millet consumption of 100 g can provide roughly half of the recommended dietary allowance (RDA) for calcium content [15]. The phosphorus content (200-339 mg/100 g) varies widely among millets. According to the recommendation of the Indian Council of Medical Research (ICMR), consumption of millets can supply about 50% of the phosphorus requirement [14]. The zinc content of finger millet, kodo millet, and little millet is found higher than the RDA of 12 mg. The consumption of 100 g of barnyard millet can meet 100% of the RDA for iron content.

Millets also have high amounts of vitamins, especially vitamin B complex and vitamin E. Among millets, proso millet has a higher amount of thiamine, riboflavin, niacin, and total carotenoids whereas highest tocopherol content is found in finger millet. The riboflavin content of millets is several times higher compared to other staple cereals. Pearl millet contains a considerable amount of vitamin E (2 mg/100 g; fat-soluble component) and vitamin A [15]. Vitamins (water-soluble vitamins) play an important role in energy production, while carotenoids and vitamin E serve as antioxidants that support immunity, prevent eye defects, and enhance anti-cancerous properties of the body [16].

Phenolic compounds such as phenolic acids, flavonoids, and tannins are commonly present in millets and play a significant role in the immune system [17]. Millets have more phenols than staple cereals. The presence of ferulic and p-coumaric acids in whole pearl millet grains has the capacity to reduce tumor cells [18]. Among millets, finger millet has the highest phenolic compound content. The major phenolic compounds present in proso millet, finger millet, and foxtail millet are hydroxycinnamic acids. The phenolic compounds extracted from finger millet seed coat slow down glucose absorption, help in ameliorating postprandial hyperglycemia, and partially inhibit pancreatic amylase and alpha-glucosidase activity in carbohydrate metabolism [19]. Flavonoids are more common in free form. Brown-colored finger millet genotypes/cultivars have the highest concentrations of flavonoids and tannins [12]. Millets contain a variety of flavonoids among which luteolin, quercetin, catechin, naringenin, dizein, apigenin, and kaempferol are the most prevalent [18,19]. Regular consumption of millets containing high amounts of flavonoids and tannins lowers the risk of developing cancer, diabetes, cardiovascular disease, and neurological illnesses.

# Millets' cancer-fighting compounds and their anti-cancer property

Millets are a source of several types of compounds that have anti-cancerous properties; they are discussed next.

Dietary Fibers

Dietary fiber is an important component present in plant cell walls. Dietary fibers are cell wall polysaccharides and play a significant role in the human gut, helping in digestion and improving gut health. Millet dietary fibers are classified into two, namely, (i) soluble dietary fiber (SDF) and (ii) crude fiber or insoluble dietary fiber (IDF) [20]. SDF is soluble in water and composed of pectins, glucans and some hemicellulose whereas IDF is insoluble in water and contains cellulose, hemicellulose, and lignin. In comparison with IDF, SDF has a higher nutritional content, lowers cholesterol (fatty substances trapped in

the GI tract) level, absorbs more water, forms a gel-like structure, ferments the gut bacteria in the large intestine, regulates the immune system, and has anti-tumor properties [21]. The formation of resistant starch (RS) contributes the dietary fiber content that ultimately provides significant health benefits [22]. RS is a functional fiber fraction that plays a significant role as it escapes the enzymatic digestion in the intestinal tract leading to several health benefits [23]. It has been shown that the production of butyrate metabolites from gut microbial fermentation helps stabilize colorectal cell proliferation [24].

Dietary fibers gained more attention as potential therapeutic agents for manipulating gut microbiota and alleviating GI tract inflammation. For example, a recent study found that foxtail millet SDF inhibited the colony formation ability of HCT116 and HT-29 cells and could significantly induce the increase of reactive oxygen species (ROS) and apoptosis of HCT116 and HT-29 cells [25]. In another study, Zhang et al. compared the effects of dietary supplementation of foxtail millet and rice on colorectal cancer; it was found that foxtail millet inhibited the phosphorylation of STAT3 and the related signaling pathway proteins involved in cell proliferation, survival and angiogenesis (mediated by the activation of gut receptors, i.e., aryl hydrocarbon receptor, or AHR, and G-protein-coupled receptors, or GPCRs). Furthermore, millet treatment increased the abundance of *Bifidobacterium* and *Bacteriodales\_S24-7*, when compared to the rice-treated mice [26].

#### Plant Protease Inhibitors

Plant protease inhibitors are multifunctional proteins. Proteases are involved in a variety of biological processes, such as inflammation, infection, extracellular matrix breakdown, blood coagulation, programmed cell death, tumor invasion and metastasis [27,28]. Protease inhibitors are classified into six groups: cysteine, serine, threonine, glutamic acid, aspartate proteases, as well as matrix metalloproteinases, based on the type of amino acid present in the active site of the protease and the mechanism of peptide bond cleavage. Numerous studies have been conducted on protease inhibitors; the plant protease inhibitors are classified into families such as Bowman-Birk, Kunitz, Potato I, Potato II, Serpine, Cereal, Rapeseed, Mustard, and Squash [29,50]. These families are distinct from one another in terms of their mass, cysteine concentration, and the number of reactive sites [31].

Among millets, a protease inhibitor is isolated and well characterized in *ragi* (finger millet, *Eleusine coracana*), which is popularly known as "ragi bifunctional inhibitor (RBI)". RBI is a 14 kDa bifunctional inhibitor purified from ragi seeds [32], is a member of cereal trypsin/ $\alpha$  amylase inhibitor family that inhibits  $\alpha$ -amylase and trypsin forming a ternary complex simultaneously [35]. It consists of single polypeptide chain containing 122 amino acids including five intramolecular disulfide bonds [34]. A study was conducted by Sen and Dutta to evaluate the anti-carcinogenic activity of RBI in K562 human chronic myeloid leukemia cells [32]. The purified RBI from finger millet seeds suppressed the proliferation and induced apoptosis of K562. The cytotoxicity test showed that the RBI has antiproliferative potential against K562 chronic myeloid leukemia cells.

#### **Bioactive** Peptides

In the recent decades, an increasing number of lifestyle-related non-communicable diseases have been found to be heavily associated with incorrect eating habits and sedentary lifestyle; therefore, there is a huge surge in demands of foods with additional health benefits. Foods that provide additional health benefits along with basic nutrition (vitamins, minerals, fiber, protein, or peptides) are known as functional foods [35]. Among these nutritional components, peptides have received significant attention due to extraordinary biological functions and health-related benefits to combat lifestyle-related degenerative diseases. Peptides are a small string of proteins consisting of 2-20 amino acid subunits (encrypted in the parent protein) linked by peptide bonds. Peptides are characterized by low molecular weight (MW, <3 kDa) compounds and become more active when released during enzymatic proteolysis of proteins and during food processing such as cooking, fermentation, and ripening [36,37]. Bioactive peptides are obtained from protein-rich plants and animals. Recently, protein-rich millets have been identified as a novel source of bioactive peptides. For instance, finger millet, barnvard millet, and proso millet are rich sources of antimicrobial proteins/peptides [38]; finger millet and pearl millet show antioxidant activity and foxtail millet peptides possess antihypertensive properties [39,40]. In addition to this, millets' bioactive peptides possess several physiological and biological functions that include antioxidant, antimicrobial, anti-hypertensive, immune modulatory, anti-inflammatory, antifungal, antiviral and anti-cancer effects [41,42]

The phytochemicals derived from millets have a variety of biological functions like anti-cancer, antimicrobial and anti-inflammatory activities, suggesting their use as a functional food in the prevention and therapy of diseases. A peroxidase of class III protein extracted from foxtail millet bran, known as "FMBP", was found to have anti-colon-cancer activity in mice in both in vitro and in vivo studies [43]. The FMBP suppressed colon cancer cell growth by arresting the G1 phase, but apparently had no effect on the normal colon epithelial cells. In the same study, the epithelial-mesenchymal transition (EMT), and FMBP reduced the phosphorylation of JAK1 and its downstream signaling molecular STAT3, followed by the reduced expression of c-Myc and Snail1 of EMT. The STAT3 overexpression could partially reverse the migration inhibition caused by FMBP [44]. Also, the anti-cancer effects of FMBP were mainly achieved through the accumulation of more ROS in colon cancer cells than normal cells, attributed to the down-regulation of NF-E2-related factor 2 (Nrf2) expression, and the reduction of catalase activities and glutathione contents [45].

In a study by Kuruburu et al., phytochemical-rich fractions (phenolics and flavonoids) were extracted from foxtail millet seeds using 70% ethanol (FTM-FP) and 10% alkali (FTM-BP). Both fractions showed antiproliferative properties against breast cancer cell lines by inducing cell cycle arrest in the G2/M phase followed by increased DNA fragmentation leading to the accumulation of more cells in the Sub-G1 phase [46]. Also, Zhang and Liu reported that the extracted phenolic and carotenoids from foxtail millet cultivars Jingu28 and Jingu34 inhibited the growth of human breast and liver cancer cells in culture [47].

The antioxidants obtained from both kodo and pearl millet bran extracts exhibited higher antiproliferative/anti-cancer activities compared with those of the dehulled extracts (Table 2) [48]. The same study also concluded that a low concentration (0.1 mg/ml) of phenolic extracts obtained from the dehulled pearl millet grain showed 1.3 times higher anti-carcinogenic activity than a high concentration (0.5 mg/ml). In addition, it was noted that phenolic compounds isolated from whole pearl millet grain (52.7%) and dehulled grain (43.8%) exhibited greater inhibitory effects against the colon cancer cell line HT-29 in comparison to their kodo millet counterparts. Vanillin (4-hydroxy-3-methoxybenzaldehyde) extracted from proso and barnyard millets showed that the proso millet extract suppressed the cellual proliferation of HT-29 cells significantly when treated with 250 µg/ml and 1000 µg/ml concentrations of phenolic extracts followed by a 48-h incubation period, while the barnyard millet extract moderately inhibited the proliferation of the HT-29 cell line at the same concentrations of phenolic extracts and within the same time period [49]. The same extracts applied to the MCF-7 cell line at the same concentrations and for the same time period showed that both the extracts had significantly inhibited the proliferation of MCF-7 cells through G0/G1 phase cell arrest and increased the apoptotic cells in the sub-G0 phase in a dose-dependent manner [50].

S. no.	Crop	Functional factor	Cancer		Mechanism of action	
			Туре	Cell line		Reference
1	Pearl millet	TPC, hydroxyl and peroxyl radical inhibition	Colon cancer	HT-29	Millet grain phenolic extracts suppressed the cancerous cell growth, which was majorly time and dose dependent. At a higher concentration (0.5 mg/ml), hulled pearl phenolic extracts exhibited 68% of cellular proliferation inhibition activity. On the other hand, at a lower concentration (0.1 mg/ml), the whole and dehulled pearl millet grain phenolic extracts had a higher inhibition activity of 53% and 44%, respectively, against HT-29 cells, compared to kodo millet counterparts.	[48]
2	Foxtail millet	35 kDa protein FMBP extracted from foxtail millet bran	Colon cancer	DLD1, SW480, HT-29 and human colon epithelial cell line FHC	FMBP, homologous to peroxidase enzyme, suppressed colon cancer cell growth through induction of G1 phase arrest. The phenolic extract induced caspase-dependent apoptosis in colon cancer cells leading to the loss of mitochondrial trans-membrane.	[43]
		BPIS from foxtail millet bran (vitexin and syringic acid)	Breast cancer	MDA- MB-231 and MCF-7	BPIS suppressed the breast cancerous cells through blocking the conversion of SFA to MUFA. BPIS also reduced the GRP78 protein that inhibits the expression of SREBP-1 and downstream-target SCD1.	[51-52]
		Phytochemical rich fractions (free and bound phenolics)	Breast cancer	MCF-7 and MDA- MB-468	Induced the cell cycle arrest in the G2/M phase; increased the DNA fragmentation and a large number of cells accumulated in the Sub-G1 phase.	[46]
3	Proso millet	Vaniilin (4-hydroxy-3 methoxybenzaldehyde)	Colon cancer	HT-29	Significantly inhibited the proliferation of HT-29 cells when treated with concentrations of 250 $\mu g/ml$ and 1000 $\mu g/ml$ for 48 h.	[49]
			Breast cancer	MCF-7	Significantly inhibited the proliferation of MCF-7 cells when treated with 250 µg/ml and 1000 µg/ml concentrations for 48 h; cell cycle arrest occurred in the G0/G1 phase and increased cellular growth inhibition in the Sub-G0 phase, in a dose-dependent manner.	[50]
		Vanillin (4-hydroxy-3 methoxybenzaldehyde)	Colon cancer	HT-29	Moderately inhibited the proliferation of HT-29 cells when treated with concentrations of 250 $\mu g/ml$ and 1000 $\mu g/ml$ for 48 h.	[49]
4	Barnyard millet		Breast cancer	MCF-7	Suppressed the tumor-producing cells in the MCF-7 line when treated with 250 µg/ml and 1000 µg/ml concentrations of phenolic extracts followed by a 48-h incubation period; cell cycle arrest occurred in the G0/G1 phase and increased cellular growth inhibition in the Sub-G0 phase, in a dose-dependent manner compared to normal cells.	[50]
5	Finger millet	Free and bound phenolics	Breast cancer	MCF-7, MDA- MB-231, and MDA- MB-468	Free phenolics (procyanidin B1, 3-O-methyl quercetin and epigallocatechin) exhibited better cytotoxicity against breast cancer cell lines compared to bound phenolics. Free phenolics induced fragmentation of DNA and apoptotic cell death in breast cancer cells.	[51]
6	Kodo millet	TPC, hydroxyl and peroxyl radical inhibition	Colon cancer	HT-29	Cell proliferation inhibition by millet grain phenolic extracts was time and dose dependent. At a lower (0.1 mg/ml) and higher (0.5 mg/ml) concentration, hulled kodo millet phenolic extracts showed 100% cellular proliferation inhibition activity. (3) At a higher concentration (0.5 mg/ml), the whole and dehulled kodo millet grain phenolic extracts showed a higher inhibition activity of 100% and 76%, respectively, against HT-29 cells, compared to their pearl millet counterparts.	[48]

### TABLE 2: Millets' bioactive peptides and their effects on cancer cell lines

TPC, total phenolic content; BPIS, bound polyphenol of inner shell; SFA, saturated fatty acids; MUFA, monounsaturated fatty acids

The phenolics and phenolic acid derivatives, flavonoids and amino acids extracted from seeds of finger millet variety KMR 301 using 70% ethanol and 10% alkali revealed that free phenolic acids induced cell death in breast and colorectal cancer cells by inducing G0/G1 or G2/M arrest in a cell line-dependent fashion and increased the fragmentation of DNA leading to the accumulation of cells in the Sub-G1 phase [51-52].

Bioavailability, Bioaccessibility, and Delivery of Millet Bioactive Peptides

The bioactivity of peptides is determined by both the bioavailability and bioaccessibility of peptides [53]. The bioavailability, bioaccessibility, and delivery of millet bioactive peptides face significant difficulties due to the physiochemical and biological properties of peptides such as molecular size, charge, lipophilicity, solubility, and route of administration [54]. So far, peptide delivery has primarily relied on parental routes (injections and infusions), which have their own limitations. Other delivery routes, such as oral,

transdermal, nasal, pulmonary, and buccal, are being considered as an alternative non-invasive strategy to deliver peptides [55].

Production and Processing of Millet Bioactive Peptides

Millet bioactive peptides should be extracted and used for further research to cure cancer. The production and processing of millet bioactive peptides involves several steps starting from extraction and purification of the protein from the source seeds, hydrolysis of the purified protein, and identification, purification, and characterization of peptides from the hydrolyzed protein. Generally, bioactive peptides are inactive within the sequence of parent proteins and become active once released through the hydrolysis process.

### Conclusions

Millets are an affordable and a nutritious option for people living in poor, low-income settings, who cannot afford expensive functional foods with high health benefits. Farmers should be encouraged to grow millets, and subsidies should be given by the government in order to boost their production and consumption. Millets' consumption should be encouraged by making people aware of the nutritional merits and overall health benefits of millets, including their potential to lower the risk of cancer in humans. The compounds found in millets that have anti-cancerous properties can be extracted and recommended for cancer cure. Anti-cancer properties of the millets further to successfully exploit the potential of these grains for the treatment of future cancer patients.

# **Additional Information**

### Author Contributions

All authors have reviewed the final version to be published and agreed to be accountable for all aspects of the work.

Concept and design: Mansha Gupta, Dina Medhanie Asfaha, Govintharaj Ponnaiah

Acquisition, analysis, or interpretation of data: Mansha Gupta, Govintharaj Ponnaiah

Drafting of the manuscript: Mansha Gupta, Dina Medhanie Asfaha

Critical review of the manuscript for important intellectual content: Mansha Gupta, Dina Medhanie Asfaha, Govintharaj Ponnaiah

Supervision: Mansha Gupta

#### Disclosures

**Conflicts of interest:** In compliance with the ICMJE uniform disclosure form, all authors declare the following: **Payment/services info:** All authors have declared that no financial support was received from any organization for the submitted work. **Financial relationships:** All authors have declared that they have no financial relationships at present or within the previous three years with any organizations that might have an interest in the submitted work. **Other relationships:** All authors have declared that there are no other relationships or activities that could appear to have influenced the submitted work.

# References

- Amadou I, Gounga ME, Le GW: Millets: nutritional composition, some health benefits and processing a review. Emir J Food Agric. 2013, 25:501-8. 10.9755/ejfa.v2517.12045
- Kumar A, Kaur A, Tomer V, Rasane P, Gupta K: Development of nutricereals and milk-based beverage: process optimization and validation of improved nutritional properties. J Food Process Eng. 2020, 43:e15025.10.1111/fne.13025
- Tomer V, Kaur A, Kuar A, Kumar A: Glycaemic index of Indian flatbreads (rotis) prepared using multigrain flour and whole wheat flour. Ann Biol. 2018, 34:143-7.
- Kumar A, Tomer V, Kaur A, Kumar V, Gupta K: Millets: a solution to agrarian and nutritional challenges. Agric Food Secur. 2018, 7:31. 10.1186/s40066-018-0183-3
- Singh RB, Watson RR, Takahashi T (eds): The Role of Functional Food Security in Global Health . Academic Press, Cambridge; 2019. 10.1016/C2016-0-04169-4
- Ambati K, Sucharitha KV: Millets review on nutritional profiles and health benefits. Int J Recent Sci Res. 2019, 10:33943-8. 10.24327/ijrsr.2019.1007.3786
- Kannan SM, Thooyavathy RA, Kariyapa RT, Subramanian K, Vijayalakshmi K: Seed Production Techniques for Cereals and Millets. Vijayalakshmi K (ed): Centre for Indian Knowledge Systems (CIKS) Seed Node of the Revitalising Rainfed Agriculture Network, Chennai, India; 2013.
- Dayakar RB, Bhaskarachary K, Arlene Christina GD, Sudha Devi G, Tonapi A: Nutritional and Health Benefits of Millets. ICAR-Indian Institute of Millets Research, Hyderabad, India; 2017.
- Saleh ASM, Zhang Q, Chen J, Shen Q: Millet grains: nutritional quality, processing, and potential health benefits. Compr Rev Food Sci Food Saf. 2013, 12:281-95. 10.1111/1541-4337.12012
   Li YO, Komarek AR: Dietary fibre basics: health, nutrition, analysis, and applications. Food Oual Saf. 2017,
- In To, Roman Criter Decumption of a constrainty matrices, and spin and appreciations in our guint our 2017 1:47-59. 10.1093/fqsafe/fyx007
   Markowiak P Śliżowska K: Effects of prohiotics prehiotics and synhiotics on human health. Nutrients
- Markowiak P, Śliżewska K: Effects of probiotics, prebiotics, and synbiotics on human health. Nutrients. 2017, 9:1021. 10.3390/nu9091021
- Singh P, Raghuvanshi RS: Finger millet for food and nutritional security. Afr J Food Sci. 2012, 6:77-84. 10.5897/AJFSX10.010
- 13. FSSAI issues directions regarding recommended dietary allowances . (2021). Accessed: September 5, 2023: https://foodsafetyhelpline.com/fssai-issues-directions-regarding-recommended-dietary.
- Phosphorus and magnesium. (2020). Accessed: September 5, 2023: https://www.nestle.in/nhw/nutritionbasics/nutrients/minerals/phosphorus-magnesium.
- Taylor J: Millet pearl. Encyclopedia of Grain Science. Wrigley C, Corke H, Walker CE (ed): Academic Press, London; 2004. 253-61. 10.1016/B0-12-765490-9/00097-5
   Asharani VT, Jayadeep A, Malleshi NG: Natural antioxidants in edible flours of selected small millets . Int J
- Asharam VI, Jayauep A, Maneshi NG, Natural antoxical structure nous of selected sharin hines . In J Food Prop. 2010, 13:41-50. 10.1080/10942910802163105
   Chandrasekara A, Shahidi F: Content of insoluble bound phenolics in millets and their contribution to
- Chandrasekara A, Shahidi F. Content of insolutio bound pitchines in innects and item controlotion to antioxidant capacity. J Agric Food Chem. 2010, 58:6706-14. 10.1021/f100868b
   Chandrasekara A, Shahidi F. Determination of antioxidant activity in free and hydrolyzed fractions of millet
- grains and characterization of their phenolic profiles by HPLC-DAD-ESI-MSn. J Funct Foods. 2011, 3:144-58. 10.1016/j.jff.2011.03.007
- Pradeep PM, Sreerama YN: Impact of processing on the phenolic profiles of small millets: evaluation of their antioxidant and enzyme inhibitory properties associated with hyperglycemia. Food Chem. 2015, 169:455-63. 10.1016/j.foodchem.2014.08.010

- Pruksasri S, Wollinger KK, Novalin S: Transformation of rice bran into single-cell protein, extracted protein, soluble and insoluble dietary fiber, and minerals. J Sci Food Agric. 2019, 99:5044-9. 10.1002/jsfa.9747
- Rose DJ, DeMeo MT, Keshavarzian A, Hamaker BR: Influence of dietary fiber on inflammatory bowel disease and colon cancer: importance of fermentation pattern. Nutr Rev. 2007, 65:51-62. 10.1111/j.1753-4887 2007 tb0028 x
- Shobana S, Malleshi NG: Preparation and functional properties of decorticated finger millet (Eleusine coracana). J Food Engg. 2007, 79:529-38. 10.1016/j.jfoodeng.2006.01.076
- Annison G, Topping DL: Nutritional role of resistant starch: chemical structure vs physiological function. Annu Rev Nutr. 1994, 14:297-320. 10.1146/annurev.nu.14.070194.001501
- Englyst HN, Kingman SM, Cummings JH: Classification and measurement of nutritionally important starch fractions. Eur J Clin Nutr. 1992, 46:S33-50.
- Ren A, Chen L, Zhao W, Shan S, Li Z, Tang Z: Extraction optimization and structural characterization of soluble dietary fiber in foxtail millet and its inhibitory activities on colon cancer. J Funct Foods. 2013, 107:105659. 10.1016/j.ff.2023.105659
- Zhang B, Xu Y, Liu S, et al.: Dietary supplementation of foxtail millet ameliorates colitis-associated colorectal cancer in mice via activation of gut receptors and suppression of the STAT3 pathway. Nutrients. 2020. 12:357. 10.3590/mu12082367
- Mosolov VV, Grigor'eva LI, Valueva TA: Plant proteinase inhibitors as multifunctional proteins. Appl Biochem Micro. 2001, 37:545-51. 10.1023/A:1012352914306
- Pandey R, Patil N, Rao M: Proteases and protease inhibitors: implications in antitumorigenesis and drug development. Int J Hum Genet. 2007, 7:67-82. 10.1080/09723757.2007.11885986
- Laskowski M Jr, Qasim MA: What can the structures of enzyme-inhibitor complexes tell us about the structures of enzyme substrate complexes?. Biochim Biophysic. 2000, 1477:524-57. 10.1016/S0167-4858/9000284-8
- De Leo F, Volpicella M, Licciulli F, Liuni S, Gallerani R, Ceci LR: PLANT-PIs: a database for plant protease inhibitors and their genes. Nucleic Acids Res. 2002, 30:347-8. 10.1093/nar/30.1.347
- Lingaraju MH, Gowda LR: A Kunitz trypsin inhibitor of Entada scandens seeds: another member with single disulfide bridge. Biochim Biophys Acta. 2008, 1784:850-5. 10.1016/j.bbapap.2008.02.013
- Sen S, Dutta SK: Evaluation of anti-cancer potential of ragi bifunctional inhibitor (RBI) from Elusine coracana on human chronic myeloid leukemia cells. Eur J Plant Sci Biotec. 2012, 6:103-8.
- Maskos K, Huber-Wunderlich M, Glockshuber R: RBI, a one-domain alpha-amylase/trypsin inhibitor with completely independent binding sites. Federat Eur Biochem Soc Lett. 1996, 397:11-16. 10.1016/S0014-5793(96)01131-3
- Campos FAP, Richardson M: The complete amino acid sequence of the bifunctional α-amylase/trypsin inhibitor from seeds of ragi (Indian finger millet, Eleusine coracana Gaertn.). FEBS Lett. 1983, 152:300-4. 10.1016/0014-5793(83)80400-1
- Jones PJ: Clinical nutrition: 7. Functional foods more than just nutrition. CMAJ. 2002, 166:1555-63.
  Chalamaiah M, Ulug SK, Hong H, Wu J: Regulatory requirements of bioactive peptides (protein hydrolysates) from food proteins. J Funct Foods. 2019, 58:123-9. 10.1016/j.jff.2019.04.050
- from food proteins. J Funct Foods. 2019, 58:123-9. 10.1016/j.jff.2019.04.050 37. Daliri EB, Oh DH, Lee BH: Bioactive peptides. Foods. 2017, 6:32. 10.3390/foods6050032
- Bish A, Thapliyal M, Singh A: Screening and isolation of antibacterial proteins/peptides from seeds of millets. Int J Curr Pharm Res. 2017, 8:96-9.
- Agrawal H, Joshi R, Gupta M: Purification, identification and characterization of two novel antioxidant peptides from finger millet (Eleusine coracana) protein hydrolysate. Food Res Int. 2019, 120:697-707. 10.1016/j.foodres.2018.11.028
- Chen J, Duan W, Ren X, Wang C, Pan Z, Diao X, Shen Q: Effect of foxtail millet protein hydrolysates on lowering blood pressure in spontaneously hypertensive rats. Eur J Nutr. 2017, 56:2129-38. 10.1007/s00394 016-1252-7
- Xu S, Shen Y, Xu J, et al.: Antioxidant and anticancer effects in human hepatocarcinoma (HepG2) cells of papain-hydrolyzed sorghum kafirin hydrolysates. J Funct Foods. 2019, 58:374-82. 10.1016/j.jff.2019.05.016
- Castro-Jácome TP, Alcántara-Quintana LE, Tovar-Pérez EG: Optimization of sorghum kafirin extraction conditions and identification of potential bioactive peptides. Biores Open Access. 2020, 9:198-208. 10.1089/biores.2020.0013
- Shan S, Li Z, Newton IP, Zhao C, Li Z, Guo M: A novel protein extracted from foxtail millet bran displays anti-carcinogenic effects in human colon cancer cells. Toxicol Lett. 2014, 227:129-38.
   10.1016 toyler 2014 03 008.
- Shan S, Li Z, Guo S, Li Z, Shi T, Shi J: A millet bran-derived peroxidase inhibits cell migration by antagonizing STAT3-mediated epithelial-mesenchymal transition in human colon cancer. J Funct Foods. 2014, 10:444–55. 10:1016/j.iff.2014.07.005
- Shan S, Shi J, Li Z, Gao H, Shi T, Li Z, Li Z: Targeted anti-colon cancer activities of a millet bran-derived peroxidase were mediated by elevated ROS generation. Food Funct. 2015, 6:2331-8. 10.1039/c5fo00260e
- 46. Kuruburu MG, Bovilla VR, Leihang Z, Madhunapantula SV: Phytochemical-rich fractions from foxtail millet (Setaria italica (L.) P. Beauv) seeds exhibited antioxidant activity and reduced the viability of breast cancer cells in vitro by inducing DNA fragmentation and promoting cell cycle arrest. Anticancer Agents Med Chem. 2022, 22:2477-95. 10.2174/1871520622666220215122141
- Zhang LZ, Liu RH: Phenolic and carotenoid profiles and antiproliferative activity of foxtail millet . Food Chem. 2015, 174:495-501. 10.1016/j.foodchem.2014.09.089
- Chandrasekara A, Shahidi F: Bioactivities and antiradical properties of millet grains and hulls. J Agric Food Chem. 2011, 59:9563-71. 10.1021/jf201849d
- Ramadoss DP, Sivalingam N: Vanillin extracted from Proso and Barnyard millets induce apoptotic cell death in HT-29 human colon cancer cell line. Nutr Cancer. 2020, 72:1422-37. 10.1080/01635581.2019.1672763
- Ramadoss DP, Sivalingam N: Vanillin extracted from proso and barnyard millets induces cell cycle inhibition and apoptotic cell death in MCF-7 cell line. J Cancer Res Ther. 2021, 17:1425-33. 10.4103/jcrt.JCRT\_1128\_19
   Kuruburu MG, Bovilla VR, Naaz R, Leihang Z, Madhunapantula SV: Variations in the anticancer activity of
- Kuruburu MG, Bovilla VR, Naaz K, Leihang Z, Madhunapantula SV: Variations in the anticancer activity of free and bound phenolics of finger millet (Eleusine coracana (L) Gaertn; Variety KMR-301) seeds. Phytomed Plus. 2022, 2:100276. 10.1016/j.phyplu.2022.100276
   Zhang L, La X, Tian J, et al.: The phytochemical vitexin and syringic acid derived from foxtail millet bran
- Zhang L, La X, Tian J, et al.: The phytochemical vitexin and syringic acid derived from foxtail millet bran inhibit breast cancer cells proliferation via GRP78/SREBP-1/SCD1 signaling axis. J Funct Foods. 2021, 85:104620. 10.1016/j.jff.2021.104620
- Sun X, Acquah C, Aluko RE, Udenigwe CC: Considering food matrix and gastrointestinal effects in enhancing bioactive peptide absorption and bioavailability. J Funct Foods. 2020, 64:103680. 10.1016/j.jff.2019.103680
- Kumar TR, Soppimath K, Nachaegari SK: Novel delivery technologies for protein and peptide therapeutics. Curr Pharm Biotechnol. 2006, 7:261-76. 10.2174/138920106777950852
- Wallis L, Kleynhans E, Toit TD, et al.: Novel non-invasive protein and peptide drug delivery approaches. Protein Pept Lett. 2014, 21:1087-101. 10.2174/0929866521666140807112148