

A review of millet grain phenolics, their health promotion, and disease risk reduction.

Abstract

Millet, an often-overlooked cereal grain, has recently garnered attention due to its rich store of health-promoting phenolic compounds. This comprehensive review delves into the myriad facets of millet phenolics, from their extraction and analysis to their implications for health and disease risk reduction. The varied methodologies for phenolic extraction, including solvent, enzymatic, and ultrasound-assisted techniques, have their own merits and limitations. Advanced analytical tools such as High Performance Liquid Chromatography and Mass Spectrometry have enabled the identification of specific phenolic compounds in different millet varieties. These phenolics have shown potential in promoting health through their antioxidant, anti-inflammatory, anti-diabetic, cardiovascular, anti-cancer, digestive, and neuroprotective effects. Mechanistic studies have illuminated their role in modulating signaling pathways, interacting with enzymes and receptors, and altering gene expression. Notably, the consumption of millet phenolics has been linked with reduced disease risks, supported by epidemiological studies and clinical trials. However, the phenolic content in millets can be influenced by various factors such as agricultural practices, post-harvest processing, and cooking methods. As we gaze into the future, there are challenges to address, especially concerning the bioavailability of millet phenolics. Efforts towards genetic modification and breeding to enhance phenolic content, along with their incorporation into functional foods and nutraceuticals, promise novel avenues for research and application. In summary, this review underscores the untapped potential of millet phenolics in promoting human health, urging further exploration and integration into the modern diet for holistic well-being.

Keywords: *Millet, Phenolics, Bioavailability, Nutraceuticals, Health-promotion*

Introduction

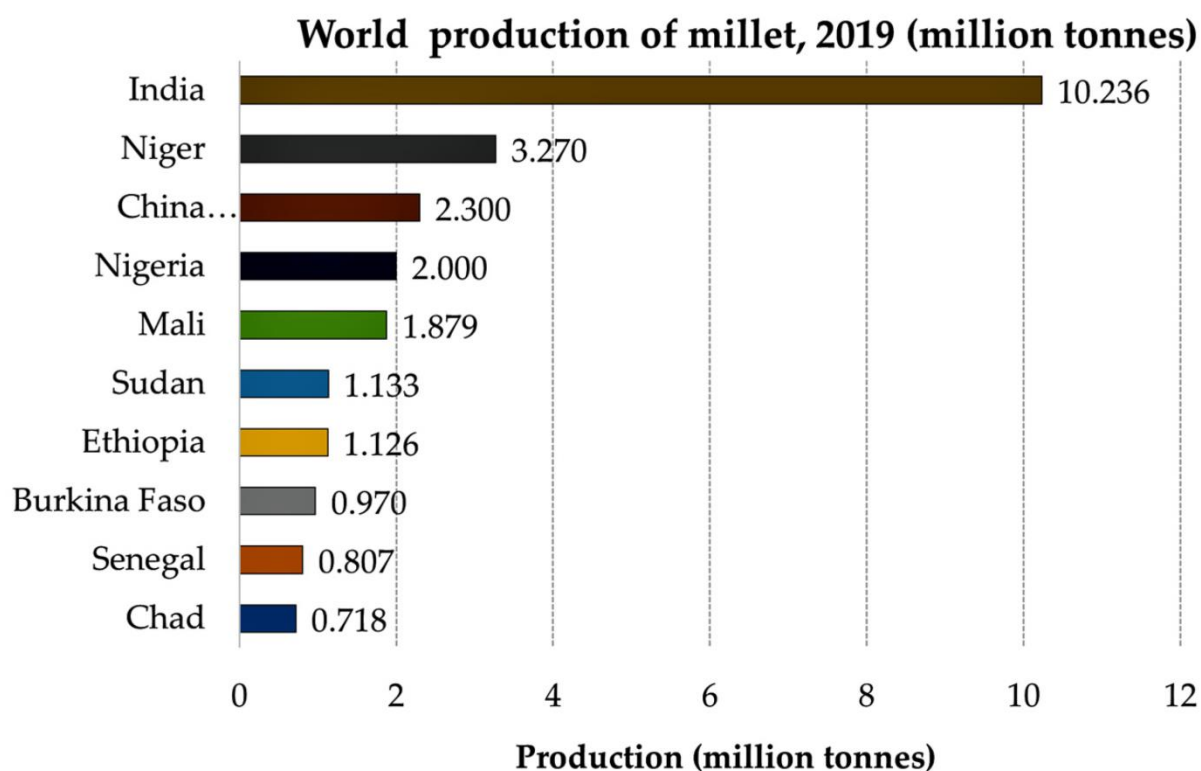
Millet is not a single grain but rather a term used to describe a diverse group of small-seeded cereals that have been cultivated since ancient times. They are primarily grown in the semi-arid tropics of Asia and Africa and have long been staple foods for these regions. Due to their ability to grow in harsh conditions, including droughts and poor soil quality, millets are considered critical for ensuring food security in many parts of the world [1]. Over the years, different species of millets have been identified, with the most popular being pearl millet (*Pennisetum glaucum*), foxtail millet (*Setaria italica*), proso millet (*Panicum miliaceum*), and finger millet (*Eleusine coracana*) (Table 1). Each variety has its unique nutritional profile, flavor, and culinary uses. What makes millets particularly interesting, aside from their hardiness, is their rich nutrient content. They are a good source of essential nutrients like dietary fiber, vitamins, minerals, and various bioactive compounds. Phenolic compounds, a major group of secondary metabolites, play vital roles in plants. These compounds serve several

functions, from contributing to the plant's defense mechanisms against pests and diseases to the development and reproduction of the plant. Moreover, they are responsible for the colors of many flowers, fruits, and leaves, which can attract pollinators or act as a protective mechanism against harmful ultraviolet radiation [2]. From a human nutritional perspective, phenolics have garnered significant attention due to their potential health benefits. They possess potent antioxidant properties, enabling them to combat oxidative stress in the body. Oxidative stress is implicated in the progression of various chronic diseases, including cardiovascular diseases, neurodegenerative conditions, and certain types of cancers. By neutralizing free radicals – unstable molecules that can damage cells – phenolic compounds can potentially mitigate the adverse effects of these diseases. Several plants have been studied for their phenolic content and associated health benefits. Among these, millet grains are emerging as an important source of dietary phenolics, especially in regions where they are staple foods. The increasing global interest in healthy and functional foods has brought millet phenolics to the forefront of nutritional research. Despite their consumption for millennia in many cultures, a comprehensive understanding of the phenolic content of millets and their potential health benefits is still evolving. This review aims to explore the various phenolic compounds identified in different millet varieties, their potential health-promoting properties, and their role in disease risk reduction. By consolidating current knowledge in this area, this review seeks to highlight the importance of millets not just as a food security crop but also as a potentially significant player in promoting public health [3].

Table 1. Different Types of Millet and Their Potential Features [4]

Millet Type	Features
Foxtail millet	Reduces risk of colon cancer. Lessen cholesterol and possesses anti-diabetic capability. Attenuates ethanol-induced hepatic damage.
Pearl millet	Gluten-free property averts celiac disease. The immune system improves by inhibiting pathogenicity induced by Shigella.
Finger millet	Reduces damage to soft tissue and facilitates the healing process. Reduces plasma triglycerides, thus reducing the risk of cardiovascular disease.
Kodo millet	Minimize glycemic index and diabetes occurrence, and have antioxidant actions as well.
Proso millet	Celiac disease can be prevented due to gluten-free properties. Being a low-glycemic index (GI) food reduces type 2 diabetes risks.
Little millet	Polyphenol content helps to prevent various metabolic disorders.
Barnyard millet	Damaging apoptotic cells reduces colorectal cancer risk. Inhibits protein glycation and glycooxidation, which improves the state of diabetes.

Figure 1. World production of millet (million tonnes) [5].



Nutritional Profile of Millets

When we talk about millets in the context of nutrition, it's an impressive narrative. These tiny grains pack a punch when it comes to health benefits. **Macronutrients:** Millets are a good source of carbohydrates, making them an excellent energy source. They also contain proteins, and while they might not rival legumes in protein content, their amino acid profile is commendable. Millets like finger millet are particularly high in protein content compared to other grains [6]. **Micronutrients:** The mineral content in millets deserves mention. They are rich in magnesium, phosphorus, iron, calcium, potassium, and zinc. Finger millet, for instance, is an outstanding source of calcium, making it beneficial for bone health. **Fiber:** Dietary fiber is another forte of millets. The high fiber content aids in digestion, helps in slowing the absorption of glucose, thus beneficial for diabetics, and can assist in weight management by promoting a feeling of fullness. **Phytochemicals:** Beyond the conventional nutrients, millets are also rich in bioactive compounds like polyphenols, which have antioxidant properties. These compounds have been linked to various health benefits, from anti-inflammatory effects to potential anti-cancer properties. **Gluten-Free:** With the rise in cases of celiac disease and gluten sensitivity, millets are gaining popularity as a gluten-free grain. This makes them an excellent choice for people with gluten-related disorders.

Phenolics

Phenolic compounds, ubiquitous in the plant kingdom, have been a focal point of interest for researchers, nutritionists, and health enthusiasts alike (Table 2). Their presence in our daily diet, from the morning cup of tea to the fruits and vegetables we consume, indicates their importance. This article delves deep into the nature, classification, and importance of phenolics in human health. Phenolic compounds, often referred to as polyphenols, are a vast group of chemicals characterized by the presence of one or more aromatic rings bearing one or more

hydroxyl groups [7]. Their structure is fundamentally derived from the phenol molecule, which comprises a benzene ring bonded to a hydroxyl group. At the molecular level, phenolic compounds can range from simple molecules, like phenolic acids, to highly polymerized substances, like tannins. Their structure predominantly influences their biological activities and their interactions within the human body. For instance, the number and arrangement of the hydroxyl groups on the aromatic rings can determine the antioxidant capacity of the compound.

Table 2. Phenolic content (mg/100 g) and reducing capacity (%) of millets and rice. [8]

Food Grain	Vanillic	Proto	Catechic	P-Hydroxy Benzoic	Syringic	Genisic	Gallic	Coumaric	Caffeic	Ferulic	Sinapic	Cinnamic	Reducing Capacity (%)
Rice	0.54	0.90	0.49	0.55	NA	1.38	1.62	0.61	11.48	2.08	0.30	-	-
Finger millet	1.52	2.31	0.89	0.77	6.15	NA	5.69	1.66	67.97	NA	3.50	5.7 ± 1.05	-
Pearl millet	1.63	1.18	2.20	1.73	9.63	NA	26.82	2.13	38.70	NA	34.53	-	-
Proso millet	NA	NA	NA	3.05	NA	NA	8.35	7.55	23.56	NA	NA	2.6 ± 0.20	-
Foxtail millet	8.71	NA	1.46	9.36	2.15	NA	213.37	1.06	75.58	NA	78.17	4.8 ± 1.15	-

Classes of Phenolics Found in Plants

The plant kingdom is teeming with a plethora of phenolic compounds. Their diversity is vast, and they can be broadly categorized based on their structural properties [9].

Simple Phenols and Phenolic Acids: These are relatively small molecules. Examples include gallic acid, often found in tea, and vanillic acid present in vanilla.

Flavonoids: Among the most researched group of phenolics, flavonoids encompass a wide range of compounds. They are recognized by their structure of two aromatic rings (A and B) bound together by three carbon atoms, forming an oxygenated heterocycle (Ring C). Flavonoids can be further divided into:

- Flavonols:** e.g., quercetin and kaempferol found in onions, broccoli, and apples.
- Flavones:** e.g., luteolin and apigenin, often located in parsley and celery.
- Flavanones:** e.g., hesperidin and naringenin found in citrus fruits.
- Flavan-3-ols:** e.g., catechin and epicatechin present in tea and cocoa.

Anthocyanins: Responsible for the vibrant colors of many fruits and flowers. Examples include cyanidin in cherries and delphinidin in blueberries.

Lignans: These are derived from phenylpropanoid units. Flaxseeds are a significant source of lignans, particularly secoisolariciresinol.

Stilbenes: These are composed of two aromatic rings connected by a two-carbon methylene bridge. Resveratrol, found in grapes, is a well-known stilbene with various studied health benefits.

Tannins: These are large polymeric phenolic compounds. Based on their structure and properties, tannins can be categorized as hydrolysable or condensed. They're common in various fruits, especially unripe ones, and in beverages like wine and tea.

Importance of Phenolics in Human Health

The growing interest in plant phenolics is fueled by a plethora of scientific evidence highlighting their manifold health benefits. Known for their strong antioxidant activity, phenolic compounds neutralize free radicals and combat oxidative stress, which has been linked to chronic diseases like cardiovascular disorders, cancer, and neurodegenerative diseases [10]. These compounds also exhibit potent anti-inflammatory effects, especially in the form of flavonoids that can inhibit various inflammatory pathways. In the realm of cardiovascular

health, phenolics bolster endothelial function, thwart LDL oxidation, and reduce blood pressure, collectively acting as cardioprotective agents. Beyond this, they also have neuroprotective qualities, with compounds like resveratrol and certain flavonoids shown to shield neural cells and possibly mitigate diseases like Alzheimer's [11]. They also offer promise in the fight against cancer, as some phenolic compounds can inhibit cancer cell growth, induce apoptosis, and forestall DNA damage. Additionally, phenolics have a significant impact on gut health; they can be metabolized by gut microbiota to produce beneficial metabolites and also influence the microbiota composition to favor beneficial bacteria. Lastly, some phenolics have been found to have anti-diabetic effects by affecting carbohydrate metabolism, inhibiting enzymes associated with glucose digestion and absorption, and improving insulin sensitivity.

Methods of Phenolic Extraction from Millets

Phenolic compounds, as previously detailed, play an integral role in human health due to their various beneficial properties. Extracting these compounds from plants, including millets, in their purest form is pivotal for both research and application. This article will explore the different methods employed in the extraction of phenolic compounds from millets, detailing the processes, and discussing the associated limitations and challenges. Solvent extraction is a traditional and widely-used method to obtain phenolics from plant materials. The principle behind it is relatively straightforward; solvents are used to dissolve the phenolic compounds, separating them from the plant matrix.

The millet samples are first dried and ground to increase the surface area for better solvent penetration. The powdered sample is then mixed with a suitable solvent, commonly ethanol, methanol, or their aqueous solutions. The mixture undergoes agitation for a specified time, followed by filtration or centrifugation to separate the liquid extract from the solid residues. The obtained extract, rich in phenolics, is then concentrated by evaporating the solvent. While solvent extraction is relatively simple and effective for a broad spectrum of phenolic compounds, it can be time-consuming. Additionally, residual solvents might contaminate the extract, demanding further purification. A more eco-friendly alternative to traditional solvent extraction is enzymatic extraction. This method leverages enzymes to break down the cell walls, facilitating the release of phenolic compounds. The millet is subjected to enzymatic hydrolysis using specific enzymes like cellulase, pectinase, or a combination thereof. These enzymes degrade the cell wall components, improving the accessibility and yield of phenolics. Post enzymatic treatment, phenolics are extracted using solvents or water and then separated from the solid residues. One of the key benefits of enzymatic extraction is the reduced need for solvents. However, the enzymes can be expensive, and maintaining optimal conditions, such as pH and temperature, makes the process more complex.

Another innovative method in the extraction space is ultrasound-assisted extraction (UAE), which employs ultrasonic waves to disrupt cell walls and improve solvent penetration, thus enhancing the extraction efficiency of phenolic compounds. In this method, millet samples, similar to solvent extraction, are dried, ground, and then mixed with a suitable solvent. This mixture is then subjected to ultrasonic waves, typically at frequencies between 20 kHz to 40 kHz. The ultrasonic waves create cavitation bubbles. Upon collapsing, these bubbles produce microjets that disrupt the cell walls. Post ultrasonication, the mixture is filtered or centrifuged to obtain the phenolic-rich extract. UAE accelerates the extraction process and often achieves higher yields and purity of phenolics. However, over-exposure can degrade certain sensitive

compounds, and the method requires specialized equipment. Despite the advancements in extraction methods, they come with several limitations and challenges. The choice of solvent can significantly impact the yield and profile of extracted phenolics. The polarity of the solvent, its toxicity, and environmental impact need thorough consideration. Each method also demands the careful optimization of parameters like temperature, pH, solvent concentration, and extraction time. There's the challenge of scale-up as processes optimized at a lab scale might not directly translate to industrial scales. Environmental concerns also arise, especially with solvent extraction methods. Advanced extraction techniques might offer better yields, but their initial setup costs are significantly higher, making them less feasible for small-scale industries. Lastly, preserving the integrity of certain phenolic compounds, sensitive to light, heat, and oxygen, remains a consistent challenge across all extraction methods.

Quantitative and Qualitative Analysis of Millet Phenolics

The presence of phenolic compounds in millets has long been established as a boon for their nutritional profile. These compounds, not only add to the health benefits associated with millet consumption but also provide insights into the diverse chemical ecology of the millet plant. Hence, the accurate quantitative and qualitative analysis of these phenolics is of paramount importance. Employing advanced analytical techniques ensures the comprehensive characterization of these bioactive compounds in millets.

High Performance Liquid Chromatography

High Performance Liquid Chromatography (HPLC) is a prevalent technique used in the analysis of phenolic compounds. A powerful and flexible method, HPLC is renowned for its ability to separate complex mixtures of compounds with high precision. When dealing with millets, a sample is first prepared by extracting the phenolic compounds, often through solvent extraction. This extract is then injected into the HPLC system. The system, equipped with a pump, pushes the solvent (or a mixture of solvents) through a column packed with a stationary phase. The phenolic compounds present in the sample then interact with this stationary phase, leading to their separation based on their affinities. The separated compounds are then detected, most often using a UV detector, given the natural UV-absorbing properties of phenolics. The peak areas in the resulting chromatogram give a quantitative measure of the phenolics, whereas the retention times can be used to identify them, especially when compared to known standards [12].

Mass Spectrometry

Complementing HPLC, Mass Spectrometry (MS) is another instrumental technique of immense value. While HPLC separates the compounds, MS provides information about the molecular weight and structural features of the phenolics. The combined approach, known as LC-MS, is especially powerful. In LC-MS, once the phenolic compounds are separated in the HPLC column, they are ionized and transferred to the mass spectrometer. Here, the compounds are subjected to an electric or magnetic field, making them follow a trajectory based on their mass-to-charge ratio. Detectors then measure the ion's flight path, and the resulting data can be used to deduce the molecular weight and sometimes, structural elements of the phenolics. This dual system offers the advantage of not only separating the compounds but also providing a deeper insight into their molecular structures [13].

Different millet varieties have been found to harbor a range of phenolic compounds, thanks to these advanced analytical techniques. For instance, pearl millet, one of the widely consumed varieties, has shown the presence of compounds like ferulic acid, p-coumaric acid, and sinapic acid. These compounds have been linked with antioxidant properties and potential health benefits. Finger millet, another variety, is rich in catechin, epicatechin, and quercetin derivatives, known for their anti-inflammatory and antioxidant properties. Kodo millet has revealed the presence of compounds like protocatechuic acid and p-hydroxybenzoic acid, which have been studied for their potential antimicrobial and anti-cancer properties. Furthermore, foxtail millet has been found to contain gallic acid and chlorogenic acid, known for their role in health promotion and disease prevention [14]. The diversity in the phenolic profile across different millet varieties underscores the importance of both quantitative and qualitative analysis. Not only does it highlight the nutritional richness of millets but also provides potential avenues for targeted nutrition. For instance, if one is looking at deriving antioxidant benefits, then choosing a millet variety rich in catechins might be advisable. Similarly, for anti-inflammatory effects, millets rich in quercetin derivatives might be preferred. The techniques like HPLC and MS offer detailed insights, challenges persist. The extraction method chosen can influence the phenolic profile. Also, environmental factors, including soil type, climate, and cultivation practices, can induce variability in the phenolic content of millets. This mandates the need for standardizing methodologies and considering the potential external influences while interpreting results [15].

Health Benefits of Millet Phenolics:

Millet, often regarded as an ancient grain, has surged in global popularity in recent times. Beyond its role as a staple in many cultures, millet holds a treasure trove of phenolic compounds that contribute to a multitude of health benefits. The health-enhancing attributes of these phenolics span from their potent antioxidant properties to their potential in safeguarding neurological health. At the forefront of these benefits is the antioxidant property of millet phenolics. Antioxidants are vital compounds that combat oxidative stress, a condition that arises when there is an imbalance between the production of free radicals and the body's ability to counteract their harmful effects (Table 3). Millet phenolics have demonstrated a pronounced free radical scavenging activity, which essentially means they can neutralize these harmful free radicals before they inflict damage on cells and tissues. This neutralization is a direct testament to their ability to donate electrons, rendering the free radicals harmless [16]. In addition to scavenging free radicals, millet phenolics possess a dual antioxidant mechanism. They exhibit reducing power, which involves donating electrons to free radicals, and also exhibit metal chelation. Some metals, when free in the body, act as catalysts in the production of free radicals. The phenolics from millets can chelate, or bind to these metals, inhibiting their involvement in free radical generation. This dual-action makes them exceptionally potent in combatting oxidative stress. Chronic inflammation is at the core of many modern-day diseases. Millet phenolics have been researched for their anti-inflammatory effects. These compounds can inhibit the release of pro-inflammatory cytokines and suppress the activation of inflammation-inducing pathways. By modulating these pathways, millet phenolics can potentially alleviate conditions driven by chronic inflammation, such as arthritis and certain auto-immune diseases [17]. Diabetes, a global health concern, has been another area where millet phenolics have shown promise. These compounds can inhibit enzymes like alpha-amylase and alpha-glucosidase, responsible for the breakdown of carbohydrates into glucose. By inhibiting these

enzymes, millet phenolics can modulate the postprandial increase in blood sugar, assisting in the management of type 2 diabetes. Heart diseases, the leading cause of global mortality, are influenced by factors such as cholesterol levels, blood pressure, and inflammation. Millet phenolics have demonstrated cardiovascular health benefits. They can inhibit the oxidation of low-density lipoproteins (LDL), a process linked with atherosclerosis.

Table 3: Antioxidants in millet. [18]

Name of Compound	Major Types	Mechanisms of Action
Phenolic acids	Kodo, Finger, Foxtail, Proso, Little, Pearl millet	Their ability to donate hydrogen atoms via hydroxyl groups on benzene rings to electron-deficient free radicals and in turn form a resonance-stabilized and less reactive phenoxyl radical.
Flavonoids	Kodo, Finger, Foxtail, Proso, Little, Pearl millet	Multiple hydroxyl groups confer upon the molecule substantial antioxidant activity. A double bond and carbonyl function in the heterocycle or polymerization of the nuclear structure increases its activity by affording a more stable flavonoid radical through conjugation and electron delocalization.
Tannins	Finger millet	Procyanidin o-quinone is capable of producing oligomeric compounds through various coupling reactions that retain the number of hydroxyl groups, and that can act as prooxidants by forming reactive oxygen species through futile redox cycling.
Xylo-oligosaccharides	Finger millet	Most oligosaccharides consist of ester linked phenolic acids. Apart from phenolic acids, the presence of sugars with > C = O (uronyl/acetyl) groups and degree/nature of polymerization impart strong antioxidant activity to the polysaccharides.
Insoluble fibers	Foxtail millet	The antioxidant properties of insoluble fiber could be attributed in part to their unique phytochemical composition.
Protein and Peptides	Pearl, Foxtail millet	The antioxidant activity of proteins is due to complex interactions between their ability to inactivate reactive oxygen species, scavenge free radicals, chelate prooxidative transition metals, reduce hydroperoxides, enzymatically eliminate specific oxidants, and alter the physical properties of food systems in a way that separates reactive species.
Carotenoid	Finger, Little, Foxtail, Proso millets	Carotenoids act as antioxidants by quenching single oxygen and free radicals.
vitamin E	Finger, Little, Foxtail, Proso millets	Biological activities of tocols are generally believed to be due to their antioxidant action by inhibiting lipid peroxidation in biological membranes.

Mechanisms of Action of Millet Phenolics

Millet, a nutritional powerhouse that has been cultivated for thousands of years, has steadily gained scientific attention due to the myriad health benefits attributed to its rich phenolic compounds. The intrigue does not merely lie in the health benefits these compounds offer, but the diverse molecular mechanisms they employ. By modulating intricate cellular signaling pathways, interacting with key enzymes and receptors, and even influencing the genetic blueprint by altering gene expressions, millet phenolics stand as a testament to the complex interplay between nutrition and human health. At the heart of cellular operations lie signaling pathways, which can be envisioned as intricate communication networks that cells employ to

respond to external stimuli. These pathways are not linear but rather a complex mesh of cross-communicating signals that ultimately result in a cellular response, whether it be growth, apoptosis, or any other function. Millet phenolics have been shown to modulate several of these pathways. For instance, the Nuclear Factor kappa-B (NF- κ B) pathway, which is central to inflammation, immune responses, and even cancer, has been shown to be influenced by millet phenolics. When activated inappropriately, NF- κ B can result in chronic inflammation and has been linked to various diseases, including cancer. Phenolic compounds from millet can inhibit this pathway, thereby reducing inflammation and potentially curbing the growth of cancer cells [19]. In a similar vein, the Mitogen-Activated Protein Kinase (MAPK) pathway, which plays roles in cell differentiation, growth, and apoptosis, can also be influenced by millet phenolics. By modulating this pathway, these phenolic compounds can have far-reaching effects on cellular functions, from controlling cell cycle progression to even influencing cell death mechanisms in cancerous cells. Moving from signaling pathways, the world of enzymes and receptors presents another layer of complexity. Every cellular function is orchestrated by enzymes, which catalyze reactions, and receptors, which are proteins that receive signals. Millet phenolics exhibit a keen ability to interact with both. For instance, alpha-amylase and alpha-glucosidase, enzymes crucial in carbohydrate metabolism, have been shown to be inhibited by phenolic compounds from millets. This inhibition can slow down the breakdown of carbohydrates into glucose, potentially assisting in the management of post-prandial blood glucose levels and offering benefits for diabetic individuals [20]. Beyond enzymes involved in metabolism, millet phenolics also target enzymes linked with neurodegenerative diseases. One such enzyme is acetylcholinesterase, whose increased activity has been linked with Alzheimer's disease. Phenolic compounds from millet can inhibit this enzyme, potentially slowing down disease progression. Receptors, on the other hand, act as gatekeepers for cells, receiving external signals and initiating a cellular response. Millet phenolics can interact with various receptors, modifying their activity. This interaction can range from enhancing the activity of certain beneficial receptors to inhibiting potentially harmful ones. These interactions have implications for a range of conditions, from inflammation to neurological health.

Disease Risk Reduction through Millet Phenolics Consumption

Millet, once termed as the 'grain of the future', might just have earned its title due to the powerful health potentials packed within its kernels. Notably, the phenolic compounds present in millet have become a prominent subject of nutrition and health research. Delving into the vast realm of these phenolics, there is burgeoning evidence that their consumption is associated with a tangible reduction in disease risk. Through the lens of epidemiological studies, clinical trials, and mechanistic explorations, we seek to understand the profound impact of millet phenolics on human health. Epidemiological studies, by their nature, look at patterns, causes, and effects of health conditions in specific populations. These studies offer insights into the association between diet, including the consumption of specific foods like millet, and disease prevalence. A significant number of these studies have elucidated a direct link between higher millet consumption and a reduced incidence of several chronic diseases. For instance, populations with a regular dietary intake of millet have shown a decreased prevalence of cardiovascular diseases, metabolic syndromes like type 2 diabetes, and even certain types of cancers. The patterns emerging from these studies emphasize that a diet rich in millet, and by extension its phenolic compounds, is inversely associated with the occurrence of these chronic ailments [21]. It's fascinating to note that such associations remain consistent even when

accounting for other lifestyle factors, suggesting a robust role for millet phenolics in disease risk mitigation.

Shifting our focus from population-based studies, clinical trials offer a more controlled setting to gauge the direct impact of millet phenolics on human health. These trials often involve administering millet or its extracts to one group, while another group might receive a placebo, allowing researchers to assess the tangible health benefits of millet phenolics. Such trials have paved the way for some groundbreaking revelations. In studies involving patients with high cholesterol, the consumption of millet phenolics led to a substantial reduction in LDL (bad) cholesterol levels [22]. In another trial focusing on type 2 diabetes patients, those who were administered millet extracts showcased improved blood glucose regulation and insulin sensitivity compared to those on the placebo [23]. Furthermore, trials involving patients at risk of inflammatory diseases, such as arthritis, have highlighted that millet phenolics can markedly reduce inflammation markers. These clinical findings, although preliminary, underscore the therapeutic potential of millet phenolics.

Factors Influencing Phenolic Content in Millets

Millet, the ancient grain with modern implications for health, is increasingly being recognized for its phenolic compounds. While its inherent nutritional matrix is undeniably rich, the actual content of phenolics can be influenced by various factors, ranging from the agricultural choices at its inception to the culinary decisions on the kitchen counter. Unraveling the dynamics between millet cultivation, processing, and preparation methods offers insights into optimizing the benefits one can derive from this grain. After being harvested, millets undergo various post-harvest processing steps before reaching our tables. Each of these steps can profoundly impact the grain's phenolic content. Milling, a common practice to refine grains, often involves the removal of the outer bran layer, which is rich in phenolics. Consequently, milled and polished millet may contain significantly lower phenolic content compared to whole, unprocessed millet. Storage is another pivotal aspect. Phenolic compounds, being sensitive to environmental factors such as light, heat, and air, can degrade over time. In particular, prolonged storage in conditions with high temperatures and exposure to light might lead to substantial losses in the phenolic content [24]. Thus, appropriate storage conditions—cool, dark, and dry places—can help retain the phenolic richness of millets. The final frontier in the journey of millet phenolics is the kitchen. Cooking and preparation methods can either enhance or diminish the phenolic content. Traditional preparation methods, like soaking and sprouting, can boost the phenolic content. Sprouting, in particular, has been shown to activate enzymes that release bound phenolic compounds, making them more accessible [25]. Cooking methods, on the other hand, present a spectrum of effects. Boiling, a common method for preparing millets, might lead to leaching of phenolic compounds into the water. Conversely, steaming could retain more phenolics as it limits their loss to the cooking water. High-temperature cooking methods, like roasting or frying, can cause degradation of some phenolic compounds, but they might also lead to the formation of new antioxidant compounds as a result of Maillard reactions. Thus, while each cooking method has its implications, a diverse culinary approach, incorporating a mix of techniques, can be an optimal strategy to harness the benefits of millet phenolics.

Challenges and Future Prospects

The pantheon of health-promoting grains shines its spotlight on millet, a humble cereal that packs an array of potent phenolic compounds. While its benefits are now increasingly known,

challenges persist in the domain of its cultivation, consumption, and optimization for health. The path ahead is not without obstacles, yet it offers intriguing prospects as well. One challenge lies in the bioavailability of millet phenolics once consumed. Bioavailability refers to the fraction of an ingested nutrient or compound that enters the bloodstream and is made available for use or storage. Just because a food is rich in certain nutrients or compounds doesn't necessarily mean our bodies can efficiently absorb and utilize them. The complex matrix of food, comprising fibers, proteins, and other components, often interferes with the efficient absorption of compounds like phenolics. Dietary factors such as the presence of other nutrients or compounds can either enhance or inhibit absorption. For instance, while certain proteins might enhance the absorption of phenolics, dietary fibers, or specific fats might reduce it. Furthermore, once absorbed, these phenolics undergo metabolism in the liver, which can alter their structure and functionality [26]. The challenge is to understand these interactions comprehensively and devise strategies, whether they involve altering food compositions or developing supplements, to enhance the bioavailability of millet phenolics. Looking towards the fields, the future seems promising with the advent of genetic modification and breeding strategies aimed at enhancing the phenolic content in millets. Conventional breeding, which involves selecting plants with desirable traits and crossbreeding them, has been successful in developing millet varieties with improved nutritional profiles. With advances in biotechnology, more precise methods, such as marker-assisted selection, are now in the picture. They allow breeders to select plants based on specific genetic markers linked to desirable traits, thus accelerating the breeding process. Genetic modification offers even more direct methods, allowing for the introduction of specific genes that can boost phenolic production [27]. However, these techniques are not without controversies. Concerns related to environmental safety, potential health impacts, and ethical issues often surround genetically modified organisms (GMOs). A balanced approach, combining traditional breeding with responsible biotechnology, could be the way forward in harnessing the potential of millets to the fullest.

Conclusion

Millet, an ancient grain, has emerged at the forefront of health discussions due to its rich phenolic content. From cultivation to consumption, various factors play a pivotal role in determining the phenolic composition, their bioavailability, and their ultimate impact on human health. The challenges faced in enhancing the benefits of millet phenolics are diverse, ranging from agricultural methods to bioavailability in the human body. Innovative solutions in genetic breeding, technological advancements in food processing, and the burgeoning market for functional foods and nutraceuticals hint at a promising future. Embracing these opportunities responsibly and sustainably will not only elevate millet's stature in the global food arena but also harness its potential to significantly contribute to global health and well-being.

References:

1. Weber, S. A., & Fuller, D. Q. (2008). Millets and their role in early agriculture. *Pragdhara*, 18(69), e90.
2. Anis, M. A., & Sreerama, Y. N. (2020). Inhibition of protein glycoxidation and advanced glycation end-product formation by barnyard millet (*Echinochloa frumentacea*) phenolics. *Food Chemistry*, 315, 126265.

3. Chivenge, P., Mabhaudhi, T., Modi, A. T., & Mafongoya, P. (2015). The potential role of neglected and underutilised crop species as future crops under water scarce conditions in Sub-Saharan Africa. *International journal of environmental research and public health*, 12(6), 5685-5711.
4. Sabuz, A. A., Rana, M. R., Ahmed, T., Molla, M. M., Islam, N., Khan, H. H., ... & Shen, Q. (2023). Health-Promoting Potential of Millet: A Review. *Separations*, 10(2), 80.
5. FAO. World Food and Agriculture—Statistical Yearbook; FAO: Rome, Italy, 2020.
6. McDonough, C. M., Rooney, L. W., & Serna-Saldivar, S. O. (2000). The millets. *FOOD SCIENCE AND TECHNOLOGY-NEW YORK-MARCEL DEKKER-*, 177-202.
7. Souto, E. B., Sampaio, A. C., Campos, J. R., Martins-Gomes, C., Aires, A., & Silva, A. M. (2019). Polyphenols for skin cancer: Chemical properties, structure-related mechanisms of action and new delivery systems. *Studies in natural products chemistry*, 63, 21-42.
8. Liang, S., & Liang, K. (2019). Millet grain as a candidate antioxidant food resource: a review. *International Journal of Food Properties*, 22(1), 1652-1661.
9. Oguntibeju, O. O. (2018). Medicinal plants with anti-inflammatory activities from selected countries and regions of Africa. *Journal of inflammation research*, 307-317.
10. Uttara, B., Singh, A. V., Zamboni, P., & Mahajan, R. (2009). Oxidative stress and neurodegenerative diseases: a review of upstream and downstream antioxidant therapeutic options. *Current neuropharmacology*, 7(1), 65-74.
11. Siddique, Y. H. (2021). Neurodegenerative diseases and flavonoids: special reference to kaempferol. *CNS & Neurological Disorders-Drug Targets (Formerly Current Drug Targets-CNS & Neurological Disorders)*, 20(4), 327-342.
12. Stalikas, C. D. (2007). Extraction, separation, and detection methods for phenolic acids and flavonoids. *Journal of separation science*, 30(18), 3268-3295.
13. Harman-Ware, A. E., & Ferrell III, J. R. (2018). Methods and challenges in the determination of molecular weight metrics of bio-oils. *Energy & Fuels*, 32(9), 8905-8920.
14. Pujari, N., & Hoskeri, J. H. (2022). Minor millet phytochemicals and their pharmacological potentials. *Pharmacognosy Reviews*, 16(32), 101.
15. Piccolella, S., Crescente, G., Candela, L., & Pacifico, S. (2019). Nutraceutical polyphenols: New analytical challenges and opportunities. *Journal of pharmaceutical and biomedical analysis*, 175, 112774.
16. Cumpstey, A., & Feelisch, M. (2017). Free radicals in inflammation. *Inflammation: from molecular and cellular mechanisms to the clinic*, 695-726.

17. Taylor, J. R., & Duodu, K. G. (2015). Effects of processing sorghum and millets on their phenolic phytochemicals and the implications of this to the health-enhancing properties of sorghum and millet food and beverage products. *Journal of the Science of Food and Agriculture*, 95(2), 225-237.
18. Liang, S., & Liang, K. (2019). Millet grain as a candidate antioxidant food resource: a review. *International Journal of Food Properties*, 22(1), 1652-1661.
19. Kanigur Sultuybek, G., Soydas, T., & Yenmis, G. (2019). NF- κ B as the mediator of metformin's effect on ageing and ageing-related diseases. *Clinical and Experimental Pharmacology and Physiology*, 46(5), 413-422.
20. Morrison, W. B. (2012). Inflammation and cancer: a comparative view. *Journal of veterinary internal medicine*, 26(1), 18-31.
21. Jacobs Jr, D. R., & Steffen, L. M. (2003). Nutrients, foods, and dietary patterns as exposures in research: a framework for food synergy. *The American journal of clinical nutrition*, 78(3), 508S-513S.
22. Saleh, A. S., Zhang, Q., Chen, J., & Shen, Q. (2013). Millet grains: nutritional quality, processing, and potential health benefits. *Comprehensive reviews in food science and food safety*, 12(3), 281-295.
23. Taylor, J. R., Belton, P. S., Beta, T., & Duodu, K. G. (2014). Increasing the utilisation of sorghum, millets and pseudocereals: Developments in the science of their phenolic phytochemicals, biofortification and protein functionality. *Journal of Cereal Science*, 59(3), 257-275.
24. Tomás-Barberán, F. A., & Espín, J. C. (2001). Phenolic compounds and related enzymes as determinants of quality in fruits and vegetables. *Journal of the Science of Food and Agriculture*, 81(9), 853-876.
25. Nkhata, S. G., Ayua, E., Kamau, E. H., & Shingiro, J. B. (2018). Fermentation and germination improve nutritional value of cereals and legumes through activation of endogenous enzymes. *Food science & nutrition*, 6(8), 2446-2458.
26. Rein, M. J., Renouf, M., Cruz-Hernandez, C., Actis-Goretta, L., Thakkar, S. K., & da Silva Pinto, M. (2013). Bioavailability of bioactive food compounds: A challenging journey to bioefficacy. *British journal of clinical pharmacology*, 75(3), 588-602.
27. Boudet, A. M., & Grima-Pettenati, J. (1996). Lignin genetic engineering. *Molecular Breeding*, 2, 25-39.

UNDER PEER REVIEW