



Mustard Seeds and Leaves: Exploring Nutritional Benefits and Therapeutic Applications

Tanzeela Shahzadi¹, Maria Noor^{1*}, Marwa Waheed², Muhammad Bilal Hussain¹, Azha Fatima¹, Mariam Islam¹, Maha Atiq¹, Aliza Naeem¹, Sawera Asif¹, Haris Khan¹ and Abid Mehmood³

¹ Department of Food Science, Government College University Faisalabad, Pakistan

² Department of Food Science and Technology, Riphah International University, Faisalabad, Pakistan

³ Department of Agronomy, The University of Haripur, Pakistan

* **Correspondence:** Maria Noor, maria.malik.noor@gmail.com

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A multipurpose crop from the Brassicaceae family, mustard has been grown since 3000 BCE and has industrial, culinary, and medicinal uses. Due to their rich lipid, phenolic, and glucosinolate content, key species, white (*Sinapis alba*), black (*Brassica nigra*), and brown (*Brassica juncea*) offer special advantages. These substances have antibacterial, anti-inflammatory, and antioxidant qualities that support heart health, diabetes control, and cancer prevention. Concerns about the erucic acid content in Brassica oil, which is used extensively in culinary and medicine, have led to attempts to create safer, lower-erucic-acid varieties. Mustard is a natural preservative due to its antibacterial qualities, especially those derived from allyl isothiocyanate, and its usage in food preservation and functional foods is further enhanced by phenolic compounds like sinapine. Meals made from mustard seeds have additional uses as soil improvers and biopesticides. Despite its potential, issues including toxicity and adulteration call for additional research and quality controls. The numerous uses of mustard in industry, sustainable agriculture, and health highlight its significance and call for further development and regulation to ensure its safer and wider usage.

KeyWords: Bio-pesticides, Erucic acid, Phenolic compounds, Sustainable agriculture,

Introduction:

Mustard has been revered for millennia as one of the most widely cultivated and versatile plants in the world, and it stands among the oldest known condiments in human history. Historical evidence suggests that mustard plants were first cultivated around 3000 B.C. [1]. This plant is related to the family Brassica and sub-family Cruciferae or Brassicaceae is the mustard seed (*Brassica nigra*). The important stage in crop development is planting the seed, and typical hands-on methods are time-consuming, ineffective and labor-intensive [2]. Almost all of the most widely grown types in this family are members of the six species which also includes 2 of the 3 most significant species of mustard [3]. The various benefits of mechanized sowing include reduced cultivation costs, improved precision, faster operations, and more efficient harvesting [4][5]. However, conventional planters often do not have the elements required to satisfy the particular requirements of sowing mustard seeds. Mustard ranks as the third-largest oil-producing crop in the world, following palm and soybean, and is widely cultivated for its valuable oil content. Around 38 million metric tons of mustard are yielded all over the world, and overall 12–14 million metric tons of oil are produced [6].

The genus *Sinapis* L. encompasses a variety of mustard species, notably including Charlock mustard and White mustard. The Brassicaceae family, which includes three genera, is primarily used for commercial purposes and mustard condiments. The cultivated variety, *S. Alba* L., is similar to *Brassica* species and has larger, white mustard seeds. Mustard is primarily cultivated for its seeds, which are valued for their high oil content, and it is also commonly used as a spice, particularly in warmer regions. It is planted as a green food crop, catch crop or green manure in cold climates. The market offers several kinds of mustard fusions created from white mustard. In this case, white seeds are used to make American ballpark mustard, which is then combined with spices and vinegar and typically colored with turmeric.

White mustard kernels have a unique glucosinolate sinalbin, which is degraded by enzymes to 4-hydroxy benzyl isothiocyanate, causing the mustard's strong flavor and unpleasant smell. However, it has a softer flavor compared to other mustard species. White mustard seeds are commonly used in pickle gherkins [3]. Several varieties of yellow mustard exhibit distinct characteristics, such as early maturation, resistance to nematodes, low erucic acid content, or modified glucosinolate levels. More than 100 variant of *S. Alba* L. are included in the collection of agricultural plant species varieties [7]. Black mustard, also known as *Brassica nigra*, is a type of mustard with blackish dark-red seeds that are slightly bitter and sharp-tasting. The main glucosinolate found in mustard seeds, sinigrin, can hydrolyze into allyl-isothiocyanate (AITC), which is responsible for the seeds' strong, pungent aroma [8]. Mustard oil is a widely used as organic oil in South Asia, Russia, and China, used to enhance food flavor and nutritional content. It is extracted from the seeds of *Brassica* species, including brown, black, and white mustard. The oil contains protein, fixed oils, and carbohydrates, with a small proportion of water, volatile oil, fiber, mineral, and phytin. It can be used internally and externally for medicinal purposes and culinary purposes [9]. Mustard seeds contain antioxidant compounds like phenolic composites like ferulic acid, synaptic acid, and 3,4-dihydroxybenzoic acid. These compounds can be extracted using a water and acetone solution to prevent food spoilage. Ultrasound extraction can minimize temperature and time required for effective extraction [10]. Since the beginning of time mustard kernels have been used for cooking purposes [11]. Mustard seeds can be used as a cause of bioactive compounds in foodstuff, because of the presence of antioxidants and glucosinolates, along with them distinguish flavor. Scientists are exploring the use of ground mustard in meat-based food storage to delay lipid oxidation and inhibit microorganism growth. White mustard has been found to be more effective in sensory assessment compared to brown and black mustard, indicating that mustard can be a more effective alternative for meat-based products [12].

Objectives:

This research aims to:

- Explore the nutritional composition and bioactive compounds of mustard seeds and leaves;
- Evaluate the therapeutic and functional applications of mustard in food and medicine;
- Discuss the challenges and future perspectives of mustard utilization in sustainable agriculture, health, and industry.

Due to the presence of physiologically progressive chemicals in the plants and their seeds, mustard is once thought of as a medical plant rather than a savory one. These so-called "bioactive compounds" and generally found in a variety of natural sources and are distinguished by particular functions. These phytochemicals have been studied in various commercial areas, leading to increased nutritional innovations and product improvements. Technological advancements have led to the production of food yields with potential medicinal and fitness benefits, resulting in a variety of naturally active substances in various

forms, such as nutritional supplements, for various health goals. However, these substances often lack a solid scientific base for their safety or efficacy [13][14].

Types of Mustard:

White Mustard:

Sinapis Alba L., a Mediterranean weed and cultivated plant species, is a popular mustard in Europe and North America for food handling and condiment industries. It contains palatable oilseeds, fast-growing salads, and can remove heavy metals from soil, making it a valuable addition to various agricultural uses. The leaves of young seedlings, which are high in vitamins A, C, and E, can be used medicinally to purify blood and can be eaten as fresh, delicious salad leaves [15][11]. White mustard kernels have important agronomic utility due to its extraordinary protein and oil content and little starch level. The seed is a nutritious protein source with a balanced amino acid configuration, used in mayonnaise, hot dog mustard, and salad dressings. It is also used as an obligatory agent and protein extender in meat handling. Kernels are also used as food preservers with strong disinfecting properties [16]. White mustard seed oil, with its potent antimicrobial properties, is valuable for food preservation, industrial use as lard and illumination, and traditional medicine due to its antiviral, anticancer, and analgesic properties. It also aids in digestive and respiratory disorders. Recently, it has gained attention as a potential biodiesel feedstock [17]. The oil that one can be used as a substitute fuel, according to researchers. The financial benefits can also be increased by using oil meal, a consequence of the biodiesel business generated from white mustard seeds, as animal feed or by further processing it to produce more oil [15].

Black mustard:

Brassica nigra L., also well-known as black mustard, is a supporter of the Brassicaceae family and the species *Brassica*. It grows normally as a yearly without any requirement for special growth circumstances. 532,769 tons of mustard seeds were produced worldwide in 2021. In 2021, Nepal accounted for more than 41.3% of the world's mustard seed output (220,250 tonnes), surveyed by Russia and Canada these three nations held a 79.8% share, while the ten largest nations held a 99.9% share. In 2021, Serbia produced 2432 tonnes of mustard seeds, placing it eighth in the world. The United States, France and Germany were the world's top traders of mustard seeds in recent years. Although black mustard seeds (BMSs) contain a high percentage of oil (more than 25%), the oil's nutritional value is limited because of the high levels of branched, long chain fatty acids and erucic acid (C22:1). However, when burning, biodiesel made from BMS oil (BMSO) releases fewer nitrogen oxides and hydrocarbons and has superior lubrication [18]. The Mediterranean and the Middle East are the two regions from where black mustard originates. It is grown throughout North and South America, China, Siberia, western India, Asia Minor, North Africa, and Europe.

Brown mustard:

The mature, dried kernels of the cruciferous plant, known as brown mustard seeds, are utilized in both cooking and medicine [19]. With a growing season spanning 70 to 120 days, brown mustard also referred to as Chinese, Indian, oriental, leaf, and vegetable mustard has its origins in India and the Middle East. Like the others, this cultivar is also more resilient to environmental stressors like high temperatures, late sowing, and little water than it is to rape. Additionally, it produces more dry matter than rapeseed.

Wild mustard:

Wild mustard, correspondingly recognized as field or charlock mustard, is a plant that grows widely over the world and is formerly used to create mustard and oil. Because of its competitive once a year growth habit, great fecundity, and extremely extensive seed germination period, it is currently one of the most troublesome weeds. Additionally, wild mustard naturally resists several pesticides [10]. The annual cruciferous (Brassicaceae) weed known as wild mustard is commonly found in Europe and further parts of the world. It has a

tenacious seed bank, modest development lifestyles, and high fertility [20]. Depending on the environment, it can grow anywhere from a few centimeters to about 80 cm in height. Its blooms can range in number from a few to several hundred per person (400-500). It develops in disturbed and nitrified environments, and its competition with winter cereals has been studied. The existence of wild mustard decreased the total nitrogen levels in wheat and triticale by 20% and 19%, respectively, but not of barley. Didon and Bostrom discovered that unmanaged wild mustard on barley caused crop damages extending from 47 to 70% after many years of appropriate management using POST herbicides. Thus, good weed management results in reduced tidy rivalry and yield loss due to wildflower occurrence [21]. Wild mustard species regularly infest wheat fields, causing severe crop losses if not effectively managed [22].

Ethiopian or Abyssinian mustard:

According to [23], around 4,000 years ago, *B. carinata* is first cultivated in Ethiopia, which is also the location of the genetic diversity of this species. Information on genetic diversity is necessary in order to use the available *B. carinata* material for various breeding objectives. Only in Ethiopia is the production of *B. carinata* seed significant. Its leaves are used as oil crops and veggies. It can be utilized for a variety of purposes, including boosting farmers' income and establishing environmentally friendly businesses. It is grown as a green vegetable in the gardens of farmers in the country's highlands. Mostly grown in East and Southern Africa, this green vegetable is less frequent in West and Central Africa. In the highlands of Ethiopia, this oilseed crop is commonly grown. Ethiopian mustard plants' leaves are low in bitterness and high in beta-carotene, vitamin C and K, and antioxidants that prevent cancer. An essential oleic plant with African origins is Ethiopian or Abyssinian mustard. It takes 150–180 days for its seeds to mature. The leaves and seeds are both utilized as food. In addition to select regions of Canada, Spain, Australia, France, China, and India, it is grown in northeastern Africa. Because of its fatty acid content and stability during oil storage, it is resilient to both biotic and abiotic anxiety and may be utilized in the generation of biofuels [24].

Table 1. Comparison of verities in terms of its cultivation of mustard.

Parameters	Black mustard	White mustard	Wild mustard 5	Brown mustard 4	Ethiopian mustard
Vegetation time	40–100 days	80–125 days	70–84 days	70–120 days	150–180 days
Drought resistant	No Information	Yes	Yes	Intermediate	No
Prevents the spread of nematode	Yes	Yes	Yes	Yes	Yes
Good aftercrop	Yes	Yes	No Information	Yes	No Information
Impact of agroclimatic conditions	No	No	No Information	No	No

Nutritional Composition:

Lipid:

Mustard seeds, with a high fat content ranging from 23% to 47%, are rarely used in edible oil due to their controversial fatty acid outline. The oil contains PUFAs (20-28%), SFAs (5-7%), and a significant amount of erucic acid (66-74%), making it a controversial choice in the edible oil industry [25]. In actuality, the quantity of erucic acid in mustard germ oil may range from 26.5% to 36.5% of the overall volume of fatty acids. The fatty acid shape of the

other mustard seed varies by species and includes linoleic acid (9%–15%) and oleic acid (19.5%–22%) [10]. The connection between the development of heart injury and erucic acid is the main cause for concern. It is brought to light by researchers, they gave male and female Sprague-Dawley pests refined oil containing three different absorptions of erucic acid (1.6%, 4.3%, and 22.3%) for a period of 112 days. Although the rates varied by gender, myocardial lipidosis, focal myocardial necrosis, and fibrosis remained shown to be proportionate to the erucic acid levels at the end of the experiment. Additional research by researchers also revealed comparable outcomes. As a result, the FDA disqualified mustard oil for use in cooking. In contrast, erucic acid consumption is restricted by Australian and New Zealand dietary standards to 2% to 5% of energy intake, or 7.5 mg/kg body weight per day. A reduced daily consumption of 7 mg/kg body weight/day is controlled by the European Food Care Authority.

Much work is done to create low erucic acid mustard strains so that mustard seed may be used in meals. Research carried out in Canada by [25]. Created a mustard with nil erucic acid by interspecifically crossing brown-seeded *B. carinata* S-67, yellow-seeded *Dodolla*, and *B. juncea* line Zem 2330. With a high percentage of oleic acid ($28.3 \pm 2.0\%$), linolenic acid ($22.9 \pm 2.4\%$), linoleic acid ($38.1 \pm 2.9\%$), and a very small level of erucic acid ($0.1 \pm 0.0\%$), the new seeds' fatty acid profile appeared encouraging. However, the paucity of scientific studies on the long-term effects of genetically engineered products makes them controversial. An effective technique for lowering the erucic acid in mustard species is the introduction of preheat treatment, which included microwave and roasting.

Glucosinolates:

Mustard seeds and other plants in the Brassicaceae family contain a class of secondary metabolites called glucosinolates [10][26]. Aliphatic, aromatic, and indole are the three distinct forms of glucosinolates that are structurally classified according to the variations in their side sequence R [26]. Only a tenth of the more than 100 distinct glucosinolates known to exist are found in plants in the Brassicaceae family, and even fewer are found in mustard seeds. However, glucosinolates continue to be crucial precursors in mustard species, influencing the formation of their odor and flavor. Although glucosinolates have no taste by themselves, thiocyanates, isothiocyanates, nitriles, and other byproducts of glucosinolate degradation give them their flavors and odors. Three primary mechanisms underlie these degradation processes: enzymatic, thermal, and chemical [25]. Despite their resistance to warmth, glucosinolates can occasionally degrade chemically and cause health issues. Enzymatic hydrolysis mediated by myrosinase is the most efficient method for breaking down glucosinolates. However, for this reaction to occur, the cell wall of the mustard seed must be disrupted, typically through crushing, cutting, or chewing to allow the enzyme to act. Mustard naturally contains myrosinase, though its concentration varies among different species. For instance, Oriental mustard has the maximum activity (2.75 units/mL), followed by black mustard (1.50 units/mL), and yellow mustard (0.63 units/mL). When the temperature hits 60°C, the myrosinase enzyme, which is thermally sensitive, could be easily deactivated. However, when pressure is added, the thermal threshold may rise. [25] reported that inactivation occurred at 300 MPa and 70°C. They also pointed out that black and oriental mustards are more resilient to heat and pressure than yellow mustard in terms of myrosinase.

Isothiocyanates, thiocyanates, epithionitriles, nitriles, and indoles are the foodstuffs of the hydrolysis of glucosinolates by myrosinase, which typically yields uneven aglycones and glucose [10]. Researchers found that sinigrin can be converted into allyl isothiocyanate (AITC) at a neutral pH, producing a strong taste and smell similar to crushed mustard seeds. AITC is also potent antibacterial, inhibiting mold, bacteria, and yeast development in both liquid and gas phases, according to a study by author2024. This substance gives yellow mustard its spicity

mouthfeel and is said to possess bacteriostatic and bactericidal qualities against gram-negative bacteria, such as *Schizosaccharomyces pombe* and *Salmonella enteritidis* [27].

Table 2. Magnesium content of Brassicaceae mustard leaves (per 100 g edible amount)

Vitamin	Mustard Species (Amount per 100 g)	References
Thiamine	<i>B. napuss</i> (0.8 mg)	[28]
Riboflavin	<i>B. napuss</i> (0.3 mg)	
Niacin	<i>B. napuss</i> (8.1 mg)	
Pyridoxine	<i>B. napuss</i> (1.9 mg)	
Vitamin C/Ascorbic acid	Vitamin C content is higher in <i>Alliaria petiolata</i> (261 mg) than oranges, <i>B. nigra</i> , <i>B. juncea</i> (72–89 mg), <i>Erysimum repandum</i> , and <i>S. officinale</i> .	
Tocopherols and vitamin E (α -, β -, γ -, and δ -tocopherols; γ tocopherol being the most common)	Canola (<i>B. napus</i>) seed oil with <i>Sinapis alba</i> : α and γ tocopherols (12 and 21.3 mg, respectively)	
Vitamin K	<i>B. juncea</i> and all other rapeseeds (0.3 mg). Oil from <i>B. napus</i> (canola) seeds (70–150 μ g)	[29]

Phenolic compounds:

Endogenous phenolic compounds found in flowers and oil-rich seed have concerned a lot of consideration in the past 20 years because of their potential for medicinal use and capacity to support functional food items. One fragrant loop with one or more hydroxyl clusters is typically seen in phenolic substances [25]. These classifications aid in the separation of phenolic chemicals into modest flavonoids and non-flavonoids [25]. Then, non-flavonoids are further divided into modest phenolics, phenolic acid and byproducts, phenylacetic acid and byproducts, tannins, phenones, and stilbenes based on their carbon skeleton. One of the most significant secondary metabolites of plants is phenolic acids and their derivatives. They are found in leaves, roots, seed coats, and seeds that have defenses against insects and the environment [30]. Phenolic acids are classified as either benzoic acid derivatives or cinnamic acid consequences, depending on how the carboxyl group and benzene ring interact [25]. However, ferulic acid, p-coumaric acid, caffeic acid, and sinapic acid are the derivatives of cinnamic acid [31]. Mustard's total phenolic concentration ranges since 2.62 to 36.5 mg/g dry weight, making it a prospective source of phenolic compounds. Sinapine, the primary phenolic component found in mustard, can be found both free and bonded in soluble forms (as conjugates or esters). The following phenolic compounds were found in mustard seed extract: syringaldehyde, 4-vinylsyringol (canolol), sinapine, sinapic acid, and sinapoyl esters.

Sinapine:

With trace levels of sinapic acid, it makes up roughly 70% to 90% of the seed's total phenolic components. The recovery and yield of sinapine from various mustard products have been reported in a number of investigations. One of the first investigations of yellow mustard's sinapine is published by [25]. Yellow mustard has the greatest concentration of sinapine, at 33 μ mol/g plant material. Sinapine is found in mustard meal extracts at about 62% (w/w) by high-performance liquid chromatography analysis, while sinapic acid is found at 16% (w/w). A recent investigation by [32] found that the greatest sinapine concentration of 8.8 ± 0.1 mg/g BDM (bran dry matter) is obtained by removing the mustard bran seed coat with ethanol at 75C for 30 minutes [25]. However, after air-frying the Oriental mustard cake for 15 minutes at 180 degrees Celsius, the oil fraction had the greatest sinapine level, measuring 117.61 ± 7.13 μ g/100 g of roasted cake. Additionally, the lipophilic portion of air-fried Oriental mustard

powder and lesser grade mustard seed had sinapine values of $62.5 \pm 2.0 \mu\text{g}/100 \text{ g}$ and $415.64 \pm 26.63 \mu\text{g}/100 \text{ g}$, respectively.

Sinapic acid and its derivatives:

More than 73% of phenoplast acids are made up of sinapic acid, a certainly happening p-Hydroxycinnamic acid, and its byproducts. According to [25], the distribution of sinapic acid in mustard seeds varies across different forms: 7%–32% exists in free form, 65%–86% in esterified form, and 71%–97% in bound or conjugated forms that are not easily extractable. In plants, sinapic acid is a component of the phenylpropanoid and shikimic acid pathways, the latter of which is where it forms. However, syringaldehyde and malonic acid can be artificially condensed via the Knoevenagel–Doebner condensation process to produce the Hydroxycinnamic acid derivatives [33][34]. Due to its single hydroxyl group, this phenolic molecule is the main water-soluble antioxidant found in mustard dishes [25].

Stability:

Like many phenolic compounds, sinapic acid and its derivatives are sensitive to heat. As stated by researchers, after the seeds were heated to 180 degrees Celsius for ten minutes, a decrease in the amount of FSA and TSAH is seen. The Oriental variety's FSA and TSAH dropped, distinctly, prior to and following preheat treatment. A more noteworthy pattern is noted by [25]. For mustard seeds, both yellow and black. The conversion of sinapic acid to canolol occurs after mustard seeds are heated, even though this process greatly lowers the concentration of sinapic acid.

Canolol:

After decarboxylation, sinapic acid is converted to canolol, often referred to as 4-vinylsyringol, by losing carbon dioxide [35]. Canolol is made from sinapine and sinapic acid. Canolol is an unstable substance that can form dimers, trimers, and oligomers when oxygen is present [25][36][37][38]. Furthermore, canolol can form complexes with proteins and polysaccharides due to its polarity. Canolol must be released from these matrices under extremely high pressure and temperature conditions. [39][36]. The impact of warmth and compression action on the extractability of the main sinapates from mustard seeds is studied by earlier researchers. The greatest recovery of canolol is achieved at 160C for 10 minutes using a new commercial RapidOxy 100 system in a solvent-free dry heat pre-treatment approach. This technique improved the yield of canolol from mustard seeds by establishing the ideal temperature and pressure for its production [37][40] increased the amount of canola and other lipophilic sinapates that were mined from mustard testers by using air frying as a roasting pre-treatment procedure. By lowering the removal solvent's surface tension and viscosity, applying air circulation at a certain pressure and temperature enhanced the phenolics' solubility and mass transfer [37].

Therapeutic Properties:

Goiterogenic Effect:

Brassica vegetables contain thioglycosides, which are converted to thiocyanates during metabolism. These elements block the transfer of iodine and stop it from being incorporated into thyroglobulin. Consequently, TSH secretion and thyroid cell development are increased. Thyroid carcinomas were thus found to be caused by these chemicals. A study conducted in Malaysia found a strong correlation between ingesting of brassica root vegetable and thyroid cancer in 293 women with the disease. Thyroid cancer is found to be more common in a group of Malaysian women with mild iodine deficit who also consumed a lot of brassica vegetables [41].

Antioxidant effect:

Antioxidant chemicals and a higher vitamin content found in brassica veggies shield us from a variety of dangerous illnesses. Brassica vegetable isothiocyanates catalyse the combination of glutathione into a substrate for glutathione S-transferase (GST). A series of stage II enzymes, commonly referred to as environmental toxins, aids in the detoxification of oxidative stress products and carcinogens. Stage II detoxification response starts raises nuclear factor Mitogen-activated protein kinase can be triggered by erythroid-derived 2-like 2(Nrf2)-kelch-like ECH-associated 1(keap1) interaction and degeneration. A cytoplasmic protein called keep 1 is required to regulate Nrf2 activity. In summary, Nrf2 regulates gene appearance through the antioxidant retort element (ARE) [41].

Anti-inflammatory Effects:

Irritation grows cellular propagation, prevents apoptosis, and upsurges the threat of emerging cancer. By stimulating the immune system, removing free radicals, and activating detoxification enzymes, bioactive compounds in Brassica plants help reduce inflammation. Numerous inflammatory processes are inhibited by isothiocyanates. Sulphoraphane has been shown to have anti-cancer properties, including lowering white blood cell secretion of inflammatory signaling molecules and lowering nuclear-powered factor kappa B (NF- κ B), a transcript feature that promotes inflammation, from binding to DNA. One of the Brassica vegetables, cabbage has anti-inflammatory and antibacterial qualities that make it a popular choice in traditional medicine for treating mastitis, tiny cuts, and wounds as well as gastrointestinal ailments [41].

Cancer Protective Effects:

The World Cancer Study Deposit claims that eating a lot of Brassica vegetables protects the body against thyroid, colon, and rectal cancers. Additionally, Brassica vegetables typically show preventive effects against cancer in other places when ingested in large quantities as part of a diet that also includes other vegetables. The chemicals that come from the hydrolysis are also responsible for this impact. Vegetable components of Brassica have the capacity to alter the expression of biotransformation enzymes. Its operations also contribute significantly to the prevention of cancer. Detoxify substances that can damage DNA by regulating their mutagenic, toxic, and neoplastic properties [41]. Oxidation, reduction, and hydrolysis processes are examples of phase I enzymes that increase the hydrophilicity of the molecules and make them suitable for detoxification. These enzymes may therefore be transformed into procarcinogens. Through conjugation or other metabolic pathways, stage II enzymes catalyze a variety of events that offer wide defense alongside electrophiles and oxidants, transforming responsive molecules into easily removed steady metabolites. Biotransformation enzymes can metabolize endogenous substances like steroid hormones in addition to their function in the metabolism of carcinogens. Therefore, it can circuitously influence the development and spread of premalignant and malignant tissues via changing hormone exposure. The beginning and development of carcinogenesis are impacted by sulforaphane and a number of isothiocyanates derived from glucosinolate breakdown products. Although no clinical research has assessed any therapeutically efficacious quantities, eating three or five rations of broccoli or cauliflower per week appears to prevent cancer. It is discovered that humans can tolerate 100 mg of glucoraphanin or 10 mg of pure sulforaphane daily [41].

Liver Cancer:

One of the foremost reasons for cancer-related deaths globally, liver cancer ranks fifth among cancers in men and ninth among cancers in women. Metastasis has been associated with a poor finding in people [42]. According to estimates, the disease's incidence will rise by 50% in 2020 [41]. Fighting infections, changing one's food, and changing one's lifestyle can all help avoid this kind of cancer [42]. Through a p53-dependent mechanism, sinigrin, one of the primary glucosinolate mechanisms originating in *B. nigra* and additional Brassicaceae seeds,

has been exposed to suggestively slow the growth of liver tumor cells. Another study showed that 4-methylthiobutyl isothiocyanate, even though normal liver cells did not exhibit necrosis or apoptosis when exposed to it [41].

In another study, mice fed broccoli showed low scores for non-alcoholic fatty liver disease and hepatic lipid. Meanwhile, it is shown that the formation and spread of hepatic neoplasm is stopped, hepatic macrophage activation is blocked, and liver damage is reduced [42]. According to a different study, when rats were given extracts of kale and cabbage during the beginning and developing stages of the hepatocarcinogenesis model, it inhibited ex vivo-induced DNA damage. Furthermore, it is shown that the antioxidant belongings of volatile fatty acids take out from white cabbage excerpt had a hepatoprotective effect [43].

In a case-control study involving 217 liver cancer cases in Shanghai, researchers investigated the relationship between urinary levels of isothiocyanates and the risk of developing liver cancer. While the overall findings were not statistically significant, most of the stratified analyses indicated inverse correlations, suggesting a potential protective role of isothiocyanates.

It has been demonstrated that 1-methoxy-3 indolylmethyl alcohol, a metabolite of neoglucobrassin that is generated from the hydrolytic yields of glucosinolates, forms DNA adducts in the liver of rats and is a genotoxic carcinogen. There is really no need to cease consuming Brassica plants because it has been demonstrated that their benefits can offset the harmful effects of certain metabolites when consumed at regular dietary levels [41].

Gastric cancer:

The fifth most frequent leading cause of demise worldwide is gastric cancer, even though its incidence has significantly decreased in many regions of the world. A panel organized by the AICR & WCRF decided that eating vegetables "probably" lowers the occurrence of stomach cancer. A meta-analysis inspecting the relationship between consuming Brassica vegetables and the incidence of stomach cancer found that doing so produced statistically significant results in prospective trials and substantial results in case-control studies. Human stomach cancer risk is revealed to be inversely correlated with high consumption of Brassica plants [41].

Colorectal Cancer:

In the US colon cancer 3rd most common cause of cancer-related deaths. Ingesting Brassica vegetables is allied with a minor risk of colon cancer in eight case-control studies, a negative suggestion in three, and a positive correlation in one. Comparing high and low intakes of Brassica vegetables reveals a lower risk of rectal cancer in five case-control studies that look at the connotation between these vegetables and the risk of the disease. Eating 250 g of broccoli and 250 g of Brussels sprouts per day meaningfully increased the excretion of 2-amino-1-methyl-6-phenylimidazo [4][5]-b pyridine (PhIP), a potentially carcinogenic chemical found in well-cooked meat, according to another clinical investigation. By increasing the elimination of PhIP and carcinogens associated with diet-related heterocyclic amines, consuming a lot of Brassica vegetables has been shown to reduce the incidence of colorectal cancer. In a multi-center experiment, the group that consumed the most broccoli had a reduced risk of colorectal cancer, according to the food frequency questionnaire. It is not possible to conclude that consuming Brassica vegetables raises the risk of colon cancer based on the information currently available [41]

Lung cancer:

Lung cancer, the important cause of death worldwide, arises from genetic lesions instigated by contact to ROS, bacteria, viruses, oestrogens, and smoking. It is hard to remove the afflicted area in many lung cancer patients due to unfavorable metastases or advanced lesions. Therefore, a different approach to reducing the risk of lung cancer is to adopt preventative eating habits. A study looked at GSTP1 and NQO1 inducers of phase II enzymes,

as well as the effects of numerous phytochemical agents on human lung cells. GSTP1 mRNA levels in bronchial epithelial cells exposed to broccoli sprouts increased little, but NQO1 mRNA levels increased in response to sulforaphane exposure. NQO1 protein expression increased 11.8 times in bronchial epithelial cells treated with sulforaphane [41]

A potential study looked at the affiliation between urine isothiocyanate levels and lung cancer risk in nonsmokers. Lung cancer risk in nonsmokers is not associated with urine isothiocyanate levels. However, based on the levels of isothiocyanate in the urine, the secondary analysis revealed an interaction between the GSTM1 genotype and lung cancer risk. In a separate study, mice were given 9 μ mol of sulphoraphane per day of oral body weight in order to examine the effects of sulphoraphane on lung carcinogenesis brought on by the oxidatively damaging polycyclic aromatic hydrocarbon family member benzo(a)pyrene [B(a)P]. In conclusion, it is shown that sulforaphane therapy reduces hydrogen production peroxide, which has an antioxidant effect. It's crucial to remember that the benefits of stopping smoking are probably greater than the benefits of eating more Brassica vegetables when evaluating the consequence of doing so on the risk of lung cancer [41]

Breast Cancer:

Current case-control studies provide epidemiological evidence that eating Brassica vegetables is associated with a lower incidence of breast cancer. Furthermore, several studies indicate that eating certain Brassica vegetables lowers the risk of breast cancer. 16aOHE1 is more estrogenic than 2OHE1, and it has been discovered that oestrogen increases sensitive breast cancer cells in vitro. The incidence of oestrogen-sensitive cancer, including breast cancer, can be decreased by switching the metabolism of 17 β -oestradiol to 2OHE1 and avoiding 16aOHE1. Another study compared the dietary habits of 2,650 healthy women and 2,832 breast cancer patients, all aged between 50 and 74 years. The findings aimed to explore the relationship between diet, particularly the intake of Brassica vegetables and the risk of developing breast cancer. One to two servings of Brassica vegetables per day have been shown to lower the threat of breast cancer by 20–40%, potentially through changes to the oestrogen metabolism pathway. However, there is no correlation between the risk of breast cancer and the total amount of fruits and vegetables consumed. According to a different study, the edible part of cauliflower contains compounds that severely slow the growth of breast cancer cells in people with and without oestrogen receptors. An excessive intake of Brassica vegetables is linked to a markedly lower risk of breast cancer in Chinese women, according to a study that used a urine isothiocyanate biomarker to measure consumption of these veggies. Consumption of Brassica vegetables, particularly broccoli, is adversely correlated with breast cancer risk in premenopausal women in a case-control study of Caucasian women with breast cancer. According to these results, premenopausal women may be at lower risk for breast cancer if they eat Brassica vegetables. However, the combined review of eight cohort studies found no link between the risk of breast cancer and the consumption of fruits and vegetables, either individually or collectively [41].

Prostate cancer:

The most prevalent solid tumor is prostate cancer. Prostate cancer risk is linked to dietary practices. The most frequently documented link between diet and prostate cancer is a higher risk of eating foods heavy in fat. Vegetable products may, however, contribute to prostate cancer, according to advances in our knowledge of the cellular processes that cause cancer and the control of metabolic and genetic alterations that cause cancer. Prostate cancer is of particular concern when it comes to the induction of Brassica vegetables and GST- π in cell culture models. The most common GST active in prostate tissue is GST- π , and prostate cancer, prostate cancer precursor lesions, and prostate intraepithelial neoplasms all exhibit decreased expression of this gene. It is believed that up regulating this gene may in some way prevent the progression of prostate cancer, as loss of GST- π seems to be a crucial stage in the

early stages of the disease. It has been discovered that giving animal's broccoli changes the amounts of biotransformation enzymes in peripheral tissues that are distant from the sites of immersion or metabolism [41].

Pancreatic Cancer:

Pancreatic cancer ranks fifth in Japan and fourth in the US in terms of cancer-related mortality. The prognosis for this type of cancer is particularly bad because it is typically discovered at an advanced stage. There is no proof in cohort research that eating vegetables is generally associated with a higher risk of pancreatic cancer. Nonetheless, high-risk people showed a negative correlation with dark green vegetable consumption. Three or more servings of Brassica vegetables per week were found to be non-significantly inversely correlated with one serving of Brassica vegetables per week among particular subgroups of fruits and vegetables by another study. When comparing individuals who consumed one or more servings of cabbage per week to those who did not, there is a statistically significant decrease in the risk of pancreatic cancer [41].

Apoptotic cells proliferated in two human pancreatic cancer cell lines after they were exposed to X-rays and treated with benzyl isothiocyanate (BITC), a member of the isothiocyanate family. The impact of BITC on the radiation sensitivity of human pancreatic cancer cells is investigated in this work. It is found that BITC could be utilised to treat pancreatic cancer in addition to the present radiation therapy.

Bladder Cancer:

Additionally, the Health Specialists Follow-up Study found a non-significant negative correlation between the incidence of bladder cancer and the overall amount of fruits and vegetables consumed. Nonetheless, it is demonstrated that eating Brassica plants, such as broccoli and cabbage, is substantially linked to an increased risk of bladder cancer. According to these findings, eating a lot of Brassica may lower your risk of bladder cancer, while other fruits and vegetables might not have a major positive impact. According to authoralpha-tocopherol beta-carotene cancer prevention trial, eating Brassica vegetables did not increase the incidence of bladder cancer.

It is demonstrated that the isothiocyanates from eating Brassica vegetables are protective against bladder cancer in a study involving 697 individuals with recently diagnosed bladder cancer and a control group. Intake of isothiocyanates is found to be negatively related with the risk of bladder cancer, and this impact is more pronounced in non-smokers than in heavy smokers. Another study discovered that giving rats freeze-dried broccoli extracts significantly and dose-dependently reduced the development of bladder cancer caused by N-Butyl-N- (4-hydroxybutyl) nitrosamine without changing the histology of the bladder. In conclusion, because Brassica crops' isothiocyanate extract is a strong activator of the bladder's GST and quinone oxidoreductase 1, it may be used as a preventative measure against bladder cancer. Moreover, the main isothiocyanate in broccoli sprout preparations, sulphoraphane, triggers apoptosis and cell cycle inhibition in addition to inducing enzymes that detoxify carcinogens [41].

Neurological diseases (Neurodegenerative diseases):

Numerous neurodegenerative illnesses are genetic, while some are caused by deadly or metabolic methods. It's still unclear what causes neurodegeneration. According to some beliefs, oxidative anxiety plays a significant role in most [44]. Meanwhile there is no treatment that can prevent the emergence of neurodegenerative illnesses, human health benefits from dietary consumption of antioxidant foods or plant extracts [45]. The glucosinolates in Brassica plants and the isothiocyanates they create during degradation have been shown to prevent some chronic illnesses, such as cancer and neurological disorders. Isothiocyanates alter inflammatory pathways, encourage apoptosis, and reduce the activation of cell death, all of

which have an active impact on the central and peripheral nervous systems. These phytochemicals have been shown to particularly induce NF- κ B translocation, which sets off the production of reactive species, proinflammatory cytokines, and pathways leading to neuronal apoptosis [44]. One study found that pretreatment with sulphoraphane reduced the cognitive effects of repeated PCP administration. It has been demonstrated that eating broccoli sprouts high in sulphoraphane during childhood and adolescence can delay the onset of psychosis, as well as have proinflammatory and therapeutic effects on cognitive impairment associated with schizophrenia [46]. In another study, raw broccoli sprout juice protected cells from β -amyloid peptide (A β)-induced cytotoxicity and apoptosis. It has been demonstrated that this protection is achieved via modifying mitochondrial activity, activating the Nrf2-ARE signalling pathway, and regulating the transcription and expression of the HSP70 gene. Additionally, through Nrf2 activation, broccoli juice has been shown to raise intracellular glutathione concentration, mRNA levels, and the activity of antioxidant enzymes like HO-1, thioredoxin, thioredoxin reductase, and NQO1. The results of the study suggest that Alzheimer's disease may be prevented and treated by pharmacologically stimulating the Nrf2 signalling system and consuming broccoli sprout juice [45].

Antidiabetic Effects:

Controlling blood sugar is essential for avoiding or postponing diabetic complications, which increase the rates of morbidity and death. New therapeutic approaches are therefore desired [47][48]. New approaches to treating type 2 diabetes mellitus and preventing its long-term effects may involve the use of nutraceutical ingredients in functional meals. It has been demonstrated that increased antioxidant capacity can help reduce diabetic issues, which are thought to be related to oxidative stress brought on by anomalies in the metabolism of fats and carbohydrates [48]. Strong Nrf2 activator sulphoraphane has been shown to prevent a number of diabetes problems, including cardiomyopathy, nephropathy, neuropathy, and retinopathy, by boosting Nrf2 activity.

[48][49] Furthermore, sulphoraphane-enabled Nrf2 inhibits oxidative stress and metabolic dysfunction brought on by hyperglycemia in human microvascular endothelium cells. One gram of cabbage extract per body weight is found to enhance glutathione and SOD activation and reduce catalase activity in diabetic kidneys in rats with diabetes. Therefore, red cabbage extract's antihyperglycemic and antioxidant qualities may offer promising therapeutic sources for the treatment of diabetes.

Additionally, it has been demonstrated that sulphoraphane reduces blood-insulin concentration and hyperglycemia, protects the amount of insulin-producing islets, prevents β -cell mortality from enhanced oxidative stress or pyogenesis, and lessens cytokine-induced cell death in rat insulinoma cells.

Patients with type 2 diabetes were divided into three groups and given either 10 g or 5 g of powdered broccoli sprouts or a placebo for four weeks in a randomized controlled double-blind clinical research. Consequently, it is found that broccoli sprout powder lowers patients' serum levels of oxidized LDL and malondialdehyde, improving oxidant and antioxidant levels and raising total antioxidant capacity. Another study using the same application revealed significant decreases in HOMA-IR and insulin levels. A prospective meta-analysis study did not find a link between citrus consumption and type 2 diabetes; however, it did find a negative correlation between type 2 diabetes and Brassica vegetables [41].

Cholesterol-Lowering Effects:

Elevated cholesterol levels cause coronary heart disease, one of the world's chief causes of mortality. Meals that lower cholesterol primarily have two characteristics: reduced endogenous cholesterol synthesis and competition for absorption sites. Broccoli sprouts have been shown to reduce fat and cholesterol levels in addition to their well-known chemoprotective properties. Twelve healthy trial participants experienced a significant increase

in HDL cholesterol and a decrease in total and LDL cholesterol after consuming 100 g of fresh broccoli for a week. After just one week of ingestion, broccoli sprouts were shown to improve oxidative stress markers and boost cholesterol metabolism. In another research, the LDL cholesterol levels of 77 males were assessed after consuming broccoli-cabbage juice. Serum LDL cholesterol levels in the test group had decreased by 8.5% at nine weeks. However, there is no decrease in the serum LDL cholesterol levels of the control group. The extract is found to lower serum cholesterol levels and increase bile acid excretion and cholesterol 7 α -hydrolase activity in the faeces of hepatoma-carrying rats with hypercholesterolaemia, which were used to study the effects of elevated cholesterol formation on cholesterol metabolism. These results show that by encouraging cholesterol catabolism, cabbage lowers hypercholesterolaemia in rats with hepatoma.

Polyphenols found in red cabbage and Brussels reduced the amount of cholesterol in hypercholesterolemic patients' erythrocytes. It is determined that these compounds' polyphenol contents had a direct impact on erythrocyte membranes, and that anthocyanins may be involved in this effect. The hypocholesterolemic exchange effects of adding broccoli sprouts to the diet of hamsters fed diets high in fat and cholesterol were examined in a different study. Simvastatin, glucoraphanin-rich broccoli sprouts extract (GRE), sulphoraphane-rich broccoli sprouts extract (SFE), and a freeze-dried broccoli extract with 2 or 20 μ mol of glucoraphanin (BSX, BS10X) were given for seven weeks. As a result, all mice receiving BS10X and SFE treatments had lower hepatic cholesterol. GRE had less of an impact on cholesterol homeostasis than glucoraphanin and broccoli sprouts (BS10X) at the same concentration.

Helicobacter pylori is closely linked to a number of gastrointestinal disorders, such as gastric infections, chronic superficial gastritis, duodenal and gastric ulcers, stomach adenocarcinoma, and non-Hodgkin's lymphoma of the stomach [41]. The urease enzyme, which is produced by *H. Pylori* and is absent from mammalian tissues, converts the host's urea to ammonia, rendering stomach acid inactive. It has been shown that sulphoraphane and other isothiocyanates inhibit the urease that *H. Pylori* generates. Purified sulphoraphane is found to kill more *H. Pylori* strains than any other, except for antibiotic-resistant strains in test tubes, and to impede the formation of cancer cells. *H. Pylori* is eradicated in eight of the eleven xenographies of human stomach tissues implanted in immunocompromised mice after they were given sulphoraphane for five days in an animal model. Additionally, after eating broccoli sprouts high in glucoraphanin (56 g/d glucoraphanin) for a week, only three out of nine gastritis patients got *H. Pylori* eradication, according to a small clinic trial.

Furthermore, it is observed that eating broccoli sprouts daily for two months (70 g/d glucoraphanin) may help reduce oxidative stress caused by *H. Pylori* and prevent gastritis in both humans and experimental animals [41]. Another study found that broccoli sprout extract containing sulphoraphane inhibits lipid peroxidation in the stomach mucosa but does not lessen the density of *H. Pylori* infection. It has been demonstrated that sulphoraphane has a cytoprotective impact on *H. Pylori*-induced gastritis [50]. In conclusion, two theories have been proposed to explain how sulphoraphane influences *H. Pylori* infection. First, it has a direct impact on *H. Pylori*, which reduces gastritis. The second of these is to stimulate cytoprotective responses in order to demonstrate an indirect influence on the host.

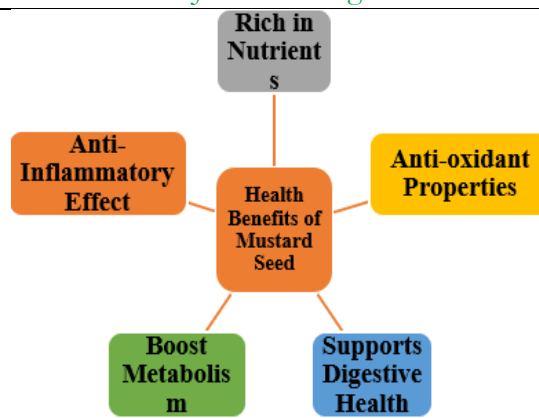


Figure 1. Health Benefits of Mustard Seed.

Applications of Mustard:

Food:

Since earliest times, mustard has been used to season and flavour food because of its aromatic properties. For instance, Indians add oil-heated whole brown or black mustard seeds to sauces, chutneys, and pickles to enhance their flavor, while Barbados and other Caribbean countries use yellow or brown mustard along with fruits and chilli peppers to create delicious sauces, marinades, and stews. All you need to do is combine mustard with vinegar, wine, or salt to create a delicious sauce [25]. Mustard is a good source of antioxidants and glucosinolates, which can be added with a number of possible benefits. Traditional meat-based products are preserved with nitrates and nitrites, which give the meat a pinkish color, a Smokey flavor, and protection from harmful microbes [51]. However, their presence might be detrimental to human health and the environment. The use of ground mustard as a food preservative in place of nitrates and nitrites in meatballs is assessed [12]. They found that by preventing bacterial growth and postponing lipid peroxidation, mustard greatly extended shelf life. The meatballs' sensory qualities were also enhanced by the addition of mustard; white or yellow mustard performed better than black or brown mustard. The efficacy of mustard as a natural supplement is confirmed by earlier studies conducted by author [52] and others. The organic fermented sausage is found to have a 90-day shelf life when acid whey and autoclaved mustard seed were added. In addition to maintaining the quality, there is a discernible decrease in oxidation-reduction potential and an increase in antioxidant capacity. This might be explained by the high-temperature pressured extraction method used in the autoclave, which significantly raised the phenolic concentration in the additive combination [25]. Using food-based products, vinegar, and sodium bicarbonate, a study [53] improved the extraction of total phenols, total flavonoids, and antioxidant activity from Oriental, black, and yellow mustard cultivars. Temperature, pressure, and pH all had a major effect on the extracted phenolic compounds, increasing their potential applications in the food and pharmaceutical sectors. In calculation to its effectiveness as a potential additive, mustard seed extracts effectively reduced the quantities of live *Listeria monocytogenes* when applied as an antibacterial film on bologna sausages. According to [25] and [54], black mustard's high Sulphur content (60 mg/kg) might be considered an antibacterial agent. Black mustard's high sinigrin concentration, according to [55], destroys membranes and inhibits enzymatic activity to prevent the growth of dangerous bacteria. Furthermore, by blocking the synthesis of the microbe's DNA, ally isothiocyanate, a result of sinigrin breakdown, accomplishes the same goal [25] [54]. Furthermore, [54] shown antifungal activity against food-borne pathogens *Staphylococcus aureus*, *Bacillus cereus*, and *Clostridium perfringens*, as well as *Aspergillus niger*, *Aspergillus ochraceus*, and *Penicillium citrinum*. Consequently, mustard has a lot of promise as a bio-preservative.

To put defatted mustard seed by-products in food-based biopolymer film, combine the meal with 0.6% glycerol. Due to its high protein content, mustard is also a good food-grade vegetable protein and bread ingredient [25] [56]. Yellow mustard mucilage has been demonstrated by researchers to be an exceptional gum with texturizing, coagulating, and stabilizing belongings that can be employed in food.

Recommended intake:

54 grams of brassica vegetables are consumed daily per person in Germany. White cabbage, red cabbage, and cauliflower are the most popular brassica vegetables. Although there aren't many gender variations, consumption of brassica has been observed to rise with age. Other studies conducted in Germany have confirmed that the recommended daily intake of glucosinolates is 36 mg and 46 mg during the summer. In Finland and Denmark, the per capita consumption of glucosinolates is 4 mg in the summer and 6 mg in the winter, according to study. The National Institute of Cancer and many other organizations recommend eating five to nine servings of fruits and vegetables each day. There has been no distinct suggestion for the brassica veggies. Furthermore, prospective cohort studies indicate that individuals should consume at least five servings of brassica vegetables [41].



Figure 2. Nutritional composition of Mustard.

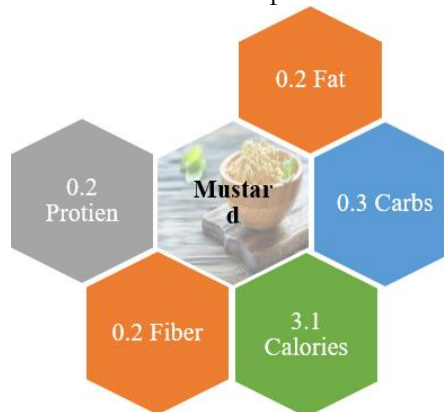


Figure 3. Nutritional composition in 500g mustard powder.

Contamination:

The contamination of palatable mustard with argemone oil causes a clinical illness known as dropsy. In humans, oil contaminated with argemone oil induces oxidative stress and the destruction of red blood cells. The method involves the development of methemoglobin, a defective type of haemoglobin that is incapable of carrying oxygen, lowering blood oxygenation and causing tissue hypoxemia. Antioxidant therapy is thought to be a treatment for argemone oil poisoning. The quaternary benzophenanthryrisine alkaloids dihydrosanguinarine and sanguinarine have been linked to argemone oil toxicity. Argemone oil adulteration in palatable mustard oil is detected utilizing an HPTLC approach that used sanguinarine as an indicator of argemone adulteration. Densitometric scanning determined the limit of detection (LOD) and limit of quantification (LOQ) of sanguinarine in argemone oil to be 1 ng/6 mm band and 3 ng/6 mm band, respectively. After 15 minutes of exposure to UV light (366 nm), dihydrosanguinarine is converted to sanguinarine. The concentration of sanguinarine in argemone oil samples ranged between 4.84 and 5.79 mg/ml of oil. The

percentage of adulteration in mustard oil testers ranged from 1.22 to 8.77%. In one study of 12 mustard oils (including both commercial and crude plants) conducted in Bangladesh, nearly all of the samples were found to be contaminated with argemone and mineral oils. Commercial oil is found to be ten times greater than raw oils. Two of the samples contained more castor oil than the limits [57].

Perspectives and Future Directions:

The cytotoxic, antibacterial, anti-inflammatory, antioxidant, antidiabetic, and hypolipidemic properties of mustard oils have been discovered. However, even when they function correctly, they may be hazardous because to their high content of erucic acid and allyl isothiocyanate. However, these oils are extensively used in Asian countries, mostly due to their nutritional value, which exceeds the toxicity. To ascertain the healthiness benefits for humans, clinical and pharmaceutical research must be carried out to validate the findings of animal studies [58].

Conclusion:

In the end, mustard is a unique crop that combines traditional uses with contemporary uses in sustainability, food, and health. It is a mainstay in functional foods and natural preservation because of its bioactive components, glucosinolates and phenolics, which support its antibacterial, antioxidant, and anticancer qualities. Despite worries about erucic acid, mustard oil is a nutritional powerhouse that has led to the development of low-toxicity strains. Beyond the plate, mustard promotes environmentally sustainable methods in agriculture by acting as a soil improver and bio-pesticide. However, issues like toxicity and adulteration necessitate strict quality controls and solutions based on research. Mustard is more than just a crop; it's a link between tradition and innovation because of its potential for therapeutic use and environmental adaptability. Mustard's potential can be expanded by tackling its issues through research and legislation, improving food security, sustainable agriculture, and global health. Its historical legacy is still thriving and helping to shape a healthy future.

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