



Review on Antioxidant and Hemagglutination Properties of Chia and Basil Seeds

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Authors' contributions

This work was carried out in collaboration between both authors. Author ASH designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors SB and ASH managed the analyses of the study. Author SB managed the literature searches. Both authors read and approved the final manuscript.

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ABSTRACT

Medicinal plants have always benefitted the human society. Traditionally many plants and their products have been used to cure many deadly diseases. It has estimated by the WHO that more than 80% of the world population relies on the natural products for their health care. It has been reported that these natural remedies are cost-efficient, environment friendly, as well as biocompatible for the human use. Chia and Basil seeds are the two plant products that have been long known for their antioxidant properties. It has been reported that both of these seeds are abundant source of healthy omega-3 as well as omega-6 fatty acid, polyphenol and antioxidants like caffeic acid, myricetin, quercetin and others. The current study is focused on reviewing the antioxidant benefits of consumption of these seeds in the daily diet. This study will further help the researchers to carry out more extensive research to find out other benefits of these seeds on the human health. These seeds then can be further used as the main ingredient in many essential foods for the marketable application.

Keywords: *Antioxidant; Basil; chia; Total Antioxidant Capabilities (TAC); health care, plants; seeds; Red Blood Cells (RBC); biocompatibility; World Health Organization (WHO).*

1. INTRODUCTION

Throughout humanity's civilization herbal medicines were known and used. Orthodox doctors use plant-derived drugs for the treatment of diseases. This comes generally from their immediate setting, where several different diseases have been cured. Plants can synthesize a number of chemical substances used to conduct essential biological functions and to guard against predator attacks, like insects, fungi and herbivores. In certain Asian and African countries, the World Health Organization (WHO) reports that 80% of people in world actually uses herbal medicinal products for healthcare services. Medicines have been used since time immemorial to treat human disease [1]. Now, most people still use their conventional prescription supplies to satisfy their everyday needs [2]. Biocompatible, biodegradable, cost-efficient, and environmentally friendly herbs are used as immunostimulants. Plants are warehouses and rich suppliers of cheaper and cleaner ingredients. As herbal medicinal products are using various herbal as well as other immuno-stimulants examined to improve the non-specific or specific immune reaction [3].

1.1 Antioxidants

The end of a free radical will occur only if it is neutralized with or scavenged with an antioxidant by interacting with other free radical or cell part. Consequently, antioxidants are molecules inhibiting oxidation. Antimicrobials have recently been commonly used as an active ingredient in dairy, pharmaceutical and cosmetic industry and to improve shelf life of the product. They also help reduce different degenerative diseases and cancers by the elimination of free cell radicals. Because most current antioxidants are produced synthetically, natural antioxidants with the same or higher total antioxidant capabilities (TAC) are continuously being searched later.

Chia seeds (*Salvia hispanica*) and basil plant (*Ocimum basilicum*) have been long known for their antioxidant properties and thereby, could be used in prevention of diseases caused due to oxidative stress. Both the seeds belong to the family Lamiaceae, which are known to have excellent phytochemical properties including antioxidant property. The effect of chia seeds in peroxidation of lipids has also been studied intensely [4].

1.2 Agglutination of Cells and Lectins

Agglutination, in biology, is the clumping of cells. Many substances in nature can cause agglutination. In our body, agglutination is used to phagocytose the pathogens with more efficiency. This property is effectively used to help diagnose various diseases, to determine blood groups and for matching donor and recipient in organ transplantation. One of the types of agglutinations seen is hemagglutination, which is the agglutination of RBCs due to antigen - antibody interaction. RBCs have surface antigens which determine its blood group. If an antibody is added to the blood, antibodies specific to the surface antigens on RBCs interact and cause agglutination. An additional antigen may or may not be present, called the Rhesus or Rh factor, which gives the RBC a '+' or '-' grouping. Absence of Rh factor can make the blood group '-' Based on this, blood group can be determined as summarized in Fig. 1.

Agglutination is possible only because of the presence of agglutinin molecules that bind to its counterparts very specifically. One of most commonly found agglutinins in nature are Lectin. These are the proteins which attaches to carbohydrate and carbohydrate containing proteins with very high degree of specificity. Lectins are not only useful in determining blood groups, but they are tools that can be used as biomarkers to detect various diseases like cancers. Lectins are known to facilitate the binding of bacteria and viruses to target cells. Disabling lectins can stop their attachment and thereby prevent the establishment of a disease [6].

1.3 Chia Seeds

The yearly plant which holds purple or white hermaphrodite flower in the summer is named as *Salvia Hispanica*. It is a short herbal plant about 1 meter in height and is about 4-8 cm thick with thin serrated leaves. Flowers are grown in clusters and the seeds are grown in the plant. It is part of the Lamiaceae mint family.

It is native to Guatemala and Mexico, but is now grown in most parts of South America, Australia and the United States. The seeds were a staple in Mayan and Aztec civilizations not only as a culinary ingredient, but also to prepare folk medicines, canvases and paints. They were utilized in various forms- whole seeds, soaked, ground and its oil extracted. They were also used

for cosmetic purposes and religious rituals. Some of the benefits of consumption of Chia seeds are represented in the Fig. 2.

The seeds' rise in prominence has its roots in its history, but scientific work done on it in recent years has unearthed its true nutritional potential, which has encouraged people to use the seeds. It is laden with omega-3 fatty acids (64%),

proteins (18.8% to 21.5%) high amounts of dietary fibre (25%), vitamins and minerals (greater than that in wheat, rice, oats and corn) [ref ?]. Various studies have found that the seeds are high in polyphenolic antioxidants, which account for around 8.8 percent of the overall dry matter chia seed extract. Chlorogenic acid, caffeic acid, and quercetin are the most popular polyphenols available [7].

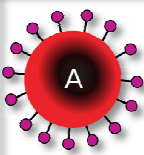
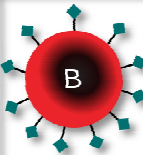
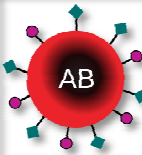



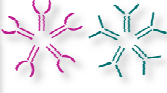



	Group A	Group B	Group AB	Group O
Red blood cell type				
Antibodies in plasma	 Anti-B	 Anti-A	None	 Anti-A and Anti-B
Antigens in red blood cell	 A antigen	 B antigen	 A and B antigens	None

Fig. 1. Determination of blood groups as well as antibodies and antigens present in the human body [5] (adapted from Wikipedia.org)

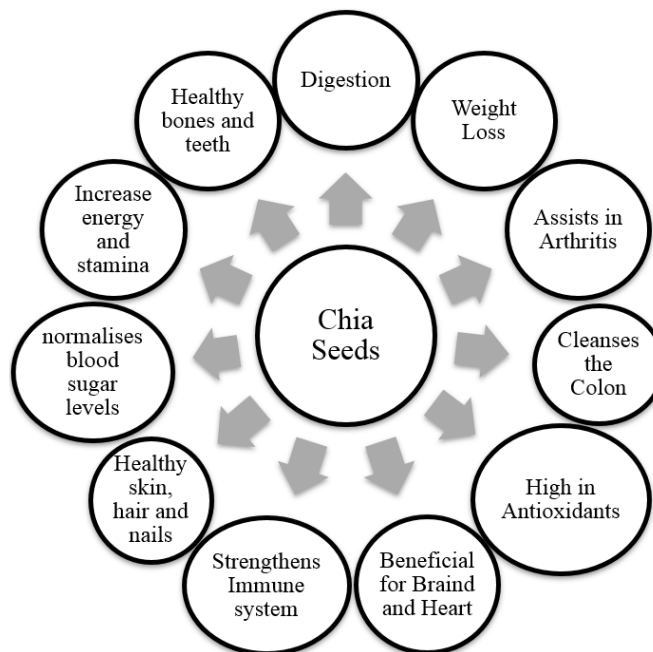


Fig. 2. Various benefits of consumption of Chia seeds on the health and well-being of humans

1.4 Antioxidants

The Chia seed is rich in phenolic compounds that show antioxidant-related functions scientifically illustrated. Health promoters and protection against degenerative illnesses, like cardiovascular disease, tumors, high blood pressure and diverticular disease have been shown to have antioxidants and phenolic compounds. Chia and oil also have several active molecules, including quercetin, myricetin, kaempferols, chlorogenic acids, and the ellagic dialdehyde-dihydroxyphenyl ethane. Various in-vitro studies proved the high antioxidant potential of such polyphenols and their existence is paired with reduced concentrations of lipid autoxidations [8].

1.5 Vitamins and Minerals

Chia is also origin of many vitamin and mineral, including niacins, magnesium, phosphorus, calcium and copper. The amount of chia niacin is greater as compared to those of other cereal (grain, soybean and rice), while its composition is close to that in maize and rice. The amount of calcium present in Chia is 6 times more than 100 g, 11 times as much phosphorus and 4 times more potassium.

1.6 Effects of Chia Seeds

A limited human volunteer trials was conducted to determine the wellbeing effects of eating chia seeds. A randomized, placebo-controlled study in 76 adults showed no substantial drop, even after feeding 50 g of chia seed per day, for 12 week in body's mass, lipids report, inflammatory markers, and blood sugars. In contrast with the reference sample, an increase of the plasma alpha linolenic acid (24.4%) was observed. Seven weeks of ingestion is tested on 10 stable postmenopausal women for 25g milled chia seeds. Chia seed intake led to a substantial rise of 30% and 138% respectively in plasma in the eicosapentaenoic acid (EPA) and alpha linolenic acid (ALA). Indeed, no big improvements were seen on Body Mass Index. Higher intakes of both aquatic and herbal omega-3 fatty acids in diets (eicosapentaenoic acid and docosahexaenoic acid) was closely correlated with decreased death risk due to coronary disorders that are becoming one of the world's main causes of mortality [9].

Vuksan et al. [10] completed a spontaneous, placebo-controlled clinic study for 20 controlled

(medically supplied) type 2 diabetics, 37 g chia seeds per day (added in white bread). The findings show that high chia fibers helped regulate hyperglycemia and lower systolic blood pressure. With chia intake, levels of ALA and EPA have risen. Chia-seeds can help to avoid stroke and heart disease on patients with diabetes type II, through anticoagulant and anti-inflammatory influence. Few tests have shown that Chia seeds have beneficial health benefits including reductions in blood glucose and systolic blood pressure [11]. 67 individuals (categorized into two parts) were provided 4g chia sown with peas, palm and soy container filled into 250 mL of drink 2 time in a day for 60 days, and a few caloric improvements in the normal diet in one dual-blind placebo controlled study with excess body weight and metabolism. In contrast with placebo participants, the research findings show a substantial drop in the body's mass, waist line perimeter as well as rate of metabolism. There has been a substantial decrease in tolerance to insulin, triglyceride and C-reactive protein. In overall cholesterol, blood sugar and plasma insulin, however, there were no major improvements [8].

1.7 Pharmaceutical Use

The ability of bilayer emulsification as delivery mechanisms for chia oil fatty acids is strong in pharmaceutical and food processes as emulsions could be utilized precisely or exposed to a dehydrating method in order to produce powder products. Advantages include ease of synthesizing and economic viability; the first alternative for supplying bioactive lipids is standard oil-in-water (O/W) emulsion. As a fat acids supply system of the foodstuff array, chia oils may be integrated into O/W emulsion. The composition of sodium caseinate and introduction of lactose have an important effect on chia O/W emulsion stability and rheological properties. Maintenance with a specific volume of emulsifiers provides mild stabilization of chia O/W emulsion and Newtonian behavior. Decreased amounts of primary and secondary oxidation products have been shown for Chia O/W emulsifiers. In M's research the use of layer-by-layer filing technique was used in studies of chia bilayers O/W emulsion. It composed of a positive-charge chitosan electrostatic accumulation on negative oil's drops. They were balanced in the inclusion or exclusion of maltodextrin utilizing altered sunflower lecithins (deoiled or enhanced by phosphatidylcholine).

A recent research [12] notes that microcapsules sprayed dry chia seed oil (CSO), chia seed gums (CSGs) and a CPI-CSGs complexes have been prepared to co-activate as shell material. The composite co-acervate CPI-CSG was considered ideal for supplying CSOs into the intestinal digestion process as most of the un-encapsulated oil became hydrolysed, while *in-vitro* digestion just 60% of the oils stored in the shell CPI-CSG were hydrolyzed. The leaves of *Salvia hispanica* L are expected to possess an important β -caryophyllenes, globulols, β -pinenes, α -humolene and widdrol oil. Such substances are reported that they contain high preventive features to a extensive variety of creatures [13].

1.8 Basil Seeds

Ocimum basilicum is an aromatic herb in the Lamiaceae family, sometimes used as a foodstuff. There are several various types of basil, such as honey basil, holy basil, African blue basil, lemon basil, and thai basil. Basil originates and is a farm crop in India, Burma, and Mediterranean regions, such as Italy and Turkey. It is indigenous to Central and Southeastern Asia. The herbs grow up to 30 cm high and have small, white, rosy, or pure flowers in clusters, every year and often continuously. The seed's

covering are tiny and strong. Some of the benefits of consumption of Basil seeds are represented in the Fig. 3 [14].

1.9 Basil as an Antioxidant

Rosemary acid from the *O. basilicum* plants was detected as a source of antioxidant properties. Jayasinghe et al. [15] investigated the existence of the rosemary acid antioxidative activity in the liposome network. The findings indicate that one rosemary acid will absorb 1.52 radicals and that the synergistic effect is present among α -tocopherol and rosmariy acid. Durga et al. [16] demonstrated that the antioxidant properties vary by levels of acetone and ethanol extracts *O. basilicum* (50, 100, 250 and 500 $\mu\text{g/ml}$). The ethanol extract concentration of 500 $\mu\text{g/ml}$ of 75.87% was very close to the average of the standard α -tocopherol concentration of 500 $\mu\text{g/ml}$ (82.14%). Extract behavior improved with a rise of polar solvent, which indicates that the activity level is impaired by polyphenols, flavanone and flavonoids. The *O* leaves ethanol extract. In addition to demonstrating strong radical superoxide and nitric oxide radical scavenging activity in the goat liver, basil demonstrated positive antilipid peroxidation impact *in-vitro* [17].

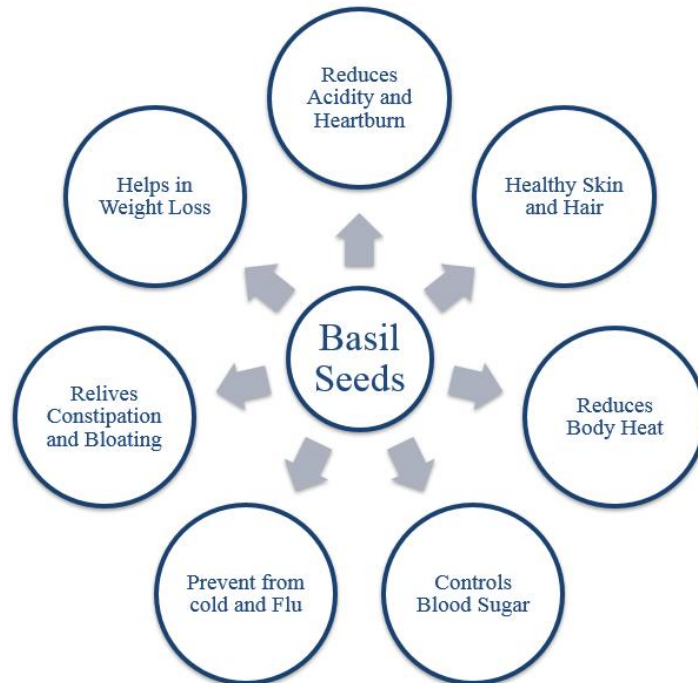


Fig. 3. Various benefits of consumption of Basil seeds on the health and well-being of humans

1.10 Hemagglutination Activity

Hemagglutinins or phytolectins can act as a cheaper source of blood typing reagents. These phytolectins can be found in many plants. Hemagglutination assay was firstly found by the Wong et al. [18] in the initial stages, this assay was tested on the erythrocytes. In this assay, Wong took the fresh samples of the blood and centrifuged them at 3000 rpm so that the erythrocytes can be centrifuged down and the left over supernatant can be discarded. After which the suspension of erythrocyte was prepared in the phosphate buffer of pH 7.2. Followed by the incubation for 30 minutes in the RT and after which the evaluation of any agglutination is evaluated [19].

1.11 Uses of Basil in Traditional Medicine

O. basilicum is commonly used in herbal remedies with the leaves or injections. The Santhal tribe of India uses sweet basil for headache, earache, cough, cold, inflammation, snake bites and rabies in certain mediterranean regions, including eastern Marocco, for reductions in plasma lipid content. Other therapeutic properties of basil leaves were recorded, including treating diarrhea, dysentery, constipation, flatulence and worms; as an antibiotic and repellent for insects; relief of bronchitis, flu, colds, coughs and sinusitis; and a remedy for rheumatism, muscular weariness, gout and fatigue. The leaves also have shown to be helpful in warts; a basil leaf ointment could be used as an insect bite therapy and can be added directly to the skin to cure acne.

The juices from the plants have quite a variety of medicinal uses: when combined with honey it relieves the effects of cold and cough and croup. It is often used as a toothache, earache and pain remedy and can be combined with camphor to avoid nasal bleeding. It also seems to make the eyes luster, and to heal ring worms, scorpion bite and serpent sting creates an outstanding nostrum. These seeds are often advised to be soaked in water and then consumed as they are found to be nourished and cooling. In various place it has been suggested that chewing of basil seeds overcomes the snake bites. The washing and bottling seeds was used for the prevention of persistent incontinence in poultices for painful sores and sinuses. A tablespoon of tea diluted with a small amount of sugar in a bowl of water serves as anti-emulgent in the case of genital-urinary disease when taken daily; it is called a

cool injection of seeds to cure birth pains; and an infusion of seed is often given in fever. As a diuretic, the aqueous extract of the seeds was used. Eventually, the plant roots are used to complain about bowels in kids [20].

1.12 Use in Ayurvedic Medicine

Basil has been used in medicine for centuries in Unani and Ayurvedic remedies as a stomachic, antipyretic, diuretic, anti-helminthic, diaphoretic, antiemetic, anti-diarrheic, aphrodisiac and an anti-dysenteric. The seeds, in particular, have been used for unhealthy sores, fever and sinuses. The seeds are chewed in case of snake-bites and parturition. Basil seeds, like chia seeds, are rich in proteins (17%), fiber (7%), carbohydrates (51%) and antioxidants (64 mg gallic acid equivalent / g). There is extensive literature on the basil planted but very limited work has been done on the seeds.

O. basilicum by Bhavamisra is known as 'barbari.' The herb is used for the disease due to Kapha and Vata aggravation, as the seeds are used to pacify the exacerbation of Pitta and Vata, as per Ayurveda. In a number of classical Ayurvedic books, the medicinal properties of *O. basilicum* are demonstrated. The entire plant is used in Ayurveda in the treatment of coughing, asthma, bronchitis, ophthalmia, giddiness, malaria fever, catarrh, otalgia, cephalgia, dyspepsia, spasmodia. The plant is known to be intestine, stimulant, Carminative, diaphoretic and expectorant [21].

1.13 Free radical and Reactive Oxygen Species (ROS)

Free radical is a molecular entity possessing an unpaired electron. ROS are radicals of oxygen formed in the body, physiologically. 1-3% of oxygen in the body, after its utilization, is converted into ROS. They can be of many types:

- Hydroxyl radical $\cdot\text{OH}$
- Peroxyl radical $\text{ROO}\cdot$
- Superoxide anion radical O_2^-
- Hydrogen peroxide H_2O_2
- Singlet oxygen O_2

They are formed due to various metabolic activities taking place in the cell, like the oxidative phosphorylation in mitochondria, oxidation of unsaturated fatty acids by cytochrome P450 enzyme, phagocytic cells producing ROS during oxidative burst etc. In

lower concentrations, free radicals are in fact, necessary for maturation of cellular structures. Phagocytes kill pathogens using free radicals, in a process called oxidative burst.

Increased accumulation of free radicals in the body, on the other hand, can have deleterious effects. Since free radicals are unstable due to presence of unpaired electron/s, they try to gain stability by attacking nearby molecules to pull an electron to itself. In this process, they convert the molecule into a free radicals instead. Such a molecule then uses a different electron, disrupts it and makes it a free radicals. Ultimately, this domino effect disrupts as well as damages the entire organism. Free radicals mediated cell injuries include lipids peroxidation, proteins oxidative activity, damages to DNA as well as damages to cytoskeleton [22].

2. DISCUSSION

As the advantages of medicinal plant materials have been well known for an exceptionally long time, the positive role of plant species and their constituents is no new growth. Basic oils in numerous plants have several biochemical properties, such as antioxidant and inflammatory proprieties.. Sweet basil is commonly used as a commercial seasoning species and is widely used in cuisine, like meats and soup. In several countries, sweet basil has an extensive medicinal importance as a plant. Especially for snake bite, cold and rhinitis therapy initial assist, the essential oil is helpful.

The plants of Lamiaceae family contain high levels of bioactive components like antioxidants. The two seeds selected for this study belonging to family Lamiaceae are Chia and Basil seeds. Chia seeds (*Salvia* spp.) are utilized for long period by the Aztec and Mayan tribe for the purpose of foodstuff and medicines. They were also used to make paints by them. They are known to include elevated level of antioxidant among other nutrient like omega-3 fatty acid, dietary fiber, protein and calcium. Phenolics like caffeic acids, chlorogenic acids and quercetins, amount to 9% of the total chia seed extracts. Due to these properties, chia seeds are being increasingly used in diet today.

Basil seeds have been consumed in India for generations, almost 3,000 years. The plant itself is holy to Indians, as they are grown in temple courtyards and at homes. The herb was then taken by Alexander the Great, in 350 B.C. to

Greece and cultivated there. Now, they are used in desserts like 'falooda' or powdered and used in flatbreads. They too, like chia, have risen to fame in recent years for their health benefits. Rosmary acids are the major phenolics found in Basil. It has anti-viral, antioxidant, anti-microbial and antiinflammatory effects. The *Ocimum basilicum* plant has been extensively studied for its antioxidant properties but there isn't a lot of work done on its seeds.

ROS causes the oxidative strain in living organisms. They are natural by-products of many physiological mechanisms taking place inside cells, such as oxidative phosphorylation taking place in mitochondria, oxidative burst performed by phagocytic cells, oxidation of unsaturated fatty acids by cytochrome P-450 etc. These ROS are unstable, free radicals that initiate a chain reaction. Living cells, therefore, produce antioxidants which donate electrons to free radicals, so they gain stability. When free radicals in the body, increase beyond the quantity of antioxidants present, they cause oxidative stress inside the cells. Oxidative stress is found to be responsible for ageing and many other diseases like Alzheimer's, Parkinson's, atherosclerosis, cancer etc.

Antioxidants are, therefore, required as external supplements to reduce the risk of oxidative pressure and reduce the making of free radical and/or break the chain reaction. They are known for playing a supportive role in prevention of diseases. The antioxidant properties of chia and basil seeds can be utilized not only as a dietary supplement, but also as a natural alternative for synthetic antioxidants used in food packaging.

Agglutination, in cell biology, means the clumping of cells. It is the basis of many medical diagnostic tests performed, whether it is a simple blood group determination or to detect and treat cancers. Lectins are one of the major agglutinins used in various field of research today. They are proteins that have the characteristics of binding to polysaccharides and polysaccharide-containing proteins with high specificity. They are of great importance in the medical and general biological fields, used for blood typing, bacterial typing, bone marrow transplantation, characterizing polysaccharides and glycoproteins, biomarkers for cancerous cells, molecular architecture of cells, affinity chromatography, affinity electrophoresis, blotting etc.

3. CONCLUSION

Chia seeds provide a healthy source of nutrient fibre, omega-3 fatty acid, bioactive protein and phytochemical. Chia has several essential physio-chemicals as well as operational characteristics that make it extra suitable in the foodstuff industries. Analysis performed *in-vivo* and *in-vitro* has demonstrated that it is healthy to eat and contain a wide array of health benefits. Investigation on Chia seeds in food and nutraceutical industries is available. Chia should also be used as a functional diet that can lead to improved mass wellbeing.

Basil is an annual spicy plant, native to India, grown for aromatic and medicinal uses for many millennia. This research describes basil before analyzing the chemical composition of the various plant varieties. Basil is defined as a gastronomic and medicinal application and its functional properties with a review of quality issues and toxicity. These seeds will thus be commercially used to produce new functional foodstuffs which contain omega-3, protein, fiber and phenolic compound-enriched products. Both seeds can help prevent, cure and treat many non-communicable diseases, improve immunity and likely alter the process of coagulation of the blood. This seeds also help to reduce the digestion of carbohydrate to slow down post-prandial blood glucose levels throughout the blood. These seeds can be utilized for frozen products, bakery, drinks, candy, baby products, spaghetti, sausages etc. Apart from applications in food and nutraceutical, these seeds have enormous benefits in physiological wellbeing. Based on the studies discussed in the present review, these seeds seem to entail physiological benefits by being key players affecting molecular targets. Therefore the review, opens novel horizons for detailed study of seeds as novel components for pharmaceutical applications.

CONSENT

It is not applicable.

ETHICAL APPROVAL

It is not applicable.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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