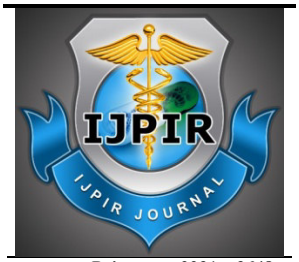
Review Article



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A review on pumpkin seed oil (PSO): Its nutritional benefits and therapeutic effects

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ABSTRACT

Pumpkin seeds are commonly dumped as agro-industrial waste. The seeds are eaten raw, roasted, or cooked in several parts of the world, but only in small quantities. Pumpkin seed oil contains high concentration of unsaturated fatty acids, phytoestrogens, and vitamins E, all of which have potential medicinal, nutraceutical, and cosmeceutical applications. In recent years, there has been a rapid increase in the amount of information available about the nutritional components and therapeutic benefits of pumpkin seeds oil. They are now regarded as valuable for the food business due to their high content of protein, fibers, minerals, polyunsaturated fatty acids, and phytosterols. Also their beneficial benefits on blood glucose levels, immunity, cholesterol, the liver, the prostate gland, the bladder, depression, learning difficulties, and parasite inhibition are also being investigated. Converting these agro- wastes into value-added components is expected to be a significant step forward in global sustainability efforts, and it merits more exploration. This article provides an updated overview of this new nutraceutical and therapeutic impact.

Keywords: Pumpkin seed oil, phytosterol, nutraceutical.

INTRODUCTION

Pumpkin including *Cucurbita moschata, Cucurbita pepo*, and *Cucurbita maxima* are gourd squashes that belong to the *Cucurbita genus* and the *Cucurbitaceae* family[1, 2]. The flower, berries, leaves, root, and seeds are all edible parts of the plant. The seeds, fruits, and leaves are used ethno botanically as functional foods or for the treatment of colds, irritable bladders, prostatic complaints, micturition, fever, exhaustion, hunger, and nausea, as well as ache, burns, gastritis, and enteritis[3]. Pumpkin is grown all over the world and has long been used as medicine in India, China, Yugoslavia, Armenia, Mexico, Brazil, and the United States. It has been used for a variety of illnesses in different schemes, including antidiabetics, antihypertensive, antitumor, immunomodulation, anti bacterial[4].

Pumpkin fruit varies in colour from orange to yellow due to the presence of vitamin A, and the pulp and seeds are contained within the protective shell[5]. Pumpkin seeds have long been used as both a food and a medicine. Similarly, pumpkin seed oil (PSO) has recently received a lot of attention as a potential nutraceutical as well as edible oil. It's viscous, dichromatic oil that's been shown to have good antioxidant properties[6]. The dark greenish- red oil is used in cooking, as a marinade, and as a salad dressing, and it has already been compared to olive oil. It's in chocolate, cereal bars, pizza, cake, muffins, broth, pesto, stew, and pasta garnish. Pumpkin seed butter is an excellent substitute for peanut butter[7]. Vitamin E content, especially 7-tocopherol, is extremely high[8]. The oil content of a pumpkin seed is approximately 50%. The seed can be eaten and has shown to be effective in treating a variety of prostate diseases. As a result, a pumpkin variety that contains a lot of vitamin E can be used as an ideal nutraceutical. The biochemical and oxidative stability properties of pumpkin seed oil will help to increase the oil's value, particularly in the pharmaceutical, cosmetic, and food industries[9]. Palmitic, stearic, oleic, and linoleic fatty acids predominate in pumpkin seed oil, accounting for 96-99 percent of total fatty acid content. Linoleic acid, also known as omega 6 polyunsaturated fatty

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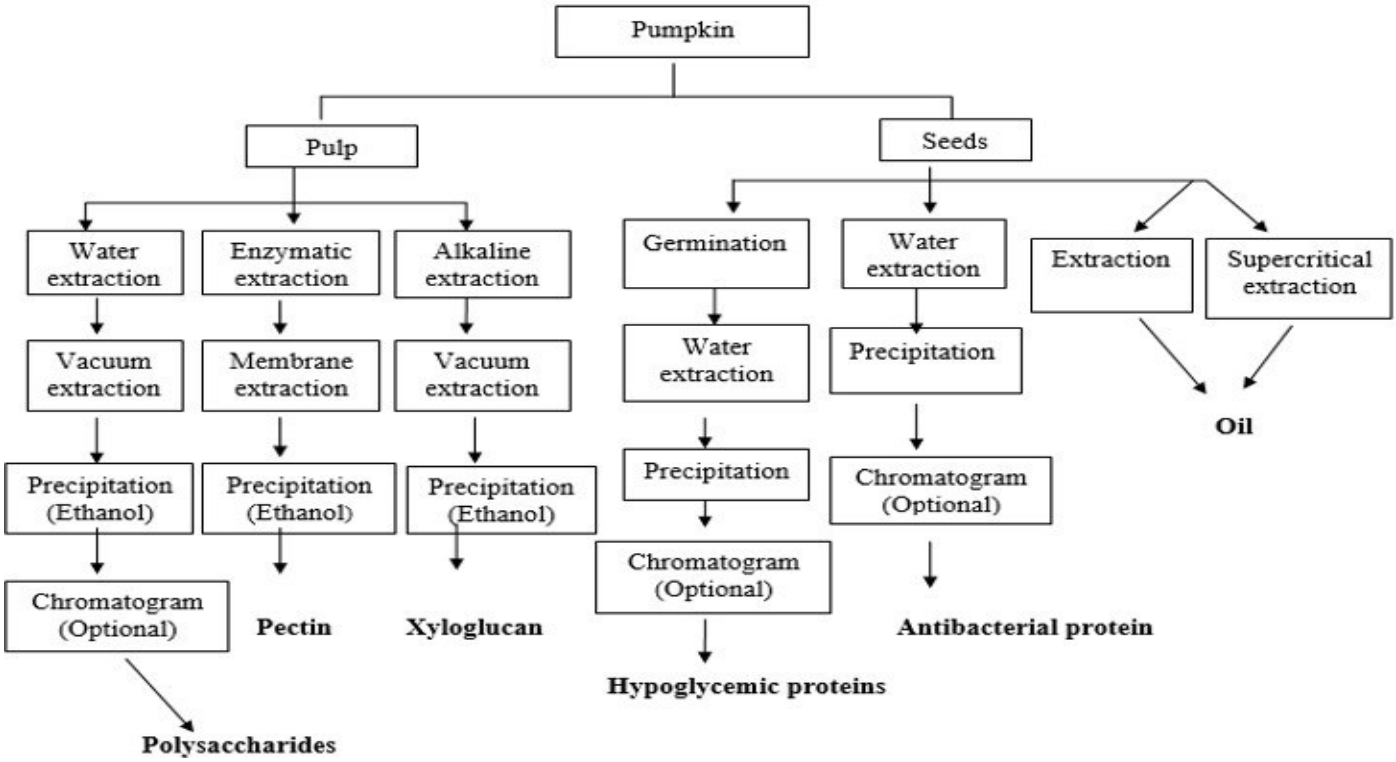
acid, is a primary fat portion of pumpkin seed oil that produces a hormone-like` substance that aids in blood clotting, prevents inflammatory responses, and boosts immunity. Pumpkin seed oil is also high in the monounsaturated oleic acid omega 9, which has been shown to improve cardiac health and reduce the risk of heart disease. These fats help to lower unhealthy LDL (low-density lipoprotein) cholesterol levels and prevent the formation of arterial plaque[10].

PSO (pumpkin seed oil) has also been shown to be an effective treatment for BPH (Benign Prostate Hyperplasia) symptoms[11]. Its activities have been attributed to phytosterols, which have been shown in rats to inhibit 5-reductase and have antiandrogenic properties[12].

EXTRACTION OF PUMPKIN SEED OIL



SOXHLET METHOD

200ml of the petroleum ether was put in the conical flask, 100g of the seed was weighed after grounding and wrapped in a muslin cloth, which was then placed in the extraction tube, which was then fixed into the conical flask and the condenser tube was then fixed. The entire setup was then put in the heating mantle and allowed to heat up to 600°C for two hours. After getting the oil, the conical flask was put in an oven dryer at 700 °C and weighed every 3 minutes until it reached a constant weight[13].

SOAKING METHOD

The weight of the seed from which the oil would be extracted was first determined. A blender was used to weigh 100g of seed for size reduction. The ground seeds sample was poured into 200ml of petroleum ether in a plastic bottle, and the container was tightly covered to prevent the petroleum ether from escaping. The mixture was shaken four times every four hours for 72 hours (3days). Filter paper, a separating tube, and a flask were used to filter the mixture. The oil was extracted from the filtrate by heating the solvent (petroleum ether) in an oven dryer at 700°C. For each of the varieties, the process is repeated[13].

COLD PRESSING OF OIL

Plant oil processing begins with the cleaning and pressing of oil seeds. Extraneous impurities such as debris, plant pieces, and damaged seeds were removed from the seeds. This ensures a steady supply of oil. A special device or a heat exchanger that uses the heat from the warm press cake pre-warms the seed to around 25°C. There is no advantage of preheating the seed to over 25°C. Cold pressed pumpkin oil was produced using screw presses. The screw rotated at a rate of 20 revolutions per minute[14].

COMPOSITION OF PUMPKIN SEED OIL

Pumpkin seeds come in a variety of organisms, each with its own set of components and biological activities[4]. Many scientists investigated the bioactive compositions of pumpkin seed oil grown in various parts of the world. Due to differences in the yield of fatty acids, sterols or phytoestrogens, and tocopherols among *Cucurbita spp*. species and/or varieties, several studies have focused on the three major components of pumpkin seeds: fatty acids, sterols or phytoestrogens, and tocopherols. However, the minor components of pumpkin seeds such as protein,

mineral, terpenic alcohol, and fiber also could not be ignored, because they have played role in the synergistic spositive effects of pumpkin seeds[15].

The pumpkin seed oil is dark green in colour and contains a high amount of free fatty acids (Palmittic acid, Stearic acid, Oleic acid, Linolic acid) with proportional distributions of 43.8 percent, 33.1 percent, 13.4 percent, and 7.8 percent respectively, representing 98 + 0.1 percent of the total fatty acids amount[16]. Dry pumpkin seeds have 47.03 percent oil content. The variability of oil content in various pumpkin species, on the other hand, is primarily due to its broad genetic diversity[17].

Pumpkin seed oil is a natural source of proteins, essential fatty acids, polyunsaturated fatty acids, omega 3, 6, and 9, carotenes, lutein, vitamins like carotenoids and beta- and gamma-tocopherols, phytosterols, chlorophyll, and trace elements like zinc and selenium[18, 19].

Pumpkin seed oil were found to have high levels of squalene, tocopherols, phytosterols[20] and carotenoids (lutein and zeaxanthin) [21]. As phytosterols present in pumpkin seed oil include desmosterol, campesterol, stigmasterol, -sitosterol, spinasterol, D7, 22, 25-stigmastatrienol, D7-stigmastenol, D7, 25-stigmastadienol, and D7-avenasterol. The predominant phytosterols contained in pumpkin seed oil are 7-sterols[22]. Pumpkin seed oil contained phenolic acids such as protocatechuic, syringic, caffeic, p-coumaric, vanillic, and ferulic[23].

Cucurbita pepo L. is the most popular pumpkin species that has piqued the attention of scientists all over the world. *Cucurbita pepo* species is high in polyunsaturated fatty acids like palmitic acid, stearic acid, oleic acid, and linoleic acid, vitamin E like alpha-tocopherols, gamma-tocopherols, and carotenoid, sphytoestrogens and phytosterols like daidzein, genistein, and secoisolariciresinol, and trace components, according to recent studies unsaturated fatty acids were the most abundant component in pumpkin seeds, accounting for up to 80% of the total percentage of ingredients. This value is higher than what has been recorded for peanut and soybean seed oils[24, 25]. The nutritional composition of pumpkin seed oil is given in the table 1 below.

Table 1: Composition of pumpkin seed oil

INGREDIENTS	CONCENTRATION IN SEVERAL VARIETIES			
	Cucurbita Pepo L.*	***Cucurbita Pepo Subsp. Pepovar Styriaka**)***	***Cucurbita maxima…)***	***Curcubita maxima,Var. Berrettina******
Palmittic acid	9.5-14.5%	10.86%	15.97%	Unquantified
Stearic acid	3.1-7.4%	8.67%	Unquantified	Unquantified
Oleic acid	21.0-46.9%	38.42%	44.11%	41.40%
Linolic acid	35.6- 60.8%	39.84%	34.77%	37.00%
Other fatty acid	<0.5%	Unquantified	Unquantified	Unquantified
α-tocopheropols	n,d-91mg/kg	882.65mg/kg	42.29%	Unquantified
γ-tocopherols	41-620mg/kg			
Daidzein	5.6-15.3ng/g	Identified as	Identified as	
Genistein	5.6-15.3ng/g	polyphenol:66.27mg/kg	Polyphenol;79.6mg/kg	
Secoisolariciresinol	210µg/g			Unquantified
Phytosterol	1.6-1.9%	1.86%	39.60%	63.20%
Protein	25.2-37%	25.40%	33.92%	1.28%
Carotenoid	Unquantified	Unquantified	Unquantified	2.5mg/g

Cucurbita maxima are the second most popular pumpkin species, and it is grown in many parts of the world. Rezig and colleagues investigated the chemical composition of pumpkin seeds as well as the oil characteristics of *Cucurbita maxima* seeds from Tunisia. Fatty acids and tocopherol were found in abundance, according to the researchers[23]. Another researcher looked at the nutritional components of various varieties of *Curcubita maxima L. var. Berrettina* and discovered that oleic acids were the most abundant fatty acids, and sterols were the most abundant component[26]. Due to the different components among the varieties of *Cucurbita* grown in the different areas of the world, all studies agreed that pumpkin seeds are a good source of many nutrients[27].

The fatty acid profile identified by Gas chromatography[28] is shown in table 2 below.

Table 2: Fatty acid profile

NAME OF THE COMPOUNDS	%w/w
Palmitic acid methyl ester	16.36
Stearic acid methyl ester	9.2
Oleic acid methyl ester	34.27
Linoleic acid methyl ester	48.02

NUTRITIONAL BENEFITS

Pumpkin seeds have a wide variety of applications as a vegetable or herbal medicine in recent years. These waste streams have value and can be used to make food and/or nutraceutical products. In their roasted shape, they can be eaten as a snack, salad, or breakfast cereal (salted or not). Furthermore, they may be used in baking as excellent bread or cake ingredients. Furthermore, their oil is excellent and has the potential to be used as edible oil as well as an additive ingredient in the food, pharmaceutical, and cosmetic industries. Pumpkin seed oil can be used for frying, slicing, baking, and dressing salads. A soft capsule supplement made from pumpkin seeds could be produced. In cosmetic industries, they usually use for skin care products such as anti-aging, free-radical scavenging, skin protection and hair care products such as hair growth stimulants and emollients. The consumption of pumpkin seeds in the oil form or roasted pumpkin seeds is proved to exhibit several positive health effects[27].

Pumpkin seeds are commonly discarded as waste, despite the fact that they are a rich source of nutrients and oil and can be eaten. After the pulp has been removed, the pumpkin seeds and rinds that remain as a waste product in vast quantities can be eaten. Researchers have discovered that pumpkin seed oil has medicinal properties. Pumpkin seeds and oil have traditionally been used as an anthelmintic agent to kill or aid in the evacuation of parasitic intestinal worms in Mexico and the United States. It's also used to treat bladder problems, as well as for vermifuge and diuretic seeds. Furthermore, due to the presence of phyto-chemical components, pumpkin oil has been shown to protect diabetic nephropathy in several studies. Pumpkin inhibits di-hydro-epi-androstenedione, which aids in the prevention of ovarian and prostate cancer[28].

An investigator and his colleagues measured the physical properties, chemical composition, and fatty acid proportion of pumpkin seeds, finding that they contained 41.59 percent oil, 25.4 percent protein, 5.2 percent moisture, 25.19 percent carbohydrates, 5.34 percent fiber, and 2.49 percent total ash. 66.25 (mg galic acid per kg oil), 1.86 percent, 1.56 percent, and 882.65 (mg tocopherol per kg oil) were the total phenolic compounds, total sterols, waxes, and total tocopherols, respectively[29]. Researchers looked at phytosterol, tocopherol, and squalene levels in pumpkin seeds. The method entails acid hydrolysis and lipid extraction, which is monitored using alkaline saponification before being analysed using HPLC (High performance liquid chromatography). The maximum normal phytosterol is beta-sitosterol, which ranges from 24.90mg per 100 gramme of pumpkin seeds. Squalene was given special attention (89mg per 100 gram) and the total content of oil material in pumpkin seeds was 42.30 percent[30].

Palmitic acid, oleic acid, linoleic acids, and stearic acid were among the fatty acids (FAs.) found in pumpkin seeds, according to Kim and colleagues. *Cucurbita moschata* and *Cucurbita pepo* seeds exhibited significantly higher c-tocopherol levels than *Cucurbita maxima* seeds, which had much higher b-carotene levels. *Cucurbita pepo* seeds had a higher concentration of b-sitosterol than pumpkin seeds. They found that pumpkin seeds (95.85 33.01ppm) had the highest iron content among 11 kinds of seeds and nuts profiled for dietary abundance[31].

Table 3: Nutritional composition of pumpkin seed oil

COMPONENTS	NUTRITIONAL VALUE	RECOMMENDED DIETARY ALLOWANCE (RDA) PERCENTAGE
Carbohyrates	10.71 grams	8 percent
Energy	559 k cal	28 percent
Total fats	49.05 grams	164 percent
Protein	30.23grams	54 percent
Fiber	6grams	16 percent
Cholesterol	-	-
MICRON NUTRIENTS (VITAMINS)		
B_9 (FOLIC ACID)	58 microgram	15 percent
B_3 (NIACIN)	4.8 mg	31.0 percent
B_5	0.7mg	15.0 percent
B_6	0.14mg	11.0 percent
B_2	0.15mg	12 percent
B_1 (THIAMIN)	0.272mg	23 percent
VITAMIN C	0.272mg	3.0 percent
VITAMIN A	16 IU	0.50 percent
VITAMIN E	35.1mg	272 percent
MAJOR MINERALS		
Na^+	7.0 mg	0.5 percent

K^+	809.0mg	17.0 percent
MINERAL DEPOSITS		
C^+	46.0mg	4.5 percent
Cu	1.43mg	148.0 percent
Fe	8.8mg	110.0 percent
Mg	592mg	148.0 percent
Mn	4.54mg	195 percent
P	1232mg	175 percent
Se	9.40 microgram	17.0 percent
Zn	7.8mg	17.0 percent
PHYTOCHEMICALS		
Beta-carotenoid	9 microgram	-
Beta-cryptoxanthin	1 microgram	-
Lutein-zexanthin	74 microgram	-

THERAPEUTIC EFFECTS

HAIR GROWTH IN MEN

Pumpkin seed oil is high in phytosterols, which have been shown to inhibit 5-reductase and have anti-androgenic properties in a rat animal model[12]. 76 patients aged 20–65 years old with mild to moderate hair loss were enrolled in a randomized placebo-controlled double blind study. Patients were randomly assigned to one of two groups: those who received 400 mg pumpkin seed oil capsules daily (n = 37) or those who received a placebo (n = 39). The capsules were given to the patients twice a day, before breakfast and dinner. In the intervention and placebo classes, respectively, about 5 and 7 patients dropped out of the study. Every clinic visit was evaluated for safety and compliance. Compliance was measured using the pill count. After 24 weeks of care, blood samples from patients were used as a baseline. The researchers looked at fasting blood sugar, serum Aspartate Amino transferase (AST), Alanine transaminase (ALT), Gamma glutamyl transferase (GGT), creatinine, and serum-free testosterone. Self-reported progress ratings, investigator tests, and phototrichography hair modifications were used to determine treatment efficacy. In terms of demographic features, there was no significant difference between the two groups. After 24 weeks of treatment by pumpkin seed oil, self-rated improvement score was significantly higher than placebo group (3.4 ± 2.9 vs. 2.1 ± 2.2) (p = 0.013). Self rated satisfaction scores were 3.5 ± 2.9 and 2.3 ± 2.0 for pumpkin seed oil and placebo groups, respectively (p = 0.003).At the 24-week visit, a blinded investigator evaluation revealed that pumpkin seed oil outperformed the placebo community (p 0.001). 44.1 percent of patients improved marginally or moderately after 24 weeks of treatment with pumpkin seed oil, while 51.4 percent and 2.7 percent remained unchanged or deteriorated, respectively. Investigators found that 64.1 percent, 7.7 percent, and 28.2 percent of the placebo sample were unchanged, slightly marginally improved, and deteriorated, respectively. When hair counts and diameters were compared at baseline, 12 weeks, and 24 weeks, there were substantial differences in hair count shifts in the intervention group compared to the placebo group. After 12 and 24 weeks of therapy, men treated with pumpkin seed oil experienced 30 percent and 40 percent mean rises in hair counts from baseline (5 percent and 10 percent in placebo group). After 12 and 24 weeks of intervention with pumpkin seed oil, a net increase of about 25% and 30% was observed, respectively. One patient experienced minor abdominal pain after receiving pumpkin seed oil. In the pumpkin seed oil group, neither the liver enzyme nor the creatinine level remained unchanged. There was no difference in blood pressure or glucose levels between the two classes. Both groups had similar levels of serum- free testosterone[32].

ANTI-DIABETIC EFFECT

The diabetes mellitus is the one of the most prevailing disorder affecting all aged people. The diabetes mellitus is the disorder of metabolic system in which the body does not construct enough insulin or insulin produces but body do not repose properly to insulin. Type I diabetes and Type II diabetes are the two main types of the diabetes. Many studies have shown that pumpkin seeds and pumpkin contain components that help to lower blood glucose levels. Many diabetics avoid pumpkin because of its high carbohydrate content, despite the fact that pumpkin consumption poses no risk. The hypoglycemic and antioxidant effects of combining pumpkin seeds and flax seeds in diabetic rats have been discovered. Histopathological changes include reduced MDA (malondialdehyde and antioxidant enzyme) and increased CAT (Chloramphenicol acetyltransferase), GSH (growth stimulating hormone), and SOD (superoxide dismutase).Glucose, total lipid, triglycerides, and total cholesterol levels in plasma were all significantly unresponsive[33]. It has been discovered that eating a diet rich in pumpkin seeds reduces the elevated levels of the enzymes Alanine aminotransferase (ALT) and Aspartate aminotransferase (AST) in the blood, thus reducing the risk of diabetes. It can be beneficial for diabetes prevention and complications when eaten in regular food[34]. The hypoglycemic effects of protein extracted from various forms of pumpkin seeds from the *Cucurbitaceae* family, including *Cucurbita moschata*, have been determined. The results of an oral glucose tolerance test performed on rats revealed that globulin was the most abundant storage protein, with a calculated 295 milligrams per grams dry matter and the ability to cause a substantial decrease in blood sugar levels (88 to 137.80 percent) [35].

ANTI-HYPERTENSIVE AND HEART PROTECTIVE EFFECT

Pumpkin seeds aid in the relaxation of blood vessels and the reduction of blood pressure. The effects of pumpkin seed oil treatment on chemical-induced hypertension in rats were studied by El-Mosallamy and colleagues[36]. For 6 weeks, the oil (40–100 mg/kg) was provided once a day It has been discovered that eating a diet rich in pumpkin seeds reduces the elevated levels of the enzymes Alanine aminotransferase (ALT) and Aspartate aminotransferase (AST) in the blood, thus reducing the risk of diabetes. Additionally, it reduced elevated levels of MDA (Malondialdehyde) and restored natural levels of NO (Nitric oxide) metabolites. The findings revealed that pumpkin seed oil has a protective effect against pathological changes in the heart and aorta, with the mechanism being due to the production of NO. The amino acid L-arginine is responsible for NO production. The high magnesium content is also linked to a lower risk of heart attack. The seed supplements were found to be as effective as the calcium channel blocker amlodipine, a prescription medication[7].

ANTI-OXIDANT ACTIVITY

Some researchers estimated the total phenolic contents of pumpkin seed oil, which ranged from 25 to 51 mg/kg of pumpkin seed oil. Vanillin, tyrosol, luteolin, sinapic acid, and vanillic acid were among the phenolic compounds found. The maximum antioxidant potential estimated with the aid of the DPPH radical reduction was 62 percent[37]. Nkosi and colleagues investigated the antioxidative properties of isolated pumpkin seed protein in rats fed a low- protein diet for five days. The isolated pumpkin seed protein was supplied to rats that had been given acetaminophen intoxication. The rats were killed 24 hours, 48 hours, and 72 hours after their treatment. The protein isolated from pumpkin seeds had around 80% radical scavenging activity, about 64% chelating activity on Fe2+ ions, and around10% xanthine oxidase inhibition. The use of pumpkin protein isolate increases the CCl_4- induced liver damage, as evidenced by the decreased level of lipid per oxidation and increased antioxidant level[38]. According to one study, pumpkin seed oil protects the small bowel of rats from methotrexate-induced damage through anti-inflammatory and anti-oxidative properties. Oral administration of the oil in combination with ellagic acid or pumpkin seed oil for five days prior to methotrexate treatment reduces small intestine damage, stimulates and increases GSH (growth stimulating hormone) levels and storage, and decreases serum myelo peroxidase, prostaglandin, tissue nitric oxide, malondialdehyde, xanthine oxidase, and adenosine deaminase[39]. Ward and Ainsworth developed the less costly weaning meals in Kenya for malnourished infants that contained enough nutrition from diet, protein, and fat. The porridge was fried, dried, and then mixed with pumpkin seeds powder before being heat treated and stored at room temperature for two months. Protein digestibility was estimated at 82.50 percent, indicating a high-quality protein meal. The food stability was determined by HPLC analysis, and no free water-soluble amino acids were found. The low peroxide value confirms the absence of rancidity, and the viscosity of the porridge is measured with a viscometer[40].

CONCLUSION

Pumpkin seeds oil has evolving bioactive compositions that promote health and human life, according to the findings of this literature review. All of these results lead to a novel concept for developing and innovating nutraceuticals, pharmaceuticals, and cosmeceuticals products from pumpkin seeds oil for a wide variety of applications. Pumpkin seeds are used for cultivation in Pakistan and then discarded as waste. More research and knowledge about their nutritional and therapeutic value are needed so that people can include them in their daily die Aside from its proven medical effects, pumpkin seed oil has other potential health benefits. They are thought to prevent kidney stones, treat incontinence, alleviate depression, prevent Osteoporosis, improves ocular health, and nourishes skin, etc; however, there are insufficient studies in these areas. Its combination with other healthy botanicals could result in supplements that are appealing[41]. More clinical research is needed to fully grasp and utilize pumpkin seed oil's nutritional potential.

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