Chapter 29
Pumpkin Bio-Wastes as Source of Functional Ingredients

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Abbreviation

ANN	Artificial Neural Networks
CSE	Conventional Solvent Extraction
DLQI	Dermatology Life Quality Index
DM	Diabetes mellitus
DPP-IV	dipeptidyl peptidase IV
DPS	Defatted Pumpkin Seeds
G-6-P DH	Glucose-6-phosphate dehydrogenase
GSH	Glutathione
HECSI	Hand Eczema Severity Index
MDA	malondialdehyde
NO	Nitric oxide
NSAID	Nonsteroidal anti-inflammatory drug
PP	Pumpkin Peel
PPAR-γ	Peroxisome proliferator-activated receptor gamma
PPE	Pumpkin Peel Extract
PPF	Pumpkin Peel Flour
PPP	Pumpkin Peel Powder
PS	Pumpkin Seeds

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PSC	Pumpkin Seed Cake
PSCF	Pumpkin Seed cake Flour
PSO	Pumpkin Seed Oil
PSP-1	Protein Bound Polysaccharide-I
PTP-1B	Protein-tyrosine Phosphatase
RSM	Response Surface Methodology
SFE	Supercritical Fluid Extraction
SWE	Subcritical Water Extraction
TBHQ	*tert*-butyl hydroquinone
UAE	Ultrasound-assisted Extraction

29.1 Introduction

Food processing wastes are those end products of various food processing industries that have not been recycled or used for other purposes. They are the non-product flows of raw materials whose economic values are less than the cost of collection and recovery for reuse; therefore discarded as wastes. These wastes could be considered valuable by-products if there were appropriate technical means and if the following products' value exceeded the reprocessing cost.

A tremendous amount of food wastes is generated worldwide, largely through the industry of fruit and vegetable, oils, fermentation, dairy products, and seafood. Wastes are generated at the different food processing stages: pre- and post-harvest, food manufacturing, packaging, retail/catering, and consumer/household. The loss can reach up to 25% or more worldwide (Parfitt et al., 2010). In the developed leading countries, such as Japan, the USA, and United Kingdom, more than 40% was discarded as food waste.

Reprocessing of food wastes offers the potential to recycle these by-products to add values rather than discarding these residues to the environment, which causes detrimental environmental pollution. Successful food waste recycling includes (a) recover of natural by-products, beneficial economic use, (b) promoting marketability, (c) the use of Hi-Tec in reprocessing, and (d) enterprise development, acceptable and economically feasible (Al-Okbi et al., 2017).

Pumpkin is a well-known edible plant; almost all pumpkin plants are consumed by humans and used as animal feed. For humans, the pulp is used as a table vegetable in pies and soups. The seeds are popular snacks, and pumpkin seed oil is used for cooking and salad dressing. Pumpkin pie is a traditional staple of the Thanksgiving holiday in Canada and the United States. The carving of the rind of the hollowed-out fruit into jack-o'-lanterns is a popular activity on Halloween. Pumpkins are believed to have originated in Mexico and Central America or Peru, Ecuador, and Colombia (Orzolek, 1995). The worldwide production of pumpkin, squashes, and gourds in the year 2018 was 27.643.932 metric tons from an area of 2.042.955 ha and 6.222.000 metric tons for the Mediterranean countries from an area of 179,928 ha (Considine & Considine, 2013).

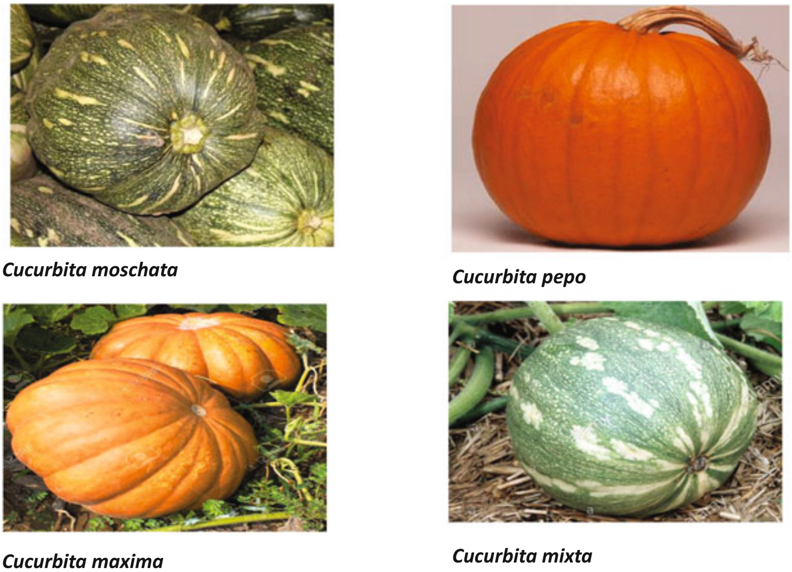

Fig. 29.1 The most common types of pumpkin fruits

Pumpkin is a cultivar of winter squash of large-fruited varieties of several species of vine plants of the genus *Cucurbita*. Pumpkin is a round, pulpy, orange, or orange-yellow fruit with a slightly thick or smooth ribbed rind. Pumpkins of the genus *Cucurbita* belong to the family Cucurbitaceae. Members of the Cucurbitaceae are commonly known as melons, gourds, or cucurbits, including crops like cucumbers, squashes (including pumpkins), luffas, melons, and watermelons. Pumpkin is referred to various varieties (Fig. 29.1) of the following species *Cucurbita pepo, C. moschata, C. mixta,* and *C. maxima*.

Pumpkin is a plump, botanical berry known as a pepo and highly nutrient-dense food. It is low in calories but rich in vitamins and minerals. It has been frequently used as functional food or herbal medicine. The fruits of pumpkins range differently in size, shape, color, and appearance; they range from less than five pounds to varieties over 40 to 60 pounds (Orzolek, 1995) or sometimes get larger.

The distinction between squash and pumpkin is not a botanical distinction; since some squash shares the same botanical classifications as pumpkins, the names are frequently used interchangeably (Considine & Considine, 2013).

Pumpkin showed a wide range of biological and medicinal values such as anti-inflammatory, antioxidant, anticancer, antiangiogenesis, hypolipidemic, and antidiabetic activities. Chemically, it showed the presence of tocopherols, carotenoids, triterpenes, phytosterols, lignans, and glycolipids (Isutsa & Mallowa, 2013; Kim et al., 2012; Wang et al., 2012; Ayyildiz et al., 2019), in addition to protein, low carbohydrates, and fixed oil (Özbek & Ergönül, 2020). All pumpkin parts, including edible seed, fruit, and greens of pumpkin, have broad nutritional values as medicine or food, mainly because of their high amount of β-carotene and a moderate amount of carbohydrates, vitamins, and minerals (Rakcejeva et al., 2011).

29.2 Chemical Composition and Bioactive Compounds of Pumpkin Fruits

Pumpkin seeds are one of the most important agro-industrial waste, attributable to the presence of interesting natural bioactive compounds with potential effects on human health, such as carotenoids, tocopherols phytosterols, phenolics, antidiabetic polysaccharides, minerals, vitamins, antifungal proteins, essential and nonessential amino acids, pectin and fibers (Fig. 29.2). Pumpkin seeds and seed oil could be considered ideal ingredients in pharmaceutics and cosmetics. Previous studies confirmed that pumpkin seed oils are interesting vegetable oils with important nutritional value, related to the presence of monounsaturated fatty acids (MUFA), polyunsaturated fatty acids (PUFA), phytosterols, and carotenoids (Ayyildiz et al., 2019; Özbek & Ergönül, 2020). Variation in the oil yield could be attributed to differences in plant variety, cultivation climate, ripening stage, and the extraction method (Nyam et al., 2009).

29.2.1 Pumpkin Nutritional and Medicinal Components

29.2.1.1 Fatty Acids

A lot of differences among the species and/or varieties of *Cucurbita* spp. grown in different world areas were reported (Al-Khalifa, 1996; Bravi et al., 2006; El-Adawy & Taha, 2001; Glew et al., 2006; Haiyan et al., 2007; Parry et al., 2008; Mitra et al., 2009). The significant variability of fatty acids (FAs) composition of pumpkin seeds

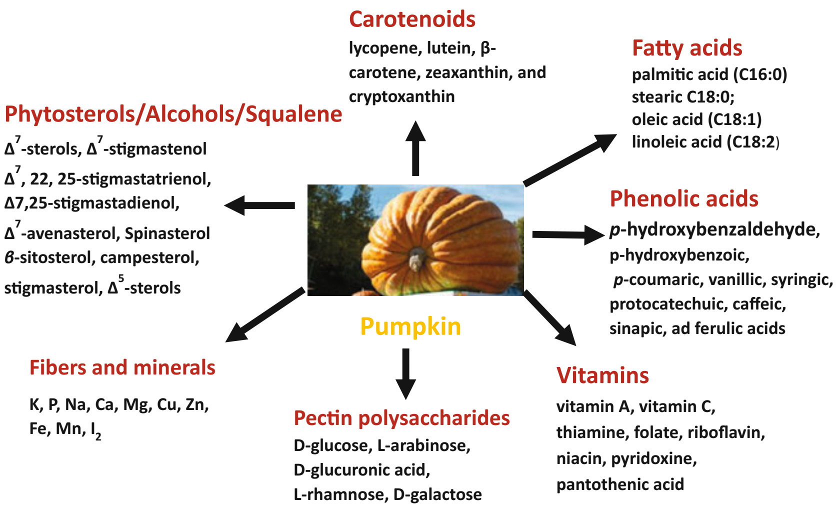

Fig. 29.2 Nutritional and medicinal components of pumpkin

is affected by the variety of the cultivar, the degree of ripeness, and the growth conditions and preparation method (Murkovic et al., 1996a). The fatty oil content of pumpkin seeds is about 45-60% (Murkovic et al., 1996a; Tsaknis et al., 1997). The main FAs are palmitic acid (C16:0), oleic acid (C18:1), and linoleic acid (C18:2), which account in total for about 90%. Pumpkin oil contains a high amount of PUFAs or essential FAs as detected by GC/MS analysis with more than 50% of the total FAs, which confirms undoubtedly the good nutritional quality of pumpkin oil (Andrikopoulos et al., 2004) and also of about 19.4% saturated fatty acids (Andrikopoulos et al., 2004). The primary saturated fatty acids were palmitic acid (9.5–14.5%) followed by stearic acid (3.1–7.7%), while the primary unsaturated fatty acids were linoleic acid followed by oleic acid with concentrations of (21–55.6%) and (20.4–60.8%), respectively. High amounts of essential linoleic acid suggest that the pumpkin seed oil is highly nutritious.

Oleic acid was the predominant FA of *Cucurbita* spp. Nigerian origin, whereas linoleic acid accounts for nearly one-third of the total FA in pumpkin seeds, and only trace amounts of α- and γ-linolenic acids were found (Glew et al., 2006). The dominant FAs found in *Cucurbita* spp. Italian origin were palmitic (C16:0), stearic (C18:0), oleic (C18:1), and linoleic acids (C18:2) (Murkovic et al., 1996a). The dominant FAs found in *C. pepo* L. convar. Citrullina-var. styriaca were palmitic (C16:0; 9.5–14.5%), stearic (C18:0; 3.1–7.4%), oleic (C18:1; 21.0–46.9%) and linoleic acids (C18:2; 35.6–60.8%). El-Adawy and Taha (2001) detected linoleic acid as the main fatty acid followed by oleic acid in pumpkin seed oil. Besides, Stevenson et al. (2007) studied the FAs composition of pumpkin seed oil extracted from twelve cultivars of *C. maxima* and reported significant differences among them ranged from 10.9% to 30.9% for gadoleic, linoleic, oleic, and stearic acid content of oil. They reported that the oil contained about 19.4% saturated FAs with two dominants; palmitic acid, C 16:0 (10.7–13.3%), and stearic acid, C 18: 0 (5.28–7.7%). At the same time, the high amounts were in unsaturated fatty acids with a total content of 80.7%. Among the monounsaturated acids (MUFA), oleic acid, C 18:1, occurs in the greatest amounts (20.4–38.4%). Regarding PUFAs, linoleic acid, C18:2 was the dominant acid (39.8–59.2%). Habib et al. (2015) determined the proximate composition of powdered seed and the lipid composition of the oil of *C. maxima* collected in Bangladesh and found that seed oil contained a high amount of oleic acid, 40.5%, while stearic acid, palmitic acid, and linoleic acid contents were found to be 27.0%, 17.3%, and 14.9% respectively. Siano et al. (2015) reported that saturated FA and MUFA of *C. maxima* produced in southern Italy showed similar values (25.2% and 25.5%, respectively), while the PUFA content was 48.1%. They found that the main FA of southern Italian pumpkin (*C. maxima*) seed oil was linoleic acid (47.4%), followed by oleic (25.5%) and palmitic (17.5%) acids. Also, Orsavova et al. (2015) reported the high content of palmitic acid (C16:0; 4.6–20.0%), oleic acid (C18:1; 6.2–71.1%) and linoleic acid (C18:2; 1.6–79%). Minor FAs (contents lower than 0.5%) of Berrettina pumpkin seed oil were myristic, palmitoleic (C16:1n-7), margaric (C17:0), heptadecenoic (C17:1n-7), arachidic (C20:0), behenic (C22:0), and lignoceric (C24:0) acids. Türkmen et al. (2017) studied the FAs composition of edible pumpkin seed collected from a different

Turkey region and reported that the unsaturated fatty acid content was 73.3% and comprised of 18.4% oleic acid, 52.6% linoleic acid, and 1.27% linolenic acid. The saturated fatty acids concentration was 27.7% and consisting of 16.4% palmitic acid and 11.1% stearic acid. Linoleic acid was the principal fatty acid, followed by oleic, palmitic, and stearic acids.

Montesano et al. (2018) investigated *C. maxima* L. (var. Berrettina) pumpkin seed oil and observed that PUFA and MUFA fractions were the most abundant (37.2% and 41.7%, respectively). The main FAs were oleic (C18:1n-9) and linoleic (C18:2n-6) acids. In another study conducted by Rezig et al. (2018), the chemical composition of *C. maxima* 'Béjaoui' Tunisian cultivar, investigated and they found that the major FAs were oleic, linoleic, and palmitic acids ranging from 28.1% to 46.6% of the total amount of fatty acids for cold-pressed seed oil and 30.5–43.8% for pumpkin seed oil. Amin et al. (2019) studied the FAs profile using GC/MS of two varieties (indigenous and hybrid) of pumpkin (*Cucurbita maxima*) in Bangladesh and reported that the hybrid type was rich in saturated fatty acid (capric, myristic, and stearic acids). In contrast, unsaturated fatty acids (oleic, linoleic, and linolenic acids) were rich in the seed of indigenous type. Morocho et al. (2019) also reported that ethanol is a good solvent to identified fatty acids in pumpkin seeds, where linoleic, oleic, stearic, and palmitic acids showed the highest peaks in pumpkin seed of *C. maxima* and might be it supported the genetic characterization of the pumpkin.

29.2.1.2 Phytosterols, Alcohols and Squalene

The unsaponifiable fraction of pumpkin seed oil showed other essential components, viz., sterols and alcohols. Sterols, up to 0.5% of the oil (55–60% of the unsaponifiable fraction) and supposed to confer to this oil a beneficial effect in the prostatic ailments' treatment and prophylaxis and the urinary bladder disorders (Sandra et al., 2006). Predominantly Δ^7-sterols are considered the key active constituents of pumpkin seed in the treatment of benign prostatic hyperplasia. Much smaller amounts of Δ^5 and Δ^8-sterols are also present. The predominant sterols of pumpkin native to central Italy, locally known as "Berrettina" (*C. maxima* L.) are Δ^7-sterols, in particular Δ^7, 22, 25-stigmastatrienol, Δ7,25-stigmastadienol, and spinasterol, which accounted for about 76.8% of the total sterols, followed by Δ^7--avenasterol and Δ^7-stigmastenol. The Δ^5-sterols were represented by campesterol, stigmasterol, and only a trace of cholesterol. In contrast, sitosterol was also the sterol marker in *C. pepo* seed oil; it ranges from 75.7% to 87.3% (Nyam et al., 2009; Rezig et al., 2019). These differences between the contents of Δ^5- and Δ^7-sterols could be attributed to the maturity stage of seeds or the solvent used in the extraction procedure.

Contrary to the other vegetable oils with Δ^5-sterols (*β*-sitosterol, campesterol, and stigmasterol) as the major components, Wenzl et al. (2002) showed the presence of specific Δ^7-phytosterols, which is typical for few plant families (e.g., Cucurbitaceae), that provide a fingerprint for detection of adulteration.

Triterpenic alcohols are also constituent of the unsaponifiable fraction of the seed oil, where butyrospermol, obtusifoliol, β-amyrine, and cycloartenol reported in pumpkin seeds oil native to central Italy, locally known as "Berrettina" (*C. maxima* L.) (Montesano et al., 2018). Additionally, squalene (39-46%) is the characteristic constituent of the unsaponifiable fraction of the seed oil and can be used as a marker for the differentiation of oils obtained from other seeds (Kreft et al., 2009).

29.2.1.3 Phenolic Acids

Pumpkin is considered to be a rich source of phenolics, where *p*-hydroxybenzaldehyde, p-hydroxybenzoic, *p*-coumaric, vanillic, syringic, protocatechuic, caffeic, sinapic, and ferulic acids were reported in *C. maxima* and *C. pep* seeds (Bombardelli & Morazzoni, 1997; Nyam et al., 2009; Krimer-Malešević, 2020). Eight phenolic glycosides, cucurbitosides F-M, were isolated and identified from the seeds of *C. pepo* (Li et al., 2005). The phenolic compounds in whole hull-less seed, its skin and oil cake meal, dehulled kernel, and hull of *C. pepo* were identified and quantified by HPLC-DAD. Ferulic, protocatechuic, *p*-coumaric, trans-sinapic, *p*-hydroxybenzoic, and vanillic acids with p-hydroxybenzaldehyde were detected. *p*-Hydroxybenzoic acid was the main phenolic acid in all samples, with 34.7%, 51.4%, 51.8%, 52.0%, and 67.4% found in hull-less seed, skin, hulls, oil cake meal, and dehulled kernels, respectively (Peiretti et al., 2017). Rezig et al. (2012) identified six phenolic acids: vanillic, caffeic, ferulic, *p*-coumaric, syringic, and protocatechuic the syringic acid was predominant phenolic acid (7.96 mg/100 g). Phenolics were extracted from *C. pepo* seeds and characterized by HPLC-DAD. The results revealed that the pumpkin seeds extract showed higher contents of total phenolics than the amaranth grains extract and the spectra of pumpkin extract (Lalnunthari et al., 2019). Kulczyński and Gramza-Michałowska (2019) investigated 15 pumpkin varieties belonging to the *C. pepo* and *C. moschata* species for their phenolic profiles, in which rutin, 4-hydroxybenzoic acid, gallic acid, caffeic acid, protocatechuic acid, chlorogenic acid, and vanillic acid were identified in all cultivars.

29.2.1.4 Carotenoids

Pumpkins are an essential source of carotenoids, where β-carotene is the major one in most species, with concentrations of more than 70 μg/g (Rezig et al., 2012). Analysis performed directly on the seed oils detected carotenoids such as lycopene, lutein, β-carotene, zeaxanthin, and cryptoxanthin (Kreft et al., 2009; Provesi & Amante, 2015; Murkovic et al., 2002). These pigments possess have a pro-vitamin A activity. Carotenoids are essential as a vitamin A precursor, which is essential to improve night vision, growth, and other functions. β-Carotene in pumpkins has an immune stimulant, cardioprotective and cytotoxic activities due to its antioxidant role (Montesano et al., 2018). Kulczyński and Gramza-Michałowska (2019) and identified zeaxanthin, β-carotene, and lutein and in 15 cultivars belonging to the *C. pepo* L. and *C. moschata* Duchesne ex Poir species.

29.2.1.5 Fibers and Minerals

C. maxima var. "Béjaoui" seeds were found to contain significant amounts of minerals and fibers. Potassium was the most known element, followed, in descending order, by phosphorus, sodium, calcium, magnesium, copper, zinc, iron, manganese, and iodine (Rezig et al., 2012; Azevedo-Meleiro & Rodriguez-Amaya, 2007). In addition to selenium, which is of particular importance, its content ranges between 0.08 and 0.4 μg/g. It is worth noting that this is one of the highest values found in plants (Kreft et al., 2009; Kreft et al., 2002), sometimes reaches up to 1.29 μg/g (Glew et al., 2006). Pumpkin seeds contain also a relatively large amounts of magnesium (5690), zinc (113), copper (15.4), molybdenum (0.805) and another mineral including phosphorus (15700), calcium (346), iron (106), manganese (49.3), aluminum (9.21), barium (1.16), cobalt (0.29), strontium (1.83), nickel (0.53), and arsenic (0.45) (μg/g dry weight), with low amount of calcium in the seeds. According to Suphakarn et al. (1987), 100 g roasted pumpkin seeds contain calcium 25.9 mg, phosphorus 955 mg, and iron 8.06 mg. *C. pepo* Kakai is rich in minerals: phosphorus 17.831, potassium 13.736, magnesium 5.688, calcium 1.643, iron 0.211, sodium 0.332, copper 0.016, zinc 0.190, and manganese 0.080 (g/kg) (Provesi & Amante, 2015). Potassium, sodium, and calcium were the main minerals in 15 pumpkin varieties belonging to the *C. pepo* and *C. moschata* species (Kulczyński & Gramza-Michałowska, 2019).

29.2.1.6 Pectin

Pumpkin contains 1.26% pectin (as calcium pectate). Two different heating methods were used for isolation of pectin from *C. moschata* Duch ex. Poir. The first method was by boiling at atmospheric pressure at 95 °C for 60 and 120 min or by autoclaving at 15 psi and 121 °C for different times using distilled water. The results revealed that autoclaving at 121 °C and 15 psi for 20 could be the appropriate method for pectin extraction from pumpkin (Ramachandran et al., 2017). To maximize pectin's yield, the effect of blanching before acid hydrolysis, time, and temperature were evaluated for *C. maxima* peels. The highest pectin content was extracted using 0.1 N hydrochloric acid for 1 h at 80 °C, 1.25 pH (Dona, 2019).

29.2.1.7 Polysaccharides

The water-soluble polysaccharides of pumpkin fruits were characterized by fourier transform infrared spectroscopy and revealed D-glucose, L-arabinose, D-glucuronic acid, L-rhamnose, and D-galactose (Junli et al., 2010). The polysaccharides from pumpkin were identified as D-arabinose, D-mannose, D-glucose, and D-galactose by gel filtration chromatography, gas chromatography, fourier transform infrared spectroscopy, and nuclear magnetic resonance spectroscopy (Zhu et al., 2015). The polysaccharides were obtained from pumpkin seeds by hot water extraction followed by precipitation with ethanol and analyzed by GC, which revealed mannose, glucose, and galactose e in the molar ratio of 1.0:12.9:5.3 (Wang et al., 2017).

29.2.1.8 Proteins

Proteins are abundantly present in pumpkin seeds, with values between 31% and 51% (7, 27) with a good content of glutamic acid (104), arginine (93.2), aspartic acid (52.8), leucine (40.9), serine (31.7), phenylalanine (31.4), glycine (28.3), valine (28.2), alanine (23.4), isoleucine (23.0), tyrosine (22.1), lysine (22.0), proline (20.2), threonine (18.4), tryptophan (15.3), histidine (13.8), methionine (12.4), and cysteine (6.73) (mg/g dry weight) (Glew et al., 2006). According to Mansour et al. (1993), the pumpkin seeds have an excellent pattern of amino acids, with high levels of most essential amino acids, including leucine 6.13; lysine 5.20; phenylalanine 4.00; histidine 3.62; valine 3.40; tyrosine 2.94; threonine 2.75; isoleucine 2.66; tryptophan 1.56; cystine 1.52; methionine 1.25 and non-essential amino acids: glutamic acid 18.13; arginine 16.70; aspartic acid 10.19; glycine 5.86; serine 5.46; proline 4.34; and alanine 4.29 (in g per 16 g). Rare amino acids have also been found in *Cucurbita pepo* seeds, such as γ-aminobutyric acid, *m*-carboxyphenylalanine, citrulline, ethyl-asparagine, and β-pyrazolalanine (Fruhwirth & Hermetter, 2007).

29.2.1.9 Vitamins

Pumpkin seeds showed the presence of several vitamins, such as vitamin A, vitamin C, thiamine, folate, riboflavin, niacin, pyridoxine, and pantothenic acid (Azevedo-Meleiro & Rodriguez-Amaya, 2007; Wang et al., 2017; Peter et al., 2013), in addition to, tocopherol or vitamin E. As reported, vitamin E content in the seed oil of *C. maxima* var. Bejaoui is higher than that of *C. pepo* L. seed oil but much lower than those of other pumpkin seed oils (Stevenson et al., 2007). In the seed oil of the Bejaoui variety, δ-tocopherol was the main component and represented about 42.2% of total tocopherols, followed by α-tocopherol γ-tocopherol (Rezig et al., 2012). The concentration of γ-tocopherol is dominant from the 100 lines of *C. pepo* L. *convar. Citrullina* var. *styriaca* range from 41 to 620 mg/kg (Murkovic et al., 1996b). Kulczyński and Gramza-Michałowska (2019) studied 15 pumpkin cultivars for the content of vitamins B1, C, and folates. The results revealed that all cultivars had a high content of vitamin C in addition to thiamine, but its concentration was relatively low (0.15–0.72 mg/100 g). Regarding the folates' content, folic acid, 5-methyltetrahydrofolate, and tetrahydrofolate were tested for their presence in the selected cultivars, and only 5-methyltetrahydrofolate was detected.

29.3 Biological and Functional Properties of Extracts and Bioactive Compounds from Pumpkin Bio-Wastes

29.3.1 Nutraceutical and Antioxidant Activity

Oxidative stress is an imbalance between the production of free radicals and the body's ability to get rid of these harmful items, contributing to several diseases such as cancer, diabetes, and cardiovascular diseases. Antioxidants can help counteract the free radical's accumulation, which prevents and manage many diseases. Pumpkin by-products are rich sources of many antioxidant components such as flavanoids, tocopherols, phenolic acids, and carotenoids, which maintain adequate oxidative stability (Abou Seif, 2014; Hernández-Santos et al., 2016; Xanthopoulou et al., 2009). For instance, Saavedra et al. (2015) revealed that pumpkin peels have a higher content of total phenolics than seeds (11 and 6.1 mg GAE/g dry weight, respectively) (Saavedra et al., 2015). Also, carotenoids are present in higher amounts in peels than seeds, particularly in *C. maxima*, where β-carotene is the most common in all pumpkin species (Cuco et al., 2019; Kim et al., 2012). Moreover, Blanco-Díaz et al. (2015) proved that pumpkin peels have chlorophylls content 21-fold higher than pulp (Blanco-Díaz et al., 2015). On the other hand, pumpkin pulp contains high content of carotenoids (171.9 mg/g d.w.), vitamin C (2–10 mg/100 g), and vitamin E (9–10 mg/100 g) (Kaur et al., 2019). To our knowledge, not all phenolic constituents of peels have been quantified yet, while most flavonoids, phenolic acids, and lignans were quantified in seeds (Ghisoni et al., 2017; Nawirska-Olszańska et al., 2013), while the main compounds quantified were vanillic and caffeic acids, with concentrations of 0.6 and 0.41 mg/100 g oil, respectively (Nyam et al., 2013) and sesamol with 47 mg/kg (Ghisoni et al., 2017). Besides, when different pumpkin species were investigated, it was observed that the seeds of *C. pepo* and *C. moschata* had higher tocopherols content than those of *C. maxima* (Kim et al., 2012).

Antioxidant activity of different pumpkin by-products was investigated by different authors (Blanco-Díaz et al., 2015; Nawirska-Olszańska et al., 2013; Saavedra et al., 2015). Yasir et al. (2016) revealed that using acidified methanol in the extraction of pumpkin seeds showed a higher increase in extraction yield by 1.4 to ten-fold, higher phenolic content (149.5–396.4 mg GAE/g) as well as higher DPPH· radical scavenging activity and enhancement of the geno-protective activity using the pBR322 plasmid assay (Yasir et al., 2016). Besides, El-Boghdady (2011) discussed the antioxidant effect of pumpkin seed oil (40 mg/kg b.w.) in methotrexate-induced bowl destruction rats, where it significantly decreased the damage of the small intestine as well as reduced serum levels of prostaglandin, nitric oxide, malondialdehyde, xanthine oxidase, and adenosine deaminase. Also, it increased the level of GSH and its storage (El-Boghdady, 2011). Nkosi et al. (2006) examined the antioxidant role of a protein isolate (1 mL/kg b.w.) from seeds oil of *C. pepo* in CCl_4-induced liver damage rats, where the protein isolate showed an increase in the activities of catalase, SOD, glutathione peroxidase enzymes and the total antioxidant capacity in plasma as well as G6Pase in liver tissues. MDA level in liver tissues was decreased (Nkosi et al., 2006). Furthermore, Elfiky et al. (2012) revealed that oral treatment with pumpkin seed oil (4 mL/kg b.w.) in azathioprine-induced genotoxicity rats decreased ROS, frequencies of Mn-PCEs, DNA fragmentation, total sperm abnormalities and increased sperm count, percentage of PCEs as well as glutathione content in testis. Besides, random amplified polymorphism of DNA (RAPD) showed distinct differences in animal groups intoxicated with azathioprine before and after pumpkin seed oil treatment, which reflected a DNA protective effect of pumpkin seed oil (Elfiky et al., 2012). In another study conducted by Gill and Bali (2011), a new isolated tetracyclic triterpenoid (19-(10 → 9β)-abeo-10α-lanost-5-ene) from pumpkin seeds oil showed the highest antioxidant activity using the DPPH· method with the percentage of inhibition of 72.8% at 300 μg/mL when compared to vitamin C (Gill & Bali, 2011). The UPLC-Q-TOF-MS was used to analyze pumpkin seeds oil of 12 varieties belonging to the species *Cucurbita maxima* Duch. *Cucurbita pepo* showed the identification of many phenolic compounds (*p*-coumaric, caffeic acid tri-hexoside, *p*-hydroxybenzoic acid, caffeic acid derivative, vanillic acid, sinapic acid, and dihydroxybenzoic acid) that might be responsible for their potent antioxidant activity shown in DPPH·, $ABTS^{+}$ and FRAP assays (Nawirska-Olszańska et al., 2013).

29.3.2 Anticancer Activity

Nowadays, the most prevailing health crisis is cancer. It is a challenge for many professionals and researchers to produce preventive and curative approaches to manage cancer. Different pumpkin by-products proved to possess cytotoxic and antitumor activities. Seeds oil of *C. pepo* showed good inhibition against MCF7 (breast carcinoma) cell line with IC_{50} of 0.4 mg (Adnan et al., 2017). In a comparative study between the cytotoxic activity of different pumpkin parts (flesh, peel, seed, and defatted seed extracts against HEPG2 (liver carcinoma) and MCF7 cell lines, all extracts exhibited potent cytotoxic activity against HEPG2 with IC_{50} values ranged between 0.60 and 5.03 μg/mL, and against MCF7 with IC_{50} in the range of 0.40–1.01 μg/mL (Badr et al., 2011).

Concerning the polysaccharide composition of pumpkin peels or pulp, pectin proved to be the most promising one, where many authors stated the bioactive pectin as a potent polysaccharide in cancer prevention (Rico et al., 2020). In a comparative study, the alkali-soluble fraction of pumpkin peels showed the highest pectin content (25 g/100 g) in contrast to water and EDTA fractions (Jun et al., 2006). Moreover, pumpkin seeds are rich in phytoestrogenic compounds that can be a good candidate for preventing hormone-dependent cancer. For instance, the anticancer activity of the seeds extract on MCF7 as well as Jeg3 and BeWo (chorionic carcinoma) was evaluated and the results revealed a good cytotoxic activity (at only low and moderate doses: 10 or 50 μg/mL) and high estradiol production. These results suggested that pumpkin seeds that are rich in lignans and flavones, exerted a biphasic effect *via* different pathways with both estrogenic and antiestrogenic actions like many phytoestrogenic compounds (Richter et al., 2013). Also, hydroalcoholic extract of *C. pepo* seeds revealed an inhibitory effect on the proliferation of cancerous prostate cell lines (IC_{50}: 177.05 μg/mL), but these effects were not mediated by hormone receptors (Medjakovic et al., 2016). Furthermore, ethanol and aqueous extracts of *C. pepo* seeds were evaluated for their cytotoxic activity in the LNCaP prostate cancer cell line. The results showed a decrease in cell viability (IC_{50}: 55 and 49 μg/mL, respectively), with inhibition of oxidative stress and an increase in cell apoptosis (Rathinavelu et al., 2013). An *in vivo* study by Gossell-Williams et al. (2006) showed that pumpkin seeds oil (2.0 mg/100 g b.w.) as a dietary complement inhibited testosterone-induced benign prostatic hyperplasia in Sprague dawley rats (Gossell-Williams et al., 2006). In a randomized, double-blind, placebo-controlled trial on 47 patients with benign prostatic hyperplasia (age: around 50 years old), the group received pumpkin seeds oil (320 mg/day), revealed a reduction in international prostate symptoms score, an improvement in quality of life score and a reduction in serum prostate-specific antigen, compared to the baseline value (Hong et al., 2009). Moreover, components like moschatin and cucurbitacins were isolated and identified in several pumpkin seeds; they proved to induce cancer cell apoptosis *via* PARP, JAK/STAT, and MAPK pathways (Lestari & Meiyanto, 2018). For instance, moschatin isolated from *C. moschata* seeds suppressed the growth of M21 melanoma cells with IC_{50} of 0.26 nM (Xia et al., 2003). In addition, MAP2 and MAP4 (microtubule-associated proteins) identified in the seeds, showed a potent inhibition of K-562 growth (leukemia cell line) (Kaur et al., 2019). Furthermore, cucurmosin isolated from the sarcocarp of *C. moschata* showed potent cytotoxicity activity against three cancer cell lines of both human and murine origin labeled from Type 1 ribosome-inactivating proteins (Yadav et al., 2010). At last, when a food frequency questionnaire was developed, and a group of women susceptible to cancer was fed on pumpkin seeds and sunflower seeds, postmenopausal risk of breast cancer was reduced (Zaineddin et al., 2012).

29.3.3 Antimicrobial Activity

Different microbes like bacteria, parasites, viruses, and fungi can develop many diseases and lead to death. The seed oil of pumpkin showed different antimicrobial activities, where cardiac glycosides, resins, terpenoids, and saponins in the seed oil may have a role in its antimicrobial action. For instance, *C. pepo* seed oil revealed a potent antifungal activity against *Fusarium oxysporium* and *Trichoderma reesei* (Sood et al., 2012). Moreover, *C. pepo* seed oil revealed a high inhibition zone (60%) against *Escherichia coli* and *Staphylococcus aureus* (Adnan et al., 2017). The seed oil (2%) inhibited the growth of *Aeromonas veronii, Candida albicans, Enterociccus feacalis, Escherichia coli, Salmonella enterica, Typhimurium,* and *Staphylococcus aureus* (Hammer et al., 1999). Furthermore, it was suggested that linoleic and oleic acids in the seed oil might have potent antibacterial activity against *S. aureus* with a zone of inhibition of 15 mm (Adnan et al., 2017). The methanol and petroleum ether extracts of pumpkin seeds with concentrations of 10, 50, 100, 200, 500, 1000 μg/mL were found to be effective against *Bacillus subtilis, S. aureus,* and *E. coli* (Adnan et al., 2017). In another study, Park et al. (2010) isolated a new protein (pr-1) from pumpkin rinds, which showed potent anti-fungal activity. However, it did not inhibit *E. Coli* and *Staphylococcus* (Park et al., 2010). Moreover, methanol and ethyl acetate extracts of pumpkin flowers were analyzed to contain many flavanoids such as quercetin-3,4`-O-β-D-diglucopyranoside, 3,4-dihydroxy methyl benzoate, isorhamnetin 4-O-β-D glucopyranoside, 3,4-dihydroxybenzoic acid, isorhamnetin, quercetin, myricetin, quercetin-4`-O-β-D-glucopyranoside and 5,7dihydroxy,3,6,3'trimethoxyflavone. These flavanoids were supposed to be responsible for the extracts' antifungal activity against *A. flavus* (Mohamed et al., 2009).

In a comparative study between the antimicrobial activity of different pumpkin parts (flesh, peel, seed, and defatted seed) using the agar disc method (100 μg/mL extract/disc), extracts of peel and flesh revealed moderate antifungal activity against *Bacillus subtilis* and *Bacillus cereus*, while the seed oil exhibited intense antifungal activity against *Saccharomyces cerevisiae*. However, the defatted seed oil did not display any antimicrobial activity (Badr et al., 2011). Furthermore, cucurmoschin an isolated peptide from pumpkin seeds, showed a potent inhibition to *Botrytis cinerea*, *F. oxysporum,* and *Mycosphaerella arachidicola* at the dose of 375 mg (Wang & Ng, 2003). In another study, pumpkin seed oil was proved to have potent antimicrobial activity against *Pseudomonas aeruginosa*, *C. albicans*, *Acinetobacter baumanii*, *Enterococcus faecalis, Klebsiella pneumonia, E. coli,* and *Staphylococcus aureus* (Ahmad & Khan, 2019). Moreover, different proteins extracted from pumpkin rinds and seeds revealed potent antimicrobial activity. The seeds showed antibacterial activity against *Staphylococcus aureus,* and *Bacillus subtilis*, the inhibition growth zone diameter was 10.0 and 8.0 mm, respectively. For the rinds, it showed antimicrobial activity (27.5 and 25.0 mm) against *Penicillium chrysogenum* and *Aspergillus flavus*, respectively, (Abd El-Aziz & Abd El-Kalek, 2011).

29.3.4 Antidiabetic and Hypocholestremic Activity

Diabetes mellitus (DM) is one of the primary disorders affecting all aged communities. It is a metabolic system disorder where; the body does not produce enough insulin (Type I diabetes), or the body does not respond appropriately to the insulin produced (Type II diabetes). Besides, hypercholesterolemia is another aging disorder, where its prevalence is determined as a risk factor for DM and glucose intolerance. Pumpkins by-products are a rich source in many nutraceuticals that can manage DM and hypercholesteremia. For instance, high dietary fibers in pumpkin peels can help prevent DM and hypercholesteremia (Rico et al., 2020).

Pumpkin kernels were found to be rich in phytosterols (265-289 mg/100 g) (Phillips et al., 2005), and these phytochemicals have a significant hypocholesterolemic effect (Rajasree et al., 2016). Also, a hypoglycemic effect was obtained for alloxan-meditated diabetic rats with flax and pumpkin seeds combination. This mixture ameliorated antioxidant enzyme activities and level of GSH as well as considerably decreased MDA level. Also, it resulted in a subsequent recovery towards the normalization of liver enzymes (Makni et al., 2011). Another mixture of pumpkin seeds with flex seeds or purslane seeds was proved to have an anti-atherogenic effect by reducing total cholesterol, triglycerides, and total lipids in both liver and serum of rats fed 2% cholesterol diet (Barakat & Mahmoud, 2011).

Furthermore, the hypoglycemic effect of tocopherol isomers in the seed oil (2 and 5 g/kg b.w.) was evaluated in PX-407-induced type 2 diabetic rats. Additional, HYBRID and FRED docking were performed for them against PTP-1B, PPAR-γ and DPP-IV. Oil-treated rats revealed a considerable amelioration of hyperglycemia and lipid dysmetabolism. A notable decrease in oxidative markers, as well as improvement in pancreatic function, was also observed. Further, the tocopherol isomers showed a good interaction prospective with the docking study's targeted proteins (Bharti et al., 2013). Moreover, unsaturated fatty acids such as oleic acid and linoleic acid in the seeds can rebuff the blood cholesterol in humans and rats, which is perhaps affiliated to depletion of cholesterol synthesis and elevated cholesterol catabolism in the liver (Al-Okbi et al., 2014). In another study, it was shown that globulin proteins isolated from different species (ranged from 295 mg/g d.w.) of pumpkin seeds showed a significant decrease in blood sugar level in diabetic rats and could act as a potent hypoglycemic agent (Teugwa et al., 2013).

29.3.5 Antihypertensive and Cardioprotective Activity

Hypertension is a long-standing health condition where blood pressure in the arteries is steadily elevated; it is also considered a risk factor for cardiovascular diseases. Pumpkin seeds oil is rich in many therapeutic ingredients such as mono and polyunsaturated fatty acids, like omega 3, omega 6, oleic acid, and linoleic acid, that are important in reducing the risk for hypertension and heart diseases (Das, 2006). For instance, pumpkin seeds supplement *in vivo* revealed an anti-atherogenic and hepato-protective action in hypercholesterolemic rats (Makni et al., 2010; Mazur & Adlercreutz, 1998). Furthermore, in a recent open-label clinical trial, oral treatment of a combination of onion and pumpkin extract (OPtain120 tab/twice daily for 12 weeks) by healthy adults with slightly elevated blood pressure revealed a decrease in systolic and diastolic blood pressures (Yoshinari et al., 2015).

The estrogen hormone plays a chief role in reproduction, bone density modulation, and cholesterol transportation in our body (Rosano et al., 2007). Phytoestrogens and sterols in pumpkin seeds oil, like lariciresinol and secoisolariciresinol, can exert estrogenic-like effects and manage many diseases (Patel et al., 2012; Phillips et al., 2005; Sicilia et al., 2003). For instance, *in vivo* supplementation of pumpkin seed oil (40 mg/kg orally) to ovariectomized rats lowered total cholesterol, LDL, and triglycerides, while increased HDL levels lowered the systolic and diastolic blood pressures (Gossell-Williams et al., 2008). Moreover, the thermal stability of acylglycerols (doses: 1, 2, and 3 mg/kg b.w.) in pumpkin seeds oil was evaluated by Zeb and Ahmad (2017), where non-oxidized acylglycerols lowered the levels of total cholesterol, triglycerides, LDL, glucose, and ALT in rabbits when compared to thermally oxidized acylglycerols (Zeb & Ahmad, 2017).

Furthermore, pumpkin seeds are rich in different valuable minerals such as potassium, calcium, and magnesium, which are essential in controlling hypertension and heartbeats (Houston, 2010). Idouraine et al. (1996) standardized eight lines of *C. pepo* seeds to contain calcium in the range of 24–279 mg/100 dry weight, potassium (2069–2838 mg/100 g dry weight), and magnesium (735–856 mg/100 g dry weight) (Idouraine et al., 1996). Additionally, the high magnesium content in the seeds oil (40 or 100 mg/kg b.w.) showed potent hypotensive and cardioprotective actions in l-NAME-induced hypertension rats. The seeds oil revealed relaxation of vessels, decreased MDA, and NO, it also showed a protective effect against heart and aorta alterations, leading to decreasing the risk of heart attacks (El-Mosallamy et al., 2012).

29.3.6 Anti-Inflammatory and Wound Healing Activity

Inflammation is a risk factor for many pathological disorders such as diabetes, cancer, and other complications, where its prevention can lead to the management of many diseases. Pumpkin seed oil was reported to prevent and manage inflammation in different diseases. For instance, treatment of Freund's complete adjuvant-induced arthritis in rats with pumpkin seed oil (100 mg/kg b.w. IM.) showed a significant rise in serum sulphhydryl groups, blood GSH as well as plasma albumin level. Also, it decreased the paw oedema and G-6-P DH activity in the liver (Fahim et al., 1995). Moreover, a new isolated tetracyclic triterpenoid (19-(10 → 9β)-abeo-10α-lanost-5-ene) from pumpkin seeds oil at dose 300 μg/mL showed optimum anti-ulcer activity, with the percentage of inhibition of 55.7%, 67.1%, and 59.1% by pyloric ligation, water immersion stress and indomethacin-induced ulcer methods in rats, respectively (Gill & Bali, 2011). Furthermore, a cucurbitacin derivative isolated and identified from the methanol extract of *C. pepo* seeds showed potent antiulcer activity in NSAID-induced ulcer rats by decreasing ulcerative index and protecting the gastric mucosa damage (Yoshinari et al., 2015). As well, oral treatment of aspirin-induced ulcer rats with pumpkin pulp (200, 300, 350, 400, 450, and 500 mg/kg b.w.) revealed an increase in alkaline phosphate activity and a decrease in mucosal thickness, as well as ulcer index representing that pumpkin pulp, might have gastroduodenal protective and antiulcer properties (Sarkar & Guha, 2008). Bardaa et al. (2016) proved that oral treatment of azathioprine-induced cutaneous wounds rats with pumpkin seed oil (4 mL/kg b.wt) showed improvement in macroscopic, morphometric, and histological data of rats' skin than the untreated group, through a reduction in DNA fragmentation (Bardaa et al., 2016).

29.4 Pumpkin Bio-Wastes and their Application

29.4.1 Food and Medical Application

The use of pumpkin wastes as functional food represents a new challenge in the last few decades. Based on human consumption or processing of pumpkins, four types of wastes are generated (Fig. 29.3).

29.4.1.1 Pumpkin Rind or Peel

Pumpkin peels (PP) are not edible and are produced from human consumption or pumpkin processing from seed oil production. They are usually discarded as agricultural by-products for livestock feed (Jun et al., 2006; Song et al., 2018). Pumpkin peels are a rich source of carotenoids and provitamin A and accounted for 10–40% of total carotenoids in pumpkin (Nuerbiya et al., 2014). Besides, the peels contain protein (9–17%), pectin, amino acids (Kim et al., 2012), and fiber, which are mainly composed of cellulose (de Escalada Pla et al., 2007; Nyam et al., 2009). Recently, pumpkin peels were used to develop nutraceutical products rich in carotenoids or as a rich source of fiber. The carotenoids in PP include lutein, α-carotene, β-carotene, and zeaxanthin, which possess the potential application value in the food industry. Song et al. (2018) investigated the use of ultrasound-assisted extraction (UAE) technique against conventional solvent extraction (CSE). It is well known that the UAE technique has been used to assist in extracting carotenoids from natural sources

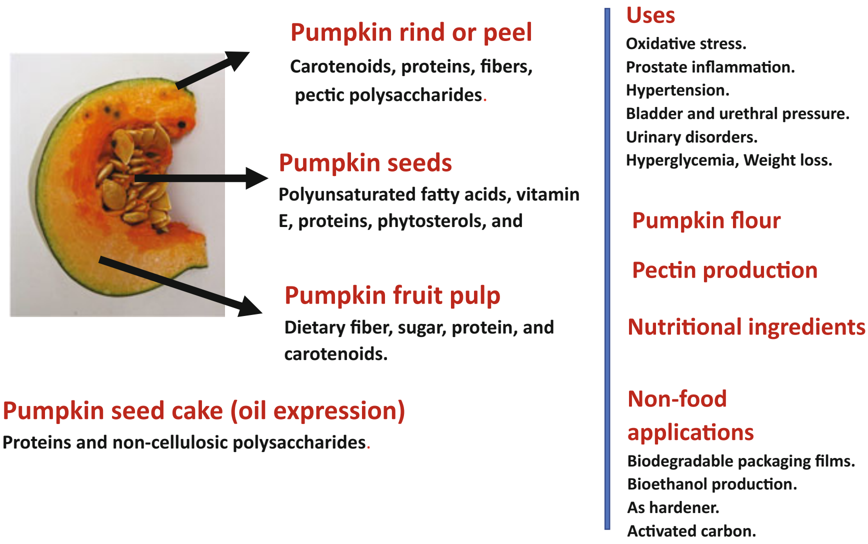

Fig. 29.3 Pumpkin bio-wastes and its applications

due to its high efficiency, short extraction duration, simple and easy operation. Song et al. (2018) studied the effect of ultrasonic power and frequency, time, solvent, and solvent to material ratio on the yields of *trans* lutein and total *trans* carotenoids. Compared to the CSE, UAE avoided degradation and isomerization, resulting in a high yield of *trans* lutein and *trans* carotenoids.

Also, pumpkin peel flour (PPF) was prepared from PP and was partially incorporated with wheat flour in bread production (Song et al., 2018; Staichok et al., 2016). The bread produced with 5% PPF showed good characteristics with high protein content, raw fiber, and a lesser amount of carbohydrate. The use of PPF represents an excellent alternative to the market by offering a new type of healthy and functional food and its contribution to reduce food waste and a new added value for the use of discarded pumpkin peels. Also, pumpkin peel was used as a natural source for the production of carotenoids. Mishra and Sharma (2019) studied nutritional value and modeling of carotenoids extraction. This work demonstrated particle size's effects on carotenoid extraction efficiency and evidenced that pumpkin by-product flour could be used as a food ingredient or natural dye.

PP also contains a considerable amount of pectic polysaccharide (Hamed et al., 2017; Hamed & Mustafa, 2018). Compositional and structural characteristics of the pectic polysaccharides from PP were studied by Jun et al. (2006). The authors revealed that pumpkin peel pectic substance possessed glucose and bile acid retardation effects and growth-promoting activities on the intestinal bacteria, suggesting that PP might be an attractive material for the development of functional foods.

Lalnunthari et al. (2019) studied the extraction of protein and pectin from pumpkin seeds and peels industry, respectively, and their application for the production of edible film. The authors found that extraction of protein and pectin was affected by the process of different variables. Such edible film can be used to apply in the food industry of snakes like cut fruits, bakery products, sweets, etc. Also, Hamed et al. (2017) and Hamed and Mustafa (2018) studied the extraction of pectin from PP using soxhlet using different acids (nitric and citric acids) to obtain the highest yield, and nitric acid was the preferred acid.

Mishra and Sharma (2019) developed a pumpkin peel-based biscuit containing 20% of PPF, which was the most accepted based on a 9-point hedonic scale of sensory evaluation. The developed biscuit is rich in nutrients and can be used as a supplement in malnutrition and pregnant women based on its proximate analysis (moisture, protein, fat, fiber, carbohydrate, energy, and ash value). George (2020) also studied the impact of PPF and pumpkin pulp flour using different ratios in the biscuit industry.

The effect of supplementation of pumpkin peel powder (PPP) on the flesh quality of Japanese quail (*Coturnix japonica*) was the main objective of an experiment carried out by (Azman & Ahmad, 2019). Feed formulation with PPP for two weeks indicated that the highest concentration (6%) of PPP showed the highest reading of carotenoid content, while treatment with 2% of PPP obtains the highest weight gain. This study showed that quail feeding with higher PPP might help improve its flesh quality with good antioxidant properties.

The use of pumpkin peel extract (PPE) as a natural antioxidant in edible oils was recommended by Salami et al. (2020). The authors compared PPE prepared by the supercritical fluid extraction (SFE) and subcritical water extraction (SWE) methods or their natural antioxidant combination. The total phenol content in the SFE extract was higher than the SWE extract; however, carotenoid content was higher in the SWE extract. The results revealed that the sample containing the combined extracts had the highest oxidative stability, higher performance in preventing oil oxidation during the frying process compared to tert-butyl hydroquinone (TBHQ).

A study was undertaken in Bangladesh to determine the chemical composition of different types of vegetable peels that could be used as potential unconventional feeds for livestock. It was found that pumpkin peels showed the highest protein content (16.5%) and a moderate amount of crude fiber (14.8%) among ten vegetable peels. The authors recommended pumpkin peel as a good feed resource for livestock (Hossain et al., 2015).

29.4.1.2 Pumpkin Seed

Pumpkin seeds (PS) are the edible part of a pumpkin but are generally considered agro-industrial wastes and discarded (Kaur & Sharma, 2017). The seeds are flat, oval, and light green. PS is used for oil production and consumed raw, cooked in culinary purposes, roasted seeds as snack in many African countries, and used for comestible and medicinal purposes (Murkovic et al., 1996a, 1996b). PS is a rich source in protein, polyunsaturated fatty acids (ω-3, −6 and − 9 fatty acids), α- and γ-tocopherols, phytosterols, β-carotene and lutein (Jiao et al., 2014; Rezig et al., 2012; Salgın & Korkmaz, 2011), fibers, substantial amounts from micronutrients (P, K, Mg, Mn and Ca) (Roy & Datta, 2015).

Pumpkin seed oil (PSO) has recently gained much attention not only as an edible oil but also as a potential nutraceutical. It is a dichromatic, viscous oil with intense antioxidant activity (Stevenson et al., 2007). The health benefits of PSO are well known in prostate inflammation (Al-Okbi et al., 2017), hypertension progression (El-Mosallamy et al., 2012), bladder and urethral pressure, urinary disorders, and as hypoglycemic agent (Nishimura et al., 2014).

In the view of high economic and nutritional values of PS, numerous studies were undertaken for the production of seed flour to be incorporated in food products (Kaur & Sharma, 2017) or the development of different extraction techniques of seed oil (Alfawaz, 2004; Rezig et al., 2018). Kaur and Sharma (2018) developed an extraction method for the bioactive peptides by probiotic microorganisms from pumpkin seed and investigated their applications to enhance their nutritional value to reduce obesity, cholesterol, infection, and it acts as an antioxidant.

A mixture of defatted pumpkin seeds (DPS) and peels (PP) generated from the pumpkin processing industry was utilized in different proportions for developing biodegradable packaging films (Lalnunthari et al., 2019). The film developed using an equal proportion of DPS and PP had the highest tensile strength and elongation percentage compared to other proportions films. This film is one of the better options for effective use of the food waste industry as a commercial model needs to developed competitive to the synthetic polymer films with proper application on food products like bread, cake, and sweets.

A protein-bound polysaccharide (PSP-I) was prepared from the crude polysaccharides of pumpkin seeds by hot water extraction and ethanol precipitation followed by purification on Q-Sepharose FF column and Sephadex G-100 gel filtration (Wang et al., 2017). PSP-I was composed of mannose, glucose, and galactose in addition to 17 general amino acids and was rich in glutamic acid, alanine, and glycine. PSP-1 displayed inhibitory activity against α-amylase and has intense antibacterial activity, in addition to moderate antioxidative activity.

The production of high-quality extract suitable for food, pharmaceutical, and cosmetic purposes was developed by extracting pumpkin seed oil together with the fruit's peel (Cuco et al., 2019). The pumpkin peel's addition in the oil extraction of the seed enriched the extract with peel bioactive compounds such as carotenoids, phytosterols, tocopherols, and antioxidant activity. The simultaneous extraction was performed using sub- and super-critical CO_2 as the solvent system. The use of optimized pressurized extraction conditions of temperature and pressure using CO_2 produced better quality extract than those obtained by classical and ultrasonic methods based on the content of β-carotene, tocopherols, phenolics, phytosterols, antioxidant activity, and thermal stability.

29.4.1.3 Pumpkin Seed Cake

Pumpkin seed cake (PSC) is a waste product produced during the production of pumpkin oil. PSC contains a large amount of protein that reaches up to 60% with a relatively constant amino acid composition (Pirman et al., 2007). The pumpkin seed protein could be an essential source of these amino acids in animal nutrition. PSC has been used for different purposes, such as dairy animal and poultry nutrition and baits for fish and human nutrition. Pumpkin seed cake flour (PSCF) was mixed with wheat flour to produce a special bread (Mansour et al., 1999) and for improving sausage quality (Mansour et al., 1996).

Košťálová et al. (2009) and his coworkers suggested that the oil pumpkin biomass could be a potential source for non-cellulosic polysaccharides (hemicelluloses and pectic polysaccharides). The advantage of this polysaccharide is its low degree of branching of the side chains of sugar moieties of arabinose and galactose, which is promising for production of pectin used in the food industry. In addition, the accompanied phenolics and other extractive compounds exert antioxidant activity.

PS and PP have a high potential for conversion and use into valuable products of higher value, like edible or biodegradable film. The film-forming ability of pumpkin oil cake and pumpkin oil cake protein isolate is evaluated by Popović et al. (2011, 2012). The composition and functional characteristics of enriched fiber products obtained from the extraction of pumpkin powdered peel were evaluated by de Escalada Pla et al. (2007).

29.4.1.4 Pumpkin Fruit Pulp

The pumpkin pulp or flesh flour (PFF) was a partial replacement for wheat flour to develop more nutritious and gluten-free products. This flour was used in the bakery, confectionary, and beverage industries. Pulp flour from pumpkin flesh of several *Cucurbita* species (*C. moschata*) has potential ingredients for developing healthy foods. Pumpkin pulp flour is characterized by high content of carotenoids, protein, fiber and low content of sodium, and high antioxidant activity. Bemfeito et al. (2020) investigated pumpkin pulp and peel flour to evaluate their nutritional and functional potential to develop healthy food products. The developed PFF is characterized by a high level of dietary fiber (21.9 g/100 g), total sugars (51.8 g/100 g), protein (11.0 g/100 g) and carotenoids (249 mg/g), low levels of sodium (27.2 mg/100 g), and high antioxidant capacity. Biscuits were prepared by replacing wheat flour with pumpkin flour (pulp and peel) at different ratios. The formula with 3% was the most acceptable for all sensory and physical evaluation parameters (George, 2020).

Kulkarni and Joshi (2013) and Mala et al. (2018) studied the nutritional, sensorial, and physicochemical effects of incorporating pulp flour in wheat flour mix for the production of muffins and biscuits with high carotene content. The inclusion of small amounts of pumpkin flour in cakes, muffins, and snacks increased β-carotene content, decreased saturated fatty acids, and increased unsaturated fatty acids in the food products (Jabeen et al., 2018).

Several food products such as noodles and pasta lacking gluten were prepared by partial substitution of corn flour with pumpkin flour (up to 50%). The sensory, physicochemical, and cooking yield attributes in the prepared formulation have been investigated (Mirhosseini et al., 2015). In another study by Malkanthi and Hiremath (2020), a Sri Lankan traditional food string hopper supplemented with 20% pumpkin pulp powder showed improvement in physical and sensory properties such as appearance, color, aroma, taste, texture, and overall acceptability.

In the milk industry, the addition of red pumpkin pulp powder to Basundi buffalo milk improved its quality (Raut et al., 2019). The incorporation of red pumpkin pulp powder up to 7.5% in Basundi buffalo milk did not affect its sensory characteristics adversely. It decreased moisture, fat, and sucrose content significantly; on the other hand, it increases protein, ash, carbohydrate, and total solid content significantly in the treated product compared to control. Also, the beef sausage's quality formulated with 30% dried pumpkin powder was improved (Ahmed et al., 2020). It reduced the moisture content, increased protein, and fat content; besides, the formula was free of *salmonella*.

In a randomized, active-controlled, double-blind clinical trial, the efficacy of pumpkin ointment in treating chronic hand eczema was investigated against betamethasone (Khademi et al., 2020). Patients' Dermatology Life Quality Index (DLQI) scores in pumpkin and betamethasone groups were significant and with a better response in quality of life. Both ointments showed significant improvement compared with almond and eucerin and reduced Hand Eczema Severity Index (HECSI) scores with no adverse effects.

29.4.2 Non-food Application

Corn, pumpkin, and carrots are cheaper, natural, and promising sources for bioethanol production. The production of ethanol from the corn, pumpkin, and carrots was performed by fermentation using yeast (Saccharomyces cerevisiae CCD) with a small amount of α-amylase enzyme to enhance the fermentation (Yesmin et al., 2020).

Pumpkin peel wastes were also used for bioethanol production, which showed the potential uses of these wastes (Chouaibi et al., 2020). The study was conducted to optimize the bioethanol production from pumpkin peel wastes using artificial neural networks (ANN) and response surface methodology (RSM). ANN was better than RSM in terms of its estimation and prediction capabilities. The fermentation process's optimum conditions were predicted based on time, temperature, loading substrate, α-amylase, and amyloglucosidase concentration. The results showed that substrate loading and fermentation temperature were the most significant factors affecting the reducing sugar and bioethanol concentration.

Pumpkin seed shell was used to produce the activated carbon using H_3PO_4 as a chemical agent (Demiral & Şamdan, 2016). The impregnation ratio and activation temperature were tuned to affect the pore structure of the activated carbon significantly. Therefore, pumpkin seed shells could be used as an alternative starting material for commercial activated carbon productions.

The pumpkin seed oil was evaluated as new low-cost alternative feedstocks for biodiesel production (Kumar & Binnal, 2012; Schinas et al., 2009). The oil was converted through an alkaline transesterification reaction using methanol to methyl esters. The quality of biodiesel produced was comparable to that of petroleum diesel according to the standard of EN 14214, although the large-scale production is still not achieved.

The pumpkin seed shell was used to produce activated carbon through chemical activation using H_3PO_4 as a chemical agent (Demiral & Şamdan, 2016). The study showed that the activation temperature and the impregnation ratio significantly affect the pore structure of the activated carbon. Therefore, pumpkin seed shells could be an alternative starting material for the production of commercial activated carbon.

The ashes of pumpkin (*C. pepo*) seeds were used to increase the corrosion resistance characteristic of the epoxy coating system based on epoxy resin as a base resin and polyamine hardener (Agrawal & Amrutkar, 2019). The coating was then applied to mild steel panels using brush coating and was tested for mechanical properties and chemical resistance. The presence of MgO, Fe_2O_3, ZnO, and CaO in ash is responsible for increasing the coating's anti-corrosive performance.

29.5 Valorization of Pumpkin Seeds and Peels into Biodegradable Packaging Films

Defatted pumpkin seeds meal and peels were successfully employed to develop the biodegradable films, where bio-based films prepared with co-products aids to reduce the environmental problem on composting. Such film can be used for application on food products like bread, cake, and sweets. These wastes are rich in carbon content and could be attractive renewable substrates to manufacture value-added products (Peiretti et al., 2017).

References

Abd El-Aziz, A., & Abd El-Kalek, H. (2011). Antimicrobial proteins and oil seeds from pumpkin (*Cucurbita moschata*). *Nature and Science, 9*(3), 105–119.

Abou Seif, H. S. (2014). Ameliorative effect of pumpkin oil (*Cucurbita pepo* L.) against alcohol-induced hepatotoxicity and oxidative stress in albino rats. *Beni Suef University Journal of Basic and Applied Sciences, 3*(3), 178–185.

Adnan, M., Gul, S., Batool, S., Fatima, B., Rehman, A., Yaqoob, S., Shabir, H., Yousaf, T., Mussarat, S., & Ali, N. (2017). A review on the ethnobotany, phytochemistry, pharmacology and nutritional composition of *Cucurbita pepo* L. *The Journal of Phytopharmacology, 6*(2), 133–139.

Agrawal, A., & Amrutkar, S. (2019). Novel use of waste fruit-seeds for enhancing anti-corrosive performance in epoxy system. *The Bombay Technologist, 66*(1), 31–36.

Ahmad, G., & Khan, A. A. (2019). Pumpkin: Horticultural importance and its roles in various forms; a review. *International Journal of Horticulture and Agriculture, 4*(1), 1–6.

Ahmed, W. M. M., Alsiddig, S. A., Abdelgadir, M. O., Eltayeb, A., Ismail, E. O. B., & Elhassan, I. H. (2020). Quality evaluation of beef sausage formulated with different levels of dried pumpkin powder. *International Journal of Multidisciplinary and Current Research, 8*, 150–154.

Alfawaz, M. A. (2004). Chemical composition and oil characteristics of pumpkin (*Cucurbita maxima*) seed kernels. *Food Science and Agriculture, 2*(1), 5–18.

Al-Khalifa, S. (1996). Physicochemical characteristics, fatty acid composition, and lipoxygenase activity of crude pumpkin and melon seed oils. *Journal of Agricultural and Food Chemistry, 44*(4), 964–966.

Al-Okbi, S., Mohamed, D. A., Kandil, E., Abo-Zeid, M., Mohammed, S., & Ahmed, E. (2017). Anti-inflammatory activity of two varieties of pumpkin seed oil in an adjuvant arthritis model in rats. *Grasas y Aceites, 68*(1), 180.

Al-Okbi, S. Y., Mohamed, D. A., Hamed, T. E., & Esmail, R. S. (2014). Rice bran oil and pumpkin seed oil alleviate oxidative injury and fatty liver in rats fed high fructose diet. *Polish Journal of Food and Nutrition Sciences, 64*(2), 127–133.

Amin, M. Z., Islam, T., Mostofa, F., Uddin, M. J., Rahman, M. M., & Satter, M. A. (2019). Comparative assessment of the physicochemical and biochemical properties of native and hybrid varieties of pumpkin seed and seed oil (*Cucurbita maxima* Linn.). *Heliyon, 5*(12), e02994.

Andrikopoulos, N. K., Chiou, A., & Mylona, A. (2004). Triacylglycerol species of less common edible vegetable oils. *Food Reviews International, 20*(4), 389–405.

Ayyildiz, H. F., Topkafa, M., & Kara, H. (2019). Pumpkin (*Cucurbita pepo* L.) seed oil. In M. F. Ramadan (Ed.), *Fruit oils: chemistry and functionality*. Springer. https://doi.org/10.1007/978-3-030-12473-1_41

Azevedo-Meleiro, C. H., & Rodriguez-Amaya, D. B. (2007). Qualitative and Quantitative Differences in Carotenoid Composition among *Cucurbita moschata*, *Cucurbita maxima*, and *Cucurbita pepo*. *Journal of Agricultural and Food Chemistry, 55*(10), 4027–4033.

Azman, M., & Ahmad, F. T. (2019). The effect of pumpkin peels (*Cucurbita Maxima*) on the quality of Japanese quail flesh. *Malaysian Applied Biology Journal, 48*(1), 185–191.

Badr, S. E., Shaaban, M., Elkholy, Y. M., Helal, M. H., Hamza, A. S., Masoud, M. S., & El Safty, M. M. (2011). Chemical composition and biological activity of ripe pumpkin fruits (*Cucurbita pepo* L.) cultivated in Egyptian habitats. *Natural Product Research, 25*(16), 1524–1539.

Barakat, L. A., & Mahmoud, R. H. (2011). The antiatherogenic, renal protective and immunomodulatory effects of purslane, pumpkin and flax seeds on hypercholesterolemic rats. *North American Journal of Medical Sciences, 3*(9), 411.

Bardaa, S., Halima, N. B., Aloui, F., Mansour, R. B., Jabeur, H., Bouaziz, M., & Sahnoun, Z. (2016). Oil from pumpkin (*Cucurbita pepo* L.) seeds: Evaluation of its functional properties on wound healing in rats. *Lipids in Health and Disease, 15*(1), 1–12.

Bemfeito, C. M., Carneiro, J. D. D. S., Carvalho, E. E. N., Coli, P. C., Pereira, R. C., & Boas, E. V. D. B. V. (2020). Nutritional and functional potential of pumpkin (*Cucurbita moschata*) pulp and pequi (*Caryocar brasiliense* Camb.) peel flours. *Journal of Food Science and Technology, 57*(10), 3920–3925.

Bharti, S. K., Kumar, A., Sharma, N. K., Prakash, O., Jaiswal, S. K., Krishnan, S., Gupta, A. K., & Kumar, A. (2013). Tocopherol from seeds of *Cucurbita pepo* against diabetes: Validation by in vivo experiments supported by computational docking. *Journal of the Formosan Medical Association, 112*(11), 676–690.

Blanco-Díaz, M. T., Font, R., Martínez-Valdivieso, D., & Del Río-Celestino, M. (2015). Diversity of natural pigments and phytochemical compounds from exocarp and mesocarp of 27 *Cucurbita pepo* accessions. *Scientia Horticulturae, 197*, 357–365.

Bombardelli, E., & Morazzoni, P. (1997). Cucurbita pepo L. *Fitoterapia, 68*, 291–302.

Bravi, E., Perretti, G., & Montanari, L. (2006). Fatty acids by high-performance liquid chromatography and evaporative light-scattering detector. *Journal of Chromatography A, 1134*(1-2), 210–214.

Chouaibi, M., Daoued, K. B., Riguane, K., Rouissi, T., & Ferrari, G. (2020). Production of bioethanol from pumpkin peel wastes: Comparison between response surface methodology (RSM) and artificial neural networks (ANN). *Industrial Crops and Products, 155*, 112822.

Considine, D. M., & Considine, G. D. (2013). *Van Nostrand's scientific encyclopedia*. Springer Science & Business Media.

Cuco, R. P., Cardozo-Filho, L., & da Silva, C. (2019). Simultaneous extraction of seed oil and active compounds from peel of pumpkin (*Cucurbita maxima*) using pressurized carbon dioxide as solvent. *The Journal of Supercritical Fluids, 143*, 8–15.

Dar, A., Sofi, S., & Rafiq, S. (2017). Pumpkin the functional and therapeutic ingredient: A review. *International Journal of Food Science and Nutrition*, 2(6), 165-170.

Das, U. N. (2006). Essential fatty acids: Biochemistry, physiology and pathology. *Biotechnology Journal: Healthcare Nutrition Technology, 1*(4), 420–439.

de Escalada Pla, M., Ponce, N., Stortz, C., Gerschenson, L., & Rojas, A. (2007). Composition and functional properties of enriched fiber products obtained from pumpkin (*Cucurbita moschata* Duchesne ex Poiret). *LWT-Food Science and Technology, 40*(7), 1176–1185.

Demiral, İ., & Şamdan, C. A. (2016). Preparation and characterisation of activated carbon from pumpkin seed shell using H3PO4. *Anadolu Üniversitesi Bilim Ve Teknoloji Dergisi A-Uygulamalı Bilimler ve Mühendislik, 17*(1), 125–138.

Dona, J. S. M. (2019). Isolation and characterization of pectin from pumpkin (*Cucurbita maxima*) waste and its food application. *Asian Food Science Journal., 13*(2), 1–9.

El-Adawy, T. A., & Taha, K. M. (2001). Characteristics and composition of watermelon, pumpkin, and paprika seed oils and flours. *Journal of Agricultural and Food Chemistry, 49*(3), 1253–1259.

El-Boghdady, N. A. (2011). Protective effect of ellagic acid and pumpkin seed oil against methotrexate-induced small intestine damage in rats. Indian Journal of Biochemistry & Biochemistry 48 (6), 380-387.

Elfiky, S., Elelaimy, I., Hassan, A., Ibrahim, H., & Elsayad, R. (2012). Protective effect of pumpkin seed oil against genotoxicity induced by azathioprine. *The Journal of Basic & Applied Zoology, 65*(5), 289–298.

El-Mosallamy, A. E., Sleem, A. A., Abdel-Salam, O. M., Shaffie, N., & Kenawy, S. A. (2012). Antihypertensive and cardioprotective effects of pumpkin seed oil. *Journal of Medicinal Food, 15*(2), 180–189.

Fahim, A. T., Abd-El Fattah, A. A., Agha, A. M., & Gad, M. Z. (1995). Effect of pumpkin-seed oil on the level of free radical scavengers induced during adjuvant-arthritis in rats. *Pharmacological Research, 31*(1), 73–79.

Fruhwirth, G. O., & Hermetter, A. (2007). Seeds and oil of the Styrian oil pumpkin: Components and biological activities. *The European Journal of Lipid Science and Technology, 109*, 1128–1140.

George, S. A. S. S. S. (2020). Preparation of pumpkin pulp and Peel flour and study their impact in the biscuit industry journal of biology. *Agriculture and Healthcare, 10*(6), 25–33.

Ghisoni, S., Chiodelli, G., Rocchetti, G., Kane, D., & Lucini, L. (2017). UHPLC-ESI-QTOF-MS screening of lignans and other phenolics in dry seeds for human consumption. *Journal of Functional Foods, 34*, 229–236.

Gill, N., & Bali, M. (2011). Isolation of anti ulcer cucurbitane type triterpenoid from the seeds of Cucurbita pepo. *Research Journal of Phytochemistry, 5*(2), 70–79.

Glew, R. H., Glew, R. S., Chuang, L. T., Huang, Y. S., Millson, M., Constans, D., & Vanderjagt, D. J. (2006). Amino acid, mineral and fatty acid content of pumpkin seeds (*Cucurbita* spp.) and *Cyperus esculentus* nuts in the republic of Niger. *Plant Foods for Human Nutrition, 61*, 51–56.

Gossell-Williams, M., Davis, A., & O'connor, N. (2006). Inhibition of testosterone-induced hyperplasia of the prostate of Sprague-dawley rats by pumpkin seed oil. *Journal of Medicinal Food, 9*(2), 284–286.

Gossell-Williams, M., Lyttle, K., Clarke, T., Gardner, M., & Simon, O. (2008). Supplementation with pumpkin seed oil improves plasma lipid profile and cardiovascular outcomes of female non-ovariectomized and ovariectomized Sprague-Dawley rats. *Phytotherapy Research, 22*(7), 873–877.

Habib, A., Biswas, S., Siddique, A. H., Manirujjaman, M., Uddin, B., Hasan, S., Khan, M. M. H., Uddin, M., Islam, M., Hasan, M., Rahman, M., Asaduzzaman, M., Rahman, M., Khatun, M., Islam, M. A., & Rahman, M. (2015). Nutritional and lipid composition analysis of pumpkin seed (*Cucurbita maxima* Linn.). *Nutrition and Food Sciences, 5*(4), 1000374.

Haiyan, Z., Bedgood, D. R., Bishop, A. G., Prenzler, P. D., & Robards, K. (2007). Endogenous biophenol, fatty acid and volatile profiles of selected oils. *Food Chemistry, 100*(4), 1544–1551.

Hamed, A., Elkhedir, A., & Mustafa, S. (2017). Effect of Soxhlet method extraction on characterization of pectin of pumpkin peels. *Journal of Experimental Food Chemistry, 3*(1), 1–3.

Hamed, A. A. R., & Mustafa, S. E. (2018). Extraction and assessment of pectin from pumpkin peels. *Biofarmasi Journal of Natural Product Biochemistry, 16*(1), 1–7.

Hammer, K. A., Carson, C. F., & Riley, T. V. (1999). Antimicrobial activity of essential oils and other plant extracts. *Journal of Applied Microbiology, 86*(6), 985–990.

Hernández-Santos, B., Rodríguez-Miranda, J., Herman-Lara, E., Torruco-Uco, J. G., Carmona-García, R., Juárez-Barrientos, J. M., Chávez-Zamudio, R., & Martínez-Sánchez, C. E. (2016). Effect of oil extraction assisted by ultrasound on the physicochemical properties and fatty acid profile of pumpkin seed oil (*Cucurbita pepo*). *Ultrasonics Sonochemistry, 31*, 429–436.

Hong, H., Kim, C.-S., & Maeng, S. (2009). Effects of pumpkin seed oil and saw palmetto oil in Korean men with symptomatic benign prostatic hyperplasia. *Nutrition Research and Practice, 3*(4), 323–327.

Hossain, M. E., Sultana, S. A., Karim, M. H., & Ahmed, M. I. (2015). Vegetable peels: A promising feed resource for livestock. *Journal of Animal Feed Research, 5*, 33–39.

Houston, M. C. (2010). Nutrition and nutraceutical supplements in the treatment of hypertension. *Expert Review of Cardiovascular Therapy, 8*(6), 821–833.

Isutsa, D. K., & Mallowa, S. (2013). Increasing leaf harvest intensity enhances edible leaf vegetable yields and decreases mature fruit yields in multi-purpose pumpkin. *Journal of Agricultural and Biological Science, 8*(8), 610–615.

Jabeen, A., Hassan, S., Masoodi, L., Ajaz, N., & Rafiq, A. (2018). Physico-chemical composition and functional properties of blended flour obtained from lentil, pumpkin and barley for development of extrudates. *Journal of Food Processing & Technology, 9*(1), 1–9.

Jiao, J., Li, Z.-G., Gai, Q.-Y., Li, X.-J., Wei, F.-Y., Fu, Y.-J., & Ma, W. (2014). Microwave-assisted aqueous enzymatic extraction of oil from pumpkin seeds and evaluation of its physicochemical properties, fatty acid compositions and antioxidant activities. *Food Chemistry, 147*, 17–24.

Jun, H.-I., Lee, C.-H., Song, G.-S., & Kim, Y.-S. (2006). Characterization of the pectic polysaccharides from pumpkin peel. *LWT-Food Science and Technology, 39*(5), 554–561.

Junli, L., YunQiang, W., De, Z., XiaoHua, H., ZhengQin, Z., & ChangPing, X. (2010). Characterization and bioactivity of water-soluble polysaccharides from the fruit of pumpkin. *Journal of Food, Agriculture & Environment, 8*(2), 237–241.

Kaur, H., Sharma, P. (2018). Extraction of bioactive peptides from pumpkin seed and their application extraction of bioactive peptides from pumpkin seed and their application. Lovely Professional University, MSc thesis.

Kaur, M., & Sharma, S. (2017). Development and nutritional evaluation of pumpkin seed (*Cucurbita moschata*) supplemented products. *Food Science, 8*(2), 310–318.

Kaur, S., Panghal, A., Garg, M., Mann, S., Khatkar, S. K., Sharma, P., & Chhikara, N. (2019). Functional and nutraceutical properties of pumpkin-a review. *Nutrition & Food Science, 50*(2), 384–401.

Khademi, A., Mansuri, P., Pahlevan, D., BOZORG, M., Nasiri, M., Hejazi, S., Azizian, Z., & Shirbeigi, L. (2020). Efficacy of pumpkin ointment in treatment of chronic hand eczema: A randomized, active-controlled, double blind clinical trial. *Iranian Journal of Public Health, 49*(7), 1339–1347.

Kim, M. Y., Kim, E. J., Kim, Y.-N., Choi, C., & Lee, B.-H. (2012). Comparison of the chemical compositions and nutritive values of various pumpkin (Cucurbitaceae) species and parts. *Nutrition Research and Practice, 6*(1), 21–27.

Košťálová, Z., Hromádková, Z., & Ebringerová, A. (2009). Chemical evaluation of seeded fruit biomass of oil pumpkin (*Cucurbita pepo* L. var. Styriaca). *Chemical Papers, 63*(4), 406–413.

Kreft, I., Stibilj, V., & Trkov, Z. (2002). Iodine and selenium contents in pumpkin (*Cucurbita pepo* L.) oil and oil-cake. *European Food Research and Technology, 215*, 279–281.

Kreft, M., Zorec, R., Janeš, D., & Kreft, S. (2009). Histolocalisation of the oil and pigments in the pumpkin seed. *Annals of Applied Biology, 154*(3), 413–418.

Krimer-Malešević, V. (2020). Chapter 37—Pumpkin Seeds: Phenolic Acids in Pumpkin Seed (*Cucurbita pepo* L.), Nuts and Seeds in Health and Disease Prevention (Second Edition). Academic Press.:533–542.

Kulczyński, B., & Gramza-Michałowska, A. (2019). The profile of secondary metabolites and other bioactive compounds in *Cucurbita pepo* L. and *Cucurbita moschata* pumpkin cultivars. *Molecules, 24*(16), 2945.

Kulkarni, A., & Joshi, D. (2013). Effect of replacement of wheat flour with pumpkin powder on textural and sensory qualities of biscuit. *International Food Research Journal, 20*(2), 587.

Kumar, D. J., & Binnal, P. (2012). Performance evaluation of a single cylinder diesel engine fueled with biodiesel produced from pumpkin oil. *Journal of Scientific & Industrial Research, 71*, 75–78.

Lalnunthari, C., Devi, L. M., Amami, E., & Badwaik, L. S. (2019). Valorisation of pumpkin seeds and peels into biodegradable packaging films. *Food and Bioproducts Processing, 118*, 58–66.

Lestari, B., & Meiyanto, E. (2018). A review: The emerging nutraceutical potential of pumpkin seeds. *Indonesian Journal of Cancer Chemoprevention, 9*(2), 92–101.

Li, W., Koike, K., Tatsuzaki, M., Koide, A., & Nikaido, T. (2005). Cucurbitosides F–M, acylated phenolic glycosides from the seeds of *Cucurbita pepo*. *Journal of Natural Products, 68*(12), 1754–1757.

Makni, M., Fetoui, H., Gargouri, N. K., Garoui, E. M., & Zeghal, N. (2011). Antidiabetic effect of flax and pumpkin seed mixture powder: Effect on hyperlipidemia and antioxidant status in alloxan diabetic rats. *Journal of Diabetes and its Complications, 25*(5), 339–345.

Makni, M., Fetoui, H., Garoui, E. M., Gargouri, N. K., Jaber, H., Makni, J., Boudawara, T., & Zeghal, N. (2010). Hypolipidemic and hepatoprotective seeds mixture diet rich in ω-3 and ω-6 fatty acids. *Food and Chemical Toxicology, 48*(8-9), 2239–2246.

Mala, S. K., Aathira, P., Anjali, E., Srinivasulu, K., & Sulochanamma, G. (2018). Effect of pumpkin powder incorporation on the physico-chemical, sensory and nutritional characteristics of wheat flour muffins. *International Food Research Journal, 25*(3), 1081–1087.

Malkanthi, A., & Hiremath, U. S. (2020). Pumpkin powder (*Cucurbita maxima*)-supplemented string hoppers as a functional food. *International Journal of Food and Nutritional Sciences, 9*(1), 2.

Mansour, E., Dworschák, E., Perédi, J., & Lugasi, A. (1993). Evaluation of pumpkin seed (*Cucurbita pepo*, Kakai 35) as a new source of protein. *Acta Alimentaria, 22*, 3–13.

Mansour, E., Dworschak, E., Huszka, T., Hovari, J., & Gergely, A. (1996). Utilization of pumpkin seed and rapeseed proteins in the preparation of bologna type sausages. *Acta Alimentaria, 25*(1), 25–36.

Mansour, E., Dworschak, E., Pollhamer, Z., Gergely, A., & Hovari, J. (1999). Pumpkin and canola seed proteins and bread quality. *Accreditation and Quality Assurance, 4*(1-2), 59–70.

Mazur, W., & Adlercreutz, H. (1998). Naturally occurring oestrogens in food. *Pure and Applied Chemistry, 70*(9), 1759–1776.

Medjakovic, S., Hobiger, S., Ardjomand-Woelkart, K., Bucar, F., & Jungbauer, A. (2016). Pumpkin seed extract: Cell growth inhibition of hyperplastic and cancer cells, independent of steroid hormone receptors. *Fitoterapia, 110*, 150–156.

Mirhosseini, H., Rashid, N. F. A., Amid, B. T., Cheong, K. W., Kazemi, M., & Zulkurnain, M. (2015). Effect of partial replacement of corn flour with durian seed flour and pumpkin flour on cooking yield, texture properties, and sensory attributes of gluten free pasta. *LWT-Food Science and Technology, 63*(1), 184–190.

Mishra, S., & Sharma, K. (2019). Development of pumpkin peel cookies and its nutritional composition. *Journal of Pharmacognosy and Phytochemistry, 8*(4), 370–372.

Mitra, P., Ramaswamy, H. S., & Chang, K. S. (2009). Pumpkin (*Cucurbita maxima*) seed oil extraction using supercritical carbon dioxide and physicochemical properties of the oil. *Journal of Food Engineering, 95*(1), 208–213.

Mohamed, G., Ibrahim, S., & Sayed, H. (2009). Phenolic constituents of Cucurbita pepo L. cvEskandrani (summer squash) flowers. *Bulletin of pharmaceutical Sciences. Assiut, 32*(2), 311–319.

Montesano, D., Blasi, F., Simonetti, M. S., Santini, A., & Cossignani, L. (2018). Chemical and nutritional characterization of seed oil from *Cucurbita maxima* L. (var. Berrettina) pumpkin. *Food, 7*(3), 30.

Morocho, K. M., Khairat, E. A. E., Manzano, P., & Choez, I. (2019). Fatty acid profile of agro industry waste of pumpkin *Cucurbita maxima*. *Journal of Food Nutrition & Health, 2*(2), 1–5.

Murkovic, M., Hillebrand, A., Winkler, J., Leitner, E., & Pfannhauser, W. (1996a). Variability of fatty acid content in pumpkin seeds (*Cucurbita pepo* L.). *European Food Research and Technology, 203*, 216–219.

Murkovic, M., Hillebrand, A., Winkler, J., & Pfannhauser, W. (1996b). Variability of vitamin E content in pumpkin seeds (*Cucurbita pepo* L.). *European Food Research and Technology, 202*, 275–278.

Murkovic, M., Mülleder, U., & Neunteufl, H. (2002). Carotenoid content in different varieties of pumpkins. *Journal of Food Composition and Analysis, 15*(6), 633–638.

Nawirska-Olszańska, A., Kita, A., Biesiada, A., Sokół-Łętowska, A., & Kucharska, A. Z. (2013). Characteristics of antioxidant activity and composition of pumpkin seed oils in 12 cultivars. *Food Chemistry, 139*(1-4), 155–161.

Nishimura, M., Ohkawara, T., Sato, H., Takeda, H., & Nishihira, J. (2014). Pumpkin seed oil extracted from Cucurbita maxima improves urinary disorder in human overactive bladder. *Journal of Traditional and Complementary Medicine, 4*(1), 72–74.

Nkosi, C., Opoku, A., & Terblanche, S. (2006). Antioxidative effects of pumpkin seed (Cucurbita pepo) protein isolate in CCl4-induced liver injury in low-protein fed rats. *Phytotherapy Research, 20*(11), 935–940.

Nuerbiya, Y., Ayinuer, R., & Abdulla, A. (2014). Optimization of extraction pigment from pumpkin skin product's stability. *Food Fermentation Industries, 12*, 216–222.

Nyam, K. L., Tan, C. P., Lai, O. M., Long, K., & Che Man, Y. B. (2009). Physicochemical properties and bioactive compounds of selected seed oils. *LWT-Food Science and Technology, 42*(8), 1396–1403.

Nyam, K., Lau, M., & Tan, C. (2013). Fibre from pumpkin (*Cucurbita pepo* L.) seeds and rinds: Physico-chemical properties, antioxidant capacity and application as bakery product ingredients. *Malaysian Journal of Nutrition, 19*, 1.

Orsavova, J., Misurcova, L., Ambrozova, J. V., Vicha, R., & Mlcek, J. (2015). Fatty acids composition of vegetable oils and its contribution to dietary energy intake and dependence of cardiovascular mortality on dietary intake of fatty acids. *International Journal of Molecular Sciences, 16*, 12871–12890.

Orzolek, M.D. (1995). Pumpkin production. Penn State Extension, College of Agricultural Sciences, Cooperative Extension. https://extension.psu.edu/pumpkin-production

Özbek, A. Z., & Ergönül, G. P. (2020). Chapter 18—Cold pressed pumpkin seed oil. In M. F. Ramadan (Ed.), *Cold pressed oils* (pp. 219–229). Academic. https://doi.org/10.1016/B978-0-12-818188-1.00018-9

Parfitt, J., Barthel, M., & Macnaughton, S. (2010). Food waste within food supply chains: Quantification and potential for change to 2050. *Philosophical Transactions of the Royal Society B: Biological Sciences, 365*(1554), 3065–3081.

Park, S.-C., Lee, J. R., Kim, J.-Y., Hwang, I., Nah, J.-W., Cheong, H., Park, Y., & Hahm, K.-S. (2010). Pr-1, a novel antifungal protein from pumpkin rinds. *Biotechnology Letters, 32*(1), 125.

Parry, J. W., Cheng, Z., Moore, J., et al. (2008). Fatty acid composition, antioxidant properties, and Antiproliferative capacity of selected cold-pressed seed flours. *Journal of the American Oil Chemists' Society, 85*, 457–464.

Patel, D., Vaghasiya, J., Pancholi, S., & Paul, A. (2012). Therapeutic potential of secoisolariciresinol diglucoside: A plant lignan. *International Journal of Pharmaceutical Sciences and Drug Research, 4*(1), 15–18.

Peiretti, P. G., Meineri, G., Gai, F., Longato, E., & Amarowicz, R. (2017). Antioxidative activities and phenolic compounds of pumpkin (*Cucurbita pepo*) seeds and amaranth (*Amaranthus caudatus*) grain extracts. *Natural Product Research, 31*(18), 2178–2182.

Peter, E. A. C., Hudson, N., Alice, O. N., Stanley, O., William, T., Ijani, A. S. M., & Anne, S. (2013). Evaluation of micronutrients in seeds of pumpkin varieties grown by smallholder farmers in the Lake Victoria Basin. *African Journal of Food Science and Technology, 4*(10), 221–228.

Phillips, K. M., Ruggio, D. M., & Ashraf-Khorassani, M. (2005). Phytosterol composition of nuts and seeds commonly consumed in the United States. *Journal of Agricultural and Food Chemistry, 53*(24), 9436–9445.

Pirman, T., Marič, M., & Orešnik, A. (2007). Changes in digestibility and biological value of pumpkin seed cake protein after limiting amino acids supplementation. *Krmiva: Časopis o hranidbi životinja, proizvodnji i tehnologiji krme, 49*(2), 95–102.

Popović, S., Peričin, D., Vaštag, Ž., Lazić, V., & Popović, L. (2012). Pumpkin oil cake protein isolate films as potential gas barrier coating. *Journal of Food Engineering, 110*(3), 374–379.

Popović, S., Peričin, D., Vaštag, Ž., Popović, L., & Lazić, V. (2011). Evaluation of edible film-forming ability of pumpkin oil cake; effect of pH and temperature. *Food Hydrocolloids, 25*(3), 470–476.

Provesi, J. G., Amante, E. R. (2015). Chapter 9—Carotenoids in pumpkin and impact of processing treatments and storage, processing and impact on active components in food, Academic Press 71–80.

Rajasree, R., Sibi, P., Francis, F., & William, H. (2016). Phytochemicals of Cucurbitaceae family-a review. *International Journal of Pharmacognosy and Phytochemical Research, 8*(1), 113–123.

Rakcejeva, T., Galoburda, R., Cude, L., & Strautniece, E. (2011). Use of dried pumpkins in wheat bread production. *Procedia Food Science, 1*, 441–447.

Ramachandran, P., Dhiman, A. K., & Attri, S. (2017). Extraction of pectin from ripe pumpkin (*Cucurbita moschata* Duch ex. Poir) using eco-friendly technique. *Indian Journal of Ecology, 44*(6), 685–689.

Rathinavelu, A., Levy, A., Sivanesan, D., Murugan, D., Jornadal, J., Quinonez, Y., Jaffe, M., & Gossell-Williams, M. (2013). Cytotoxic effect of pumpkin (*Curcurbita pepo*) seed extracts in LNCaP prostate cancer cells is mediated through apoptosis. *Current Topics in Nutraceuticals Research, 11*(4), 137.

Raut, M., Shinde, A., & Wakde, P. (2019). Effect of red pumpkin pulp on quality of buffalo milk. *Basundi, 8*(10), 34–35.

Rezig, L., Chouaibi, M., Meddeb, W., Msaada, K., & Hamdi, S. (2019). Chemical composition and bioactive compounds of Cucurbitaceae seeds: Potential sources for new trends of plant oils. *Process Safety and Environmental Protection, 127*, 73–81.

Rezig, L., Chouaibi, M., Msaada, K., & Hamdi, S. (2012). Chemical composition and profile characterization of pumpkin (*Cucurbita maxima*) seed oil. *Industrial Crops and Products, 37*, 82–87.

Rezig, L., Chouaibi, M., Rm, O.-A., Gomez-Alonso, S., Salvador, M. D., Fregapane, G., & Hamdi, S. (2018). Cucurbita maxima pumpkin seed oil: From the chemical properties to the different extracting techniques. *Notulae Botanicae Horti Agrobotanici Cluj-Napoca, 46*(2), 663–669.

Richter, D., Abarzua, S., Chrobak, M., Vrekoussis, T., Weissenbacher, T., Kuhn, C., Schulze, S., Kupka, M. S., Friese, K., & Briese, V. (2013). Effects of phytoestrogen extracts isolated from pumpkin seeds on estradiol production and ER/PR expression in breast cancer and trophoblast tumor cells. *Nutrition and Cancer, 65*(5), 739–745.

Rico, X., Gullón, B., Alonso, J. L., & Yáñez, R. (2020). Recovery of high value-added compounds from pineapple, melon, watermelon and pumpkin processing by-products: An overview. *Food Research International, 132*, 109086.

Rosano, G., Vitale, C., Marazzi, G., & Volterrani, M. (2007). Menopause and cardiovascular disease: The evidence. *Climacteric, 10*(sup1), 19–24.

Roy, S. & Datta, S. (2015). A comprehensive review of the versatile as a valuable natural medicine. International Journal of Current Research 7 (8), 19355–19361.

Saavedra, M., Aires, A., Dias, C., Almeida, J., De Vasconcelos, M., Santos, P., & Rosa, E. (2015). Evaluation of the potential of squash pumpkin by-products (seeds and shell) as sources of antioxidant and bioactive compounds. *Journal of Food Science and Technology, 52*(2), 1008–1015.

Salami, A., Asefi, N., Kenari, R. E., & Gharekhani, M. (2020). Addition of pumpkin peel extract obtained by supercritical fluid and subcritical water as an effective strategy to retard canola oil oxidation. *Journal of Food Measurement and Characterization, 14*, 2433–2442.

Salgın, U., & Korkmaz, H. (2011). A green separation process for recovery of healthy oil from pumpkin seed. *The Journal of Supercritical Fluids, 58*(2), 239–248.

Sandra, N., Desanka, R., Dubravka, S., Dubravka, S., Željko, M., & Martina, B. (2006). Chemical characteristics of oils from naked and husk seeds of *Cucurbita pepo* L. *European Journal of Lipid Science and Technology, 108*, 936–943.

Sarkar, S., & Guha, D. (2008). Effect of ripe fruit pulp extract of Cucurbita pepo Linn. In aspirin induced gastric and duodenal ulcer in rats. *Indian Journal of Experimental Biology, 46*(9), 639–645.

Schinas, P., Karavalakis, G., Davaris, C., Anastopoulos, G., Karonis, D., Zannikos, F., Stournas, S., & Lois, E. (2009). Pumpkin (*Cucurbita pepo* L.) seed oil as an alternative feedstock for the production of biodiesel in Greece. *Biomass and Bioenergy, 33*(1), 44–49.

Siano, F., Straccia, M. C., Paolucci, M., Fasulo, G., Boscaino, F., & Volpe, M. G. (2015). Physico-chemical properties and fatty acid composition of pomegranate, cherry and pumpkin seed oils. *Journal of the Science of Food and Agriculture, 96*(5), 1730–1735.

Sicilia, T., Niemeyer, H. B., Honig, D. M., & Metzler, M. (2003). Identification and stereochemical characterization of lignans in flaxseed and pumpkin seeds. *Journal of Agricultural and Food Chemistry, 51*(5), 1181–1188.

Song, J., Yang, Q., Huang, W., Xiao, Y., Li, D., & Liu, C. (2018). Optimization of trans lutein from pumpkin (*Cucurbita moschata*) peel by ultrasound-assisted extraction. *Food and Bioproducts Processing, 107*, 104–112.

Sood, A., Kaur, P., Gupta, R., 2012. Phytochemical screening and antimicrobial assay of various seeds extract of Cucurbitaceae family. Journal of Applied Biology and Pharmaceutical Technology 3(3):401-409.

Staichok, A. C. B., Mendonça, K. R. B., dos Santos, P. G. A., Garcia, L. G. C., & Damiani, C. (2016). Pumpkin peel flour (*Cucurbita máxima* L.)-characterization and technological applicability. *Journal of Food Nutrition Research, 4*, 327–333.

Stevenson, D. G., Eller, F. J., Wang, L., Jane, J.-L., Wang, T., & Inglett, G. E. (2007). Oil and tocopherol content and composition of pumpkin seed oil in 12 cultivars. *Journal of Agricultural and Food Chemistry, 55*(10), 4005–4013.

Suphakarn, V. S., Yarnnon, C., & Ngunboonsri, P. (1987). The effect of pumpkin seeds on oxalcrystalluria and urinary compositions of children in hyperendemic area. *American Journal of Clinical Nutrition, 45*, 115–121.

Teugwa, C. M., Boudjeko, T., Tchinda, B. T., Mejiato, P. C., & Zofou, D. (2013). Anti-hyperglycaemic globulins from selected Cucurbitaceae seeds used as antidiabetic medicinal plants in Africa. *BMC Complementary and Alternative Medicine, 13*(1), 63.

Tsaknis, J., Lalas, S., & Lazos, E. S. (1997). Characterization of crude and purified pumpkin seed oil. *Grasas y Aceites, 48*(5), 267–272.

Türkmen, Ö., Özcan, M. M., Seymen, M., Paksoy, M., Uslu, N., & Fidan, S. (2017). Physico-chemical properties and fatty acid compositions of some edible pumpkin seed genotypes and oils. *Journal of Agroalimentary Processes and Technologies, 23*(4), 229–235.

Wang, L., Liu, F., Wang, A., Yu, Z., Xu, Y., & Yang, Y. (2017). Purification, characterization and bioactivity determination of a novel polysaccharide from pumpkin (*Cucurbita moschata*) seeds. *Food Hydrocolloids, 66*, 357–364.

Wang, H., & Ng, T. (2003). Isolation of cucurmoschin, a novel antifungal peptide abundant in arginine, glutamate and glycine residues from black pumpkin seeds. *Peptides, 24*(7), 969–972.

Wang, S.-Y., Huang, W.-C., Liu, C.-C., Wang, M.-F., Ho, C.-S., Huang, W.-P., Hou, C.-C., Chuang, H.-L., & Huang, C.-C. (2012). Pumpkin (*Cucurbita moschata*) fruit extract improves physical fatigue and exercise performance in mice. *Molecules, 17*(10), 11864–11876.

Wenzl, T., Prettner, E., Schweiger, K., & Wagner, F. S. (2002). An improved method to discover adulteration of Styrian pumpkin seed oil. *Journal of Biochemical and Biophysical Methods, 53*(1-3), 193–202.

Xanthopoulou, M. N., Nomikos, T., Fragopoulou, E., & Antonopoulou, S. (2009). Antioxidant and lipoxygenase inhibitory activities of pumpkin seed extracts. *Food Research International, 42*(5-6), 641–646.

Xia, H. C., Feng, L., Zhen, L., & Zhang, Z. C. (2003). Purification and characterization of Moschatin, a novel type I ribosome-inactivating protein from the mature seeds of pumpkin (*Cucurbita moschata*), and preparation of its immunotoxin against human melanoma cells. *Cell Research, 13*(5), 369–374.

Yadav, M., Jain, S., Tomar, R., Prasad, G., & Yadav, H. (2010). Medicinal and biological potential of pumpkin: An updated review. *Nutrition Research Reviews, 23*(2), 184–190.

Yasir, M., Sultana, B., Nigam, P. S., & Owusu-Apenten, R. (2016). Antioxidant and genoprotective activity of selected cucurbitaceae seed extracts and LC-ESIMS/MS identification of phenolic components. *Food Chemistry, 199*, 307–313.

Yesmin, M., Azad, M., Kamruzzaman, M., & Uddin, M. (2020). Bioethanol production from corn, pumpkin, and carrot of Bangladesh as renewable source using yeast saccharomyces cerevisiae. *Acta Chemica Malaysia, 4*(2), 45–54.

Yoshinari, O., Udani, J., Moriyama, H., Shiojima, Y., & Chien, X. (2015). The efficacy and safety of a proprietary onion-pumpkin extract (OPtain120) on blood pressure: An open-label study. *Functional Foods in Health and Disease, 5*(6), 224–242.

Zaineddin, A. K., Buck, K., Vrieling, A., Heinz, J., Flesch-Janys, D., Linseisen, J., & Chang-Claude, J. (2012). The association between dietary lignans, phytoestrogen-rich foods, and fiber intake and postmenopausal breast cancer risk: A German case-control study. *Nutrition and Cancer, 64*(5), 652–665.

Zeb, A., & Ahmad, S. (2017). Changes in acylglycerols composition, quality characteristics and in vivo effects of dietary pumpkin seed oil upon thermal oxidation. *Frontiers in Chemistry, 5*, 55.

Zhu, H. Y., Chen, G. T., Meng, G. L., & Xu, J. L. (2015). Characterization of pumpkin polysaccharides and protective effects on streptozotocin-damaged islet cells. *Chinese Journal of Natural Medicines, 13*(3), 199–207.