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# Apple (*Malus domestica* Borkh.) seed: A review on health promoting bioactivities and its application as functional food ingredient

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## ABSTRACT

Recent trends in food industry focus on utilization of fruit processing waste and by-products as novel ingredients due to their nutritional benefits. Apple seed is one such novel ingredient that can find application as functional ingredient in food, nutraceutical and pharmaceutical industry. The existing studies suggest that apple seed is a rich source of amino acids, fatty acids and polyphenolic compounds. This review discusses the nutritional and phytochemical profile as well as biological activities exhibited by the apple seed. It also highlights the current application of apple seed in food, pharmaceutical and nutraceutical industries along with the safety aspects considering the presence of amygdalin. The apple seed meal, extracts or seed oil as novel ingredient can find

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various industrial applications given their dense nutritional profile. Besides their functionality, rigorous and extensive research is required to promote its usage at industrial level for harnessing the benefits of apple seeds.

## 1. Introduction

Apple (*Malus domestica Borkh.*), a member of the Rosaceae family, is an ancient fruit grown all over the world in more than 93 countries making it one of the popularly grown fruits crops (http://.kew.org/-taxon/urn:lsid:ipni.org:names:726282-1powo.science accessed on January 10, 2021). Central Asia including Asia Minor, Caucasus, Kazakhstan, and Western China are the likely regions where apple was originated (http://.kew.org/taxon/urn:lsid:ipni.org:names:726282-1powo.science accessed on January 10, 2021). It is extensively cultivated in the temperate region of the globe (Spengler, 2019; Pandey et al., 2020). In India commonly called wild or crab apple, it is mainly grown in Himalayan regions and Himachal Pradesh (Dadwal et al., 2018). Among top ten countries, China is a major producer of apples (42 million tonnes (mT)) followed by USA (4.9 mT), Turkey (3.6 mT), Poland (3.1 mT), India (2.31 mT), Italy (2.3 mT), Iran (2.2 mT), Russian Federation (1.95 mT), France (1.75 mT), and Chile (1.6 mT) (https://www..com/en-in/countries-by-apple-productionatlasbig accessed on December 08, 2021). Top twenty apple producing countries are shown in Fig. 1. Water constitutes 83–84% of fruit's portion followed by carbohydrates, fibre, protein, fat, vitamins (A & C) and minerals (Omoyajowo et al., 2017). Apple fruit is well known for its nutritional profile, it is a store house of many bioactive compounds including polysaccharides, polyphenols, organic acids, pentacyclic triterpenes, and sterols (Boyer & Liu, 2004; Patocka et al., 2020). The presence of diverse bioactive compounds accords to its beneficial effect on health. Besides this, myriad of biological activities like antioxidant, anti-inflammatory, anti-cancer, and neuroprotective effects are reported in literature (Can et al., 2014; Pandey et al., 2020; Patocka et al., 2020). Perishability of apple introduces the need of its processing. Market is flooded with a variety of processed apple products like juices, jams, ciders, wine, dry apples tea, and compotes. Seed and peel wastes contribute approximately 20% of the total weight of apple (Ferrentino et al., 2020). Flourishing salad businesses that sell fresh cut fruits also contribute to fruit waste generation (Górnaś, 2015). Apple industries per year generate approximately 20 million tons of waste (Guardia et al., 2019). Domestic waste as well as waste generated from the apple processing industries is a matter of concern for environment. The apple waste generated in processing including pomace, peels, seeds is a good source of nutrients and phytochemicals conferring them with antioxidant and biological activities; thereby making their extracts potential sources for further applications (Blidi et al., 2015; He & Liu, 2008).

Seeds form the smallest portion of the waste generated during apple processing. The apple seeds constitute approximately only 0.7–4% of total mass of the fresh fruit (Fromm et al., 2012a; Górnaś et al., 2014; Senica et al., 2019) but it is densely rich in nutrients viz., protein, amino acids, fat, fatty acids, minerals, vitamins and polyphenols. From past few years, researchers are showing keen interest in valorization of apple seeds as it is documented as an unconventional source of proteins, lipids and phytochemicals. Apple seeds are good source of proteins (38–50%), fibers (3.92–4.32%) and ashes (3.7–5.20%) (Tim 2012; Ferrentino et al., 2020). The amino acid profiling of apple seeds showed balanced composition of all essential amino-acids. Protein from apple seed exhibits good functional properties i.e., water absorption capacity, emulsification capacity, foaming expansion and foaming stability (El-Safy et al., 2012). Apart from having high protein content, apple seeds are also a good source of lipids (10.1–29.4%) (Abbas et al., 2019; Alves et al., 2021; Górnaś et al., 2014). Fatty acid profile of apple seed indicates that linoleic acid (45–60%) and oleic acid (27–41.7%) are present in abundance. Other triglycerides present in minor concentrations include, palmitic acid, stearic acid, and gondoic acid (Matthäus and Özcan 2015; Abbas et al., 2019). This distinctive fatty acid composition makes it a potential raw material for food, cosmetics and pharmaceutical industries. In addition to triglycerides, minor bioactive constituents; tocopherols and phytosterols are also present in apple seed oil. These bioactive constituents help in increasing the stability of oil at high temperatures and also delay/hinder the polymerization reaction. β-sitosterol lowers the cholesterol level and reduces the risk of cardiovascular diseases. Tocopherols are natural antioxidants which protect polyunsaturated fatty acids (PUFA) from lipid peroxidation; a major concern as it is linked to several heath disorders (Górnaś, 2015; Górnaś et al., 2014). Furthermore, apple seed oil also have antibacterial activity (Tian et al., 2010).

Apple seeds are reported to contain phloridzin as the major polyphenolic compound (Xu et al., 2016). Several literatures have shown effectiveness of phloridzin in treating obesity, diabetes mellitus type 2 and stress hyperglycemia. Besides this, hyperin, quercetin-3-*O*-galactoside, phloretin-2′-xyloglucoside, 5-caffeoylquinic acid (chlorogenic acid), *p*-coumaroyl- quinic acid, and (−)-epicatechin are also present in apple seeds (Fromm et al., 2012a). However, the concentration of these polyphenols may vary in seed according to cultivar. Polyphenols in apple seeds are positively and strongly corelated with its high antioxidant activity. Polyphenols extracted from apple seeds are used as food additives. Epidemiological studies indicated that phytochemicals present in apple seed have various biological activities like anti-inflammatory, anti-atherosclerotic, and anti-cancerous, anti-microbial, cholesterol-lowering effect, and anti-diabetic properties (Patocka et al., 2020). The exceptional combination of nutritionally valuable lipids, proteins and structurally diverse phytochemicals makes apple seed a potential functional food. Ample studies have been focused recently on the utilization of the fruit and vegetable processing by-products given their nutritional significance and harnessing their nutraceutical benefits. Some of the by-products studied and reviewed include tomato seeds, litchi seeds, jamun seeds, guava leaves, pomegranate, grapes and so on (Andrade et al., 2019; Bangar et al., 2021; Kumar et al., 2021a, 2021b; Kumoro et al., 2020; Punia & Kumar, 2021; Raza et al., 2017); however, limited literature is available on the apple seeds.

Therefore, this review accentuates the nutritional and phytochemical composition of apple seeds with an objective of improvising its utilization as food supplements. In addition to this, review also provides a detailed discussion on biological activities of apple seed extract and utilization of apple seeds as functional component in food or raw material for pharmaceutical, nutraceutical and cosmetic industries.

## 2. Nutritional profile of apple seeds

### 2.1. Proximate composition

Apple seeds obtained as by-products have good proximate composition indicating that they can be used in food and non-food industry. It is reported that apple seeds have good amounts of moisture, protein, fat, fiber, ash and carbohydrate in the ranges of 3–18.03 g/100 g, 33.79–49.55 g/100 g, 10.1–29.4 g/100 g, 3.92–20.6 g/100 g, 3.66–5.20 g/100 g and 23.50–24.0 g/100 g respectively (Abbas et al., 2019; Arain et al., 2012; Dadwal et al., 2018; El-Safy et al., 2012; Madrera & Valles, 2018; Tian et al., 2010; Yu et al., 2007; Yukui et al., 2009).

### 2.2. Protein

Apple seeds contain substantial amounts of protein. Proximate analysis of apple seeds revealed protein content in the range of

33.79–49.55 g/100 g depending upon the cultivar and agro-climatic conditions (El-Safy et al., 2012; Kamel et al., 1982; Madrera & Valles, 2018; Yu et al., 2007). Potential value of apple seeds protein can be predicted from its amino acid composition. Apple seed protein is comparatively rich in sulphur containing amino acids and it also contains fairly balanced amounts of all the essential amino-acid except tyrosine. Among all the essential amino acids, glycine is found in highest percentage followed by arginine, asparagine, glutamic acid & leucine (Yu et al., 2007). Another study discovered the presence of histidine (3.96 μg/g), glycine (3.16 μg/g), tyrosine (4.35 μg/g) and cysteine (15.43 μg/g) in apple seed (Dadwal et al., 2018).

A study has also assessed the amino acid profile and it was revealed that apple seed had higher total indispensable amino acids than orange and papaya seed; even the dispensable amino acid were higher in apple seed than seeds of papaya, watermelon, guava, orange and pear. The indispensable amino acids identified were leucine (6.72 g/100 g), isoleucine (3.28 g/100 g), methionine (0.92 g/100 g), phenylalanine (4.21 g/100 g), lysine (2.44 g/100 g), threonine (2.56 g/100 g), tyrosine (3.62 g/100 g), valine (3.92 g/100 g) and cystine (1.44 g/100 g). Amongst the indispensable amino acids aspartic (8.21 g/100 g), glutamic (18.62 g/100 g), serine 3.59 g/100 g), proline (4.88 g/100 g), glycine (5.09 g/100 g), alanine (3.87 g/100 g), histidine (2.12 g/100 g) and argnine (11.82 g/100 g) were identified. While the *in-vitro* protein digestibility for apple seed was recorded as 79.45% and amino acid scores were 140.00, 78.10, 41.82, 150.36, 58.10, 64.00, 88.29 and 93.33 for leucine, isoleucine, methionine, phenylalanine, lysine, threonine, tyrosine and valine (El-Safy et al., 2012). Although very few studies reported the protein profile of apple seed it was evident that there is balanced amount of essential amino acids in apple seed, making it a good source of protein and a potential ingredient in developing protein rich food formulations.

### 2.3. Lipid/oil profile

Plant lipids contain derivatives of fatty acids, like triacyl glycerides (TAGs), glycerophospholipids (GPLs) and various further non-polar compounds like sterols, carotenoids, and terpenes. Plant oils find application in various food and non-food industries due to their beneficial properties. Furthermore, health-conscious consumers and growing concern towards sustainable use of resources is motivating researchers to find novel alternative sources for extracting edible oil. By-products generated during processing of fruits and vegetables are therefore investigated for oil extraction. Ongoing studies suggest that seeds of pomegranate, grapes, cherries etc. have potential to be used for edible oil extraction due to their exceptional phytochemical, antioxidant and fatty acid profile (Ferrentino et al., 2018; Paul & Radhakrishnan, 2020).

One of the novel ingredients that can be promoted and researched further is apple seed that has an oil content of 12–34% (Górnaś et al., 2014; Tian et al., 2010; Walia, Rawat, Bhushan, Padwad, & Singh, 2014; Yu et al., 2007). The total fatty acids found in apple seed were 79.34% (Yukui et al., 2009) out of which major share i.e., 70.6–90.3% were unsaturated fatty acids (UFAs) and only 8.74–13.94% were saturated fatty acids (Marek et al., 2015; Pieszka et al., 2015; Walia et al., 2014; Yu et al., 2007; Yukui et al., 2009). The monounsaturated fatty acids (MUFA) were reported as 26.9% whereas poly unsaturated fatty acids (PUFA) were recorded to be 58.30% (Yukui et al., 2009; Walia et al., 2014). One of the important techniques used to characterize the fatty acid profile from unknown sources is gas chromatography-mass spectroscopy (GC-MS). The GC-MS profile of apple seed oil revealed that five major fatty acids are present including oleic acid (C18:1) 26.5–46.50%, linoleic acid (C18:2) 43–43.81%, palmitic acid (C16:0) 5.6–7.25%, stearic acid (C18:0) 1.5–1.72% and arachidic acid 0.72–1.311% (Yukui et al., 2009; Walia et al., 2014). In another investigation, GC analysis reported predominance of linoleic acid (55.54%) in apple seed followed by oleic acid (26.36%), palmitic acid (9.50%), stearic acid (1.82%) and arachidic acid (1.56%) (Marek et al., 2015). Yu et al. (2007) in their study reported that extracted apple seed oil contains oleic acid (39.7%), linoleic acid (49.6%), palmitic acid (7.1%) and stearic acid (1.5%). Similarly, Górnaś et al. (2014) and Abbas et al. (2019) demonstrated that fatty acids viz., linolenic acid (C18:3), heptadecanoic/margaric acid (C17:0), palmitoleic acid (C16:1) and 11-ecosenoic acid (C20:0), lauric acid (C12:0), myristic acid (C14:0), and docosanoic acid (C22:0) were

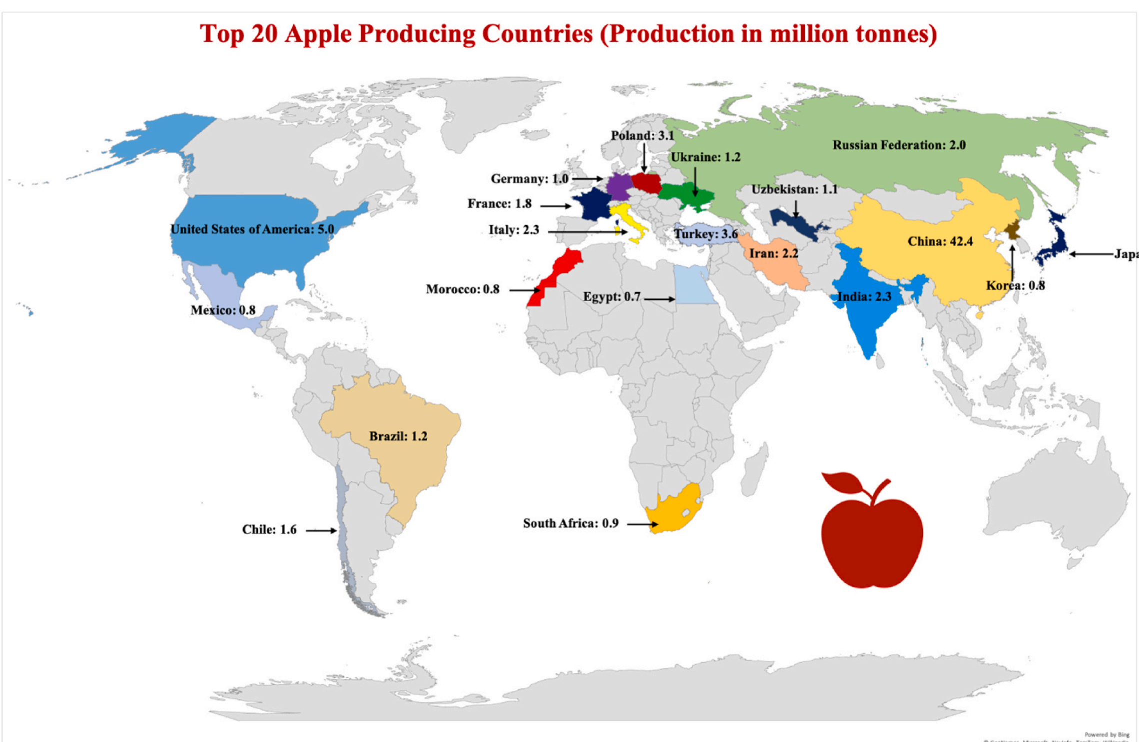



**Fig. 1.** Top twenty apple producing countries with total production in million tonnes.

present in trace amounts (0.01–1.75%). Fromm et al. (2012b) analyzed seventeen oils extracted from different apple seeds and indicated that the range of predominant (C16:0, C18:1 and C18:2) fatty acids present in apple seed oil (6.3–8.33, 20.68–44.50, and 40.5–66.29) found similarity with some commercial oils including maize (12.3, 30.2, and 53.6%), sunflower (6.0, 30.0, and 55.4%) and pumpkin oil (10.7, 34.1, and 47.1%), respectively (Tuberoso et al., 2007).

Another study confirmed that apple seeds obtained from cider industry had oleic acid (C18:1, 32.2–34.2%), linoleic acid (C18:2, 53.7–57.0%), α-linolenic acid (C18:3, 1–1.4%), arachidic acid (C20:0, 0.2–0.3%), palmitic acid (C16:0, 7.1–7.6%), palmitoleic acid (C16:1, 0.05–0.07%), stearic acid (C18:0, 1.7–2.2%), gondoic acid (C20:1, 0.3–0.4%) and behenic acid (C22:0, 0.2%); with more of unsaturated fatty acids than saturated ones (Madrera & Valles, 2018). Dadwal et al. (2018) described that GC-MS quantification of apple revealed the presence of saturated fatty acids viz., palmitic acid (0.89%) and esters such as ethyl palmitate (0.56%), methyl petroselinate (0.90%) and linolein (3.93%).

It was also noted that different apple cultivars had significantly different fatty acid composition (Table 1). Various factors responsible for these variations in fatty acid composition of apple seed oil can be variety, environmental conditions, soil nutrients, horticultural practices, exposure to sunlight, temperature, water availability (Fromm et al., 2012b). Apart from the varietal differences the use of different extraction protocol also influences the amount of fatty acid obtained. Ferrentino et al. (2020) communicated that supercritical fluid extraction (SFE) at 24 MPa, 40 °C, 1 L/h after 140 min, yielded oil with higher linoleic acid (63.76 g/100 g of oil) and better oxidative stability (21.4 h) than Soxhlet extraction (49.03 g/100 g of oil; 12.1 h) respectively. The reason for better oxidative stability was associated with higher quantity of total phenolic content (TPC) in SFE (TPC: 1.61 mg gallic acid equivalent (GAE)/g of oil) extracted oil resulting in higher antioxidant activity (DPPH: 0.71 mg Trolox/g of oil; FRAP: 0.63 mg TE/g of oil) than Soxhlet extraction (TPC: 0.65 mg GAE/g of oil; DPPH: 0.25 mg Trolox/g of oil; FRAP: 0.24 mg Trolox/g of oil) respectively. The fatty acids (g/100 g of oil) present in apple seed oil were linoleic acid (SFE: 63.76; Soxhlet: 49.03), oleic acid (SFE: 34.84; Soxhlet: 24.15), palmitic acid (SFE: 13.39; Soxhlet: 6.24), stearic acid (SFE: 7.69; Soxhlet: 2.97), linolenic acid (SFE: 1.61; Soxhlet: 1.87) and arachidic acid (SFE: 2.04; Soxhlet: 1.22) for both SFE and Soxhlet extracted oils respectively (Ferrentino et al., 2020).

Other than the fatty acid profile, limited reports are available on physicochemical properties of apple seed oil. The seeds from pomace of different varieties of apple (Red Delicious, Golden Delicious, Royal Delicious and Red Chief) were extracted to obtain oil and it was noted that they were yellow (Walia et al., 2014). The reports of Tian et al. (2010) and Walia et al. (2014) on physicochemical properties of apple seed oil extracted from Red Delicious, Golden Delicious, Royal Delicious, Red Chief, Fuji and New Red Star indicated that it had relative density (0.902–0.97 mg/mL at 25 °C), refractive index (1.465–1.47 at 40 °C $n^{20}$) being close to sunflower and mustard seed oil. Despite being within the acceptable range, the acid value of apple seed oil (4.036–4.323 mg KOH/g oil) was higher than mustard seed oil (1.21 mg KOH/g oil) and sunflower oil (3.89 mg KOH/g oil), indicating that had more unsaturated fatty acids. The iodine value also indicated that unsaturation in fatty acids was high in apple seed oil (94.140–121.8 g/100 g oil). The iodine and saponification value (179.010–197.250 mg KOH/g oil) of apple seed oil was similar to sunflower (128.0 g/100 g oil) and mustard (108.0 g/100 g oil) seed oil (Tian et al., 2010; Walia et al., 2014). The peroxide value of Fuji and New Red Star apple seed oil was 2.430–2.520 mmol/kg oil) (Tian et al., 2010).

In general, most of the physicochemical properties of apple seed oil are similar to existing literature on mustard or sunflower seed oil indicating that it can be used as edible oil. Furthermore, apple seed oil falls under the oleic-linoleic acid group of oil which helps to reduce the low-density lipoprotein (LDL) level and reduce the risk for coronary heart diseases (CVD) (Yu et al., 2007). Overall, it can be concluded that apple seed oil contains more UFAs that are regarded as healthy for human health; hence, it can act as important raw material for functional food industry.

### 2.4. Carbohydrate profile

Carbohydrate content in apple seeds ranges between 22 and 26% (Purić et al., 2020), while the pectin content ranges betweeen 3.5 and 14.32 g/100 g (Antonic et al., 2020). The carbohydrate analysis by Xie et al. (2008) revealed that just 4.9% (w/w) of the total seed weight contained starch, making it unsuitable for industrial use. The carbohydrate content of thorn apple seeds was estimated to be 26.20–29.80% by Oseni, Olarinoye, and Amoo (2010). The apple seed carbohydrates mainly consist of galacturonic acid (49%–64%), arabinose (14%–23%) and galactose (6%–15%), with minor amounts of rhamnose, xylose and glucose (Bhushan et al., 2008). The soluble carbohydrate content in apple seeds was estimated to be 36.30 ± 2.00% by Hassan et al. (2008).

### 2.5. Vitamins and minerals

Vitamins and minerals are regarded as micronutrients essential for normal body functioning. They are required in trace amounts. Even, certain specific minerals are required for absorption and assimilation of vitamins in human body (Buturi et al., 2021). Deficiency of either of them affects human health. About 66.67% of world's population suffers from micronutrient deficiency. Apple seeds contain good quantity of vitamins and minerals, which are necessary for optimum health and physiological functioning of body. Apple seeds are an excellent source of water-soluble vitamin C and B6 (pyridoxine) and fat-soluble vitamin E. Vitamins act as a coenzyme or a component of enzyme and participates in various metabolic functions. Vitamin B6 acts as a catalyst in various biochemical reactions like decarboxylation, deamination and transamination. Vitamin C possess an antioxidant activity and plays vital role in protecting against viral infections (Yaman et al., 2021).

Vitamin E has antioxidant properties, it includes the tocopherols i.e., α-, β-, γ- and δ-tocopherols. All these α- (439 mg/kg), β- (795 mg/kg), γ- (26.4 mg/kg) and δ- (19.9 mg/kg) tocopherols were identified in apple seed oil with highest quantity of β-tocopherol and least of δ-tocopherol. The authors described that total tocopherol (1127–1454 mg/kg) was abundant in apple seed oil and it was similar or higher than some commonly used edible oils (Madrera & Valles, 2018). Pieszka et al. (2015) confirmed the presence of α-, β-, γ- and δ-tocopherols in apple seed oil with concentrations of 41.7, 62.7, 13.6, and 21.8 mg/100 g respectively and α-(1.21 mg/100 g) and γ-(3.05 mg/100 g) tocotrienols. Vitamin-E-active compounds in apple seed (Golden and Starking variety) were reported to be 96 mg/100 g. Vitamin-E-active compounds in apple seed oil (golden variety apple) were 51.4, 28.3, 6.8 and 3.5 mg/100 g for α-, β-, γ- and δ-tocopherols respectively. Similarly, for starking variety apple seed oil it was 54.4–60.5, 30.9–34.3, 0.5 and 1.7 respectively for α-, β-, γ- and δ-tocopherols (Matthäus and Özcan 2015).

Studies also revealed that oil extracted using ultrasound treatments from different cultivars of crab and dessert apple seed had tocopherol content in the ranges of 130.6–202.6 mg/100 g and 191.1–379.1 mg/100 g respectively (Górnaś, 2015). Additionally, tocopherols, ascorbic acid was present in apple seed in the ranges of 11.66–19.68 mg/g, 104.1–161.3 mg/g respectively (Sharma & Nath, 2016).

The mineral composition of apple seeds includes excellent amounts of calcium (210 mg/100 g), magnesium (510 mg/100 g), sodium (214.1 mg/100 g), potassium (650 mg/100 g), phosphorous (666.5 mg/100 g) and iron (27.1 mg/100 g) (El-Safy et al., 2012). However, Yu et al., (2007) reported that apple seed flour had 110 mg/kg – iron, 270 mg/kg – calcium, 44 mg/kg – zinc, 510 – mg/kg magnesium, 650 mg/kg – potassium, 4.6 mg/kg – manganese, and 2 mg/kg – copper. These cations are paramount for various metabolic functions.

**Table 1**
Fatty acid composition of apple seed oils in different cultivars of apple.

| Variety | Oil (%) | Palmitic (C16:0) (%) | Palmitoleic acid (C16:1) (%) | Margaric acid (C17:0) (%) | Stearic acid (C18:0) (%) | Oleic acid (C18:1) (%) | Linoleic acid (C18:2) (%) | Linolenic acid (C18:3) (%) | Arachidic acid (C20:0) (%) | Behenic acid (C22:0) (%) | References |
|---|---|---|---|---|---|---|---|---|---|---|---|
| New Red Star | 24.32 | 6.60 | – | – | 1.96 | 38.55 | 50.70 | 0.19 | 1.49 | – | Yukui et al. (2009); |
| Fuji | 20.69–29.10 | 5.60–6.51 | 0.05–0.06 | – | 1.46–1.75 | 26.47–37.49 | 43.03–51.40 | 0.30–0.60 | 1.31–1.54 | 0.270–0.40 | Tian et al. (2010) |
| **Royal Gala** | 27.2 | 7.4 | 0.1 | 0.1 | 2.5 | 41.7 | 45.1 | 0.3 | 1.7 | 0.4 | Arain et al. (2012) |
| **Red Delicious** | 27.6 | 6.7 | 0.1 | 0.1 | 2.3 | 39.3 | 47.8 | 0.3 | 2.0 | 0.5 | |
| **Pyrus Malus** | 28.9 | 6.1 | 0.2 | 0.0 | 2.0 | 38.7 | 49.6 | 0.4 | 0.9 | 0.7 | |
| **Golden Delicious** | 26.8 | 7.1 | 0.1 | 0.1 | 3.1 | 44.5 | 40.5 | 0.3 | 2.0 | 0.6 | |
| **Kerr** | 21.77–27.49 | 7.80 | 0.06 | 0.07 | 1.60 | 28.88 | 59.37 | 0.45 | 1.09 | 0.06 | Górnaś et al. (2014) |
| **Kuku** | | 5.85 | 0.09 | 0.06 | 1.75 | 26.86 | 63.17 | 0.51 | 1.10 | 0.06 | |
| **Quaker Beauty** | | 6.92 | 0.11 | 0.06 | 1.59 | 26.25 | 63.21 | 0.42 | 0.96 | 0.07 | |
| **Riku** | | 5.78 | 0.07 | 0.06 | 0.04 | 28.88 | 61.88 | 0.50 | 1.27 | 0.27 | |
| **Ritika** | | 6.14 | 0.06 | 0.05 | 1.61 | 27.79 | 62.27 | 0.43 | 1.01 | 0.14 | |
| **Ruti** | | 5.84 | 0.09 | 0.06 | 1.60 | 26.82 | 63.35 | 0.40 | 1.22 | 0.06 | |
| **Beforest** | 12.06–23.03 | 7.14 | 0.17 | 0.08 | 1.26 | 20.68 | 67.94 | 1.35 | 0.79 | 0.07 | |
| **Antej** | | 8.25 | 0.18 | 0.07 | 1.30 | 21.62 | 66.18 | 0.78 | 0.96 | 0.12 | |
| **Kent** | | 7.10 | 0.09 | 0.07 | 1.63 | 29.00 | 59.67 | 0.46 | 1.23 | 0.04 | |
| **Sinap Orlovskjj** | | 8.33 | 0.11 | 0.07 | 1.62 | 24.95 | 62.73 | 0.68 | 1.00 | 0.06 | |
| **Zarja Alatau** | | 7.60 | 0.12 | 0.07 | 1.53 | 21.45 | 66.29 | 1.08 | 1.15 | 0.06 | |
| **Starking Apple** | 23.5–25.6 | 6.3–6.8 | 0.1 | – | 2.0–2.1 | 38.8–40.4 | 48.1–49.6 | 0.3 | – | – | Matthäus and Özcan (2015) |
| **Golden apple** | 21.9 | 7.0 | 0.1 | – | 1.9 | 35.7 | 51.7 | 0.6 | – | – | |
| **Apple seed (Mega-Sort company, Poland)** | 20.22 | 9.50 | – | – | 1.82 | 29.36 | 55.54 | 0.34 | 1.56 | 0.18 | Pieszka et al. (2015) |
| **Apple seed (cider industry)** | 16.2–21.1 | 7.1–7.6 | 0.5–0.7 | – | 1.7–2.1 | 32.2–34.2 | 53.7–57 | 1.0–1.4 | 0.2–0.3 | 0.2 | Madrera and Valles (2018) |

### 2.6. Phytochemical profile

Phytochemicals are secondary metabolites in fruits and vegetables that contribute to their antioxidant activity. Exploiting phytochemicals as natural source of antioxidant over synthetic ones has gained a lot of attention from past few years. Further it was noted that, waste or by-products generated in form of seeds, peels, pomace during fruit processing are concentrated sources of these phytochemicals (Sonja et al., 2009). Thus, focus and research on utilization of processing waste for value addition is the latest trend in food processing industry.

One such valuable by-product – apple seed, generated in ample during processing is a rich source of phenolic compounds (Xu et al., 2016). The concentration of total phenolic compounds (TPCs) in defatted apple seeds obtained from 12 different cultivars of dessert and cider apple was in the range of 18.4–99.8 mg/g. Amongst all TPCs, phloridzin (3256.3–22351.8 mg/kg of defatted dry matter) was predominantly present; it is known to be useful in treating type II diabetes mellitus and obesity (Fromm et al., 2012a). Another finding suggested that in comparison with its peel and pulp, apple seeds have higher polyphenols especially phloridzin (256.97–438.89 mg/100 g) thus, exhibiting highest antioxidant activity (Idared seeds: 3000 mg Trolox/100 g of fresh weight (f.w.) (Duda-Chodak et al., 2011). Seeds obtained from crab apple also called as wild apple (*Malus baccata (Lin.)* contained 112.23 ± 1.1 mg GAE/g total phenolic content and 154.16 ± 0.8 mg retinol equivalent (RE)/g of total flavonoids (Dadwal et al., 2018). When comparing between wild crab (*Malus baccata*) and 'Red Delicious' (*Malus domestica*) apple seed total phenolic compound was approximately same (1.78 mg/g) on fresh weight basis in both, while it increased on dry weight basis (15.92–14.56 mg/g respectively). Similarly, ascorbic acid content on fresh and dry weight basis was 11.66–19.68 mg/g, 104.13–161.27 mg/g respectively (Sharma & Nath, 2016). It was also revealed that defatted apple seeds had extractable polyphenols (2.7–6.7 mg gallic acid/g defatted matter), condensed tannins (2.4–3.6 mg gallic acid/g defatted matter), hydrolysable tannins (34.5–44.4 mg gallic acid/g defatted matter) (Madrera & Valles, 2018).

Additionally, hydroxycinnamic acid derivatives, 5-caffeoylquinic acid (chlorogenic acid), and phloretin-2′-xyloglucoside were also recovered from apple seeds (Fromm et al., 2012a). Other polyphenols identified in seeds included chlorogenic acid, hyperin, (+) Catechin, (−) epicatechin, caffeic acid, procyanidin B2, procyanidin B1, procyanidin C1, phloretin xyloglucoside and p-Coumaryl-quinic acid (Duda-Chodak et al., 2011; Fromm et al., 2012a; Xu et al., 2016). Smaller quantities of ascorbate in the range of 42.9–54.0 μg/g f.w. and some thiol compounds (L-cysteine, γ-glutamylcysteine, reduced glutathione, oxidized glutathione) were also detected in apple seeds of four different cultivars (Šampion, Jonagold, Gloster and Elise) commonly cultivated in Poland (Łata et al., 2005).

Further, (*Malus baccata (Lin.)* quantification using ultra-performance liquid chromatography (UPLC) and high-performance liquid chromatography (HPLC), showed that amongst phenolic compounds phloretin (88.39 μg/mg) and phloridzin (83.03 μg/mg) were in higher amounts along with traces of gallic acid, caffeic acid and protocatechuic acid in crab apple seeds (Dadwal et al., 2018). Coumarin derivatives were also discovered from the methanol and chloroform extracts of Creston apple seeds (Mustafa et al., 2018). It was also determined that apple seed contains sterols (≈3.8 mg/g) including β-sitosterol, stigmasterol, and campesterol (Patocka et al., 2020). Thus, due to presence of bioactive phytochemicals, apple seed extracts of different varieties exhibited good antioxidant activity when measured using different assays like DPPH, FRAP, $ABTS^+$, metal chelating, OH radical antioxidant, reducing power, superoxide anion scavenging, 2-deoxyribose degradation, hydroxyl free radical scavenging activity (Dadwal et al., 2018; Sharma & Nath, 2016; Xu et al., 2016).

Senica et al. (2019) confirmed presence of 23 phenolic compounds (which were grouped under six phenolic groups i.e., hydroxycinnamic acids, flavones, flavanols, flavonols and dihydrochalcones), in Golden Delicious variety harvested from orchards of four different locations (Azerbaijan, Russia, Serbia and Slovenia). The sum of phenolic content at time of harvest was highest in apple seed obtained from Russia (6.22 mg/g), followed by Serbia (5.91 mg/g), Slovenia (5.31 mg/g) and Azerbaijan (4.24 mg/g). However, it was observed that the phenolics content increased for all the seeds during storage of 8 months with highest values in Slovenia (7.9 mg/g) and least in Azerbaijan (5.3 mg/g). This variation or increase in the phenolic compounds can be attributed to the increased enzyme activity in the fruit during storage time, where the apple seeds get prepared during fruit senescence for germination. Also, some processes allocate these compounds into seeds resulting in their decrease in other parts of fruits (Senica et al., 2019).

Oil extracted from apple seed was also found to possess phytochemicals/bioactive compounds like phytosterols, tocopherols and phytols (Abbas et al., 2019). Among sterols, primarily β-sitosterol (ranging from 16.17 to 13.60%) and some other sterols like cycloartenol (9,19-Cyclolanost-24-en-3-ol), stigmast-4-en-3-one, avenasterol and campesterol were detected in apple seed oil extracted from different varieties of apple (Royal Gala, Red Delicious, Pyrus Malus and Golden Delicious) using GC-MS (Arain et al., 2012). In a study six phytosterols (β-sitosterol, cycloartenol, campesterol, stigmasterol, avenasterol and Δ7-stigmasterol) were identified in apple seed oil extracted from dessert and crab apple cultivars (Górnaś et al., 2014). Further, Pieszka et al. (2015), discovered presence of campesterol (219.8 μg/g), stigmasterol (13.2 μg/g), sitosterol (2629.3 μg/g), sitostanol (249.9 μg/g) and avenasterol (347.8 μg/g). Tocopherols (α and β) were also discovered in all seeds obtained from four apple varieties (Royal Gala, Red Delicious, Pyrus Malus and Golden Delicious) with Royal Gala and Golden Delicious having highest α-tocopherols (6.4 ± 1.1%) and β-tocopherols (1.8 ± 0.7%) respectively (Arain et al., 2012).

With changes in variety, production area, climatic conditions, sample storage, the composition of bioactive compounds in seed changes. However, the bioactive compounds present in these tiny apple seeds can be harnessed in formulating nutraceuticals and health promoting functional food products; thus, finding application in food processing industry. Bioactive compounds present in apple seeds are shown in Table 2.

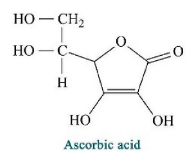
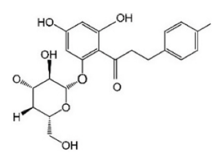
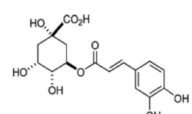
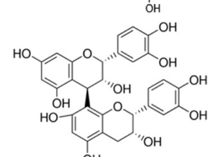
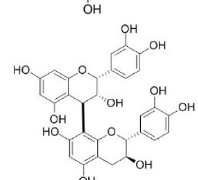
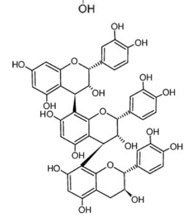
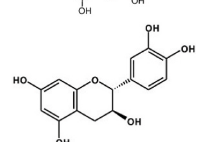
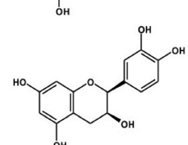
## 3. Bioactivities of the apple seed extract (ASE)

Extraction of apple seeds is done using different extraction techniques viz. conventional method, KM (kinetic maceration), UAE (ultra sound assisted extraction), MAE (microwave assisted extraction). This is the primary step in assessing the bioactive compounds and their bioactivities. These bioactive compounds can find application in direct or indirect ways in food processing industry for development of bioactive enriched novel food products or food packaging like edible coatings and nutraceutical formulations. These biological activities of apple seed extracts are summarized in Table 3.

### 3.1. Antioxidant activity

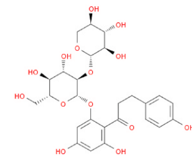
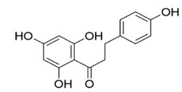
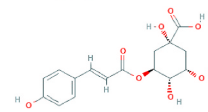
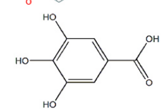
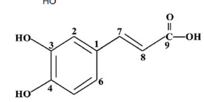
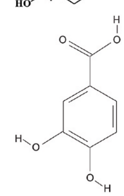
The antioxidant activity of apple seed is attributed to the higher phenolic compounds. ABTS (2,2′-azino-bis (3-ethylbenzothiazoline-6-sulfonic)) and DPPH (diphenyl-1-picrylhydrazyl) assays revealed that based on fresh weight (f.w.) apple seed extract of Idared seeds had antioxidant activity of 300 mg Trolox/100 g f.w. (ABTS) and 53 mg Trolox/100 g f.w. (DPPH) while for the Šampion seed extract it was ~210 mg Trolox/100 g f.w. and ~50 mg Trolox/100 g f.w. respectively. The study also indicated that both in Idared (Peel: ABTS 120 mg Trolox/100 g f.w., DPPH ~24 mg Trolox/100 g f.w.; Flesh: ABTS ~48 mg Trolox/100 g f.w., DPPH <10 mg Trolox/100 g f.w.) and Šampion (Peel: ABTS ~120 mg Trolox/100 g f.w., DPPH ~25 mg Trolox/100 g f.w; Flesh: ABTS~50 mg Trolox/100 g f.w., DPPH <15 mg Trolox/100 g f.w.) cultivars, seed extracts had better antioxidant activity as compared to peel and flesh due to presence of higher phenolic compounds in seeds

**Table 2**
Phytochemical profile of apple seeds.

| Region: variety/species/cultivars | Group | Structure of the compound | Composition | References |
|---|---|---|---|---|
| **Shimla, Himachal Pradesh, India: Crab apple, Red Delicious;**<br>**Hohenheim, Stuttgart, Germany: Dessert and Cider apple; Himachal Pradesh, India: Crab apple.** | Total phenolic compounds (mg/g) | – | 1.78 (f.w.)<br>15.92–14.56 (d.w.)<br>18.38–99.76 (defatted DM)<br>112.23 mg GAE/g | (Dadwal et al., 2018; Fromm et al., 2012a; Sharma & Nath, 2016) |
| **Himachal Pradesh, India: Crab apple.** | Total flavonoid (mg RE/g) | – | 154.16 mg RE/g | Dadwal et al. (2018) |
| **Pakistan (Quetta, Pishin, Ziarat, Mustang, Kalat, Kashmir, Chitral, Swat, Hunza and Gilgit): Royal Gala, Red Delicious, Pyrus Malus and Golden Delicious.** | α-tocopherol | – | 6.4% | Arain et al. (2012) |
| **Pakistan (Quetta, Pishin, Ziarat, Mustang, Kalat, Kashmir, Chitral, Swat, Hunza and Gilgit): Royal Gala, Red Delicious, Pyrus Malus and Golden Delicious.** | β-tocopherols | – | 1.8% | Arain et al. (2012) |
| **Shimla, Himachal Pradesh, India: Crab apple, Red Delicious.** | Ascorbic acid (mg/g) |  | 11.66–19.68 (f.w.)<br>104.13–161.27 (d.w.) | Sharma and Nath (2016) |
| **Warsaw, Poland: Šampion, Jonagold, Gloster and Elise.** | Total ascorbate (AA + DHAA) | – | 54.0–42.9 (μg/g f.w.) | Łata et al. (2005) |
| Concentration of polyphenols compounds | | | | |
| **Garlica Murowana, Krakow, Poland: Idared and Šampion; Hohenheim, Stuttgart, Germany: Dessert and Cider apple; Himachal Pradesh, India: Crab apple (*Malus baccata*); Yangling, Shaanxi, China: Gale Gala, Starking, Honeycrisp, Fuji, Qinguan, Golden Delicious and Qinyang.** | Phloridzin |  | 438.89–256.97 (mg/100 g of f.w.)<br>3256.3–22351.8 (mg/kg of defatted DM)<br>83.09 (μg/mg)<br>240.45–864.42 (mg/100 g of d.w.) | (Dadwal et al., 2018; Duda-Chodak et al., 2011; Fromm et al., 2012a; Xu et al., 2016) |
| **Garlica Murowana, Krakow, Poland: Idared and Šampion; Yangling, Shaanxi, China: Gale Gala, Starking, Honeycrisp, Fuji, Qinguan, Golden Delicious and Qinyang.** | Chlorogenic acid |  | 50.29–51.80 (mg/100 g of f.w.)<br>15.74–32.90 (mg/100 g of DW) | (Duda-Chodak et al., 2011; Xu et al., 2016) |
| **Garlica Murowana, Krakow, Poland: Idared and Šampion; Hohenheim, Stuttgart, Germany: Dessert and Cider apple.** | Procyanidin B2 |  | 11.39–9.71 (mg/100 g f.w.)<br>16.6–78.7 (mg/kg of defatted DM) | (Duda-Chodak et al., 2011; Fromm et al., 2012a; |
| **Garlica Murowana, Krakow, Poland: Idared and Šampion.** | Procyanidin B1 |  | 4.47–3.81 (mg/100 g of f.w.) | (Duda-Chodak et al., 2011; |
| **Garlica Murowana, Krakow, Poland: Idared and Šampion.** | Procyanidin C1 |  | 4.14–2.32 (mg/100 g of f.w.) | (Duda-Chodak et al., 2011; |
| **Garlica Murowana, Krakow, Poland: Idared and Šampion; Yangling, Shaanxi, China: Gale Gala, Starking, Honeycrisp, Fuji, Qinguan, Golden Delicious and Qinyang.** | (+) Catechin |  | 1.25–1.19 (mg/100 g of f.w.)<br>1.73–3.04 (mg/100 g of d.w.) | (Duda-Chodak et al., 2011; Xu et al., 2016) |
| **Garlica Murowana, Krakow, Poland: Idared and Šampion; Hohenheim, Stuttgart, Germany: Dessert and Cider apple; Yangling, Shaanxi, China: Gale Gala, Starking, Honeycrisp, Fuji, Qinguan, Golden Delicious and Qinyang.** | (−) Epicatechin |  | 3.87–2.08 (mg/100 g of f.w.)<br>4.2–51.6 (mg/kg of defatted DM)<br>4.11–7.43 (mg/100 g d.w.) | (Duda-Chodak et al., 2011; Fromm et al., 2012a; Xu et al., 2016) |

(*continued on next page*)

**Table 2** (*continued*)

| Region: variety/species/cultivars | Group | Structure of the compound | Composition | References |
|---|---|---|---|---|
| **Garlica Murowana, Krakow, Poland: Idared and Šampion; Hohenheim, Stuttgart, Germany: Dessert and Cider apple.** | Phloretin xyloglucoside |  | 5.83 26.97 (mg/100 g of f.w.) 103.6–1334.3 (mg/kg of defatted DM) | (Duda-Chodak et al., 2011; Fromm et al., 2012a) |
| **Himachal Pradesh, India: Crab apple.** | Phloretin |  | 88.39 (μg/mg) | Dadwal et al. (2018) |
| **Garlica Murowana, Krakow, Poland: Idared and Šampion.** | p-Coumaryl-quinic acid |  | 0.16–0.36 (mg/100 g of f.w.) | Duda-Chodak et al. (2011) |
| **Himachal Pradesh, India: Crab apple.** | Gallic acid |  | 5.79 (μg/mg) | Dadwal et al. (2018) |
| **Himachal Pradesh, India: Crab apple; Yangling, Shaanxi, China: Gale Gala, Starking, Honeycrisp, Fuji, Qinguan, Golden Delicious and Qinyang.** | Caffeic acid |  | 1.25 (μg/mg) 1.01–1.09 (mg/100 g d.w) | (Dadwal et al., 2018; Xu et al., 2016) |
| **Himachal Pradesh, India: Crab apple; Yangling, Shaanxi, China: Gale Gala, Starking, Honeycrisp, Fuji, Qinguan, Golden Delicious and Qinyang.** | Protocatechuic acid |  | 2.15 (μg/mg) 2.43–9.41 (mg/100 g d.w.) | (Dadwal et al., 2018; Xu et al., 2016) |

(f.w.: fresh weight, d.w.: dry weight, DM: dry matter, GAE: gallic acid equivalents, RE, retinol equivalents).

(Duda-Chodak et al., 2011). Similarly, Xu et al. (2016) reported that amongst the seven cultivars (Gale Gala, Starking, Honeycrisp, Fuji, Qinguan, Golden Delicious and Qinyang) analyzed, phenolic content was higher in seeds than peel or flesh. As the phenolic compounds were concentrated in seeds, it conferred higher antioxidant activity to the seeds than peels or flesh (on fresh weight basis) for DPPH (seed: 37.56–64.31 μM TE/g; peel: 17.87–18.01 μM TE/g; flesh: 16.16–18.12 μM TE/g), FRAP (seed: 57.59–397.70 μM TE/g; peel: 52.90–110.96 μM TE/g; flesh: 36.55–81.08 μM TE/g) and ABTS (seed: 220.52–708.02 μM TE/g; peel: 180.00–340.83 μM TE/g; flesh: 147.72–274.47 μM TE/g) assay.

However, Łata, et al. (2005) demonstrated that apple seeds from all four cultivars (Šampion, Jonagold, Gloster and Elise) had lowest antioxidant activity in terms of ascorbate peroxidase (18.3–38.5 nkat/g f.w.), and catalase (2.3–3.8 nkat/g f.w.) than peels (ascorbate peroxidase: 67.7–90.9 nkat/g f.w. and catalase: 5.4–7.4 nkat/g f.w.). This was attributed to lower ascorbate content in seeds than peel. While, the glutathione reductase activity was higher in the seeds (7.0–11.6 nkat/g f.w.) compared to peels (5.3–9.0 nkat/g f.w.) and this could be due to the presence of low molecular weight thiol compounds. Among the different cultivars the total ascorbate was more in Elise (54.0 μg/g f.w.), followed by Jonagold (53.9 μg/g f.w.), Šampion (45.9 μg/g f.w.) and least in Gloster (42.9 μg/g f.w.) (Łata et al., 2005).

Sharma and Nath (2016) corroborated that peel (71.24%, 65.51%) had higher reducing power than seed (58.42%, 54.13%) or pulp (51.40%, 56.54%) in crab and 'Red Delicious' apples respectively. Percent inhibition was higher in crab apple (Peel: 29.2%, seed: 24.74%, pulp: 24.12%) than 'Red Delicious'apple (Peel: 15.8%, seed: 18.23%, pulp: 19.94%) when measured using superoxide anion scavenging activity. The 2-deoxyribose degradation assay indicated that, per cent inhibition in crab apple seeds was 25.37% while it was 33.46% for 'Red Delicious' apple seeds. Dadwal et al. (2018) also confirmed that apple seed exhibited antioxidant activity in terms of DPPH (8.45–30.47% at conc. of 20–200 μg/mL), ABTS (8.28–72.42% at conc. of 20–25 μg/mL), hydroxyl radical scavenging activity (63.46% at conc. of 1 mg/mL) and metal chelating (35.11% at conc. of 1 mg/mL). This antioxidant capacity was attributed to the presence of total phenolic compounds like protocatechuic acid (2.15 μg/mg) and gallic acid (5.79 μg/mg), phloretin (88.39 μg/mg), Phloridzin (83.09 μg/mg), and flavonoid content.

Likewise, slightly higher DPPH radical scavenging activity in the range of 49.2–86.9% at different concentrations (20, 50 and 100 μg/mL) for red delicious and golden delicious apple seed flour was reported. Further, reducing power in the range of 29.8–61.0% and inhibiting the lipid peroxidation in the range of 29.1–86.3% was reported by Manzoor et al. (2021b). Apart from apple seeds, oil extracted from the seeds also had DPPH scavenging activity with $IC_{50}$ values of 7.91 μg/mL and 8.34 μg/mL for Fuji and New Red Star apple seeds oil respectively (Tian et al., 2010). Supporting this another researcher outlined that apple seed oil had DPPH activity with $IC_{50}$ value of 4.8–5.8 mg/mL (Madrera & Valles, 2018). Walia et al., (2018) also indicated that apple seed oil had good antioxidant capacity and the DPPH assay confirmed percentage inhibition as 38.12%, 60.34%, 71.23% and 88.74% for 25, 50, 75 and 100 μg/mL respectively. However, antioxidant activity of apple seed oil (IC50: 40.06 μg/mL) was slightly lower than the standard antioxidant viz., butylated hydroxy anisole (BHA) (IC50: 9.97 μg/mL). Overall, the phytochemicals in the apple seeds could scavenge reactive oxygen species by either binding or degradation.

### 3.2. Anti-diabetic properties

One of the approaches for treatment of diabetes is reducing the activities of α-glucosidase, which in turn delays the carbohydrate digestion leading to slower release of glucose in blood stream post lunch. Ci et al. (2018) investigated α-glucosidase and α-amylase of core, flesh, peel and

**Table 3**
Summary of biological activities of apple seed extracts.

| Variety (Region) | Method of extraction | Compounds/ component studied | Study type/cell type | Major findings | References |
|---|---|---|---|---|---|
| Antimicrobial potential | | | | | |
| **Granny Smith and Red Delicious apples (Mosul, Iraq)** | Non-serial and serial style: KM, UAE and MAE extraction methods | Apple seed extract | *Disc diffusion method against Klebsiella pneumonia, Pseudomonas aeruginosa, Escherichia coli and Haemophilus Influenzae* | Exhibited antimicrobial activity against bacterial strains at 5 and 10% **Inhibition zone range (mm):** **Granny Smith apple seed extract (5%):** 5.26–8.33 mm, 8.37–11.36 mm, 8.84–10.03 mm and 6.92–9.75 mm for *K. pneumonia, P. aeruginosa, E. coli,* and *H. influenzae* **Granny Smith apple seed extract (10%):** 7.05–10.09 mm, 11.12–13.57 mm, 11.68–13.10 mm, 9.30–13.55 mm for *K. pneumonia, P. aeruginosa, E. coli,* and *H. influenzae* **Red Delicious apple seed extract (5%):** 5.37–8.43 mm, 8.23–11.52 mm, 9.06–10.07 mm, 7.03–9.89 mm for *K. pneumonia, P. aeruginosa, E. coli* and *H. influenzae.* **Red Delicious apple seed extract (10%):** 7.63–10.30 mm, 11.45–13.97 mm, 11.88–13.39 mm, 9.63–13.80 mm for *K. pneumonia, P. aeruginosa, E. coli* and *H. influenzae*. | Mustafa et al. (2020) |
| **Granny Smith and Red Delicious apples (Mosul, Iraq)** | Non-serial and serial style: KM, UAE and MAE extraction methods | Apple seed extract | Disc diffusion method against *Aspergillus niger* and *Candida albicans* | Exhibited antimicrobial activity against fungal strains at 5 and 10% **Inhibition zone range (mm):** **Granny Smith apple seed extract (5%):** 3.12–4.40 mm, 3.79–5.49 mm, for *A. niger* and *C albicans* **Granny Smith apple seed extract (10%):** 5.90–8.11 mm, 6.96–8.74 mm, for *A. niger* and *C albicans* **Red Delicious apple seed extract (5%):** 3.14–4.58 mm, 4.02–5.57 mm, for *A. niger* and *C albicans* **Red Delicious apple seed extract (10%):** 5.86–8.26 mm, 7.07–8.85 mm, for *A. niger* and *C albicans* | Mustafa et al. (2020) |
| **Fuji and New star red apple (Xinjiang, China)** | Oil extracted using Soxhlet method | Apple seed oil | Disc diffusion method for bacterial (*E. coli, Salmonella* sp.*, Bacillus subtilis, S. aureus)* and fungal (*Candida* sp.*, S. cerevisiae, A. flavus, Penicillium citrinum, Mucor* sp., and *Rhizopus* sp.*)* strains | Both apple seed oils showed strong antimicrobial activity against bacterial strains that fungal strains. **Inhibition zones (mm):** **Fuji apple seed oil:** 15.00 (*E. coli, Salmonella* sp.*, S. aureus*); 15.50 (*Bacillus subtilis)*, 10.50 (*Candida* sp.*, S. cerevisiae);* 8.00 (*Penicillium citrinum)*, 7.50 (*A. flavus*), 6.00 (*Rhizopus* sp.*)* and 5.00 (*Mucor* sp.). **New star red apple seed oil:** 18.00 (*Salmonella* sp.); 17.50 (*S. aureus*); 17.00 (*E. coli, Bacillus subtilis)*, 10.50 (*S. cerevisiae);* 9.50 (*Candida* sp.*),* 7.50 (*A. flavus, Penicillium citrinum)*, 4.50 (*Rhizopus* sp.*)* and 6.50 (*Mucor* sp.). | Tian et al. (2010) |
| **Golden delicious and Red delicious, Srinagar, Jammu and Kashmir** | Milled in laboratory mill | Apple seed flour | Inhibition against *Bacillus subtilis* and *S. aureus*. | The zone of inhibitions for *Bacillus subtilis and S. aureus* Golden delicious: 8.00 mm, 10.06 mm Red delicious: 7.59 mm, 8.02 mm respectively | Manzoor et al. (2021b) |
| **Red Delicious apples** | Solvents used: n-hexane, chloroform, methanol and water. Non-serial and serial | Coumarins extracted from apple seed | Antibacterial activity against *Pseudomonas aeruginosa, Klebsiella pneumonia, Haemophilus influenzae and* | The coumarins exhibited antifungal and anti-bacterial activity Minimum inhibitory concentrations: | Mohammed and Mustafa (2020) |

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**Table 3** (*continued*)

| Variety (Region) | Method of extraction | Compounds/ component studied | Study type/cell type | Major findings | References |
|---|---|---|---|---|---|
| | style: KM, UAE and MAE extraction methods | | *Escherichia coli, Candida albicans, Aspergillus niger* | *Pseudomonas aeruginosa:* 2.15–7.50 μg/mL,<br>*Klebsiella pneumonia*: 2.05–7.50 μg/mL,<br>*Haemophilus influenzae*: 2.25–9.0 μg/mL,<br>*Escherichia coli*: 2.45–9.0 μg/mL,<br>*Candida albicans*:9.0–24.0 μg/mL,<br>Aspergillus niger: 13.0–32.0 μg/ | |
| Antioxidant effect | | | | | |
| **Idared and Šampion (Garlica Murowana, Krakow, Poland)** | Methanolic extraction | Apple seed | Antioxidant activity using ABTS, DPPH assay | Idared seed extracts:<br>ABTS: 300 mg Trolox/100 g f.w.<br>DPPH: 53 mg Trolox/100 g f.w.<br>Šampion seed extract:<br>ABTS: ~210 mg Trolox/100 g f.w.<br>DPPH: ~50 mg Trolox/100 g f.w. | Duda-Chodak et al. (2011) |
| **Šampion, Jonagold, Gloster and Elise (Poland)** | HPLC | Apple seed | Antioxidant enzyme activity: ascorbate peroxidase, glutathione reductase, catalase, total ascorbate and total glutathione | Total ascorbate: 42.9–54.0<br>L-cysteine: 6.8–19.6<br>γ- -glutamylcysteine: 1.0–2.3<br>Total glutathione: 126.3–150.9<br>Glutathione reductase: 7.0–11.6<br>Ascorbate peroxidase:18.3–38.5<br>Catalase: 2.3–3.8 | Łata et al. (2005) |
| **Apple pomace, obtained from Himachal Pradesh Horticultural Produce Marketing and Processing Corporation Ltd. juice industry** | Seeds were separated from pomace and extracted using hexane in Soxhlet apparatus to obtain oil | Apple seed oil | DPPH activity | The percent DPPH inhibition by apple seed oil at 25, 50, 75 and 100 μgmL/L was 38.12%,<br>60.34%, 71.23% and 88.74% respectively<br>When compared to BHA ($IC_{50}$: 9.97 μgmL/L), apple seed oil ($IC_{50}$: 40.06 μgmL/L) had low antioxidant capacity. | Walia et al., (2018) |
| **Seeds supplied by Martinez Sopeña Hermanos S. L.** | Hexane extraction | Apple seed oil | DPPH | $IC_{50}$: 4.8–5.8 mg of oil/mL | Madrera and Valles (2018) |
| **Wild crab apples- *M. baccata* (Himalayan region at Keylong; Lahaul-Spiti, Himachal Pradesh, India)** | Ultrasonic extraction technique | Apple seed | Hydroxyl radical,<br>ABTS,<br>DPPH,<br>Metal chelating | Results ranging at different concentrations<br>Hydroxyl radical: 63.46% at conc. of 1 mg/mL; DPPH: 8.45–30.47% at conc. of 20–200 μg/mL; ABTS: 8.28–72.42% at conc. of 20–25 μg/mL; Metal chelating: 35.11% at conc. of 1 mg/mL | Dadwal et al. (2018) |
| **Crab apple (*M. baccata*) (Shimla, Himachal Pradesh)** | Ethanol extraction | Apple seed | Reducing power, Superoxide Anion Scavenging Activity, 2-deoxyribose degradation assay, Total phenols, Ascorbic acid | **Reducing power:** 58.42%<br>**Superoxide Anion Scavenging Activity:** 24.74%<br>**2-deoxyribose degradation assay:** 25.37%<br>**Total phenols:** 1.78 mg/g f.w. and 15.92 mg/g d.w. (dry weight)<br>**Ascorbic acid:** 11.66 mg/g f.w. and 104.13 mg/g d.w. | Sharma and Nath (2016) |
| **Red Delicious seeds (Shimla, Himachal Pradesh)** | Ethanol extraction | Apple seed | Reducing power, Superoxide Anion Scavenging Activity, 2-deoxyribose degradation assay, Total phenols, Ascorbic acid | **Reducing power**: 54.13%<br>**Superoxide anion scavenging activity**: 18.23%,<br>**2-deoxyribose degradation assay**: 33.46%<br>**Total phenols:** 1.78 mg/g f.w. and 14.56 mg/g d.w.<br>**Ascorbic acid:** 19.68 mg/g f.w. and 161.27 mg/g d.w. | Sharma and Nath (2016) |
| **Golden delicious and Red delicious, Srinagar, Jammu and Kashmir** | Milled in laboratory mill | Seed flour | Antioxidant potential using: DPPH assay, reducing power, lipid peroxidation assay | **DPPH radical scavenging activity**: 49.2–86.9%<br>Golden delicious: 42.7, 55.3, 82.3% at 25, 50 and 100 μg/mL.<br>Red delicious: 49.2, 68.2, 86.9% at 25, 50, and 100 μg/mL.<br>**Reducing power:** 29.8–61.0%<br>Red delicious: 29.8%, 38.8%, 58.2%, at 25, 50, and 100 μg/mL.<br>Golden delicious: 33.5%, 44.3%, 61.0% at 25, 50, and 100 μg/mL.<br>**Inhibiting the lipid peroxidation**: 29.1–86.3%<br>Golden delicious: 29.1%–78.1% at 25–100 μg/mL. | Manzoor et al. (2021b) |

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Table 3 (*continued*)

| Variety (Region) | Method of extraction | Compounds/ component studied | Study type/cell type | Major findings | References |
|---|---|---|---|---|---|
| | | | | Red delicious: 38.2%–86.3% at 25–100 μg/mL. | |
| Antidiabetic potential | | | | | |
| **Golden delicious and Red delicious, Srinagar, Jammu and Kashmir** | Milled in laboratory mill | Seed flour | Anti-diabetic activity: inhibition of α-glucosidase | Exhibited inhibition against α-glucosidase at 20, 50 and 100 μg/mL concentrations<br>Red delicious: 34.5%, 53.7% and 86.6% golden delicious:29.3%, 46.7% and 79.9% respectively | Manzoor et al. (2021b) |
| **Mini apple (*Malu domestica* cv. 'Alps Otome')** | Ultrasound assisted extraction using 80% ethanol as solvent | Core, flesh, peel and seeds | Anti-diabetic activity: inhibition of α-glucosidase and α-amylase | It was revealed that seeds had highest inhibitory potential (IC50: 1.0 μg/mL, and 1.1 μg/mL). Amongst polyphenol fractions viz. Fra.I (monomeric polyphenols), Fra.II (oligomeric polyphenols), and Fra.III (polymeric polyphenols) obtained after Sephadex LH-20 column chromatography, Fra.III had highest α-Glucosidase inhibitory activity with IC50: 0.6 μg/mL, followed by Fra.II (IC50: 1.0 μg/mL), and Fra. I (IC50:11.8 μg/mL). | Ci et al. (2018) |
| Anti-cancer effect | | | | | |
| **Creston apple seeds, Mosul, Iraq** | Water, methanol, chloroform or n-hexane extracts obtained by soxhlet extraction and kinetic maceration | Coumarins isolated from apple seed and | Effect of natural (N) coumarins (N1: 5,6-dihydropyrone(4,3-g) coumarin; N2: 5-Ethyl-6,7-dimethylcoumarin; N3: 7-Hydroxy-4-methoxycoumarin) and semisynthetic (S) derivatives from N3 (S4: 7-Acetoxy-4-methoxycoumarin; S5: 8-Acetyl-7-hydroxy-4-methoxycoumarin; S6: 4,7-Dimethoxycoumarin) on three cancer cell lines i.e., MCF-7 (breast cancer), AMN3 (mammary gland cancer cell line), and HeLa (cervical cancer) using MTT cell viability assay. | **MCF-7:** S6 (8.22 μg/mL) had lower $IC_{50}$ values than that of 5- Fluorouracil (12.50 μg/mL) used as positive control, followed by S4 (8.30 μg/mL), N3 (9.83 μg/mL), and S5 (12.48 μg/mL) indicating their cytotoxic potential against treatment for breast cancer.<br>**AMN3:** All the compounds (N1, N2, N3, S4, S5, S6) had higher $IC_{50}$ (21.22–72.20 μg/mL) values than positive control (15.20 μg/mL), however, S6 (21.22 μg/mL) could be used as potential candidate for treatment of mammary gland cancer.<br>**HeLa:** All the compounds had higher $IC_{50}$ (21.50–42.30 μg/mL) values than positive control (11.90 μg/mL), however, S5 (21.50 μg/mL) could be used as potential candidate for treating cervical cancer. | Mustafa et al. (2018) |
| **Apple pomace, obtained from Himachal Pradesh Horticultural Produce Marketing and Processing Corporation Ltd. juice industry** | Seeds were separated from pomace and extracted using hexane in Soxhlet apparatus to obtain oil | Apple seed oil | Cytotoxic activity | Apple seed oil exhibited cytotoxic activity (*in vitro*) against Chinese hamster (CHOK1), A549 (human lung carcinoma) and human cervical cancer cells (SiHa) at different concentrations (0.05–2 mg/L) | Walia et al., (2018) |
| Cardioprotective, Anti-obesity effect | | | | | |
| **Golden delicious and Red delicious, Srinagar, Jammu and Kashmir** | Milled in laboratory mill | Seed flour | Inhibition of pancreatic lipase | Percent inhibition of lipase activity at 25, 50 and 100 μg/mL:<br>**Red delicious apple seed flour**: 24.9%, 47.0%, 72.6% percent inhibition<br>**Golden delicious apple seed flour**: 18.5%, 38.6%, 65.8% | Manzoor et al. (2021b) |
| **Mini apple (*Malu domestica* cv. 'Alps Otome')** | Ultrasound assisted extraction using 80% ethanol as solvent | Seed, flesh, core and peel | Inhibition of lipase | Lipase inhibitory activity was highest in seeds ($IC_{50}$: 3.8 μg/mL), followed by peel ($IC_{50}$: 6.2 μg/mL), flesh (IC50: 8.3 μg/mL), and core ($IC_{50}$: 8.9 μg/mL) | Ci et al. (2018) |

seeds. It was revealed that seeds had higher inhibitory potential ($IC_{50}$: 1.0 μg/mL, and 1.1 μg/mL), than peel ($IC_{50}$: 2.6 μg/mL, and 10.7 μg/mL), flesh ($IC_{50}$: 2.7 μg/mL, and 17.6 μg/mL), and core ($IC_{50}$: 3.9 μg/mL, and 58.2 μg/mL) respectively. Authors also confirmed that amongst polyphenol fractions viz. fraction I (monomeric polyphenols), fraction II (oligomeric polyphenols), and fraction III (polymeric polyphenols) obtained after Sephadex LH-20 column chromatography, fraction III had highest α-glucosidase inhibitory activity with $IC_{50}$: 0.6 μg/mL, followed by fraction II ($IC_{50}$: 1.0 μg/mL), and fraction I ($IC_{50}$:11.8 μg/mL).

Manzoor et al. (2021b) demonstrated anti-diabetic potential of Red Delicious and Golden Delicious apple seed flour extracts. Both the extracts i.e., red delicious and golden delicious, inhibited α-glucosidase at different concentrations (μg/mL) viz., 20 (34.5% and 29.3%), 50 (53.7% and 46.7%) and 100 (86.6% and 79.9%) μg/mL respectively. It was also noted that extracts of Red Delicious apple seeds had better inhibition (Fig. 1). This anti-diabetic activity of apple seed is attributed to the phenolic composition, especially, the presence of proanthocyanidins like (+)epicatechin and (−)catechin.

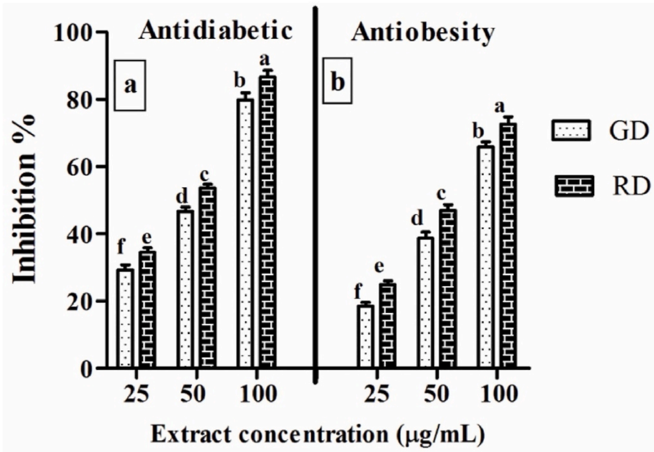


**Fig. 2.** Antidiabetic and antiobesity effects of apple seed flour [golden delicious (GD) and red delicious (RD)] demonstrated by inhibition of α-glucosidase (a) and pancreatic lipase (b). (Source: reproduced with permission from Elsevier).

### 3.3. Antimicrobial activity

There is an increasing microbial resistance developed against the antibiotics that is aggravating the problems of infections risking human health. This microbial resistance is thereby posing a challenge for researchers to discover new antimicrobial compounds. Existing literature suggests that plant based bioactive compounds/phytochemicals possess antimicrobial properties. Even in ancient medicine like ayurveda, plant extracts were widely used for treating various ailments. Thus, there is rising trend in utilization of wide range of plant extracts as antifungal or anti-bacterial agents given their phytochemical properties (Cheesman et al., 2017; Gupta & Birdi, 2017).

Red Delicious and Granny Smith apple seeds were extracted using different techniques (KM, UAE, MAE) and two styles (serially and non-serially sorted polarity) were followed in each technique. All these extracts were tested for their antimicrobial activity against both bacteria (*Klebsiella pneumonia, Pseudomonas aeruginosa, Escherichia coli,* and *Haemophilus influenzae*) and fungus (*Aspergillus niger* and *Candida albicans*) using agar disc diffusion method. When treated with 10% Granny Smith apple seed extract inhibition zones of 7.05–10.09 mm, 11.12–13.57 mm, 11.68–13.10 mm, 9.30–13.55 mm for *K. pneumonia P. aeruginosa, E. coli,* and *H. influenzae* were noted; while at 5% they were 5.26–8.33 mm, 8.37–11.36 mm, 8.84–10.03 mm and 6.92–9.75 mm respectively. Similarly, in Red Delicious apple seed extract they were in the following ranges 5.37–8.43 mm (5% extract), 7.63–10.30 mm (10% extract) for *K. pneumonia*, 8.23–11.52 mm (5% extract), 11.45–13.97 mm (10% extract) for *P. aeruginosa*, 9.06–10.07 mm (5% extract), 11.88–13.39 (10% extract) for *E. coli* and 7.03–9.89 mm (5% extract), 9.63–13.80 mm (10% extract) for *H. influenzae*. Amongst all the extracts in both varieties, extraction with UAE (non-serially sorted polarity) at 10% concentration had highest inhibition zones towards all the bacterial species tested. In the same way for fungal strains, UAE (non-serially sorted polarity) at 10% concentration exhibited highest inhibition zones for both Red Delicious (*A. niger*: 8.26 mm and *Candida albicans*: 8.85 mm) and Granny Smith (*A. niger*: 8.11 mm; *C albicans*: 8.74 mm) apple seed extracts. Overall, researchers concluded that, UAE extraction could exhibit highest antimicrobial activity and this could be due to the better extraction of phenolic compounds (Mustafa et al., 2020).

Manzoor et al. (2021b) confirmed antimicrobial properties of apple seed flour against *Bacillus subtilis and S. aureus*. The zone of inhibitions for Golden Delicious (8.00 mm, 10.06 mm) and Red Delicious (7.59 mm, 8.02 mm) against *Bacillus subtilis and S. aureus* respectively (Fig. 3) indicating that the phenolic compounds (quercetin, quercitrin, phlorizdin proanthocyanidin oligomers, procyanidin B2, epicatechin) help in rupturing the cells of bacteria hindering their growth. Mohammed and Mustafa (2020) reported that coumarins extracted from Red Delicious apple seeds possessed antimicrobial properties against *Pseudomonas aeruginosa, Klebsiella pneumonia, Haemophilus influenzae* and *Escherichia coli*, and *Candida albicans* and *Aspergillus niger*. This antimicrobial activity exhibited by different furanocoumarins viz., (E)-12-(2′-Chlorovinyl)bergapten, 12-(1′,1′-dihydroxyethyl)bergapten, 12-(2′-chloropropan-2′-yl)-8-hydroxybergapten, and 12-Hydroxy-11--chloromethylbergapten purified and identified from apple seed was measured as minimum inhibitory concentration and it was in the following ranges 2.15–7.50 μg/mL, 2.05–7.50 μg/mL, 2.25–9.0 μg/mL, 2.45–9.0 μg/mL, 9.0–24.0 and 13.0–32.0 μg/mL for *Pseudomonas aeruginosa, Klebsiella pneumonia, Haemophilus influenzae, Escherichia coli, Candida albicans* and *Aspergillus niger* respectively. These results suggest that apple seed coumarins have both anti-bacterial and anti-fungal activity.

Oil extracted from apple seed also exhibited antimicrobial activity when tested against *E. coli, Salmonella* sp.*, Bacillus subtilis, Staphylococcus aureus, Candida* sp.*, Saccharomyces cerevisiae, Aspergillus flavus, Penicillium citrinum, Mucor* sp., and *Rhizopus* sp. using disc diffusion method. It was noted that the diameters of inhibition zones for Fuji and new red

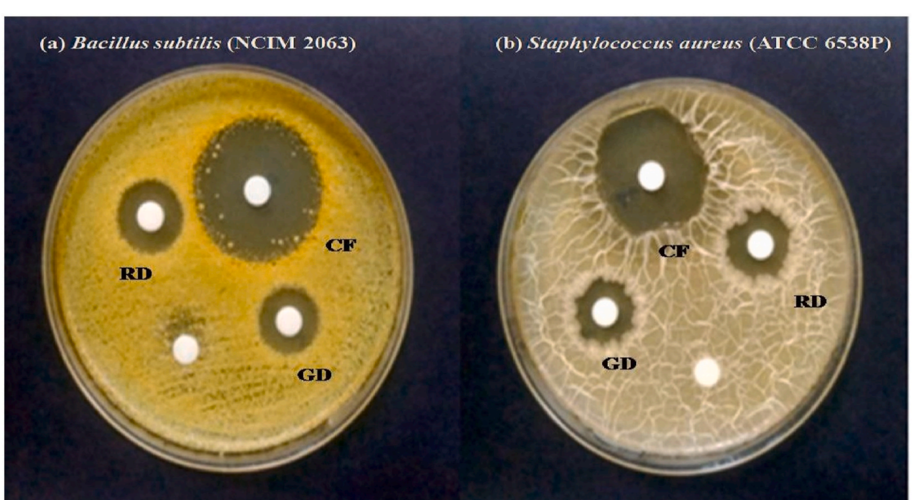


**Fig. 3.** Antimicrobial activity of red delicious (RD), golden delicious (GD) seed extracts and ciprofloxacin (CF) against (a) *S. aureus* growth (b) *B. Subtils* (Source: reproduced with permission from Elsevier). (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

star apple seed oil were in the ranges of 5.00–15.50 mm and 4.50–18.00 mm while the minimum inhibitory concentrations were 0.3–0.4 and 0.3–0.6 mg/mL respectively. Their results also indicated that both apple seed oils were more effective against the bacterial strains (Fuji apple seed oil: 15.00–15.50 mm; New star red apple seed oil: 17.50–18.00 mm) than the fungal (Fuji apple seed oil: 5.00–10.50 mm; New star red apple seed oil: 4.50–10.50 mm) strains (Tian et al., 2010).

### 3.4. Cardio-protective and anti-obesity

Cardiovascular diseases (CVDs) are on rise every year, due to the unhealthy life style and food habits. Obesity is one of the major factors contributing towards increasing threat of CVD. Inhibiting pancreatic lipase activity can arrest the rise in postprandial increase in plasma triglyceride levels; thus, one perspective in managing obesity can be inhibition of the pancreatic lipase enzyme.

Lipase inhibition ability of mini apple seeds was demonstrated by Ci et al. (2018). The authors confirmed that as compared to peel ($IC_{50}$: 6.2 μg/mL), core ($IC_{50}$: 8.9 μg/mL) and flesh ($IC_{50}$: 8.3 μg/mL), seeds ($IC_{50}$: 3.8 μg/mL) had higher inhibition activity indicated by lowest $IC_{50}$ values. Manzoor et al. (2021b) also indicated that Red Delicious and Golden Delicious apple seed flour extracts effectively inhibited pancreatic lipase, in dose dependent fashion at 25, 50 and 100 μg/mL. Red delicious apple seed flour (24.9%, 47.0%, 72.6%) had higher percentages of inhibition than Golden Delicious apple seed flour (18.5%, 38.6%, 65.8%) at all concentrations (25, 50 and 100 μg/mL) respectively (Fig. 2.). This inhibition potential of apple seed flour was attributed to the presence of phenolic compounds and their protein binding affinity. Especially phenolic compounds such as oligomeric proanthocyanidins (possessing lipase inhibition activity), were plenty in 'Red delicious' apple seed than 'Golden delicious'. Thus, apple seed polyphenols or apple seed flour can be used as alternative solutions in prevention of obesity and related CVDs.

Another study revealed that supplementation of defatted apple seed meal (containing 0.24% amygdalin) in male Wistar rats for two weeks significantly increased the HDL cholesterol level (55%), when compared to control group (41%) and rats fed with amygdalin (43%). The dietary fiber present in apple seed might be one main reason for the positive influence blood lipid profile with increase in HDL levels. The apple seed meal supplementation also resulted in decreased lipid peroxidation in liver (65.2 nmol/g tissue) than amygdalin group (76.4 nmol/g tissue) and control group (76.1 nmol/g tissue); this could be due to presence of phenolic compounds like phlorizin (Opyd et al., 2017).

### 3.5. Anticancer activity

Mustafa et al. (2018) indicated that coumarins (natural (N) coumarins (N1: 5,6-dihydropyrone(4,3-g)coumarin; N2: 5-ethyl-6,7-dimethylcoumarin; N3: 7-hydroxy-4-methoxycoumarin) and semisynthetic (S) derivatives from N3 (S4: 7-acetoxy-4-methoxycoumarin; S5: 8-acetyl-7-hydroxy-4-methoxycoumarin; S6: 4,7-dimethoxycoumarin)) isolated from Creston apple seeds when analyzed using MCF-7 (breast cancer), AMN3 (mammary gland cancer), and HeLa (cervical cancer) cell line exhibited cytotoxic potential against various cancer cells. Compounds S6 ($IC_{50}$: 8.22 μg/mL), S4 ($IC_{50}$: 8.30 μg/mL), N3 ($IC_{50}$: 9.83 μg/mL), and S5 ($IC_{50}$: 12.48 μg/mL) were identified to have potential in treating breast cancer. Compound S6 ($IC_{50}$: 21.22 μg/mL) was also identified effective against mammary gland cancer cell line; while S5 (21.50 μg/mL) was potent against cervical cancer cell lines. However, the mechanisms behind this anti-cancer activities were not identified. Another study also confirmed the anti-cancer activity of novel coumarins extracted from apple seeds. Researchers indicated that novel coumarins viz., officinalin (R1), 8-(*tert*-butyl)officinalin (R2), 8-hydroxyofficinalin (R3), officinalin-8-acetic acid (R4), and 8-(2′-hydroxypropan-2′-yl) officinalin (R5) exhibited an anti-tumor activity with $IC_{50}$ values of 104.45 μM, 50.40 μM, 25.11 μM, 52.47 μM and 25.90 μM against HeLa cancer cell line; while they were 12.46 μM, 50.69 μM, 45.44 μM, 24.17 μM, 45.70 μM and 26.39 μM respectively for MCF-7 cell line (Khalil & Mustafa, 2020).

Walia et al., (2018) indicated that apple seed oil can inhibit the growth of cancer cells. The ability of apple seed oil was tested *in vitro* against Chinese hamster (CHOK1), A549 (human lung carcinoma) and human cervical cancer cells (SiHa) at different concentrations. The cytotoxicity was measured at 24, 48 and 72 h. It was observed that as the concentration and the hours increased, the cytotoxicity also increased. The control (Vinblastine at 1 μg/g concentration) had highest cytotoxicity against CHOK1 (90%), while apple seed oil exhibited 88.6% cytotoxicity in 72 h. Similarly, the apple seed oil also inhibited growth of SiHa cells by 56% at 2 mg concentration, and they were similar to Vinblastine at 1 μg/g concentration (54.2%) after 72 h. The percent inhibition exhibited by apple seed oil ranged between 52.9 and 64.4 mg/mL for A549 (human lung carcinoma) during 24–72 h. The authors suggested that given the phenolic composition, apple seed oil possesses beneficial phenolic and phytonutrient properties and can be used as pharmacological agent. These findings indicate that phenolic compounds like coumarin present in apple seed are imparting potential anticancer activity; but the exact mechanism still remains undiscovered.

## 4. Applications of apple seed in foods

### 4.1. Functional food properties and applications as ingredient in foods

As discussed in the earlier sections, apple seed and its oil are good source of protein, lipids, essential fatty acids, vitamins, minerals, phytochemicals and antioxidants; thus, it can be explored as functional ingredient in food preparation. Very limited studies are conducted in application of apple seed in food formulations. However, given their nutritional profile they can be a promising ingredient for value addition and enrichment of calorie-dense foods.

Purić et al. (2020) incorporated defatted apple seed cake (DASC) of Golden Delicious, Idared and Šumatovka varieties at 5% and 20% for enrichment of wheat bread. The nutritional profile in terms of protein, ash, fat, soluble dietary fiber and insoluble dietary fiber increased for both 5% and 20% DASC incorporated breads of all 3 varieties viz., Golden Delicious (12.46–15.31 g/100 g, 3.19–3.20 g/100 g, 2.24–3.42 g/100 g, 2.58–2.08 g/100 g, 2.94–5.80 g/100 g), Idared (12.42–15.58 g/100 g, 2.89–3.13 g/100 g, 2.33–3.40 g/100 g, 2.27–1.59 g/100 g, 2.37–7.24 g/100 g) and Šumatovka (12.76–15.99 g/100 g, 3.0–3.03 g/100 g) compared to control breads made with only wheat flour (11.48 g/100 g, 2.99 g/100 g, 1.81 g/100 g, 1.01 g/100 g, 2.57 g/100 g) respectively. Carbohydrate content decreased from 49.64 g/100 g in control to 37.81–47.85 g/100 g in breads made with DASC (5 and 20%); this was due to the increased fiber content by the apple seeds of all varieties. The total phenolic content and antioxidant activity in terms of DPPH were highest in 20% DASC incorporated breads (≈65–100 mg GAE/100 g; 1.18–1.32 μmol TE/g) followed by 5% (≈42–52 mg GAE/100 g; 0.71–0.86 μmol TE/g) compared to control breads (≈30 mg GAE/100 g; 0.69 μmol TE/g). The nutritional profile indicated that there is a scope for developing 'high-fiber' and antioxidant-enriched breads with the addition of apple seed. However only breads made from flour incorporated with 5% DASC showed best textural and sensory properties compared to control.

Manzoor et al. (2021a) analyzed the seed and flour properties of 'Golden Delicious' and 'Red Delicious' apples. The dimensions (length, width, thickness, arithmetic mean, geometric mean) of apple seeds varied significantly between golden delicious (7.69 mm, 4.02 mm, 2.01 mm, 4.58 mm, 5.44 mm) and red delicious (9.43 mm, 4.87 mm, 2.03 mm, 3.97 mm, 4.51 mm) respectively indicating the cultivar differences. The surface area and volume of apple seeds was 64.19 $mm^2$, 29.08 $mm^3$ (Red Delicious) and 49.57 $mm^2$, 20.22 $mm^3$ (Golden Delicious) respectively. The seeds were resembling nearer to sphere with sphericity around 48.18% (Red Delicious) to 51.72% (Golden Delicious). The information regarding all these parameters can assist in developing machinery at an industrial level for processing of apple seed. Further, authors also studied the flowability of apple seed flours. It was noted that Red and Golden Delicious apples had 0.38 g/mL and 0.60 g/mL bulk density, While the true density was 0.58 g/mL and 0.69 g/mL respectively. The higher bulk density in Golden Delicious apples was attributed to its higher moisture content. The apple seed flour has poor flowability (34.51% and 37.70%) with Carr index above 25%. For industrial level application of apple seed, it becomes necessary to understand their engineering and geometric properties for ease of designing processing machinery. Further the apple seed flour had satisfactory white color, and both large and small starch particles. Starch present in red delicious apple seed exhibited rough surface with polygonal, oval-, semi-spherical shape and 4.56–15.27 μm in size. While for golden delicious it was 2.04–14.32 μm in size and the shape was irregular spherical with smooth surface, indicating its potential application in food product development.

Manzoor et al. (2021a) reported that both Red and Golden Delicious apple seeds had good water holding capacity due to their high protein (especially hydrophilic polar amino acids) and polysaccharide contents; with Red Delicious (327.65 g/100 g) having higher capacity than Golden Delicious (312.58 g/100 g) seed flour. While the oil holding capacity was in the range of 92.97 g/100 g–86.76 g/100 g. However, El-Safy et al. (2012) reported slightly higher values for water (358 g/100 g) and oil (119 mL/100 g) absorption capacity for apple (*Malus sylvestris*) seeds; and one of the reasons behind this could be their varietal differences. Manzoor et al. (2021a) disclosed that emulsion activity was 72.01 (Golden Delicious) – 79.61% (Red Delicious) indicating the presence of polar and non-polar protein in the apple seed flours; and emulsification stability was in the range of 46.56%–51.81%. As reported by El-Safy et al. (2012) the emulsification stability (43.60%) and emulsification capacity (78.11 mL oil/g protein) of apple seeds were 43.60% and 78.11 mL oil/g protein respectively. This indicates that apple seed flour can be used in modifying product formulation where emulsion is desired. Foam expansion and stability of apple seed flour were 13.60% and 79.45% respectively. All these properties of apple seed convey its potential for being administered as functional ingredient or additive in food industry.

## 5. Safety aspects of apple seeds

Despite all the nutritional and therapeutic benefits, that apple seed can confer, there is a presence of a toxic compound like amygdalin, prunasin in seeds of different apple varieties. These cyanogenic glycosides can liberate toxic hydrocyanic acid (HCN) when the seeds are processed or damaged, which when consumed can cause health issues. The presence of cyanogenic glycoside in plant seed could be due to the environmental stress, drought conditions, high soil salinity. As of now, there are no strong or confirmatory evidences regarding the poisonous effect of apple seed on human health, thus more studies are required on this area (Bolarinwa et al., 2015; Mustafa et al., 2018; Senica et al., 2019).

Presently, the reports suggest that, average amygdalin content in seeds of fifteen different varieties ranged between 1.0 mg/g to 4.0 mg/g (Bolarinwa et al., 2015). The amygdalin, neoamygdalin and prunasin belonging to natural toxicants viz. cyanogenic glycosides were identified in golden delicious apple seeds harvested from four different locations (Azerbaijan, Russia, Serbia, Slovenia). Amongst the four different locations, apple seeds belonging to Russia and Azerbaijan had prunasin (>50%) as the main cyanogenic glycoside; while amygdalin and prunasin were the main ones in Serbian apple seeds and only amygdalin with 54–67% of total cyanogenic glycosides in seeds from Slovenia. Amongst all three cyanogenic glucosides (amygdalin, neoamygdalin and prunasin), lowest quantities (1–3%) were observed of neoamygdalin (Senica et al., 2019).

Furthermore, the quantities of amygdalin in golden delicious apple harvested from Azerbaijan, Russia, Serbia, Slovenia were in the ranges of 852.59–1466.15 μg/g, 577.34–745.28 μg/g, 521.66–834.13 μg/g, 499.66–996.14 μg/g respectively. While for Neoamygdalin they were 24.11–60.62 μg/g, 15.16–33.95 μg/g, 15.99–32.18 μg/g and 14.40–31.58 μg/g and for prunasin the values were in the ranges of 1316.17–2009.46 μg/g, 914.37–1543.48 μg/g, 435.21–1137.99 μg/g and 219.30–857.65 μg/g respectively for apple seeds from Azerbaijan, Russia, Serbia, Slovenia. This variation in the content of the cyanogenic glycoside can be due to differences in environmental condition of the geographical locations. However, there are limited studies determining their detrimental effect on human health (Senica et al., 2019). Besides the tiny amount in which these cyanogenic glycosides are present, these are unlikely to cause any toxic effect on human health. Processing these apple seed before utilization to reduce the cyanogenic glycoside improves its edibility and usage in food and nutraceutical industry (Bolarinwa et al., 2015).

## 6. Conclusion and future perspectives

Apple seeds being a rich source of nutrients like fatty acids, amino acids, polyphenols can be explored as a new functional ingredient. Therefore, this review tried to compile all the available literature regarding nutritional, phyto-nutritional properties, therapeutic benefits, and application of apple seed, its extracts, seed oil as novel ingredient in food, nutraceutical and pharmaceutical industries.

Studies undertaken till now indicate that the apple seed have good amount of protein, fat, fibers, minerals, and phytochemicals especially phloridzin, phloretin, coumarins and its derivatives conferring antioxidant properties and biological activities like anti-obesity, anti-diabetic, anti-microbial, anti-cancer and so on. However, limited *in vitro* and *in vivo* studies exist to confirm these health benefits. Also, the application of apple seed is scarce in food industry considering insufficient evidence on effect upon consumption of cyanogenic glycosides present in apple seeds. Thus, there is need for thorough systematic analysis of apple seed as novel ingredient.

## Authors contributions

Manoj Kumar, Mrunal D. Barbhai, Baohong Zhang, and Sangram Dhumal conceptualized the idea and drafted the outline. Mrunal D. Barbhai, Tuba Esatbeyoglu, Radha, Vijay Sheri, Deepak Chandran, and Marthandan Vishvanathan, drafted the original manuscript and corrected portions of the manuscript. Ravi Pandiselvam, Marisennayya Senapathy, Abhijit Dey, Marthandan Vishvanathan, Sangeetha Kizhakkumkara Sathyaseelan, Sangram Dhumal, Sabareeshwari Viswanathan, Eman Mohammad Said Al Masry, and Pran Mohankumar, wrote the portions and contributed in tabular content, illustrations in the manuscript and revising the manuscript. Jose M Lorenzo, Nadeem Rais, Eman Mohammad Said Al Masry, Sheetal Vishal Deshmukh, Mohamed El Sayed Negm, and Baohong Zhang gave the critical suggestions in successive revisions of the manuscript. Jose M. Lorenzo, Mrunal D. Barbhai, Manoj Kumar, Nadeem Rais, Sheetal Vishal Deshmukh, and Mohamed El Sayed Negm contributed in the revising the manuscript as per the comments of the reviewers. All the authors contributed significantly in the preparation of the final version of the manuscript.

## Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

## Data availability

No data was used for the research described in the article.

## References


Abbas, A., Anwar, F., & Ahmad, N. (2019). Apple (*Malus pumila*) seed oil. In M. Ramadan (Ed.), *Fruit oils: Chemistry and functionality*. Switzerland AG: Springer Nature.

Alves, E., Simoes, A., & Domingues, M. R. (2021). Fruit seeds and their oils as promising sources of value-added lipids from agro-industrial byproducts: Oil content, lipid composition, lipid analysis, biological activity and potential biotechnological applications. *Critical Reviews in Food Science and Nutrition, 61*(8), 1305–1339.

Andrade, M. A., Lima, V., Silva, A. S., Vilarinho, F., Castilho, M. C., Khwaldia, K., & Ramos, F. (2019). Pomegranate and grape by-products and their active compounds: Are they a valuable source for food applications? *Trends in Food Science & Technology, 86*, 68–84.

Antonic, B., Jancikova, S., Dordevic, D., & Tremlova, B. (2020). Apple pomace as food fortification ingredient: A systematic review and meta-analysis. *Journal of Food Science, 85*(10), 2977–2985.

Arain, S., Sherazi, S. T. H., Bhanger, M. I., Memon, N., Mahesar, S. A., & Rajput, M. T. (2012). Prospects of fatty acid profile and bioactive composition from lipid seeds for the discrimination of apple varieties with the application of chemometrics. *Grasas Y Aceites, 63*(2), 175–183. https://doi.org/10.3989/gya.082811

Bangar, S. P., Kumar, M., Whiteside, W. S., Tomar, M., & Kennedy, J. F. (2021). Litchi (Litchi chinensis) seed starch: Structure, properties, and applications-A review. *Carbohydrate Polymer Technologies and Applications, 2*, Article 100080.

Bhushan, S., Kalia, K., Sharma, M., Singh, B., & Ahuja, P. S. (2008). Processing of apple pomace for bioactive molecules. *Critical Reviews in Biotechnology, 28*(4), 285–296.

Blidi, S., Bikaki, M., Grigorakis, S., Loupassaki, S., & Makris, D. P. (2015). A comparative evaluation of bio-solvents for the efficient extraction of polyphenolic phytochemicals: Apple waste peels as a case study. *Waste and Biomass Valorization, 6* (6), 1125–1133.

Bolarinwa, I. F., Orfila, C., & Morgan, M. R. (2015). Determination of amygdalin in apple seeds, fresh apples and processed apple juices. *Food Chemistry, 170*, 437–442.

Boyer, J., & Liu, R. H. (2004). Apple phytochemicals and their health benefits. *Nutrition Journal, 3*, 5. https://doi.org/10.1186/1475-2891-3-5

Buturi, C. V., Mauro, R. P., Fogliano, V., Leonardi, C., & Giuffrida, F. (2021). Mineral biofortification of vegetables as a tool to improve human diet. *Foods, 10*(2), 223.

Can, Z., Dincer, B., Sahin, H., Baltas, N., Yildiz, O., & Kolayli, S. (2014). Polyphenol oxidase activity and antioxidant properties of Yomra apple (*Malus communis L.*) from Turkey. *Journal of Enzyme Inhibition and Medicinal Chemistry, 29*(6), 829–835.

Cheesman, M. J., Ilanko, A., Blonk, B., & Cock, I. E. (2017). Developing new antimicrobial therapies: Are synergistic combinations of plant extracts/compounds with conventional antibiotics the solution? *Pharmacognosy Reviews, 11*(22), 57–72.

Ci, Z., Kikuchi, K., Hatsuzawa, A., Nakai, A., Jiang, C., Itadani, A., & Kojima, M. (2018). Antioxidant activity, and α-glucosidase, α-amylase and lipase inhibitory activity of polyphenols in flesh, peel, core and seed from mini apple. *Advance Journal of Food Science and Technology, 6*(6), 258–262.

Dadwal, V., Agrawal, H., Sonkhla, K., Joshi, R., & Gupta, M. (2018). Characterization of phenolics, amino acids, fatty acids and antioxidant activity in pulp and seeds of high altitude Himalayan crab apple fruits (*Malus baccata*). *Journal of Food Science & Technology, 55*(6), 2160–2169. https://doi.org/10.1007/s13197-018-3133-y

Duda-Chodak, A., Tarko, T., & Tuszyński, T. (2011). Antioxidant activity of apples – an impact of maturity stage and fruit part. *Acta Scientiarum Polonorum, Technologia Alimentaria., 10*(4), 443–454.

El-Safy, F. S., Salem, R. H., & Abd El-Ghany, M. E. (2012). Chemical and nutritional evaluation of different seed flours as novel sources of protein. *World Journal of Dairy & Food Sciences, 7*(1), 59–65. https://doi.org/10.5829/idosi.wjdfs.2012.7.1.61215

Ferrentino, G., Asaduzzaman, M., & Scampicchio, M. M. (2018). Current technologies and new insights for the recovery of high valuable compounds from fruits by-products. *Critical Reviews in Food Science and Nutrition, 58*(3), 386–404.

Ferrentino, G., Giampiccolo, S., Morozova, K., Haman, N., Spilimbergo, S., & Scampicchio, M. (2020). *Supercritical fluid extraction of oils from apple seeds: Process optimization, chemical characterization and comparison with a conventional solvent extraction* (Vol. 64). Innovative Food Science & Emerging Technologies.

Fromm, M., Bayha, S., Carle, R., & Kammerer, D. R. (2012a). Characterization and quantitation of low and high molecular weight phenolic compounds in apple seeds. *Journal of Agricultural and Food Chemistry, 60*(5), 1232–1242. https://doi.org/10.1021/jf204623d

Fromm, M., Bayha, S., Carle, R., & Kammerer, D. R. (2012b). Comparison of fatty acid profiles and contents of seed oils recovered from dessert and cider apples and further Rosaceous plants. *European Food Research and Technology, 234*(6), 1033–1041. https://doi.org/10.1007/s00217-012-1709-8

Górnaś, P. (2015). Unique variability of tocopherol composition in various seed oils recovered from by-products of apple industry: Rapid and simple determination of all four homologues (α, β, γ and δ) by RP-HPLC/FLD. *Food Chemistry, 172*, 129–134.

Górnaś, P., Rudzińska, M., & Segliņa, D. (2014). Lipophilic composition of eleven apple seed oils: A promising source of unconventional oil from industry by-products. *Industrial Crops and Products, 60*, 86–91. https://doi.org/10.1016/j.indcrop.2014.06.003

Guardia, L., Suárez, L., Querejeta, N., Rodriguez, M. R., Suarez, B., & Centeno, T. A. (2019). Apple waste: A sustainable source of carbon materials and valuable compounds. *ACS Sustainable Chemistry & Engineering, 7*(20), 17335–17343.

Gupta, P. D., & Birdi, T. J. (2017). Development of botanicals to combat antibiotic resistance. *Journal of Ayurveda and Integrative Medicine, 8*(4), 266–275.

Hassan, L. G., Muhammad, M. U., Umar, K. J., & Sokoto, A. M. (2008). Comparative study on the proximate and mineral contents of the seed and pulp of sugar apple (*Annona squamosa*). *Nigerian Journal of Basic and Applied Sciences, 16*(2), 179–182.

He, X., & Liu, R. H. (2008). Phytochemicals of apple peels: Isolation, structure elucidation, and their antiproliferative and antioxidant activities. *Journal of Agricultural and Food Chemistry, 56*(21), 9905–9910.

powoscience. http://powo.science.kew.org/taxon/urn:lsid:ipni.org:names:726282-1. (Accessed 10 January 2021).

atlasbig. https://www.atlasbig.com/en-in/countries-by-apple-production. (Accessed 8 December 2021).

Kamel, B. S., DeMan, J. M., & Blackman, B. (1982). Nutritional, fatty acid and oil characteristics of different agricultural seeds. *International Journal of Food Science and Technology, 17*(2), 263–269.

Khalil, R. R., & Mustafa, Y. F. (2020). Phytochemical, antioxidant and antitumor studies of coumarins extracted from Granny Smith apple seeds by different methods. *Systematic Reviews in Pharmacy, 11*(2), 57–63.

Kumar, M., Tomar, M., Amarowicz, R., Saurabh, V., Nair, M. S., Maheshwari, C., Sasi, M., Prajapati, U., Hasan, M., Singh, S., Changan, S., Prajapat, R. K., Berwal, M. K., & Satankar, V. (2021). Guava (*Psidium guajava* L.) leaves: Nutritional composition, phytochemical profile, and health-promoting bioactivities. *Foods, 10*(4), 752.

Kumar, M., Tomar, M., Bhuyan, D. J., Punia, S., Grasso, S., Sa, A. G. A., Carciofi, B. A. M., Arrutia, F., Changan, S., Radha, Singh, S., Dhumal, S., Senapathy, M., Satankar, V., Anitha, T., Sharma, A., Pandiselvam, R., Amarowicz, R., & Mekhemar, M. (2021). Tomato (Solanum lycopersicum L.) seed: A review on bioactives and biomedical activities. *Biomedicine & Pharmacotherapy, 142*, Article 112018.

Kumoro, A. C., Alhanif, M., & Wardhani, D. H. (2020). A critical review on tropical fruits seeds as prospective sources of nutritional and bioactive compounds for functional foods development: A case of Indonesian exotic fruits. *International journal of food science*. , Article 4051475. https://doi.org/10.1155/2020/4051475, 2020.

Łata, B., Trampczynska, A., & Oles, M. (2005). Antioxidant content in the fruit peel, flesh and seeds of selected apple cultivars during cold storage. *Folia Horticulturae, 17*(1), 47–60.

Madrera, R. R., & Valles, S. B. (2018). Characterization of apple seeds and their oils from the cider-making industry. *European Food Research and Technology, 244*, 1821–1827. https://doi.org/10.1007/s00217-018-3094-4

Manzoor, M., Singh, J., & Gani, A. (2021a). Assessment of physical, microstructural, thermal, techno-functional and rheological characteristics of apple (*Malus domestica*) seeds of Northern Himalayas. *Scientific Reports, 11*, Article 22785. https://doi.org/10.1038/s41598-021-02143-z

Manzoor, M., Singh, J., & Gani, A. (2021b). Characterization of apple (*Malus domestica*) seed flour for its structural and nutraceutical potential. *LWT - Food Science and Technology, 151*, Article 112138. https://doi.org/10.1016/j.lwt.2021.112138

Marek, P., Władysław, M., Robert, G., Magdalena, R., Dorota, B.-Ł., Magdalena, P., & Paulina, S. (2015). Native oils from apple, blackcurrant, raspberry, and strawberry seeds as a source of polyenoic fatty acids, tocochromanols, and phytosterols: A health implication. *Journal of Chemistry*, 1–8. https://doi.org/10.1155/2015/659541

Matthäus, B., & Musazcan Özcan, M. (2015). Oil content, fatty acid composition and distributions of vitamin-e-active compounds of some fruit seed oils. *Antioxidants, 4* (1), 124–133.

Mohammed, E. T., & Mustafa, Y. F. (2020). Coumarins from Red Delicious apple seeds: Extraction, phytochemical analysis, and evaluation as antimicrobial agents. *Systematic Reviews in Pharmacy, 11*(2), 64–70.

Mustafa, Y. F., Khalil, R. R., & Mohammed, E. T. (2020). Antimicrobial activity of aqueous extracts acquired from the seeds of two apples' cultivars. *Systematic Reviews in Pharmacy, 11*(2), 382–387.

Mustafa, Y. F., Najem, M. A., & Tawffiq, Z. S. (2018). Coumarins from creston apple seeds: Isolation, chemical modification, and cytotoxicity study. *Journal of Applied Pharmaceutical Science, 8*. https://doi.org/10.7324/JAPS.2018.8808 (08): 049-056.

Omoyajowo, K. O., Njoku, K. L., Babalola, O. O., & Adenekan, O. A. (2017). Nutritional composition and heavy metal content of selected fruits in Nigeria. *Journal of Agriculture and Environment for International Development (JAEID), 111*(1), 123–139.

Opyd, P. M., Jurgoński, A., Juśkiewicz, J., Milala, J., Zduńczyk, Z., & Król, B. (2017). Nutritional and health-related effects of a diet containing apple seed meal in rats: The case of amygdalin. *Nutrients, 9*(10), 1091.

Oseni, O. A., Olarinoye, C. O., & Amoo, I. A. (2010). Studies on chemical compositions and functional properties of thorn apple (*Datura stramonium* L) Solanaceae. *African Journal of Food Science, 5*(2), 40–44.

Pandey, J., Bastola, T., Tripathi, J., Tripathi, M., Rokaya, R. K., Dhakal, B., … Poudel, A. (2020). Estimation of total quercetin and rutin content in *Malus domestica* of Nepalese origin by HPLC method and determination of their antioxidative activity. *Journal of Food Quality, 2*, 1–13. https://doi.org/10.1155/2020/8853426, 8853426.

Patocka, J., Bhardwaj, K., Klimova, B., Nepovimova, E., Wu, Q., Landi, M., Kuca, K., Valis, M., & Wu, W. (2020). Malus domestica: A review on nutritional features, chemical composition, traditional and medicinal value. *Plants, 9*(11), 1408. https://doi.org/10.3390/plants9111408Paul, A., &. MDPI AG.

Radhakrishnan, M. (2020). Pomegranate seed oil in food industry: Extraction, characterization, and applications. *Trends in Food Science & Technology, 105*, 273–283.

Pieszka, M., Migdał, W., Gąsior, R., Rudzińska, M., Bederska-Łojewska, D., Pieszka, M., & Szczurek, P. (2015). Native oils from apple, blackcurrant, raspberry, and strawberry seeds as a source of polyenoic fatty acids, tocochromanols, and phytosterols: A health implication. *Journal of Chemistry*, 2015.

Punia, S., & Kumar, M. (2021). Litchi (*Litchi chinenis*) seed: Nutritional profile, bioactivities, and its industrial applications. *Trends in Food Science & Technology, 108*, 58–70.

Purić, M., Rabrenović, B., Rac, V., Pezo, L., Tomašević, I., & Demin, M. (2020). Application of defatted apple seed cakes as a by-product for the enrichment of wheat bread. *LWT–Food Science and Technology*, 130.

Raza, A., Butt, M. S., & Suleria, H. A. R. (2017). Jamun (*Syzygium cumini*) seed and fruit extract attenuate hyperglycemia in diabetic rats. *Asian Pacific Journal of Tropical Biomedicine, 7*(8), 750–754.

Senica, M., Stampar, F., Veberic, R., & Mikulic-Petkovsek, M. (2019). Cyanogenic glycosides and phenolics in apple seeds and their changes during long term storage. *Scientia Horticulturae, 255*, 30–36.

Sharma, R., & Nath, A. K. (2016). Antioxidant levels and activities of reactive oxygen-scavenging enzymes in crab apple fruits (*Malus baccata*). *Proceedings of the National Academy of Sciences, India - Section B: Biological Sciences, 86*, 877–885.

Sonja, Đ., Jasna, Č.-B., & Gordana, Ć. (2009). By-products of fruits processing as a source of phytochemicals. *Chemical Industry and Chemical Engineering Quarterly, 15*(4), 191–202.

Spengler, R. N. (2019). Origins of the apple: The role of megafaunal mutualism in the domestication of Malus and rosaceous trees. *Frontiers of Plant Science, 617*.

Tian, H.-L., Zhan, P., & Li, K.-X. (2010). Analysis of components and study on antioxidant and antimicrobial activities of oil in apple seeds. *International Journal of Food Sciences & Nutrition, 61*(4), 395–403.

Tuberoso, C. I. G., Kowalczyk, A., Sarritzu, E., & Cabras, P. (2007). Determination of antioxidant compounds and antioxidant activity in commercial oilseeds for food use. *Food Chemistry, 103*(4), 1494–1501.

Walia, M., Rawat, K., Bhushan, S., Padwad, Y. S., & Singh, B. (2014). Fatty acid composition, physicochemical properties, antioxidant and cytotoxic activity of apple seed oil obtained from apple pomace. *Journal of the Science of Food and Agriculture, 94*(5), 929–934.

Xie, F., Yu, L., Chen, L., & Li, L. (2008). A new study of starch gelatinization under shear stress using dynamic mechanical analysis. *Carbohydrate Polymers, 72*(2), 229–234.

Xu, Y., Fan, M., Ran, J., Zhang, T., Sun, H., Dong, M., Zhang, Z., & Zheng, H. (2016). Variation in phenolic compounds and antioxidant activity in apple seeds of seven cultivars. *Saudi Journal of Biological Sciences, 23*(3), 379–388.

Yaman, M., Çatak, J., Uğur, H., Gürbüz, M., Belli, İ., Nur Tanyıldız, S., Yıldırım, H., Cengiz, S., Burak Yavuz, B., Kişmiroğlu, C., Özgür, B., & Cihan Yaldız, M. (2021). The bioaccessibility of water-soluble vitamins: A review. *Trends in Food Science & Technology, 109*, 552–563.

Yu, X., Van De Voort, F. R., Li, Z., & Yue, T. (2007). Proximate composition of the apple seed and characterization of its oil. *International Journal of Food Engineering, 3*(5).

Yukui, R., Wenya, W., Rashid, F., & Qing, L. (2009). Fatty acids composition of apple and pear seed oils. *International Journal of Food Properties, 12*, 774–779.