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# Nutritional and functional attributes of hairless canary seed groats and components and their potential as functional ingredients

El-Sayed M. Abdel-Aal

Agriculture and Agri-Food Canada, Guelph Research and Development Centre, Guelph, N1G 5C9, Ontario, Canada

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## ABSTRACT

**Background:** Demands for novel foods, plant proteins, small starch granules and functional ingredients are steadily increasing due to the growing world population and health-conscious consumers. Hairless canary seed (*Phalaris canariensis* L.) newly received regulatory food approvals from Health Canada and US-FDA holds considerable potential in meeting these demands due to its unique nutrient profile and attributes. It is a true cereal grain which can be processed into flour, starch, protein and oil for food and non-food uses.

**Scope and approach:** This review summarizes studies on hairless canary seed as a novel food with emphasis on its unique nutrient composition and attributes. In addition, its potential as a functional ingredient for celiac disease, diabetes and health of eye and brain is discussed.

**Key findings and conclusions:** The hairless canary seed groats are fairly rich in macronutrients (starch, protein, oil) and bioactive compounds (polyphenols and carotenoids). The groats contain small starch granules which are able to form firm and stable gels under cooling and freeze-thaw treatments. The non-gluten protein of canary seed is exceptionally high in tryptophan and bioactive peptides exhibiting beneficial health effects. The canary seed oil belongs to the group “oils low in palmitic acid and high in oleic and linoleic acids” with a high ratio of unsaturated to saturated fat. The groats could offer great potential as a sustainable source of small starch granules, non-gluten proteins and highly unsaturated oil. The groats and/or their fractions could also be utilized as functional ingredients in dietary strategies aiming at the prevention of celiac disease, obesity and diabetes.

## 1. Introduction

Hairless or glabrous canary seed (*Phalaris canariensis* L.) was developed through a successful breeding program at the university of Saskatchewan in order to mitigate the skin irritation encountered by farmers during the harvest process and to eliminate the potential health threats associated with the hairy (pubescent) canary seed (Hucl et al., 2001). Later, the hairless canary seed was approved by Health Canada as a novel food (Health Canada, 2016) and received GRAS status (GRAS Notice No. GRN 000529) from the US-Food and Drug Administration (US-FDA, 2015). The traditional hairy canary seed is not safe for human consumption due to the presence of tiny siliceous hairs on the surface of hull (palea and lemma) of the kernel. The hairs, also called trichomes or spicules, can contaminate other edible crops grown adjacent to the canary seed field which could provoke health problems to consumers (Abdel-Aal, Hucl, & Sosulski, 1997a). The hairs can also pollute canary seed products during the de-hulling process causing health issues. It has been reported that the hairs are potential carcinogens if present as contaminants in foods and have been linked to esophagus cancer (O'Neil

et al., 1980). Thus, it is essential to entirely remove the harmful siliceous hairs in order to make canary seed safe for human consumption.

Toxicity studies using rats fed brown or yellow hairless canary seed have shown that the grains are safe for human consumption (Magnuson et al., 2014). The hairless canary seed has also been found to be safe for poultry feeding (Classen, Cho, Hucl, Gomis, & Patterson, 2014). Canary seed is a minor crop with a production of about 250,000 tonnes per year (Cogliatti, 2012) and is considered a special crop for birds and biodiversity (Small, 2015). The recent regulatory approvals of hairless canary seed as a novel food should open new market opportunities besides the existing birdseed market. Furthermore, the availability of rapid breeding techniques using the marker-assisted selection based on the microsatellite or simple sequence repeat markers specific for canary seed would support the development of new hairless canary seed varieties with desired traits (Li, Baga, Hucl, & Chibbar, 2011). Without a doubt, the new food regulations along with the development of more hairless canary seed varieties with a wide spectrum of traits would expand its food and non-food markets.

Canary seed is a true cereal grain belonging to the grass family,

E-mail address: [elsayed.abdelaal@canada.ca](mailto:elsayed.abdelaal@canada.ca).

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Poaceae or Gramineae similar to wheat, barley, oat, etc. It could be considered as an oat cousin since both grains share the same tribe (*Poaeae*) and contain non-gluten protein (Boye et al., 2013), and a relatively high amount of oil (Abdel-Aal, Hernandez, Rabalski, & Hucl, 2020; Mason, L'Hocine, Achouri, Pitre, & Karboune, 2020). Canary seed is a hulled or covered cereal grain (i.e. the hulls stay attached to the kernel after the harvest process) and once de-hulled, the hull-free grains are called groats (Abdel-Aal et al., 1997a). The appearance of de-hulled brown and yellow hairless canary seed is shown in Fig. 1. Several studies reported that hairless canary seed groats hold great potential as a wholegrain food ingredient and/or sustainable source of starch, protein and oil (Abdel-Aal & Hucl, 2005; Abdel-Aal, Hucl, Miller, Patterson, & Gray, 2011; Abdel-Aal, Hucl, Patterson, & Gray, 2010).

Fractionation of hairless canary seed into starch, protein and oil by wet milling extractions with ethanol followed by alkaline solution and water produces efficient separations with recoveries of about 92% pure starch, 75% pure protein and 75% pure oil (Abdel-Aal et al., 2010). A scale up of the wet fractionation process optimized for protein isolation also results in highly purified canary seed protein isolates (91–93% purity) with a protein yield of 65–69% along with good yields for oil (6–7%), starch (48–54%) and fiber (15–20%) (Achouri et al., 2020). Functional properties of hairless canary seed protein exhibit enhanced fat and water holding capacities and higher foaming and emulsifying capacities than soy protein isolate. In addition to the separation efficiency of hairless canary seed components, they also hold unique properties and great potential for several applications, i.e. starch (Irani, Abdel-Aal, Razavi, Hucl, & Patterson, 2017; Irani, Razavi, Abdel-Aal, Hucl, & Patterson, 2016; Irani, Razavi, Abdel-Aal, Hucl, & Patterson, 2019), protein (Moura et al., 2020; Mason, L'Hocine, Achouri, & Karboune, 2018; Abdel-Aal et al., 1997a) and oil (Abdel-Aal et al., 2020; Ben Salah et al., 2017). Details about their attributes are discussed in the following sections.

Recently a couple of review articles have been published, one review summarizes the potential of hairless canary seed as a novel food ingredient for several food segments such as bakeries, pastas, breakfast cereals and snack foods (Patterson et al., 2018) and the second article shows its potential as a novel cereal grain with health-enhancing properties (Mason et al., 2018). This review is intended to give insights regarding hairless canary seed as a novel food, alternative source of unique starch, protein, and oil, and its potential as a functional ingredient in dietary strategies aiming at mitigating celiac disease, obesity and diabetes and enhancing health of eye and brain.

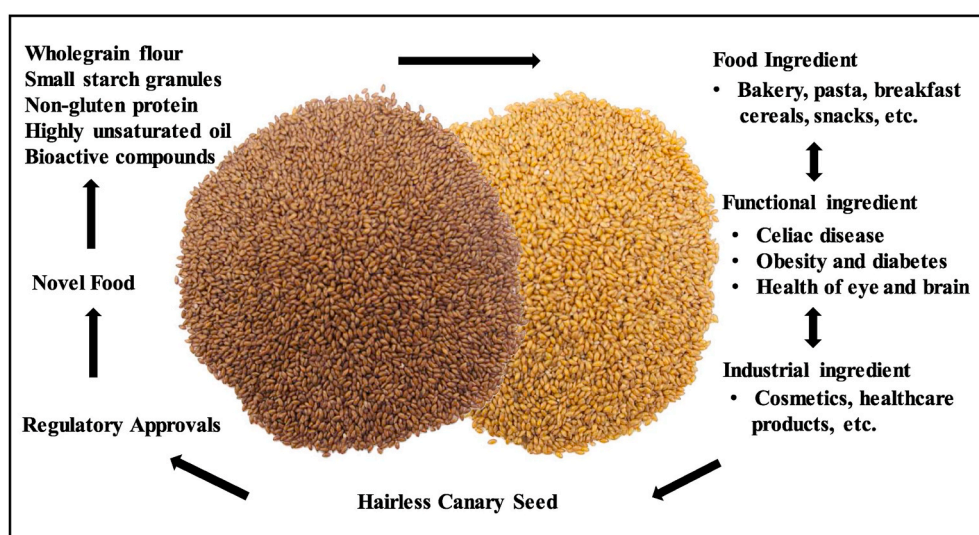
## 2. Basic nutrients

Table 1 shows content of macro-nutrients, minerals and vitamins in hairless canary seed in comparison with traditional hairy canary seed and regular wheat. The three grains were grown in plots in the same field to eliminate effects of environmental factors and soil composition on grain nutrients (Abdel-Aal, Hucl, Miller, et al., 2011). Hairless canary seed has a unique nutritional profile being relatively rich in protein (22.7%) and oil (7.7%) as a cereal grain but its starch content is comparable to wheat and other cereal grains at a level of 57%. There are no significant differences between hairless and hairy canary seed in starch content but significant differences are found in the content of protein and oil (Abdel-Aal, Hucl, Miller, et al., 2011). The hairless canary seed groats are also good sources of minerals and vitamins (Table 1). Hairless and hairy canary seed wholegrain flours have similar levels of minerals but they are significantly richer than wheat in their contents of phosphorus, sulphur, magnesium, calcium, iron, manganese and zinc, and

**Table 1**  
Average content of nutrients in hairless canary seed in comparison with hairy canary seed and hard red spring wheat grown in the same environment.

Component	Hairless canary seed	Hairy canary seed	Wheat
<b>Basic nutrients (g/100g)</b>			
Starch	57.2	54.3	59.9
Protein (Nx5.7)	22.7	24.7	17.3
Crude oil	7.7	8.0	2.5
Total dietary fiber	5.9	8.6	12.8
Soluble sugars	1.7	1.8	2.9
Total ash	2.3	2.3	1.6
<b>Minerals (mg/100g)</b>			
Phosphorus	640	590	430
Potassium	385	340	355
Sulphur	305	300	200
Magnesium	200	195	155
Calcium	40	40	20
Sodium	10	10	10
Iron	6.5	5.9	4.2
Manganese	6.3	7.1	5.9
Zinc	3.9	3.5	2.5
Copper	0.2	0.2	0.3
<b>Vitamins (mg/100g)</b>			
Thiamine (B1)	0.85	0.79	0.44
Riboflavin (B2)	0.16	0.16	0.15
Niacin	0.68	0.89	7.29

Source: Abdel-Aal, Hucl, Miller, et al., 2011.



**Fig. 1.** Appearance of brown and yellow hairless canary seed groats and their nutritional attributes and potential applications. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

exceed oat in phosphorus and iron contents (Abdel-Aal, Hucl, Miller, et al., 2011). As the role of minerals in human health are well established, hairless canary seed has great potential to boost daily intake of minerals. The three B-vitamins reported are similar in the two types of canary seed but different from wheat. The level of thiamine in canary seed is about twice as that in wheat but riboflavin is similar in the three grains. Thus, incorporation of hairless canary seed into diet would boost these two vitamins. However, wheat contains substantially higher levels of niacin than canary seed, making canary seed a poorer source of niacin. In general, the incorporation of hairless canary seed wholegrain flour in food formulations would boost the daily consumption of nutrients especially protein, unsaturated fat, minerals and thiamine. Additionally, hairless canary seed groats contain comparable levels of phytate and trypsin and amylase inhibitors to wheat, and the groats are free from alkaloids and condensed tannins (Abdel-Aal, Hucl, Miller, Patterson, & Gray 2011b).

### 3. Functional properties of starch

Starch is the principal component in cereal grains averaging about 57% of the total components in hairless canary seed (Abdel-Aal, Hucl, Miller, et al., 2011). Starch is a semi-crystalline substance which can be transformed into an amorphous state when heated in an excess amount of water, forming a viscous paste of gelatinized starch which is more digestible than native starch. Starch paste can be converted into gel structure through network formation when cooled, and upon storage it partially recrystallizes to form retrograded starch (less digestible than gelatinized starch). These starch transformations occur during thermal processing of cereals which make starch a key ingredient with multiple techno-functionalities, e.g. thickener, gelling agent, fat replacer, stabilizer, etc. Functionality of starches is determined by a number of factors including the size and architecture of starch granules, amylose/amylopectin content, molecular structure (e.g. chain length of amylose and amylopectin) and minor constituents of starch granules, e.g. lipid, protein and phosphate.

Canary seed starch granules are polygonal, small and uniform with an average diameter of 2.6  $\mu\text{m}$  and A-type crystal structure (Abdel-Aal, Hucl, & Sosulski, 1997b; Irani et al., 2017). Small starch granules are promising ingredients in fat replacement, cosmetic and healthcare applications (Lindeboom, Chang, & Tyler, 2004; Malinski, Daniel, Zhang, & Whistler, 2003). Hairless canary seed starch is able to form a strong and stable gel under refrigerated storage and freeze-thaw treatment (Abdel-Aal et al., 2019). The study has also shown that incorporation of small concentrations of xanthan gum, salt or sugar into canary seed starch can modify and improve its paste and gel characteristics especially for use in cold and frozen foods. For example, xanthan gum at concentrations of 0.1–0.5% is effective in enhancing the stability of canary seed starch gels during refrigerated storage and freeze-thaw treatment. Salt or sugar at concentrations of 0.5–1.0% is able to improve canary seed starch paste stability during thermal processing by enhancing breakdown and setback properties. The hairless canary seed starch gels exhibit shear-thinning and thixotropy behavior at concentrations of 4–8% (w/w) and a constant temperature of 25 °C (Irani, Razavi, Abdel-Aal, & Taghizadeh, 2016). Similarly this behavior is also obtained at a temperature range of 25–70 °C and a constant concentration of 6% (w/w). But, increasing the concentration from 4 to 8% (w/w), causes more shear-sensitive thixotropic nature and faster rupture rate, compared with increasing the temperature from 25 to 70 °C which shows the same shear-thinning behavior. The intrinsic viscosities of hairless canary seed and wheat starches decrease significantly with increasing ionic strength or salt concentration (5–100 mmol) (Heydari, Razavi, & Irani, 2018a), increasing temperature (25–55 °C) and sugar (5–15%) (Heydari, Razavi, & Irani, 2018b). The intrinsic viscosity of canary seed starch is lower than that of wheat starch but higher than that of rice starch as measured by Higirol model (Irani et al., 2016).

The molecular weight of hairless canary starch in a dilute solution

(0.5 g/dL) is in the range of 8.83–24.6  $\times 10^6$  g/mol which is lower than that of wheat starch at 33.4  $\times 10^6$  g/mol (Irani, Razavi, Abdel-Aal, & Taghizadeh, 2016). The same study has reported lower coil radius and coil volume in comparison with wheat starch. The canary seed starch maintains its crystal type-A structure but exhibits a stronger amylose-lipid complex, lower viscosity and retrogradation rate when naturally fermented for 45 days compared with the non-fermented starch (Batista et al., 2020). Blending of hairless canary seed starch with wheat starch at different ratios (5:75, 50:50, and 75:25, w/w) produces starch blends with improved functional properties (water and oil absorption), freeze-thaw stability and syneresis (Heydari & Razavi, 2020). Increasing the ratio of hairless canary seed starch reduces pseudo-plasticity of starch gels. The unique properties of hairless canary seed starch based on its small granules, strong amylose-lipid structure and robust gel quality and stability would make it a promising non-conventional starch source. The starch could be useful in several applications such as cold and frozen foods, fat replacement, cosmetics and pharmaceutical products. Research in those areas would expedite the developmental process.

### 4. Nutritional properties of protein

#### 4.1. Amino acids

Hairless canary seed is exceptionally high in protein (23%) for a cereal grain with unique amino acid composition. The prolamin is the major storage protein in canary seed accounting for approximately 46% of the total proteins, together with glutelin, they make up 78% of the total storage proteins (Abdel-Aal et al. 1997a). Canary seed proteins are extremely high in the essential amino acid tryptophan at levels of 2.0, 2.8, 3.3 and 4.6 g/100g protein as reported by several studies, Holt, 1988; Abdel-Aal, Hucl, Miller, et al., 2011; Robinson, 1978; Newkirk, Ram, Hucl, Patterson, & Classen, 2011, respectively. Tryptophan is commonly deficient in dietary protein sources such as dairy products, in which hairless canary seed would be an excellent complementary protein for other plant or animal proteins which barely meet the human requirement of tryptophan. Tryptophan plays several roles in the health of animals and humans (Agus, Planchais, & Sokol, 2018; Yao et al. 2011). It is essential for the synthesis of several bioactive molecules such as serotonin, melatonin, kynurenine and niacin, and plays important roles in gut-brain functions, bone health, immune modulation and mitochondrial function (Nayak, Singh, & Buttar, 2019). In this regard, hairless canary seed could be a very promising source of tryptophan based on the recommended daily intake of tryptophan (250–425 mg or 3.5–6.0 mg/kg body weight) (Richard et al. 2009). For example, the consumption of 3 servings (50 g each) of wholegrain canary seed muffin that contains 13% protein on a fresh weight basis (27.5% moisture content) would provide about 546 mg tryptophan (based on an average of 2.8 mg tryptophan/100 g protein). This amount exceeds the high end of the recommended daily intake of tryptophan. A study on normotensive and hypertensive rats administered with aqueous extract of canary seed suggests that the metabolic derivative of tryptophan or kynurenine could act as a mediator of the volume-independent antihypertensive effect in rats (Passos et al. 2012). The aqueous extract of canary seed which is rich in tryptophan was prepared by using 100 g of crushed seeds in 1 L of distilled water.

The canary seed proteins also exhibit high concentrations of cysteine, phenylalanine and arginine combined with low levels of lysine and proline, which suggest a unique composition and functionality for canary seed proteins (Abdel-Aal & Hucl, 2005). The low concentration of lysine and high concentration of arginine provide a low lysine-to-arginine ratio that has been reported to elicit a hypocholesterolemic effect based on soybean protein compared with casein (Kritchevsky, 1979). The ratio in soybean protein is 0.9 compared to 2.0 in casein but it is only 0.2 in canary seed protein which may suggest a hypocholesterolemic effect for canary seed protein. The protein in



canary seed are deficient in lysine and threonine for human and animal requirements, similar to other cereal proteins.

The apparent ileal amino acid digestibility of canary seed protein is similar to wheat protein except for lysine, with tryptophan having the highest digestibility (93%) (Newkirk et al., 2011). The *in vitro* protein digestibility (IVPD) and *in vitro* protein digestibility corrected amino acid score (IVPDCACS) of yellow and brown hairless canary seed flours are fairly similar ranging between 78.1–80.6% and 29.7–31.4%, respectively, but their protein isolates possess higher IVPD (85.7–87.4%) and IVPDCACS (55.7–64.1%) values than flours (Moura et al., 2020). Lysine is the first limiting amino acid in both flours, while histidine and threonine are the first limiting amino acid in the yellow (cv. Amarillo) and brown (cv. Café) hairless canary seed protein isolates, respectively. The study used a variety from each type of canary seed, therefore further studies are required to further probe differences between flours and isolates in protein quality. Hairless canary seed proteins have been found to be easily digested after thermal treatment (e.g. roasting at 176 °C for 12 min and boiling for 12 min) and under sequential gastric-duodenal conditions than under gastric or duodenal conditions alone (Rajamohamed, Aryee, Hucl, Patterson, & Boye, 2013). In addition, roasting of canary seed has a greater influence than boiling on the *in vitro* protein digestibility due to its larger impact on protein structure in which some of the protein subunits are not found in the roasted samples. In general, hairless canary seed proteins have a unique amino acid composition and can be considered a good source of the essential amino acid tryptophan in dietary protein supplements and products.

#### 4.2. Bioactive peptides

Studies have shown that cereal protein hydrolyzates and peptides could elicit beneficial health effects in humans. The ability of cereal hydrolyzed proteins and peptides to reduce oxidative stress has recently been summarized in a recent review (Esfandi, Walters, & Tsopmo, 2019). The majority of bioactive peptides have short chain length (<20 amino acids) and their antioxidant activity is affected by the composition and sequence of amino acids. The sequence of antioxidant peptides from oat (Du, Esfandi, Willmore, & Tsopmo, 2016), wheat (Babini, Tagliazucchi, Martini, Piu, & Gianotti, 2017), rice (Zhang et al., 2010), corn (Wang et al., 2014) and rye (Leung, Venus, Zeng, & Tsopmo, 2018) protein hydrolyzates have been identified. The antioxidant activity of peptides is based on several mechanisms including hydrogen or electron transfer, metal chelating and regulation of oxidation-reduction enzymes as determined by chemical, animal and/or cell culture assays. Peptides from wheat, oat, barley and rice have also been reported to inhibit angiotensin-converting enzyme and dipeptidyl peptidase indicating their potential to reduce the risk of hypertension and diabetes (Cavazos & de Mejia, 2013). Cereal peptides also possess anti-tumour activity with therapeutic potential against cancer (Diaz-Gomez, Castorena-Torres, Preciado-Ortiz, & Garcia-Lara, 2017; Ortiz-Martinez, Winkler, & Garcia-Lara, 2014). These studies support the role of cereal protein digests in the prevention of chronic disease and generally in the promotion of human health. Similarly, antidiabetic and antihypertensive potential of canary seed protein derived peptides has been studied (Estrada-Salas, Montero-Moran, Martinez-Cuevas, Gonzalez, & Barba de la Rosa, 2014; Valverde, Orona-Tamayo, Nieto-Rendon, & Paredes-Lopez, 2017). More information regarding the preparation of food-derived bioactive peptides and their role in human health can be found in the review article by Daliri, Oh, and Lee (2017).

Peptides prepared from canary seed albumin, globulin, prolamin and glutelin digests inhibit angiotensin-converting enzyme with IC<sub>50</sub> of 505, 443, 217 and 349 µg peptide/mL, respectively (Valverde et al., 2017). The results indicate that prolamin peptides are the most effective antihypertensive compounds. A similar trend has also been observed for peptides prepared from the protein digests of canary seed milk substitutes (prepared by soaking the seeds in water for 12 and 24 h). The

flour and milk peptides possess antioxidant activity against ABTS and DPPH radicals, but the milk substitute peptides exhibit stronger antioxidant potential than that of flour for all the protein fractions. In the same study, five peptides from the canary seed milk substitute prolamin fraction have been identified having a molecular weight range of 664–1019 Da. In another study, canary seed peptides exhibit 74% inhibition against angiotensin-converting enzyme and 44% inhibition against dipeptidyl peptidase IV, indicating their potential in antihypertensive and antidiabetic therapies (Estrada-Salas et al., 2014). The peptides at a concentration of 1 µg/mL are able to induce the production of nitric oxide (12.2 µM) in amounts similar to those induced by captopril (a high blood pressure drug). A recent study investigating the bioactive and health-promoting properties of 4 hairless canary seed varieties has shown their superior health-enhancing properties compared to that of wheat and oat (Mason et al., 2020). The cereal protein peptides were prepared using the standard INFOGEST *in vitro* human gastrointestinal digestion model and assessed based on antioxidant, chelating, antihypertensive and antidiabetic *in vitro* assays. Canary seed peptides exhibit superior or equivalent antioxidant and health-promoting activities compared with wheat or oat peptides. They also show potential anamnestic, antithrombotic, immuno-stimulating, opioid and neuroprotective activities. Since human studies are the golden standard for health benefits of foods further research on hairless canary seed is required to substantiate the health effects of its peptides and the impact of different processing technologies on protein digestibility and bioactive peptides.

#### 5. Compositional properties of oil

Monounsaturated and polyunsaturated fatty acids (MUFAs & PUFAs) are known as health-promoting nutrients as they are able to lower total and LDL cholesterol and reduce the risk of heart disease compared with saturated fatty acids (SFAs) which are considered harmful dietary components if over-consumed because of their ability to raise LDL cholesterol and increase the risk of heart disease (DiNicolantonio & O'Keefe, 2018; Kris-Etherton, Hecker, & Binkoski, 2004). Canary seed oil belongs to the oil group “oils low in palmitic acid and high in oleic and linoleic acids” based on its fatty acid composition, on average, it comprises of linoleic (53.8%), oleic (24.2%) and palmitic (11.5%) in addition to small levels of linolenic acid (2.8%) (Abdel-Aal et al. 2020). A comparable fatty acid profile has also been reported for Tunisian canary seed oil but it is higher in oleic acid, e.g. linoleic (52.0%), oleic (31.8%) and palmitic (11.1%) (Ben Salah et al., 2017). The hairless canary seed oil contains, on average, low amounts of SFAs than MUFAs and PUFAs (Abdel-Aal et al. 2020). The average unsaturated to saturated FAs (UFAs:SFAs) ratio of canary seed oil is 6.4 which is comparable to olive oil (5.8) but it is lower than canola oil (12.8). This ratio indicates its exceptionally high content of unsaturated FAs (PUFAs and MUFAs) which is favourable in human nutrition and health due to their roles in the reduction of serum cholesterol and prevention of heart diseases. The Tunisian canary seed oil possesses antioxidant, acetylcholine esterase (AChE) inhibitory and antibacterial properties perhaps due to its content of phytosterol and phenolic compounds (Ben Salah et al., 2017). About 16 phytosterols are present in the oil with β-sitosterol (48.0%) being the main compound, followed by campesterol (24.4%) and α-cholestanol (13.4%). In general, the hairless canary seed groats hold potential to be a good source of healthy oil.

Similar to rice, the oil can be concentrated in the bran fraction, i.e. 12.7 g per 100 g in bran versus 5.6 g per 100 g in the wholegrain flour (Abdel-Aal et al., 1997a), and the oil can be effectively recovered from the bran. Alternatively, the germ could be separated from the groats followed by oil extraction from the germ. In his regard, further research is needed to develop a process for the separation of germ especially the seeds are relatively too small (4.0–5.1 mm length and 1.5–2.0 mm width) in comparison with corn. The unique composition of oil makes hairless canary seed groats, bran or germ a promising source of healthy

oil for multiple uses including nutrition, cosmetics and pharmaceutical products.

## 6. Hairless canary seed as a functional ingredient

A diagram delineating nutritional attributes of hairless canary seed and its potential as a functional food ingredient is presented in Fig. 1. In this section, therapeutic properties of hairless canary seed in the areas of celiac disease, obesity, diabetes and health of eye and brain are discussed.

### 6.1. Celiac disease

Research has shown the potential of hairless canary seed as an alternative source of gluten-free food ingredient due to the absence of celiac provoking gluten epitopes (QP Signature) or the harmful amino acid sequence based on the mass spectrometry (MS) and immunoblotting analyses (Boye et al., 2013). The MS data have also shown the presence of protein homologous with the non-gluten proteins of rice, oat, corn, carrot, tomato, radish, beet and chickpea. Additionally, the ELISA results demonstrate no cross-reactivities between canary seed proteins and the major allergenic proteins including gluten, soy, peanuts, almonds, hazelnut, walnut, mustard, peanut, sesame and soy (Boye et al., 2013). This study indicates that hairless canary seed flour or its protein fraction can be used in gluten-free food formulations. But as hairless canary seed can be grown in a close proximity to gluten-containing cereal crops such as wheat, rye, barley and triticale the risk of cross-contamination during production increases, unless appropriate crop management practices are employed. In this regard, hairless canary seed should be grown, harvested and processed in a way that keeps it away from other grain culprits to minimize the risk of gluten contamination. A dedicated system of production, cleaning, grading, inspection and processing has been developed for oat used in gluten-free diets (Burrows, 2005). Similarly, a system can be adopted for hairless canary seed production to ensure the grains are free from gluten and safe for celiac patients. The official CODEX standard recommends that the foods contain a gluten level not exceeding 20 ppm (20 mg/kg) to be labeled as gluten-free (Codex Alimentarius Commission, 2008). This

might allow minute contaminations of gluten-containing grains.

Approximately 1.4% of the world population is affected by celiac disease based on a meta-analysis of 96 studies (Singh et al., 2018) which indicates massive demands for gluten-free ingredients and foods. In fact, the gluten-free market is rapidly growing with total retail sales in Canada of about 812 million CD in 2017, and the market in USA is estimated at 23.9 billion USD in 2020 (Statista, 2019). This indicates growing demands for gluten-free ingredients, and without doubt hairless canary seed can contribute a fair share to the gluten-free market due to its high content of protein along with the uniqueness of amino acid composition and health-promoting characteristics of bioactive peptides as described in section 4.

### 6.2. Obesity and diabetes

Table 2 shows the potential therapeutic properties of canary seed components. Canary seed is traditionally used as a folk medicine for diabetes in Mexico (Perez Gutierrez et al., 2014). A product for diabetes, known as *Alpiste*, is commercially available in the market. It is made by soaking the canary seed grains in water, then they are drained to remove the hairs and dried to make canary seed powder. The powder is reconstituted in water to make canary seed milk substitute before consumption. A number of components in the seeds could be linked to the prevention of obesity and/or diabetes. The above section (section 4.2) shows the ability of canary seed bioactive peptides to inhibit dipeptidyl peptidase IV indicating its potential as an antidiabetic ingredient. Additionally, canary seed extracts (e.g. hexane, chloroform and methanol) have been found to inhibit enzymes related to obesity and diabetes mellitus type II (Perez Gutierrez, Ahuatz, & Victoria, 2016). Hexane extract shows significant inhibitory effects against  $\alpha$ -amylase,  $\alpha$ -glucosidase, rat intestinal sucrase, pancreatic lipase, lipoprotein lipase and lipolysis of 3T3-L1 adipocytes. The IC<sub>50</sub> values for  $\alpha$ -amylase and  $\alpha$ -glucosidase are 2.13 and 1.25 mg/mL compared with 0.49 and 0.68 mg/mL for acarbose (an antidiabetic drug), respectively. The other two solvent extracts (chloroform and methanol) do not show inhibitory activities against  $\alpha$ -amylase and  $\alpha$ -glucosidase enzymes but they inhibit lipases. The study has also shown that oral administration of *Alpiste* (400 mg/kg) decreases serum glucose after 4 h in streptozotocin

**Table 2**  
Potential therapeutic properties of canary seed components.

Component	Health condition	Therapeutic mechanism	Efficacy	Reference
Peptides	Oxidative Stress	Inhibition of free radicals	IC <sub>50</sub> = 133 and 125 $\mu$ g/mL against ABTS and DPPH radicals	Valverde et al. 2017
Peptides	Hypertension	Inhibition of angiotensin-converting enzyme	IC <sub>50</sub> = 505 $\mu$ g/mL	Valverde et al. 2017
Peptides	Hypertension	Inhibition of angiotensin-converting enzyme	Inhibition = 73.5%	Estrada-Salas et al. (2014)
Peptides	Diabetes	Inhibition of dipeptidyl peptidase IV	Inhibition = 43.5%	Perez Gutierrez et al. (2014)
Peptides	Oxidative Stress	Inhibition of free radicals	ORAC 1.77–1.99 $\mu$ mol TE/mg; ABTS IC <sub>50</sub> = 117.5 $\mu$ g/mL; DPPH IC <sub>50</sub> = 77.9–96.4 $\mu$ g/mL	Mason et al. (2020)
Peptides	Hypertension	Inhibition of angiotensin-converting enzyme	IC <sub>50</sub> = 321.9–405.0 $\mu$ g/mL	Mason et al. (2020)
Peptides	Diabetes	Inhibition of dipeptidyl peptidase IV	IC <sub>50</sub> = 1.01–1.35 mg/mL	Mason et al. (2020)
Hexane extract	Diabetes	Inhibition of starch metabolism enzymes	IC <sub>50</sub> = 2.13 and 1.25 mg/mL for $\alpha$ -amylase and $\alpha$ -glucosidase	Perez Gutierrez et al. (2016)
Non-gluten protein	Celiac disease	Removal of toxic gluten peptides	Diets contain >20 mg gluten per kg	Boye et al. (2013)
Tryptophan	Hypertension	Antihypertensive effect	ND	Passos et al. (2012)
Low lysine-arginine ratio	Coronary heart disease	Hypocholesterolemic effect	ND	Abdel-Aal and Hucl (2005)
PUFAs and MUFAs	Coronary heart disease	Lowering cholesterol effect	ND	Ben Salah et al. (2017)
Phenolic acids (e.g. ferulic)	Oxidative Stress, Inflammation, diabetes	Antioxidative and anti-inflammatory effects & enzymes inhibitors	ND	Li, Baga, Hucl, & Chibbar, 2011; Prabhakar et al., 2013 & Adisakwattana et al., 2009
Carotenoids	Eye health	Antioxidative and protective effects	ND	Li & Beta, 2012 & Abdel-Aal et al., 2013

PUFAs and MUFAs stand for polyunsaturated and monounsaturated fatty acids; ND stands for not determined.

nicotinamide-induced diabetic mice without side effects such as flatulence and diarrhea. It has been reported that the mode of action of canary seed extract is close to that of acarbose or miglitol drugs based on the inhibition of  $\alpha$ -glucosidase, thus these drugs can be replaced with canary seed powder under similar circumstances if the powder is proven to be safe (Rayburn, 2016). The use of drugs can cause various adverse side effects, and thus demands for natural health products to treat obesity and diabetes are of increasing interest. The hairless canary seed received regulatory approvals for food use, and the seeds or their fractions or extracts can be a promising alternative for obesity drugs once the effective inhibitory dose is established. More research in this area is needed to determine efficiency of canary seed products in the treatment of obesity and diabetes with emphases on the role of polyphenols and bioactive peptides in the prevention of obesity and diabetes.

Dietary polyphenols could elicit therapeutic potential against chronic disease including obesity and diabetes. Phenolic acids are the major polyphenols in cereal grains which are predominantly found in the bound form (Ragaee, Seetharaman, & Abdel-Aal, 2014). Ferulic acid (4-hydroxy-3-methoxycinnamic acid), a non-flavonoid polyphenol and the dominant phenolic compound in cereals including canary seed, has been reported to reduce blood glucose in streptozotocin-treated diabetic rats (Prabhakar, Prasad, Ali, & Doble, 2013), increase plasma insulin and lower blood glucose in db/db mice (a well accepted type-II diabetes model) (Jung, Kim, Hwang, & Ha, 2007) and inhibit rat intestinal maltase and sucrose and  $\alpha$ -glucosidase (Adisakwattana, Chantarasinlapin, Thammarat, & Yibchok-Anun, 2009). Additionally, several review articles have discussed the beneficial health effects of ferulic acid (Mancuso & Santangelo, 2014; Ou & Kin-Chor, 2004; Srinivasan, Sudheer, & Menon, 2007).

Similar to other cereal grains, hairless canary seed is a good source of phenolic acids (Table 3). Phenolic acids are primarily present in the bound form (~90%), i.e. esterified to low- and high-molecular weight components (e.g. oligo- and poly-saccharides) in the cell wall of the grain. Thus they require base, acid or enzyme hydrolysis to release them and become available for analysis. Ferulic is the predominant phenolic acid in brown and yellow hairless canary seed and wheat with wheat having a higher concentration of ferulic acid compared with hairless canary seed. But hairless canary seed contains higher concentrations of caffeic and *p*-coumaric acids (Table 3). The average contents of caffeic, *p*-coumaric and ferulic acid are 102, 37 and 212 mg/kg, respectively in brown-seeded hairless canary seed and 73, 32 and 154 mg/kg in yellow-seeded varieties (Li, Qiu, Patterson, & Beta, 2011). It is not easy to make a valid comparison among studies due to the absence of standard methods for the analysis of phenolic acids and/or polyphenols in foods but the content of phenolic acids in hairless canary seed is comparable to other cereal grains. It has been reported that the content of phenolic acids and antioxidant properties of hairless canary seed can be improved by germination (Chen, Yu, Wang, Gu, & Beta, 2016). Other processing

technologies such as fermentation and thermal treatments can also be used to improve bioavailability of phenolic acids in grains (Abdel-Aal & Rabalski, 2013). The composition of flavonoids (other class of polyphenols) in brown and yellow canary seed varieties is similar with flavone derivatives being the dominant compounds. Research on the efficacy of phenolic acids especially ferulic, caffeic and *p*-coumaric acids in hairless canary seed products should provide a better understanding regarding their physiological effects in humans.

### 6.3. Health of eye and brain

Hairless canary seed is considered a good source of carotenoids particularly lutein, zeaxanthin and  $\beta$ -carotene compared with regular wheat (Table 3). Other cereal grains such as ancient einkorn wheat, durum wheat and corn are also good sources of carotenoids (Abdel-Aal, Young, Rabalski, Frégeau-Reid, & Hucl, 2007). Hairless canary seed and corn varieties that are rich in lutein could be used to boost the daily consumption of this essential compound since lutein, along with its isomer zeaxanthin, constitute the macular pigment in the eye, protect eyes against blue light, and reduce the risk of age-related macular degeneration and cataract (Abdel-Aal, Akhtar, Zaheer, & Rashida, 2013). This indicates the importance of lutein and zeaxanthin for eye health especially for the elderly. Lutein is also associated with the cognitive function in children (Johnson, 2014; Hammond, 2014). In general, the role of lutein in human nutrition and health, especially the health of eye and brain has been well documented. At present there is no recommended daily intake for lutein but there is a dietary gap between the daily intake (1.0–1.8 mg/day) (Johnson, 2010) and the suggested effective dose of lutein (6 mg/day) (Abdel-Aal et al. 2013). In this regard, high lutein varieties of hairless canary seed and corn could play significant roles to fill in this gap. The availability of grain-based high lutein functional foods would support the development of dietary strategies that aim at improving brain development and cognitive function in children and/or managing cataracts and AMD in elderly people (Abdel-Aal, 2014). More work is underway to quantify and identify carotenoids in hairless canary seed groats and to study their bioavailability in food prototypes made from hairless canary seed alone or in blends with other grains. Preliminary data indicate that lutein and lutein isomers are the major carotenoids in hairless canary seed products which could be considered a good source of lutein.

## 7. Conclusions

As consumers become more health conscious and the world population is growing, demands for plant proteins and functional ingredients are rising. Hairless canary seed groats have unique nutritional attributes based on their high content of non-gluten protein, bioactive peptides, tryptophan and unsaturated fat in addition to unique small and uniform strach granules. In addition, the groats also contain a diverse array of bioactive compounds particularly polyphenols and carotenoids. These nutritional and functional attributes make hairless canary seed a promising source of plant protein and functional ingredient for use in many food and non-food applications. For instance, hairless canary seed groats and/or protein fraction can be used in dietary strategies that aim to prevent celiac disease, obesity and diabetes. The unique properties of hairless canary seed starch based on small granules and strong gel properties would make it a promising non-conventional starch source in several applications such as cold and frozen foods, fat replacement, cosmetics and pharmaceutical products. The grain and/or its bran fraction is a promising source of healthy oil for multiple uses including nutrition, cosmetics and pharmaceutical products. The lutein-rich varieties could hold potential for the promotion of eye and brain health. More research on hairless canary seed groats and components is required to assess these applications and to understand how processing affects bioactive compounds in foods made from hairless canary seed in terms of their bioavailability and bioactivity. The growing demands for

**Table 3**

Composition of major phenolic acids and carotenoids in brown and yellow hairless canary seed in comparison with hard red spring wheat (mg/kg).

Component	Brown hairless canary seed	Yellow hairless canary seed	Wheat
Phenolic acids <sup>a</sup>			
Ferulic	211–268	136–190	367–728
Caffeic	83–96	58–94	1–2
<i>p</i> -Coumaric	33–54	28–43	10–38
Total	327–418	222–327	378–768
Carotenoids <sup>b</sup>			
Lutein	1.72–2.82	1.53–3.01	1.89
Zeaxanthin	0.43–0.61	0.27–0.57	0.05
$\beta$ -carotene	4.61–5.32	4.56–5.35	0.05
Total	6.76–8.75	6.36–8.93	1.99

Source.

<sup>a</sup> Li, Baga, et al., 2011; Ragaee, Guzar, Abdel-Aal, & Seetharaman, 2012.

<sup>b</sup> Li, Beta 2012; Abdel-Aal et al., 2007.



gluten-free proteins and natural health products particularly for celiac disease, obesity and diabetes treatment would provide the opportunity for hairless canary seed industry to grow.

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