



## FOOD VALUES OF TWO ENDEMIC WILD ALMOND SPECIES FROM IRAN

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**ABSTRACT:** Iran is one of the centers of *Amygdalus* genus origins. *Amygdalus lycioides* locally called “Badamak” is one of the endemic ones found in some parts of Iran especially in south regions. It is traditionally used as antidiabet, antiinflammation, antibacterial and laxative agent. *A. wendelboi* is the other endemic species distributed just in south of Iran, in Mountains of Gnow protected area. The fruits locally called “Archen”, has a wide application in folk medicine to treat cold, cough, headache and skin burns. It is locally used as an edible kernel by people. Literature survey revealed that no investigation has been done on this species by now. Results showed the proximate composition of 8.23% and 9.66% moisture; 11.06% and 13.29% ash; 38.26% and 34.23% lipid; 11.00% and 9.46% crude fiber; 4.01% and 3.93% crude protein; 27.44% and 29.43% carbohydrate for *A. lycioides* and *A. wendelboi* kernels respectively. Large amount of minerals were found in both of kernels.

**Keywords:** proximate composition, mineral elements, *A. lycioides*, *A. wendelboi*

## INTRODUCTION

Almond (Rosaceae family) with wide applications in pharmaceutical, oleo chemicals, food and cosmetic industries is considered as a pleasant nut throughout the world. Other than the regular almond, there are more than thirty wild or partially cultivated almond species in the world of which twenty species have been reported in Iran [1]. *A. lycioides* locally called “Badamak” is one of the endemic species found in some parts of Iran especially in south regions [2]. It is traditionally used as antidiabet [3] antiinflammation, antibacterial and laxative agent [4]. After removing the bitterness from the kernel oil, it could be used as edible nutritive oil. Several studies have been reported on *A. lycioides* different parts. Phenolic and flavonoid contents of this plant determined as 233 and 13.4 mg/g [5]. Six flavonoids, i.e. quercetin 3-O-rhamnoside, luteolin 7-O-rhamnoside, isorhamnetin 3-O-rutinoside, kaempferol 3-O-rhamnoside, apigenin, and naringenin have been isolated from the aerial parts of this plant [3]. Four chiral flavanones including (2R, 3R)-Taxifolin, (2R, 3R)-aromadendrin, (S)-5,7,3, 5'-tetrahydroxyflavanone and (S)-naringenin of which (S)-naringenin has been reported as the responsible constituent for the significant TNF $\alpha$  blocking effect of *A. lycioides*. TNF $\alpha$  blockers represent a major advance in the treatment of chronic inflammatory diseases, such as rheumatoid arthritis, bowel diseases and psoriasis [6]. Antioxidant and vasorelaxant properties of *A. lycioides* have been shown [3,5].

*A. wendelboi* is another endemic species distributed just in south of Iran, in Mountains of Gnow protected area [2]. The fruits locally called “Archen”, has a wide application in folk medicine to treat cold, cough, headache and skin burns. It is locally used as an edible kernel by people. Literature survey revealed that no investigation has been done on this species by now. Although, these species possess edible kernels eaten by both young and old people especially in south of Iran, there are no reports on the physicochemical properties, mineral elements, lipid, protein and carbohydrate contents of them.

The mechanisms by which particular elements or their compounds may affect heart disease risk are not clear, but it is likely that they involve effects on enzymes, hormones, and messenger molecules. After all, as Mildvan [7,8] observed, more than 27% of known enzymes contain mineral elements and/or require minerals for activity. It has been suggested that chromium may produce some of its effects by potentiating insulin [9]; zinc deprivation decreased serum thymulin and interleukin-2 [10]; and prostaglandin metabolism appears to be modified by dietary copper [11,12]. Other mechanisms may involve the complexity of interactions among the various mineral elements. For example, zinc is known to inhibit copper utilization [13,14,15,16] and increase plasma cholesterol [13,17], and cadmium can induce hypercholesterolemia that can be relieved by extra copper [18]. Lead produced increased urinary losses of copper [19] and can antagonise copper in hematopoiesis [20]. Pekelharing et al. [21] found that amounts of tin similar to those found in human diets can decrease copper status. Calcium can modify the utilization of other dietary elements. Romasz et al. [22] fed rats a wide range of dietary calcium; as dietary calcium increased, the concentration of cholesterol in serum decreased, and the concentrations of copper and zinc in liver increased and decreased, respectively, although the dietary concentration of the trace elements was constant. As plain part of the regions where these two species are grown includes much of the southern, eastern and northern part of the strip consisted of alkaline and saline soils contain large amounts of soluble salts such as chloride, sulfate and carbonate of Ca, Mg, sodium, and potassium, the mineral elements content could be high.

The aim of this study was determination of proximate composition, mineral elements (Calcium, Potassium, iron, Lithium, Zinc, Sodium, Selenium, Magnesium and Copper), carbohydrate, crude fiber and protein and lipid contents in these two endemic Iranian almond kernels.

## MATERIALS AND METHODS

### Plant material

*A. lycioides* and *A. wendelboi* fruits were collected in June 2013 from Sirmand mountains near Hadji-Abad County, Hormozgan Province and the mountains in Gnow protected area, Bandar-Abbas, Hormozgan Province respectively. Both samples were identified by R. Asadpour. The Geno Biosphere Reserve, with a total area of 27,500 hectares, situated in the Hormozgan province of Iran. It has been designated as a protected area by the Iranian Department of Environment in 1976. The area is mountainous region that located among plains and hills. The region's geographical location "18 ° 27 to" 29 ° 27 north latitude and "18 ° 56 to" 56 ° 55 'east longitude, is located in the north of Bandar Abbas (Figure-1). Plain part of the region includes much of the southern, eastern and northern part of the strip consisted of alkaline and saline soils contain large amounts of soluble salts such as chloride, sulfate and carbonate of Ca, Mg, sodium, and potassium [23].



Figure-1: Location of Samples collection

**Moisture Content**

All samples were oven dried at 60°C for 36-48 hours until a constant weights were obtained. The moisture contents were expressed as loss in weights of the wet samples [24, 25, 26, 27].

**Crude Fiber**

Five grams of the ground samples were digested in 100ml of 1.25% H<sub>2</sub>SO<sub>4</sub>. The solutions were boiled for 45 minutes and then were filtered and washed with hot distilled water. The filtrates were digested in 100 ml of 1.25% Sodium Hydroxide solutions. These solutions were heated for 45 minutes, filtered and washed with hot deionized water and oven dried. Finally the oven-dried residues were ignited in a furnace at 550°C. The fiber contents were measured by the weights of the left after ignition and were expressed in term of the weights of the samples before ignition.

**Crude Protein**

The protein nitrogen in one gram of the dried samples were converted to ammonium sulphate by digestion with concentrated H<sub>2</sub>SO<sub>4</sub> (Merck 96.5%) and in the presence of CuSO<sub>4</sub> and K<sub>2</sub>SO<sub>4</sub>. The solutions were heated and the ammonia evolved were steam distilled into Boric acid 2%. The nitrogens from ammonia were deduced from the titrations of the trapped ammonia with 0.1M HCl with Tashirus indicator (methyl red: methylene blue 2:1) until a purplish pink color were obtained. Crude proteins were calculated by multiplying the value of the deduced nitrogens by the factor 6.25mg [27,28,29].

**Ash Content**

One gram of the oven-dried samples in powder form was placed in crucible of known weight. They were ignited in a muffle furnace for 5 hours at 550°C. After cooling crucibles they were weighed and the ash contents were expressed in terms of the oven-dried weight of the sample.

**Oil (Lipid) Content**

The lipid contents of five grams of *A. wendelboi* and *A. lycioides* kernels by petroleum ether in a soxhlet apparatus were extracted. The weight of the lipid obtained after evaporating off the petroleum ether from the extracts gave the weights of the crude fat in the samples [26, 27].

**Carbohydrate Content**

The carbohydrate content of the samples were estimated as the differences obtained after subtracting the values of organic proteins, lipids, ashes and fibers from the total dry matter for both of kernel samples.

**Zinc, Manganese, Copper and Potassium Determination**

For Zinc, Manganese, Copper and Selenium concentration almond kernel samples were dried in oven for 72 hours at a temperature of 85°C. The samples were then ground and sieved through 0.5 mm sieve. The powdered samples then subjected to the acid digestion using nitric acid ( 65% Merck ), Sulfuric acid ( 96.5% Merck ) and per chloric acid ( 70% sigma ) . One gram of air-dried of each homogeneously almond samples accurately weighed and 20.0 mL of the digestion mixture ( 3 parts by weight of nitric acid: 2 parts of Sulfuric acid & 3 parts by weight perchloric acid ) and heated slowly by an oven and then rise the temperature. The remaining dry inorganic residues were dissolved in 25.0 mL of nitric acid and the solution used for the determination of mineral elements. Blanks and samples were also processed and analyzed simultaneously. All the chemicals used were of analytical grade (AR). Standardized international protocols were followed for the preparation of material and analysis of heavy metals contents [ 30,31,32,33,34] . The samples were analyzed by Flame Emission Spectrophotometer Model AA-6200 (Shimadzu, Japan) using an air-acetylene flame, using at least five standard solutions for each metal and determination of potassium content was followed by FDA Elemental analysis [35]( ORA LABORATORY MANUAL, 2013). Also, periodic testing of standard solutions was performed in order to verify of reliability of the measuring apparatus. The accuracy was checked using quality control test for fungi and their substrate samples to show the degree of agreement between the standard values and measured values; the difference was less than 5%. The samples were analyzed by Flame Emission Spectrophotometer Model AA-6200 (Shimadzu, Japan) using an air-acetylene, flame temperature: 2800°C, acetylene pressure: 0.9–1.0 bar, air pressure: 4.5–5 bar, reading time: 1–10 sec (max 60 sec), flow time: 3–4 sec (max 10 sec).

**Iron Determination**

The aliquot was passed through the atomic absorption spectrophotometer to read the iron concentration. Standards were prepared with a standard stock of 10 mg/L using ferrous ammonium sulphate where 3 - 60 ml of iron standard solution (10 mg /L) were placed in stepwise volumes in 100 ml volumetric flasks. 2 ml of hydrochloric acid were added and then brought to the volume with distilled water. The concentration of iron in the aliquot was measured using the atomic absorption spectrophotometer in mg/L. The whole procedure was replicated three times.

### Calcium, Sodium and Magnesium Determination

5 ml of the aliquot were placed in a titration flask using a pipette and diluted to 100 ml with distilled water and subsequently 15 ml of buffer solution, ten drops of Eriochrome black T indicator and 2 ml of triethanolamine were added. The mixture was titrated with Ethylene-Diamine-Tetra-Acetate (EDTA) solution from red to clear blue [36].

### Selenium Determination

Stock standard solutions for selenium were 1000 g /mL solution. All reagents and standards were of analytical grade (Merck, Germany). The palladium matrix modifier solution was prepared by the dilution (10 g/ L) Pd(NO<sub>3</sub>)<sub>2</sub> and iridium AA standard solution, 1000 g/ mL in 20% HCl, 0.1 % V/V nitric acid prepared by dilution trace pure 65 % nitric acid and 0.1 % Triton X-100 were used. Doubly distilled water was used in all operations. The samples were analyzed by Flame Emission Spectrophotometer Model AA-6200 (Shimadzu, Japan). The analyze performed according by Analytical Method ATSRD [36].

## RESULTS

### Proximate Composition

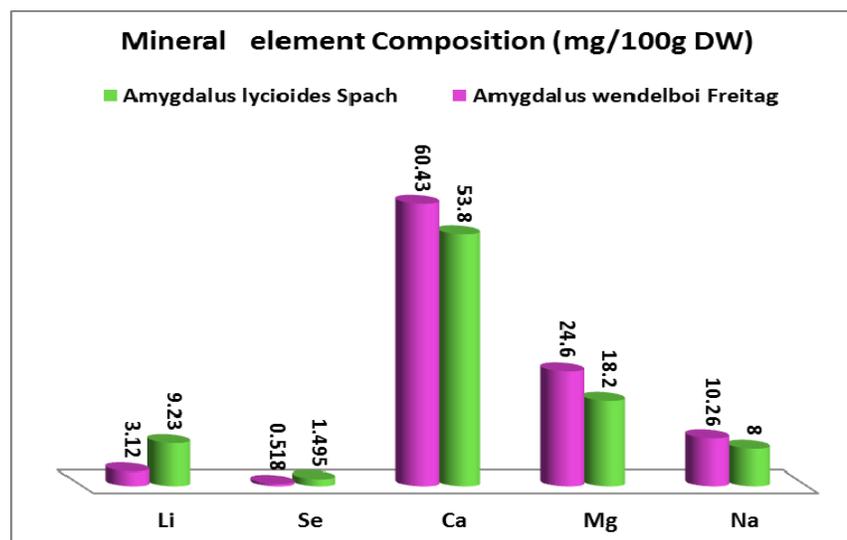
The moisture content of *A. lycioides* kernel was 8.23% and 9.66% for *A. wendelboi* kernel which shows that *A. wendelboi* kernel has high moisture content and in comparison by 25.23% for almond (*Terminalia catappa*) kernels [27] both of them can be preserved for a long time. The ash content of almond kernels: *A. lycioides* kernel was 11.06% and 13.29% for *A. wendelboi*

**Table 1- Proximate composition of *A. lycioides* and *A. wendelboi* kernel samples.**

Proximate composition	<i>A. lycioides</i>	<i>A. wendelboi</i>
Moisture	8.23	9.66
Ash	11.06	13.29
Lipid	38.26	34.23
Crude Fiber	3.93	3.93
Crude Protein	11.00	9.46
Crude Carbohydrate	27.44	29.43

### Mineral Elements Composition

The mean content of mineral elements (mg/100g DW): Sodium, Calcium and lithium in the samples are shown in figure-2. The samples were analyzed by wet digestion method and standardized international protocols were followed for the preparation of material and analysis of heavy metals contents and analyzed by Atomic Absorption Spectrophotometer in Research Laboratory in Pharmaceutical Sciences Branch, Islamic Azad University. Obviously the amount of these mineral elements are high in comparison by other almond kernels studied in other countries [27].



**Figure 2- The Mean content (mg/100g DW) of Na, Mg, Ca, Se and Li composition in *A. lycioides* and *A. wendelboi* kernel samples.**

## Selenium

Selenium is an essential component of the enzyme tetra thiodothyronone 5'-deiodinase I and is thus involved in iodine metabolism. A lack of both selenium and iodine in rats results in severe hypothyroidism and goiter [49]. Selenium deficiency can increase hypothyroid stress associated with iodine deficiency. Other data suggest that selenium deficiency impedes urinary iodine loss, thus supplementation with selenium alone may exacerbate the situation where there is combined iodine and selenium deficiency [50]. Absorption and thus bioavailability can be affected by the physical or chemical form of the selenium compound or the dosing regimen. In general, the degree of selenium absorption is independent of the exposure but in some instances, absorption can be greater where selenium deficiency exists. It is thought that 55-60% of the selenium in food is absorbed following ingestion [51]. States Environmental Protection Agency [52] US EPA, used the Reference Dose (RfD) method to establish a maximum safe level for selenium of 5 µg Se/kg bw/day and The World Health Organization [53] recommended that the upper limit of the safe range of population intake was 400 µg Se/day.

## Zinc

Zinc is an essential trace element and plays an important role in various cell processes including normal growth, brain development, behavioral response, bone formation and wound healing. Zinc deficient diabetics fail to improve their power of sensitivity and it cause loss of sense of touch and smell [54,55,56]. Zinc deficiency is common in people suffering from Chrohn's disease, hypothyroidism and gum disease, and probably plays a part in susceptibility to viral infections and diabetes mellitus. It can be beneficial in the treatment of viral infections, including those of AIDS, prostate gland enlargement, rheumatoid arthritis, healing of wounds, acne, eczema and stress [55]. Results showed that zinc concentration is high in both of studied almond kernel (figure 3).

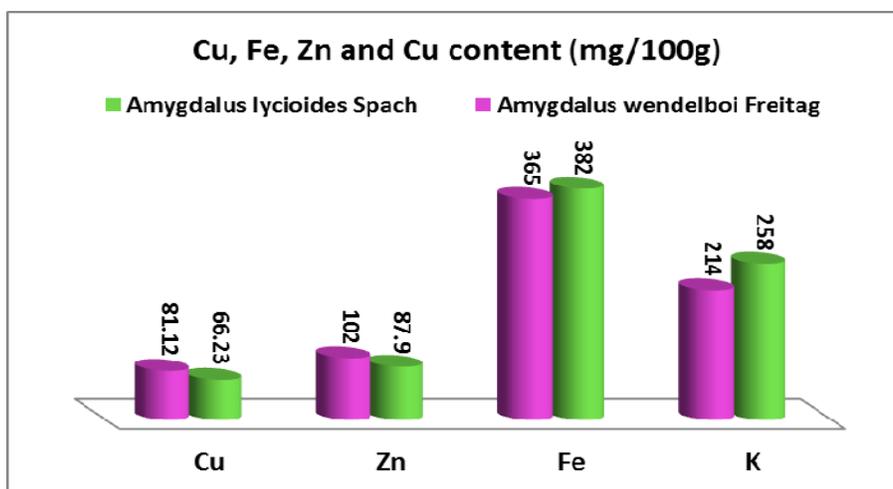


Figure 3- The Mean content (mg/100g DW) of K, Zn and Cu composition in *A. lycioides* and *A. wendelboi* kernel samples.

## Iron

The average daily intake of iron from consumption of 100 g of *A. lycioides* and *A. wendelboi* is 382 and 365 mg/day/person respectively. These intakes represent high amounts of ADI recommended by FAO/ WHO for iron [57] and as the dietary allowance for iron is 10g for 70kg [41]. Therefore, almond kernel could be recommended as a dietary supplement for people who need iron. The Iron content is high when compared to 1.94 mg/100g obtained for cocoa bean [56] and 37.5mg/100g for almond *Terminalia catappa* kernels [38] and 50 mg/100g obtained for Benni kernels[59,60]. The dietary allowance for iron is 10g for 70kg [41].

Iron is an essential element for human beings and animals and is an essential reported by component of hemoglobin. It facilitates carbohydrates, protein and fat to control body weight, which is very important factor in diabetes [61]. Iron is necessary for the formation of hemoglobin and also plays an important role in oxygen transfer in human body and low iron content causes gastrointestinal infection, nose bleeding myocardial infection [61, 62]. The effects of toxic doses of iron in animals include depression, coma, convulsions, respiratory failure, and cardiac arrest [63].

## Copper

Copper is involved in the activity of many enzymes and metabolic functions. It is necessary for the growth and maintenance of bones and is involved in the production of red blood cells, connective tissue and in metabolism of fats [54]. There is no permissible limit prescribed in local food law or by WHO, but WHO (1996) has recommended the lower limit of the acceptable range of Cu as 20 µg/mg body weight per day [64]. Gastrointestinal tract is the most sensitive target of copper toxicity. In human studies, involving a single exposure to copper following an overnight fast, adverse gastrointestinal effects (nausea, vomiting, abdominal pain, and/or diarrhea) were observed at doses of 0.011-0.03 mg/kg [63,64,65,66,67,68,69]. The Minimal Risk Level of Cu is 0.01 mg/kg /day, which has been derived for acute-duration (1-14 days) oral exposure to copper [71]. The average daily intake of copper from consumption of 100 g of *A. lycioides* and *A. wendelboi* is 66.23 and 81.12 mg/day/person respectively, therefore long term intake for everyday consumption of *A. wendelboi* is not recommended.

Therefore, both of these wild almond kernels could be recommended as a dietary supplement for people who suffer iron deficiency.

## Potassium

The value of potassium in *A. lycioides* was 258 and 214 in *Amygdalus wendelboi* Freitag. These values when compared to 350.00 mg/100g for almond *Terminalia catappa* kernels [27] and 330 mg/100g obtained for cocoa bean [58] were not so high. The dietary allowance for potassium is (1875 mg-5625 mg) for adults [41]. The kernel could not be recommended as source of dietary supplement for potassium.

## DISCUSSION

Ash content signifies the level of mineral present in the sample. The ash content of almond kernel is high when compared to 5.0% for *Terminalia catappa* [27, 38], 3.3% recorded for cashew nut [39]; it is also higher than the value of 2.7% obtained for African oil bean [27, 40]. Proximate compositions of these two endemic Iranian Badams were indicated in table 1. The dietary allowance for protein is 56g for a 70kg man [41]. The protein content for these two almond kernels are high and they could be used as dietary supplements for people who need a lot of protein and most importantly for those who require plant protein especially people suffering from hypertension. They can also be incorporated into animal feed to increase the protein content. The Recommended Dietary Allowance (RDA) for protein is 0.8 g/kg body weight for adults, set by the Institute of Medicine, and is based on the consumption of good-quality protein [42]. According to the most recent statistics from the American Cancer Society, more than 1.5 million new cancer cases are diagnosed annually. ACS reported that For decades, researchers have pursued the theory that dietary protein enhances cancerous tumor growth, particularly colon cancer [43]. In 2007 [44], a World Cancer Research Fund/American Institute for Cancer Research report on diet and cancer recommended limiting red meat intake based on limited, but suggestive, evidence that red meat consumption increases the risk of colorectal cancer. World Cancer Research Fund 2007 [45] Animal studies have found that tumor growth is greatly enhanced by diets that contain more than 10 percent animal protein and is repressed with either 5 percent animal protein or more than 20 percent plant protein. Dunaif and Youngman [46] Overall, however, studies on protein and cancer are inconsistent. A recent study found that increasing dietary protein reduced the risk of the animals developing mammary tumors [42].

It is now clear that dietary trace elements are associated with heart disease risk. Knox [47] found a negative correlation between dietary calcium intake and heart disease risk in England and Wales. Varo [48] found a highly positive correlation between death rates for ischemic heart disease and the dietary ratio of calcium to magnesium in the European Union. In fact, Finnish children with the highest concentrations of cholesterol in serum, and presumably the highest risk of IHD, consumed significantly less calcium than those with the lowest serum concentrations of cholesterol [17]. Therefore according to our findings consuming the *A. lycioides* by 53.8 and *A. wendelboi* by 60.43 mg/100g calcium is recommended.

## CONCLUSION

The results of the study revealed that almond tree kernels have higher levels of most of the chemical components. They could be not only very promising raw materials for various industries but also would serves as useful dietary supplements. Therefore, these kernels should not be overlooked anymore. The high protein and oil values of the kernels and high levels of mineral elements indicate their potentials usefulness in animal and poultry feed supplements. Almond kernel oil can also be recommended for cosmetics industries.

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