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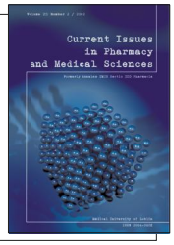

A comparison of the antioxidant properties of selected edible sprouts of the Legumes family

IWONA ZYCH-WĘŻYK*, ANNA KRZEPIŁKO

University of Life Sciences in Lublin, Faculty of Agricultural Sciences in Zamość, Poland

ABSTRACT

Germinating seeds of the Legumes family are a natural source of valuable nutrients. The aim of the study was to compare the antioxidant properties of selected edible sprouts from the Legumes family. Lentil and mung bean seeds purchased from PNOS Ożarów Mazowiecki S.A. were used for the study. The seeds were moistened in distilled water and germinated in Petri dishes lined with filter paper, in natural light conditions, at 22°C. Two methods were used to measure the total antioxidant capacity of aqueous extracts of the seedlings: the ABTS method based on reduction of ABTS cation radical and the FRAP method based on iron (III) ion reduction. The antioxidant properties were measured using the ABTS method on days 1-6 of growth. Similar antioxidant content per 1 g of fresh weight was obtained for the two kinds of sprouts (66.34 to 55.95 μmol trolox•g^{-1} FW for mung beans, 72.55 to 56.98 μmol trolox•g^{-1} FW for lentils). Then antioxidant content per seedling was calculated and found to increase with the time of growth; on day 6 it attained a value of 5.00 μmol trolux in the case of mung beans and 7.60 μmol trolox in the case of lentils. Similar dependencies were obtained using the FRAP method. Antioxidant content per seedling was highest on day 6 - 8.58 μmol Fe^{2+} equivalent for mung beans and 2.23 μmol Fe^{2+} equivalent for lentils. The results obtained in this study can be used to identify which sprouts have the highest antioxidant properties and to choose the optimal time for consuming them.

Keywords: Legumes family, antioxidant properties, ABTS, FRAP.

INTRODUCTION

Edible sprouts are an excellent source of protein, amino acids, fibre, vitamins and minerals. They also contain phenolic compounds and organic selenium derivatives, which are beneficial to human health [17]. The variety of seeds and their sprouts makes them a rich source of taste sensations. Sprouts are consumed in the initial stage of growth of the plant, during which the concentration of nutrients is very high. Most sprouts contain greater amounts of vitamin C, polyphenols and B vitamins than mature plants, and exhibit higher antioxidant activity [10].

In this study, two different methods were used to compare the antioxidant properties of sprouts of two plants of the Legumes family - mung beans and lentils. The seeds of these plants, like other pulses, are a rich source of protein with a beneficial amino acid composition [11]. Mung bean sprouts contain essential microelements - iron, copper, magnesium, sodium, potassium, calcium and zinc, as well as substantial amounts of dietary fibre, linolenic acid and tocopherols [1]. Mung bean sprouts are mainly used in Chinese and Vietnamese cuisine, in both hot and cold dishes. Lentil sprouts are rich in B vitamins and amino acids, as well as calcium, zinc, iron, potassium, magnesium and phosphorus.

Regular consumption of sprouts provides protection against many serious illnesses, activates the immune system, corrects vitamin and mineral deficiencies, and most importantly, has anticarcinogenic effects.

MATERIALS AND METHODS

We used for the study mung bean and lentil seeds purchased from PNOS Ożarów Mazowiecki S.A. The seeds were moistened with distilled water and germinated in Petri dishes lined with filter paper, in natural light conditions, at 22°C [2]. The sprouts were collected on days 1 - 6 of the culture. Aqueous extracts from the seedlings were prepared according to Zieliński and Kozłowska [19]. Total antioxidant capacity in the seedling extracts was determined by spectrophotometry, using ABTS cation radical [13, 4] and the FRAP assay proposed by Benzie et al. w 1996 [5].

The ABTS method for assessing antioxidant activity is based on a reaction between ABTS cation radical and antioxidants present in an extract, which is accompanied by a decrease in intensity of the colour of the solution. Absorbance was measured 30 minutes after the reaction had been initiated, at a wavelength of 414 nm [4]. Antioxidant activity was expressed as μmoles of Trolox per 1 g fresh weight and as μmoles of Trolox per seedling.

Corresponding author
* University of Life Sciences in Lublin, Faculty of Agricultural Sciences in Zamosc, 102 Szczebrzeska Str., 22-400 Zamosc, Poland
e-mail address: iwona.zych@up.lublin.pl

In the FRAP assay, the antioxidants contained in the extract reduce Fe^{+3} TPTZ (2,4,6-tris(2-pyridyl)-1,3,5-triazine) complex to Fe^{+2} ions. Absorbance was measured after 15 minutes of incubation at 37°C at a wavelength of 593 nm. An analytical curve was obtained for $FeSO_4x7H_2O$ [3]. The results were expressed in μmoles of iron reduced by the antioxidants contained in 1g fresh weight of seedlings and in μmoles of iron per seedling.

All determinations were made in at least three independent replications.

RESULTS AND DISCUSSION

Table 1. Measurement of the antioxidant properties of germinating seeds of the Legumes family

Time [day]	Antioxidant properties of the extract measured by the ABTS method		Antioxidant properties of the extract measured by the FRAP method	
	[μmol trolox •g^{-1}] (± standard deviation)	[μmol trolox in sprouts] (± standard deviation)	[μmol $FeSO_4x7H_2O$ •g^{-1}] (± standard deviation)	[μmol $FeSO_4x7H_2O$ in sprouts] (± standard deviation)
Mung Beans sprouts (*Phaseolus aureus*)				
1	60.12 (± 1.54)	2.31 (± 0.06)	25.68 (± 5.63)	1.98 (± 0.43)
2	63.24 (± 0.59)	2.84 (± 0.24)	30.00 (± 2.59)	2.73 (± 0.15)
3	62.63 (± 0.83)	3.97 (± 0.53)	33.03 (± 2.51)	4.18 (± 0.60)
4	55.95 (± 0.65)	3.83 (± 0.25)	34.39 (± 4.20)	4.72 (± 0.76)
5	59.84 (± 3.66)	4.49 (± 0.13)	40.23 (± 3.29)	6.03 (± 0.15)
6	66.34 (± 1.09)	5.00 (± 0.34)	57.44 (± 8.25)	8.58 (± 0.68)
Lentil sprouts (*Lens culinaris*)				
1	72.55 (± 0.54)	4.27 (± 0.11)	31.45 (± 3.23)	1.85 (± 0.23)
2	69.15 (± 0.93)	4.72 (± 0.22)	20.22 (± 2.77)	1.38 (± 0.15)
3	61.82 (± 1.37)	5.16 (± 0.18)	18.58 (± 1.74)	1.55 (± 0.10)
4	58.17 (± 0.61)	6.65 (± 0.22)	18.58 (± 0.68)	2.05 (± 0.17)
5	57.25 (± 0.91)	7.16 (± 0.11)	18.05 (± 2.58)	2.26 (± 0.32)
6	56.98 (± 1.34)	7.60 (± 0.18)	17.88 (± 2.28)	2.23 (± 0.13)

Observation of the germination process showed that the seeds differed in terms of the time, energy and rate of germination, as well as the sensory values of the sprouts. Testing of antioxidant content was begun after one day of growth; at this time most of the lentil seed coats had split and more than half of the seeds had produced a radicle. Most of the mung bean seeds had already produced a radicle after the first day of growth, and the mean length of the seedling was 15.83 mm. According to the producer's recommendations, the sprouts are ready for consumption after 3-4 days of growth. According to our observations, mung bean sprouts were best for consumption after 3 days, when the mean length of the seedling, measured from the root to the cotyledon, were 50.17 mm. Lentil sprouts are best consumed after the 4th day of growth, when the mean seedling length, measured from the root to the cotyledon, is 56.56 mm. A study by Gajewski confirms the high quality of sprouts of lentils, clover, broccoli, peas, wheat, radishes and alfalfa collected on the 3rd day of growth and stored for 2 days. Organoleptic testing has shown that the dominant sensory characteristics of lentil sprouts are their pea-like odour, grainy texture, and sweet taste [7].

Varieties of solvents are used to obtain plant extracts: water [19] or aqueous extracts of ethanol, methanol or acetone. A comparative study of how the type of solvent affects the phenolic compound content and antioxidant properties of a solution was presented by Xu and Chang. The authors determined that different types of food require different kinds of solvents, and following extraction different composition of phenolic compounds and other antioxidants is obtained. The authors suggest that the bioaccessibility of the extract should be one of the criteria used in research [18]. For the present study, aqueous extracts were prepared due to the low cost of this method. Moreover, aqueous extracts can be used directly for consumption or further technological processes, as there is no need to remove substances that are often harmful to human health, such as methanol, n-hexane or acetone.

The antioxidant activity of aqueous extracts of the sprouts was measured using two methods, ABTS and FRAP. Due to differences in the reactions that take place in each method, the measurements had different results. The antioxidant content of the seedlings increased with the time of growth (Table 1). Antioxidant activity was measured using ABTS on days 1-6 of growth, with similar antioxidant content per 1 g fresh weight obtained for the two kinds of sprouts (66.34 to 55.95 μmol trolox•g^{-1} fresh weight for mung beans, 72.55 to 56.98 μmol trolox•g^{-1} fresh weight for lentils). Antioxidant content per seedling was found to increase with time of growth. The antioxidant content in one seedling measured by the ABTS method was about twice as high on day 6 as in the one-day seedlings, in the case of both mung beans and lentils. Similar dependencies were obtained with the FRAP method. Antioxidant content per seedling on day 6 of growth increased more than 4-fold for mung beans and 1.2 times for lentils.

In the case of the mung beans, the greatest increase in antioxidant activity per seedling was noted between days 2 and 3 of growth (antioxidant activity increased by 28.46% in the ABTS method and by 34.69% in the FRAP method). In the case of the lentils, the greatest increase in antioxidant activity per seedling was noted between days 3 and 4 of growth (antioxidant activity increased by 22.41% in the ABTS method and by 24.39% in the FRAP method). The increase in antioxidant activity on succeeding days was smaller.

The methods used confirmed the high content of antioxidant compounds in the extracts tested. When Samotyja et al. tested the antioxidant properties of ethanol extracts of sprouts using the DPPH and FRAP methods, they observed that the extracts of lentil and mung bean sprouts exhibited considerably lower antioxidant activity than extracts of sunflower or radish sprouts [15].

The chemical composition of seeds of Legumes family plants changes during germination. Mung bean sprouts contain considerably fewer antinutrients than seeds; for example, decreases occur in the concentration of haemagglutinins, starchyose and total cyanide, as well as in trypsin inhibitor activity [12]. Lentil seeds germinating in light were also found to lack procyanidins, which are present in ungerminated seeds [9]. Enzyme activity during germination affects sugar, protein and fat content. During germination of

lentil seeds, changes occur in their amino acid composition, including lysine, histidine and tyrosine concentrations [14].

The antioxidant properties of plant extracts are positively correlated with phenolic compound content [18]. In germinating buckwheat seeds, changes are observed in the concentration and composition of phenolic compounds, particularly rutin [8]. Phenolic compound content also increases during lupin seed germination. Their concentration affects the free-radical scavenging ability of the extract [6].

In lentils germinating in light, concentration of trans p-coumaric acid and trans ferulic acid increases [9].

Flour and bran from germinated barley seeds contain considerably more antioxidants than seeds. After only 24 hours of the germination process the antioxidant activity of the products obtained increases by 15-22 % [16].

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