



EFFECT OF STEAMING ON THE ANTIOXIDANTS OF BRASSICA OLERACEA SPROUT, A COOL-WEATHER CROP COLLECTED FROM NILGIRIS

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Green leafy vegetables are high in minerals and antioxidant vitamins, as well as other antioxidant components like polyphenols and carotenoids, which can scavenge free radicals. In the present study, the effect of steaming was studied on the antioxidant activity, the content of vitamin C, calcium, and oxalate antinutrient of Brassica oleracea sprout (BOS) which is commonly known as Broccoli. The physicochemical and preliminary phytochemical screening was carried out according to standard methods. The antioxidant potential was assessed by using a 2,2 diphenyl -1- picryl hydrazine (DPPH) scavenging assay. Calcium, oxalate, and vitamin C were determined by simple titration methods. BOS steam heating did not affect vitamin C levels. BOS didn't show any considerable loss in vitamin C and calcium, but oxalate was reduced to a great extent as a result of steaming. The steaming method seemed to preserve better the vitamin C and calcium with a very less decrease after 10 and 20 min. of steaming. The fresh weight after steaming, in comparison to the raw one, showed a decrease due to a water loss of the vegetable tissue. The steaming method showed better results in preserving the nutrients of BOS and the desired pharmacological property can be expected.

INTRODUCTION

Broccoli is an edible plant that belongs to the italic cultivar group of the *Brassica oleracea* species¹. It is a cruciferous vegetable that belongs to the Brassicaceae family. It is thought to have originated in Italy around 2000 years ago. Broccolo is the diminutive form of broccoli, which means "small nail" or "sprout" in Italian. It signifies the "flowering crest of a cabbage." Broccoli has a huge green blossom head that looks like a tree and branches out from a sturdy eating stalk. Broccoli, which resembles cauliflower, has long been regarded as a particularly valuable food among Italians. Chou broccoli, common broccoli, calabrese, and cruciferous vegetable are some of its common names. Broccoli comes in three varieties. Calabrese broccoli, sometimes known as "broccoli," is the most well-known. The stems are thick and the heads are green. Sprouting broccoli is made up of a lot of heads

with a lot of stalks. Purple cauliflower is shaped like a head and has small flower buds. Broccoli is a cool-weather crop that thrives at temperatures between 18 and 23 degrees.

Natural antioxidants and physiologically active components can be found in abundance in vegetables². They aid in the delivery of specific nutrients that are lacking in other diets. According to epidemiological research, a diet high in cruciferous vegetables like broccoli, kale, kai Choi, cabbage, Brussels sprouts, and watercress can lower cancer risk. Glucoraphanin, glucoiberin, sulforaphane, carotenoids, vitamins, and myrosinase are all chemical components of broccoli^{3&4}. Broccoli has a lot of vitamin C and fiber. It includes a variety of elements, including diindolylmethane (DIM) and a trace quantity of selenium, both of which have anticancer potential. Broccoli includes the anticancer chemical glucoraphanin, which can be converted to sulforaphane⁵⁻⁸.

Generally, leafy vegetables contain a high concentration of nutrients and low to high levels of antinutrients as well. Nutrients have been linked to improvements in human health. Antinutrients are very less common among today's men which can result in nausea, bloating, headaches, rashes, and nutritional deficiencies⁹. Toxic amino acids, chlorogenic acid, lectins, phytic acid, oxalates, protease, and amylase inhibitors are among the few antinutrients present in plant sources¹⁰. Antinutrient levels can be reduced to acceptable levels as a result of processing. Antinutrients could be reduced to some degree by the use of soaking, cooking, boiling, and blanching techniques. Soaking reduces the amount of phytate and oxalate in foods. But these cooking methods will easily remove the nutrients to a certain extent. So a suitable method has to be followed to retain the nutrients in the leafy vegetables. Broccoli (*Brassica oleracea* var. *Italica*) is a vegetable with excellent nutritional value, containing high quantities of vitamins, antioxidants, and anticarcinogenic substances^{11&12}.

The majority of vegetables are prepared before being eaten. Vegetables are typically prepared at home for convenience and taste rather than for nutrition and health-promoting components retention¹³. Cooking is known to change the chemical composition of vegetables, changing their bioavailability and content of chemopreventive chemicals. The contents of nutritional and health-promoting substances such as vitamin C and vitamin E have been proven to be affected by cooking methods¹⁴⁻¹⁷.

In this view, our present study aims to evaluate the impact of steam cooking on the vitamin C content, calcium, oxalate, and antioxidant activity of the traditional vegetable Broccoli (*Brassica oleracea*) collected in the Nilgiris district which has enormous therapeutic properties.

MATERIALS AND METHODS

Plant materials

Brassica oleracea sprout (BOS), a cool-weather crop was procured from the local region of the Nilgiris district, Tamilnadu, India.

Processing treatments

The sample (300 g) was divided into three portions (100 g each), one of which was kept as a control (unsteamed, stored at 4° C in the refrigerator until use within 24 hrs.), and the other two were steamed for 10 minutes and 20 minutes on a perforated tray in a stainless steel steamer covered over boiling water, and then quickly cooled on ice (Fig. 1a & 1b).



Fig. 1a : Broccoli crude drug



Fig. 1b: Broccoli powdered drug

Determination of Vitamin C

The redox titration method was used to determine vitamin C levels in the dried BOS vegetable¹⁸. In a vegetable solution containing strong acid and potassium iodide, potassium iodate was added (KI). Molecular iodine is produced when potassium iodate reacts with potassium iodide. Iodine oxidizes vitamin C, resulting in dehydroascorbic acid and iodide ion (I⁻). In a 250 ml beaker, 25 ml of Vitamin C standard solution, 5 ml potassium iodide, 0.1 M hydrochloric acid, and 1 ml of the starch indicator solution were added. The potassium iodate solution was put into the burette and titrated against the beaker sample until a persistent blue-black color was observed. The

method was repeated three times, yielding an average titer value for the sample. The amount of time spent steaming differed (i.e. 10 & 20 min. respectively) Using the formula, the percentage of ascorbic acid lost owing to steaming was computed.

$$\% \text{ lost} = \frac{\text{AAF} - \text{AAH}}{\text{AAF}} \times 100$$

Where, AAF = the ascorbic acid content in the raw sample AAH = the ascorbic acid content of the steamed sample

Antioxidant activity by DPPH assay

When the DPPH free radical combines with hydrogen donors, it is converted to hydrazine. The DPPH radical is purple and turns yellow when it reacts with a hydrogen donor. It's a discoloration test that involves adding the antioxidant to a DPPH solution in ethanol or methanol and measuring the decrease in absorbance at 490 nm. The experiment was conducted on a 96-well microtitre plate. In the wells of the microtitre plate, 10 µl of each of the test samples or the reference solution were added separately to 200 µl of DPPH solution. The test and standard solutions had final concentrations ranging from 1000 to 250 µl/ml. The absorbance of each solution was measured at 517 nm with an ELISA reader against the appropriate test and standard blanks after a 20-minute incubation time at 37°C, and the remaining DPPH was estimated¹⁹.

Determination of Physicochemical properties

A standard method was used to analyze the powdered drug for physicochemical parameters. Chemical tests were performed to identify several phytoconstituents²⁰.

Estimation of calcium and oxalate

Calcium was determined by titration against the sample solution with 0.01M EDTA

as per the procedure given by AOAC/BIS/FSSAI²¹. The material is boiled in dilute sulphuric acid to remove the oxalate ions (0.5N). Then, by titrating the extract with a standard KMnO₄ solution, the oxalate content was calculated volumetrically. 1 g of material was weighed in an electronic weighing scale, then transferred to 30 ml of 0.5 N sulphuric acid and placed in a water bath to heat for 15 minutes. To remove the oxalate ions, the material is cooked in dilute sulphuric acid (0.5N). The oxalate content was then estimated volumetrically by titrating the extract with a standard potassium permanganate solution²².

RESULTS AND DISCUSSION

Minerals, vitamins, antioxidant polyphenols, and carotenoids that can scavenge free radicals, are abundant in green leafy vegetables²³⁻²⁵. The effect of steaming on antioxidant activity, the content of vitamin C, calcium, and oxalate of *Brassica oleracea* sprout (BOS) was investigated in this study. BOS steam heating did not affect vitamin C levels. BOS didn't show any considerable loss in vitamin C and calcium, but oxalate was reduced to a great extent as a result of steaming. Vallejo et al. found a similar effect of twenty minutes of steaming on BOS vitamin C concentration²⁶.

After 10 and 20 minutes of steaming, the steaming approach appeared to preserve vitamin C and calcium better, with a much smaller decline. The fresh weight after steaming was lower than the raw weight due to water loss in the vegetable tissue. The steaming method showed better results in preserving the nutrients of BOS and the desired pharmacological property can be expected. The results are shown in Table 1.

Table 1: Effect of Steaming on Vit. C, Calcium and Oxalate of BOS

S.No	Parameter	Standard value	Unsteamed	Steamed (10 min)	Steamed (20 min)
1	Vitamin C	89 mg	80 mg	78 mg	69 mg
2	Calcium	47 mg	40 mg	38 mg	36 mg
3	Oxalate	21 mg	23 mg	14 mg	11 mg

Table 2: Effect of Steaming on antioxidant activity of BOS by DPPH method

S.No	Concentration (µg/ml)	Unsteamed (I%)	Steamed (10 min) (I%)	Steamed (20 min) (I%)
1	250	28.21	27.08	29.86
2	500	47.66	48.98	49.71
3	1000	68.50	72.12	73.34

BOS that has been steam-cooked is shown to have a mild increase in antioxidant activity. Carotenoids which are powerful antioxidants and free radical scavengers that help to prevent coronary heart disease are more in BOS.

The scavenging activities are shown in Table 2. The table showed that the concentration of all the crude drugs was directly proportional to the % scavenging. All the goods have shown good antioxidant potential with better activity for BOS steamed for 20 minutes. The results show a good antioxidant potential in both steamed and unsteamed BOS indicating no considerable change as a result of steaming. When steamed BOS was compared to fresh BOS, the antioxidant activity increased.

The physicochemical properties such as total ash, water-soluble ash, acid insoluble ash, and sulfated ash, water-soluble extractive, and alcohol soluble extractives were performed for the steamed BOS. All the results comply with quality standards of Indian Medicinal plants (ICMR) and show considerable quality. Both the extracting solvents proved the efficiency in extracting the constituents. The results are shown in Table 3. The moisture content was well within the limits (less than 2%). Phytochemical screening was carried out by standard methods. The results reveal that the crude drug showed carbohydrates, alkaloids, proteins, amino acids, phenolic compounds, tannins, and flavonoids in the steamed BOS powder. The results are shown in Table 4.

Table 3: Physicochemical properties of (BOS)

S No.	Parameters	<i>Brassica oleracea</i> (BOS)
1	Total ash value	4.50 % w/w
2	Water soluble ash value	3.00 % w/w
3	Acid insoluble ash value	1.90 % w/w
4	Sulphated ash value	1.25 % w/w
5	Alcohol soluble extractive	10.30 % w/w
6	Water-soluble extractive	8.00 % w/w

Table 4: Phytochemical screening of (BOS)

S No.	Name of the Test	<i>Brassica oleracea</i>
1	Alkaloids	+ve
2	Carbohydrates	+ve
3	Glycosides	-ve
4	Saponins	+ve
5	Proteins and Amino acids	+ve
6	Phytosterols	-ve
7	Fixed oils and fats.	-ve
8	Phenolics & Tannins	+ve
9	Flavonoids	+ve
10	Gum and Mucilage	-ve

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نشرة العلوم الصيدلانية جامعة أسيوط



تأثير التبخير على مضادات الأكسدة في براعم براسيكا أوليراسيا ، محصول بارد الطقس تم جمعه من نيلجيريس

رامو جوفيندان

قسم العقاقير ، ج س س كلية الصيدلة ، ج س س أكاديمية التعليم العالي والبحث ، أوتي ، نيلجيريس ،
تاميل نادو ، الهند

تحتوي الخضراوات الورقية الخضراء على نسبة عالية من المعادن والفيتامينات المضادة للأكسدة، بالإضافة إلى المكونات الأخرى المضادة للأكسدة مثل البوليفينول والكاروتينات ، والتي يمكن أن تزيل الجذور الحرة. في هذه الدراسة ، تمت دراسة تأثير التبخير على النشاط المضاد للأكسدة ، ومحتوى فيتامين ج ، والكالسيوم ، والأوكزالات لبراعم براسيكا أوليراسيا (BOS) المعروف باسم البروكلي. تم إجراء الفحص الفيزيائي والكيميائي الأولي و وفقاً للطرق القياسية. تم تقييم إمكانات مضادات الأكسدة باستخدام ٢،٢ ثنائي فينيل -١- بيكريل هيدرازين (DPPH). تم تحديد الكالسيوم و الأوكزالات وفيتامين ج بطرق المعايرة البسيطة. لم يؤثر التبخير BOS على مستويات فيتامين سي. لم يُظهر BOS أي خسارة كبيرة في فيتامين ج والكالسيوم ، ولكن تم تقليل الأوكزالات إلى حد كبير نتيجة التبخير. يبدو أن طريقة التبخير تحافظ على فيتامين ج والكالسيوم بشكل أفضل مع انخفاض قليل بعد ١٠ و ٢٠ دقيقة من التبخير. أظهر الوزن الطازج بعد التبخير ، مقارنة بالوزن الخام ، انخفاضاً بسبب فقدان الماء من الأنسجة النباتية. أظهرت طريقة التبخير نتائج أفضل في الحفاظ على العناصر الغذائية لـ BOS وبالتالي الخاصية العلاجية المطلوبة.