



# RESEARCH ARTICLE

# Assess the impact of cultivation substrates for growing sprouts and microgreens of selected four legumes and two grains and evaluation of its nutritional properties

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#### **Abstract**

Many people suffer from a deficiency of essential micronutrients. Sprouts and microgreens can transform the whole idea of vegetables to resolve the need for a diet with fresh, nutrient-rich, and high content of phytocompounds necessary for a healthy body. The study's main objective is to evaluate the growth of 6 different seeds, such as four legumes; fenugreek, mung bean, cowpea, horse gram and two grains, wheat, sorghum microgreens. All the seeds were cultivated in soil, water and coco peat, to estimate and compare the nutritional properties of the selected sprouts vs. microgreens. The growth of microgreens in each medium was evaluated, and the proximate and nutritional properties were analysed. In terms of the growth of microgreens, coco peat medium serves the best, as it retains water for a long and it is porous to provide better aeration for the roots and also the day of harvest is shorter. In terms of the nutritional property of microgreens, soil serves the best, as it contains more nutrients than any other medium. The study results showed sprouts are better sources of proteins and carbohydrates than microgreens. However, microgreens were characterized by a high content of carotenoids, chlorophylls and ascorbic acid. It also exhibiting higher anti-diabetic and anticholinergic activity than sprouts. In addition, the microgreens have more micronutrients like zinc, copper, iron, magnesium, potassium etc., than the sprouts. Finally, microgreens were better growing with coco peat and also sources for functional components for dietary supplements and sustainable agriculture.

# **Keywords**

Sprouts, microgreens, coco peat, hydroponics, nutritional comparison

#### Introduction

Globally, about 1 billion people are chronically malnourished and approximately 2 billion suffer from a deficiency of essential micronutrients (1). Hence, to address the need for a diet with fresh, nutrient-rich and high phyto-compounds necessary for the healthy development of the body, sprouts and microgreens can be considered to transform the whole idea of vegetables. Sprouts and microgreens can quickly be grown in urban or peri-urban settings; given their short growth cycle, they can be grown without soil, fertilizers and pesticides around residential areas. Furthermore, food supplementation through sprouts and microgreens modulates weight gain and cholesterol metabolism and protects against cardiovascular diseases (2).

Sprouts are seeds that have germinated very young plants measuring 1/8-2 inches (2-5 cm) long. Despite being low in calories, sprouts are a rich

source of nutrients and beneficial plant compounds. Sprouting tends to increase nutritional levels in the sprouted grain, legume, vegetable, nut or seed. Sprouts also contain lower levels of anti-nutrients, making it easier for the body to absorb all the nutrients they contain. Studies have shown that when seeds are sprouted, the fiber they contain increases and becomes more available (3). The nutritional value of sprouts is rich. While the specific ratio of nutrients varies depending on the type of sprout, they generally contain high folate, magnesium, phosphorus and vitamins. It has a low amount of carbohydrates, and most of the fats in sprouts are mono and polyunsaturated. Sprouts are a good source of protein. Sprouts are effective in their antioxidant capacity due to their high polyphenols and L-ascorbic acid content. Additionally, sprouts are better sources of amino acids, pectin and sugars than microgreens (4).

Microgreens are tender, immature greens produced from the seeds of vegetables and legumes, having two fully developed cotyledon leaves with or without the emergence of a rudimentary pair of first true leaves with 2.5-7.6 cm (1-3") in height, harvested at 7-14 days after germination. It has outstanding nutritional and antioxidant properties and is also considered a "functional food" (5). In recent years, the microgreens market has been proliferating (6) and has also been sold as a "living product" with the growing media.

Microgreens are a new food for the 21st century attributing them a potential role as anti-inflammatory, anticarcinogenic, anti-obesogenic and anti-atherosclerotic (7). Increasing culinary demand and the ease of microgreens cultivation have generated much interest from both growers and consumers (8). However, microgreens take longer to grow and are more challenging to harvest. Microgreens contain widely differing amounts of functional compounds like antioxidants, minerals, vitamins and phenol (9, 10). Microgreen cotyledon leaves possess higher nutritional value than mature leaves. They are good sources of macro elements (potassium and calcium) and microelements (iron and zinc). Microgreens were characterized by a high content of carotenoids, chlorophylls and organic acid, without any sugars, exhibiting higher anti-diabetic and anticholinergic activity than sprouts (11).

Microgreens and sprouts, the significant difference that separates the two is the growth period. Typically, sprouts are grown for 3-5 days until harvest, but there are some sprouts that people tend to grow older until day 6-7th. For microgreens, it is usually 7-14 days, but farmers prefer to grow them for up to 25 days or until the first set of true leaves expands for a more robust flavor. Sprouts need to be given time to mature into plants. They are essentially eaten as exploded seeds, still pale because they cannot photosynthesize. They do not taste anything like the plant they are trying to grow into without being able to pull nutrients from the soil or the sun. Microgreens have more flavor and versatility than sprouts. They bring crunchiness to sandwiches, flavor and variety to leafy salads, and a sneaky dose of vitamins. Microgreens are usually grown in soil, although they can also be grown hydroponically. Sprouts are only grown hydroponically. Microgreens are taller and usually measure 4-7". Sprouts are considerably shorter and grow only to measure 2-3".

The choice of the growing medium represents one of the most critical aspects with a considerable impact on microgreens' productivity, quality and safety. Microgreens may be grown by many methods, but the growing medium determines its yield and quality (12). Soil cultivation is the most commonly used cultivation technique. It breaks down the soil crust, quickly penetrating water, nutrients and air, making it available for the plant. However, the shrinkage of land for cultivation has led to adoption of several alternative cultivation methods, such as terrace gardening, hydroponics and coco peat. Hydroponic is an agricultural method that depends on growing plants in water or mineral-based solutions without soil. With hydroponics, nutrients are more readily available for the plant to absorb. In addition, the grower can control light, heat, nutrients, hydration, pests and all other aspects of the growing process. Another alternative method of cultivation is coco peat. Coir fiber is extracted from coconut husk, a significant by-product of coconut. It is an excellent soil conditioner and is being extensively used as a soil-less medium for agro-horticultural purposes. During the process, a large quantity of dusty material called coir pith is generated. Composting of coco peat reduces its bulkiness, improves the water-holding capacity and provides nutrients in readily available form (13).

In this context, the current phyto-compounds and growth pattern among the cultivated sprouts and microgreens with the following objectives; evaluate the growth of microgreens cultivated in soil, water and coco peat. Furthermore, compare the nutritional properties of the selected sprouts and microgreens and evaluate and recommend these sprouts and microgreens for nutritional supplements to meet our future healthy food needs.

## **Materials and Methods**

#### Selection of plant seeds

Six different types of seeds, 4 legumes (Dicots); *Trigonella foenumgraecum* L., *Vigna radiata* (L.) R. Wilczek., *Vigna unguiculata* (L.) Walp., *Macrotyloma uniflorum* (Lam.) Verdc., 2 grains (Monocots); *Triticum aestivum* L., *Sorghum bicolor* (L.) Moench was selected to cultivate sprouts and microgreens (Fig. 1, 2). All the seeds were procured from the local market at Coimbatore.

## **Preparation of sprouts**

The selected seeds were first soaked for 24 h in water overnight and, the next day was tied in a cloth for sprout development.

# **Preparation of microgreens**

For microgreens, the soaked seeds (30 g) were collected, weighed and cultivated in three different mediums, *i.e.*, soil, coco peat and water.

# **Growth and harvest**

The days of cultivation were noted, and growth



 $\textbf{Fig. 1.} \ \, \textbf{Sprouts and microgreens of a)} \ \, \textbf{Mung bean, b)} \ \, \textbf{Fenugreek and c)} \ \, \textbf{Horse gram left and right respectively}.$ 

measurements were detected in each medium during the study. Thirty grams of seeds were used to measure the growth concerning substrate/days of harvest. Each value was a mean of five replicates±SE. Approximately 0.5 kg of the sample was separated into 2 parts. The first part of fresh material was used to measure contents for dry weight and total weight (14). The second part was used to find out the nutrients present.

## **Proximate analysis**

Fresh weight, dry weight, ash% and moisture % were determined according to the Association of Official Analytical Chemists (AOAC) (15).

# **Nutritional analysis**

# Qualitative and quantitative analysis

Qualitative analysis was completed using Brain and

Turner's procedure (16). First, the extracts were made by grinding the sample in a mortar and pestle with equal amounts of water to test the carbohydrates and proteins in the sample.

The quantitative assay was carried out for carbohydrates, proteins, chlorophyll and ascorbic acid. Quantitative analysis was done (17) for carbohydrates, proteins, ascorbic acid and chlorophyll.

## Statistical analysis

Analysis of variance (ANOVA) was performed on all parameters to compare the microgreen plants vs. substrates and sprouts. Means were separated using Duncan's Multiple Range Test (DMRT) (IBM SPSS 20 version).



 $\textbf{Fig. 2.} \ \textbf{Sprouts and microgreens of d)} \ \textbf{Cowpea, e)} \ \textbf{Wheat, f)} \ \textbf{Sorghum left and right respectively}$ 

## **Results**

## **Growth measurements of microgreens**

The growth measurement of 6 microgreens about substrate for growth/days of harvest are shown in Table 1. Wheat showed the maximum growth (7.42 cm), fenugreek showed minor growth (5.54 cm) on the 10<sup>th</sup> day of harvest in the soil as substrate and coco peat as a substrate on the eighth day of harvest. Wheat showed the maximum growth (9.42 cm) and fenugreek showed minor growth (5.66 cm) when water is taken as a substrate on the 12th day of harvest; wheat showed the maximum growth (6.22 cm) and fenugreek here also showed less growth (3.26 cm). There is a significant relationship existed in the growth measurement of the substrates like soil (F=12.028; P<0.001), coco peat (F=26.039; P<0.001) and water (F=14.942; P<0.001). It is assumed that wheat showed the more essential growth

measurements in all three substrates, followed by sorghum. The coco peat showed the maximum growth in microgreens compared with the three substrates. There is a significant relationship that existed in the plant (F=47.259; P<0.001), substrate (F=154.394; P<0.001) and interactions of plant vs. substrate (F=3.309; P<0.01).

# **Proximate analysis**

The values of dry weight, fresh weight, % of moisture and ash content of both sprouts and microgreens are shown in Tables 2 and Supplementary Table 1. The sprouts ranged in weight from 4.02 to 3.91 g while fresh and from 0.38 to 0.37 g when dried. All the sprouts had an average moisture content of 90.60% and an average ash content of 9.39%. There is no significant relationship existed in sprouts of all the plants between the proximate analysis like fresh weight (F=0.729; P>0.05), dry weight (F=0.144; P>0.05), moisture% (F=0.772; P>0.05) and ash % (F=0.794; P>0.05) (Table 2).

**Table 1.** Growth measurement of the different microgreens after harvest

	Growth measurement (cm)			
Microgreens	Substrate for growth /Days of harvest (days)			
_	Soil /10	Coco peat/ 8	Water/ 12	
Fenugreek ( <i>Trigonella foenum-graecum</i> L.)	#5.54±0.09d	5.66±0.12c	3.26±0.25c	
Mung bean (Vigna radiata (L.) R. Wilczek)	6.96 ± 0.34ab	6.92± 0.20b	4.10 ± 0.19bc	
Cowpea (Vigna unguiculata (L.) Walp)	6.40 ± 0.13bc	7.66 ± 0.18 b	4.64 ± 0.23b	
Horse gram (Macrotyloma uniflorum (Lam.) Verdc)	5.82 ± 0.06cd	6.36 ± 0.10 c	3.93 ± 0.34bc	
Wheat (Triticum aestivum L.)	$7.42 \pm 0.34a$	9.42 ± 0.65a	6.22 ± 0.46a	
Sorghum (Sorghum bicolor (L.) Moench)	7.16 ± 0.16 a	8.66 ± 0.13 a	5.70 ± 0.14a	
F5, 29	12.028***	26.039***	14.942***	

Plant  $(5,72) = 47.259^{***}$ , Substrate  $(2,72) = 154.394^{***}$ , P × S  $(10,72) = 3.309^{**}$ 

"Each value is a mean of five replicates ±SE (standard error). This means that a column followed by the same superscript (P>0.05) differs according to Duncan's multiple range test., \*\*\*, \*\*Significant at P<0.001 and P<0.01 respectively.

Table 2. Proximate analysis of sprouts

Sprouts	Fresh weight (g)	Dry weight (g)	Moisture content %	Ash %
Fenugreek	#3.91 ± 0.09a	$0.38 \pm 0.01a$	90.28 ± 0.15a	9.71 ± 0.15a
Mungbean	4.00 ± 0.01a	$0.38 \pm 0.01a$	90.58 ± 0.28a	9.40 ± 0.27a
Cowpea	3.96 ± 0.06a	0.37 ± 0.01a	90.56 ± 0.19a	9.44 ± 0.19a
Horse gram	4.01 ± 0.02a	0.37 ± 0.01a	90.74 ± 0.29a	9.26 ± 0.29a
Wheat	3.92 ± 0.07a	0.37 ± 0.01a	90.58 ± 0.19a	9.42 ± 0.19a
Sorghum	4.02 ± 0.04a	0.37 ± 0.01a	90.88 ± 0.23a	9.12 ± 0.23a
F 5,17	0.729 ns	0.144 ns	0.772ns	0.794ns

<sup>\*</sup> Each value is a mean of five replicates ±SE (standard error). This means in a column followed by the same superscript (P>0.05) is different according to Duncan's multiple range test. ns is non-significant.

The fresh weight of the microgreens grown in soil ranged from 5.08 g to 5.03 g, coco peat ranged between 4.73 g and 5.71 g and in water ranged from 5.39g to 4.39g, the dry weight of the microgreens grown in the soil which ranged (0.50-0.47 g), in coco peat (0.55-0.45 g) and water ranged from (0.52 -0.38 g). The % of moisture content of microgreens grown in soil (90.34-92.21 %), in coco peat (89.64-90.60 %), in water (89.61-90.47 %), the % of ash content of microgreens grown in soil ranged from 9.28 to 9.82%, coco peat ranged from 9.23 to 9.54 % and those grown in water ranged from 9.43 to 9.80%. There is no significant relationship existed in microgreens between the proximate analysis of fresh weight grown in soil (F=1.736; P>0.05), water (F=2.912; P>0.05) and it shows significance in the case of coco peat (F=3.132; P<0.05). There is a significant relationship existed in microgreens between the proximate analysis of dry weight grown in all 3 substrates, in soil (F=4.509; P<0.05), coco peat (F=4.350; P<0.05) and water (F=3.237; P<0.05). There is a significant relationship existed in microgreens between the proximate analysis of the percentage of moisture content grown in soil (F=67.64; P<0.001) and in coco peat (F=4.905; P<0.01) and in the case of microgreens grown in water, it is not significant (F=1.608; P>0.05). Regarding the % of ash content, there is a significant relationship existed in microgreens grown in soil (F=4.959; P<0.05), in water (F=4.903; P<0.05), and not significant in microgreens grown in coco peat (F=0.788; P>0.05). In comparison with the sprouts, microgreens showed the maximum proximate analysis of all the plants (Supplementary Table 1).

#### **Nutritional analysis**

#### **Carbohydrate content**

The carbohydrate content in both sprouts and microgreens is given in Table 3a and b. Sorghum sprouts had the maximum carbohydrate value (10.50 mg/g), followed by wheat sprouts (9.17 mg/g) and mung bean sprouts (7.27 mg/g). The least amount of carbohydrates was observed in cowpea sprouts (4.73 mg/g). It was of the following order: sorghum > wheat > mung bean > fenugreek > horse gram > cowpea. There is a significant relationship in carbohydrate values of sprouts (F=1170.757; P<0.001) (Table 3a).

 Table 3a.
 Estimation of carbohydrates in sprouts

Sprouts	Carbohydrate value (mg/g)	
Fenugreek	#6.83 ± 0.03d	
Mung bean	7.27 ± 0.03c	
Cowpea	4.73 ± 0.03f	
Horse gram	5.73 ± 0.03e	
Wheat	9.17 ± 0.03b	
Sorghum	10.50 ± 0.10a	
F5, 17	1170.757***	

\*Each value is a mean of three replicates  $\pm$ SE (standard error). This means that a column followed by the same superscript (P>0.05) differs according to Duncan's multiple range test. \*\*\* Significant at P<0.001.

The carbohydrate content for microgreens grown in soil ranged from 5.13 mg/g to 1.17 mg/g. The mung bean microgreens grown in soil show the highest amount of carbohydrates (5.13 mg/g), followed by cowpea (3.87 mg/g)

and fenugreek (3.43 mg/g), the least amount was noted in sorghum microgreens (1.17 mg/g). The carbohydrate content for microgreens grown in coco peat ranged (from 4.50 mg/g-0.63 mg/g). The mung bean microgreens grown in coco peat show the maximum value for carbohydrates (4.50 mg/g), followed by cowpea (1.27 mg/g) and fenugreek (1.03 mg/g). The least amount of carbohydrates was noted in sorghum microgreens (0.63 mg/g). The carbohydrate values for microgreens grown in water ranged from 2.33 mg/g to 0.33 mg/g. Here, the mung bean microgreens grown in water showed the maximum value (2.33 mg/g), and the least was noted in sorghum microgreens (0.33 mg/g). The Carbohydrate content of microgreens of all the plants was of the following order in all three substrates: mung bean > cowpea > fenugreek > horse gram> wheat > sorghum. There is a significant relationship in carbohydrate values of microgreens grown in soil (F=1972.667; P<0.001), coco peat (F=526.160; P<0.001) and water (F=447.767; P<0.001). It was observed that the mung bean microgreens grown in all 3 substrates showed full carbohydrate content. Comparing with all 3 substrates, the soil showed the maximum value of carbohydrates in microgreens than coco peat and water. There is a significant relationship existed in the plant (F=2124.406; P<0.001), in the substrate (F=2822.469; P<0.001) and the interaction of plants and substrates (F=111.819; P<0.001) (Table 3b). When compared with the sprouts, microgreens showed a lesser amount of carbohydrates. Sprouts contribute to a rich source of carbohydrates than microgreens.

 $\textbf{Table 3b.} \ Estimation \ of \ Carbohydrates \ in \ microgreens \ (different \ substrates)$ 

Microgreen	Carbohydrates (mg/g)	Carbohydrate (mg/g)	Carbohydrates (mg/g)
	in soil	in coco peat	in water
Fenugreek	$3.43 \pm 0.03c$	2.07 ± 0.03c	1.03 ± 0.03c
Mung bean	5.13 ± 0.03a	4.50 ± 0.06a	2.33 ± 0.03a
Cowpea	$3.87 \pm 0.03b$	2.47 ± 0.03b	1.27 ± 0.03b
Horse gram	2.77 ± 0.03d	1.77 ± 0.12d	0.83 ± 0.03d
Wheat	1.57 ± 0.03e	$0.83 \pm 0.03e$	0.57 ± 0.03e
Sorghum	1.17 ± 0.03f	$0.63 \pm 0.03  f$	$0.33 \pm 0.03f$
F5, 17	1972.667***	526.160***	447.767***

Plant (5,36) =2124.406 \*\*\* ,Substrates (2,36) =2822.469 \*\*\* ,  $P \times S$  (10,36) = 111.819 \*\*\* ,

"Each value is a mean of three replicates ±SE (standard error). This means that a column followed by the same superscript (P>0.05) differs according to Duncan's multiple range test. \*\*\* Significant at P<0.001.

#### **Protein content**

The protein content in both sprouts and microgreens is given in Table 4a and b. The maximum protein content was observed in mung bean sprouts (5.83 mg/g), followed by Fenugreek sprouts (5.73 mg/g) and horse gram sprouts (5.63 mg/g). The minimum protein content was observed in sorghum sprouts (2.40 mg/g). It was of the following order: mung bean>fenugreek >horse gram >cowpea> wheat>sorghum. There is a significant relationship in the protein content of sprouts (F=2087.400; P<0.001) (Table 4a).

The protein content of microgreens grown in soil

Table 4a. Estimation of proteins in sprouts

	Protein content (mg/g)	
Fenugreek	#5.73 ± 0.03ab	
Mung bean	5.83 ± 0.03a	
Cowpea	2.77 ± 0.03c	
Horse gram	$5.63 \pm 0.03b$	
Wheat	2.43 ± 0.03d	
Sorghum	2.40 ± 0.06d	
F5, 17	2087.400***	

<sup>&</sup>lt;sup>#</sup> Each value is a mean of three replicates ±SE (standard error). This means that a column followed by the same superscript (P>0.05) differs according to Duncan's multiple range test. \*\*\* Significant at P<0.001.

Table 4b. Estimation of proteins in microgreens (different substrates)

Microgreen	Protein in soil (mg/g)	Protein in coco peat (mg/g)	Protein in water (mg/g)
Fenugreek	#2.33 ± 0.03b	1.73 ± 0.09b	0.82 ± 0.01ab
Mung bean	2.63 ± 0.03a	2.03 ± 0.03a	0.85 ± 0.01a
Cowpea	2.43 ± 0.03b	1.77 ± 0.03b	0.77 ± 0.02b
Horse gram	$1.83 \pm 0.03c$	1.43 ± 0.03c	$0.47 \pm 0.02c$
Wheatgrass	1.53 ± 0.03d	1.23 ± 0.03d	0.50 ± 0.02c
Sorghum	1.23 ± 0.03e	0.85 ± 0.00e	0.24 ± 0.02d
F5, 17	276.000***	89.464***	193.298***

Plant (5,36) =426.505 \*\*\*, Substrates (2,36) =2597.703\*\*\*,  $P \times S$  (10,36) = 24.657\*\*\*

\*Each value is a mean of three replicates ±SE (standard error). This means in a column followed by the same superscript (P>0.05) is different according to Duncan's multiple range test. \*\*\* Significant at P<0.001.

ranged from 2.63 mg/g to 1.23 mg/g. The mung bean microgreens grown in soil show the highest amount of proteins (2.63 mg/g), followed by cowpea (2.43 mg/g) and fenugreek (2.33 mg/g), the least amount was noted in sorghum microgreens (1.23 mg/g). The protein content of microgreens grown in coco peat ranged from (2.03 mg/g-0.85 mg/g). The mung bean microgreens grown in coco peat show the maximum value for protein (2.03 mg/g), followed by cowpea (1.77 mg/g) and fenugreek (1.73 mg/g). The least amount of protein was noted in sorghum microgreens (0.85 mg/g). The protein content of microgreens grown in water ranged from 0.85 mg/g to 0.24 mg/g. Here, the mung bean microgreens grown in water showed the maximum protein content (0.85 mg/g) and the minimum was noted in sorghum microgreens (0.24 mg/g). It was of the following order in all 3 substrates: mung bean > cowpea > fenugreek > horse gram > wheat > sorghum. There is a significant relationship in protein content of microgreens grown in soil (F=276.000; P<0.001), coco peat (F=89.464; P<0.001), and water (F=193.298; P<0.001). It was observed that the mung bean microgreens grown in all 3 substrates showed the most significant value for protein content. On comparison with all 3 substrates, the soil showed the maximum value of proteins in microgreens than coco peat and water. There is a significant relationship existed in the plant (F=426.505; P<0.001), in the substrate (F=2597.703; P<0.001) and interaction of plant and substrate (F=24.657; P<0.001) (Table 4b). When compared with the sprouts, microgreens showed a lesser amount of proteins. Sprouts

contributed to the maximum amount of proteins than the microgreens.

## Chlorophyll

The chlorophyll content in both sprouts and microgreens is given in Tables 5 and Supplementary Table 2. Chlorophyll a in sprouts ranged from (3.73 mg/mL - 0.53 mg/mL), Chlorophyll b in sprouts ranged from (1.63 mg/mL-0.00 mg/mL) and total chlorophyll in sprouts ranged from (5.37 mg/mL-0.53 mg/mL). Mung bean sprouts had more significant amounts of chlorophyll a (3.73 mg/mL), chlorophyll b (1.63 mg/mL) and total chlorophyll (5.37 mg/mL). The least was found in horse gram sprouts, chlorophyll a (0.53 mg/mL), chlorophyll b (0.00 mg/mL) and total chlorophyll (0.53 mg/mL). There is a significant relationship existed in chlorophyll an (F=1385.786; P<0.001), chlorophyll b (F=507.200; P<0.001) and total chlorophyll (F=1288.498; P<0.001) (Table 5).

Table 5. Estimation of Chlorophyll content in sprouts

Sprouts	Chlorophyll a (mg/ml)	Chlorophyll b (mg/ml)	Total chlorophyll (mg/ml)
Fenugreek	1.87 ± 0.03b	1.27 ± 0.03b	3.13 ± 0.07b
Mung bean	3.73 ± 0.03a	1.63 ± 0.03a	5.37 ± 0.03a
Cowpea	1.07 ± 0.03d	$0.53 \pm 0.03c$	$1.60 \pm 0.06c$
Horse gram	0.53 ± 0.03e	$0.00 \pm 0.00e$	0.53 ± 0.03e
Wheat	1.33 ± 0.03c	$0.17 \pm 0.03d$	$1.50 \pm 0.06c$
Sorghum	1.01 ± 0.01d	0.07 ± 0.03e	1.07 ± 0.04d
F5,17	1385.786***	507.200***	1288.498***

 $<sup>^{\</sup>sharp}$  Each value is a mean of three replicates ±SE (standard error). This means that a column followed by the same superscript (P>0.05) differs according to Duncan's multiple range test. \*\*\* Significant at P<0.001.

The microgreens grown in all three substrates show more significant levels of chlorophyll than sprouts. The values of chlorophyll a, b and total chlorophyll vary in the 3 substrates. The maximum amount of chlorophyll a was found in Sorghum microgreens (31.40 mg/mL) grown in soil. The maximum chlorophyll a was found in wheatgrass (31.56 mg/mL) in coco peat and (10.22 mg/mL) in water. The maximum amount of chlorophyll b was found in wheatgrass (6.22 mg/mL) in soil and (7.92 mg/mL) in water. In coco peat, a more significant amount of chlorophyll b was found in Fenugreek microgreens (31.83 mg/mL). The maximum amount of total chlorophyll was found in Sorghum microgreens (21.45 mg/mL) grown in soil. Fenugreek microgreens (27.75 mg/mL) in coco peat and wheatgrass (10.33 mg/mL) in water. There is a significant relationship existing in chlorophyll a of microgreens grown in soil (F=4385.956; P<0.001), coco peat (F=2235.889; P<0.001), and water (F=70.765; P<0.001), chlorophyll b of microgreens grown in soil (F=87.682; P<0.001), coco peat (F=2937.423; P<0.001) and water (1162.232; P<0.001), and total chlorophyll of microgreens grown in soil (F=2181.101; P<0.001), coco peat (F=20.629; P<0.001) and water (F=156.141; P<0.001). Comparing all the values of chlorophyll a, chlorophyll b, and total chlorophyll in all three substrates, wheatgrass showed the most significant amount, followed by fenugreek microgreens and sorghum microgreens. It was of the following order: wheatgrass > fenugreek > sorghum > mung bean > cowpea > horse gram.

Microgreens grown in soil and coco peat showed greater levels of chlorophyll. There is a significant relationship existing in the plant (F=2225.553; P<0.001), substrate (F=7534.223; P<0.001) and interaction of plant vs. substrate (F=1037.580; P<0.001) (Supplementary Table 2). When compared to sprouts, microgreens showed the maximum amount of chlorophyll.

#### Ascorbic acid

The ascorbic acid content in both sprouts and microgreens is given in Table 6a and b. The ascorbic acid content in sprouts ranged from 1.63 mg/g to 0.03 mg/g. From the table, it was observed that Horse gram sprouts had the maximum ascorbic acid content (1.63 mg/g) and the least was observed in Wheat sprouts (0.03 mg/g). There is a significant relationship existing in the ascorbic acid of sprouts (F=432.667; F<0.001) (Table 6a).

Table 6a. Estimation of ascorbic acid content in sprouts

Sprouts	Ascorbic acid (mg/g)	
Fenugreek	0.33 ± 0.03d	
Mung bean	0.57 ± 0.03c	
Cowpea	1.43 ± 0.03b	
Horse gram	1.63 ± 0.03a	
Wheat	0.03 ± 0.03e	
Sorghum	0.07 ± 0.03e	
F5, 17	432.667***	

\*Each value is a mean of three replicates  $\pm$ SE (standard error). This means that a column followed by the same superscript (P>0.05) differs according to Duncan's multiple range test. \*\*\* Significant at P<0.001.

**Table 6b.** Estimation of Ascorbic acid content in microgreens (in different Substrates)

Microgreens -	Ascorbic acid (mg/g)			
	Soil	Coco peat	Water	
Fenugreek	12.14 ± 0.03d	10.17± 0.03d	6.07 ± 0.03d	
Mungbean	15.28 ± 0.05c	13.92 ± 0.07c	9.27 ± 0.05c	
Cowpea	28.66 ± 0.04a	26.72 ± 0.06a	15.17 ± 0.03b	
Horse gram	8.58 ± 0.05e	6.45 ± 0.04e	5.28 ± 0.03e	
Wheat	21.25 ± 0.03b	19.16 ± 0.04b	17.71 ± 0.06a	
Sorghum	0.91 ± 0.05f	0.99 ± 0.06f	0.03 ± 0.03f	
F5,17	50571.207***	31801.328***	27975.609***	

Plant (5,36) =103874.374 \*\*\*, Substrates (2,36) =24246.230\*\*\*, P × S (10,36) = 2955.067\*\*\*

\*Each value is a mean of three replicates ±SE (standard error). This means that a column followed by the same superscript (P>0.05) differs according to Duncan's multiple range test. \*\*\* Significant at P<0.001.

The microgreens grown in all 3 substrates show more significant ascorbic acid levels than sprouts. The cowpea microgreens are grown in soil (28.66 mg/g), in coco peat (26.72 mg/g), and in water (17.71 mg/g), showing the highest amount of ascorbic acid, followed by wheatgrass (21.25 mg/g) and mung bean microgreens (15.28 mg/g) in soil and wheatgrass (19.16 mg/g) and mung bean microgreens (13.92 mg/g) in coco peat. The least amount of ascorbic acid was noted in sorghum microgreens in soil (0.91 mg/g), in coco peat (0.99 mg/g), and water (0.03 mg/g). It was of the following order in all three substrates: cowpea > wheat > mung bean > fenugreek > horse gram >

sorghum. There is a significant relationship in the ascorbic acid content of microgreens grown in soil (F=50571.207; P<0.001), coco peat (F=31801.328; P<0.001) and water (F=27974.609; P<0.001). It was observed that the cowpea microgreens grown in all 3 substrates showed the maximum ascorbic acid. Compared with all 3 substrates, the soil showed the maximum ascorbic acid content in microgreens than coco peat and water. There is a significant relationship existed in a plant (F=103874.374; P<0.001), substrate (F=24246.230; P<0.001) and in the interaction of plant and substrate (F=2955.067; P<0.001) (Table 6b). When compared with the sprouts, microgreens showed a more significant amount of ascorbic acid.

#### Discussion

Nowadays, in a society that is more aware and interested in healthy lifestyles and the prevention of diseases, sprouts and microgreens seem highly desirable products. Apart from offering the mentioned health benefits, they can be easily and quickly produced and used in many different ways. Nevertheless, sprouts and microgreens are still considered innovative culinary ingredients (4). The nutritional properties of both sprouts and microgreens were found in this study. However, each has its properties, and it is necessary to find the best one for human consumption by considering the effects of food-borne illness and its storage.

The growth measurement of 6 microgreens concerning substrates such as soil, coco peat, and water was taken. Our study showed that the coco peat has more growth than the other substrate. The results were similar to an earlier study that the coco peat growing medium gave the best response, followed by rock wool, sand and husk charcoal growing media (18). The broccoli microgreen grown in the coco peat growing medium increased the microgreen height, significantly different from the other growing media. Coco peat planting media can bind water because it has the nature of crumbs, so air, water and roots easily enter the planting media and bind water (19).

The proximate analysis was carried out for fresh weight, dry weight, moisture and ash content in both sprouts and microgreens. The proximate analysis of this study showed a maximum in microgreens. The result was similar to an earlier study, which reported that the microgreens accumulated a higher portion of plant total dry matter in the shoot compared to vegetables grown hydroponically to an adult stage (20). Furthermore, the increase in ash content in microgreens, observed in the present study, can be associated with the translocation of minerals from the seed to the vegetative mass (21).

The nutritional analysis was conducted for carbohydrates, proteins, chlorophyll and ascorbic acid in sprouts and microgreens. In this study, the sprouts showed higher levels of carbohydrates than microgreens. Our result was similar to an earlier study in that grain legumes are known to have a high carbohydrate content of up to 65% (22). When compared to microgreens, which also deplete carbohydrates, sprouting breaks down the starch, which lowers

carb content. However, the cotyledon cells in the leaves metabolize stored carbohydrates until they drain them. As a result, microgreens have significantly fewer carbohydrates than sprouts. It shows that sprouts have a maximum carbohydrate content than microgreens.

The protein content was determined for both sprouts and microgreens. This study showed higher levels of protein content in sprouts than in microgreens. Although this was similar to an earlier study (23), it was reported that the protein concentrations were adjusted to dry mass basis, and it emerged that sprouts of legumes were considerably protein-richer. The increase in protein content during sprouting is attributable to the synthesis of enzyme proteins or a compositional change following the degradation of other constituents (24).

Chlorophyll was determined in both the sprouts and microgreens. However, microgreens had greater levels of chlorophyll when compared to sprouts in this study, which corroborates the previous study (25), that the pigments are found to be richer in microgreens than in sprouts. Chlorophyll and carotenoids are primary photosynthetic pigments responsible for the specific coloration of microgreens (26).

Ascorbic acid content was determined in both sprouts and microgreens. The results of this study showed that microgreens had more significant amounts of ascorbic acid when compared to sprouts. Reports are on the highest amount of ascorbic acid was ascertained for the microgreens of broccoli, while the lowest was for the sprouts of broccoli (27). It was evident that microgreens had maximum ascorbic acid.

Additionally, sprouts are better sources of amino acids, pectin, and sugars than microgreens. On the other hand, microgreens were characterized by the high content of carotenoids and chlorophylls and organic acid, without any sugars, exhibiting higher anti-diabetic and anticholinergic activity than sprouts. Therefore, consuming sprouts and microgreens can be of magnificent importance for humans to stay healthy and avoid civilization diseases associated with oxidative stress (4).

Microgreens serve better than sprouts by considering the safety of human consumption and the risk of causing food-borne illness. In addition, microgreens have low carbohydrates and anti-nutrient content. However, they have high vitamin and mineral content, so these can be recommended as dietary supplements, especially for those who prefer less carbohydrate-containing food (28). Finally, their growing environment is safer than the sprouts.

Microgreens come from the attractiveness of their shapes, colors, crispy texture and unique flavor to children and adults and can easily be incorporated into the diet. It can be an addition to sandwiches, salads, soups, desserts and drinks. They are also rich in dietary antioxidants, commonly linked with lower risks of serious illness, including cardiovascular diseases, hypertension and diabetes (29). Thus, it would be recommended to complement microgreens with that sprouts and mature leafy counterparts to

obtain an adequate amount of nutrients and phytochemicals for a wholesome diet. First, however, it is vital to create awareness in familiar people about its health claims, nutritional facts and importance in daily life.

#### Conclusion

Eating microgreens is generally considered safe. The potential for bacterial growth is much smaller in microgreens than in sprouts as it grows under proper ventilation, giving fewer infections. Microgreens have low carbohydrates and anti-nutrient contents; but high vitamin contents, so these can be recommended as a dietary supplement, especially for those who prefer less carbohydrate-containing food supplements. In addition, microgreens can add more flavor and enhance food taste.

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## **Authors contributions**

Concept, planning, supervision and statistical analysis were done by UE. Experimental work and manuscript draft were completed by PM. The manuscript review and editing were done by the PK. All the authors have read, review and final approval for the completed manuscript.

## **Compliance with ethical standards**

**Conflict of interest**: Authors do not have any conflict of interests to declare.

Ethical issues: None.

# **Supplementary data**

Table 1. Proximate analysis of micro greens after harvest

Table 2. Estimation of Chlorophyll content in microgreens (in different substrates)

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