

Short Communication

Assessment of nutritional value in selected edible greens based on the chlorophyll content in leaves**Pandi Vivek, Sam Prabhakaran and Sadagopan Ravi Shankar ***Department of Plant Biology and Plant Biotechnology, Madras Christian College
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Chlorophyll is a kind of precious nutrition. It plays an indispensable and crucial role in the life of human beings. In this study, chlorophyll pigments were estimated in selected underutilized edible greens to avail the nutritional values of this photosynthetic pigment. Chlorophyll estimation showed that *Cardiospermum halicacabum* contain higher chlorophyll content among the ten green leaf samples investigated followed by *Amaranthus spinosus* & *Chenopodium album* and least amount of chlorophyll was reported in *Oxalis corniculata*. Our results indicate that direct consumption of these edible greens either in raw or cooked state would benefit more than instead of going on for chlorophyll supplements or chlorophyll liquid solutions or chlorophyll tablets for nutritional benefits.

Keywords: chlorophyll; nutrition; edible greens

Plants are very important in health care. Most of the people in under-developed countries still rely only on traditional medicines obtained from local plants. Medicinal plants are consumed either directly as part of diet or in the form of dietary supplements. In the recent years, there is awareness among people towards usage of plant based products mainly because of lesser side effects and high nutritive value. Greens are important sources of protective food which are highly beneficial for the maintenance of good health and prevention of diseases (Sheela *et al.*, 2004; Mensah *et al.*, 2008 and Kwenin *et al.*, 2011). Chlorophyll is the name of the leaf pigment of the higher plants and green algae. It gives them the green colour and enables the photosynthesis. Chlorophyll exists in a, b, c and d forms. Chlorophyll a & b are seen in

the green plants. Chlorophyll molecule consists of a porphyrin head consisting of tetrapyrrole ring and hydrocarbon containing phytol tail. The molecular structure of Chlorophyll is quite similar to the molecular structure of hemoglobin (Dougherty *et al.*, 1966).

Chlorophyll is known to possess numerous health benefits ranging from the health of the circulatory, digestive, immune and detoxification systems of the body. Chlorophyll counteracts toxins and inhibits the activities of cancer causing elements (Ferruzzi and Blakeslee, 2007). Chlorophyll increases iron levels in human blood, which is especially useful for pregnant or nursing women. Chlorophyll also helps to purify the liver. Chlorophyll regulates blood sugar levels in human body which is good for general health and wellness. Chlorophyll is

known to be reported for its antioxidant potential (Hoshina *et al.*, 1988). Chlorophyllin (E141), is a green-coloured derivative of chlorophyll pigment used as a food additive and in alternative medicine (Levent İnanç, 2011). Recent research shows that chlorophyll has promising medicinal and health benefits (Ferruzzi and Blakeslee, 2007; Kizhedath and Suneetha, 2011 and Levent İnanç, 2011).

Identification of local greens with high chlorophyll content will enable us to utilize them in fresh state into our diet. These greens can be a cheap source of this pigment along with their medicinal value. Hence, an attempt is made to estimate the amount of chlorophyll pigments in less commonly used edible greens mainly to justify the nutritive and medicinal values of chlorophyll in greens.

Materials and Methods

Identification and Collection of Edible Greens

Healthy fresh leaves of selected commonly available under-utilized edible greens such as *Amaranthus spinosus* L. & *A. viridis* L. (Amaranthaceae); *Barringtonia acutangula* (L.) Gaertn. (Lecythidaceae); *Cardiospermum halicacabum* L. (Sapindaceae); *Chenopodium album* L. (Chenopodiaceae); *Oxalis corniculata* L. (Oxalidaceae); *Portulaca oleracea* L. (Portulacaceae); *Salvadora persica* L. (Salvadoraceae); *Sauropus androgynus* (L.) Merr. (Euphorbiaceae) and *Trianthema portulacastrum* L. (Aizoaceae) were identified and collected from various localities in and around Tambaram, Chennai, India. Some of the edible greens were also procured from the greens vendor of the local market. Based on the local information and scientific information (Gamble and Fischer, 1915-1936; Livingstone and Henry, 1994) the above edible greens were authenticated and subjected to the quantification of chlorophyll pigments as per the standard procedure.

Extraction and Estimation of Total Chlorophyll Pigments

Chlorophyll estimation was carried out according to Arnon (1949) and Brown and Hooker (1977). This procedure was carried out in dim light in order to reduce photo-destruction of the pigments. Healthy fresh green leaves of the above mentioned green species were collected and 1 gram of each will be weighed and finely cut and grinded to a fine pulp with the addition of 20 ml of 80% acetone using a mortar and pestle. This paste is centrifuged for 5 minutes at 5000 rpm and the supernatant is transferred to a 50 ml beaker. The residue is grinded with 20 ml of 80% acetone, and is centrifuged for 5 minutes at 5000 rpm and the supernatant is transferred to the same beaker. This process is repeated for four times till the residues became almost colourless. The inside of the mortar and pestle is also washed with 80% acetone and the clear washings are collected in the beaker. The volume is made up to 100 ml with 80% acetone. This process is repeated for all the leaf samples. The absorbance of the extract solutions is read at 645 and 663nm against the solvent (80% acetone) blank and calculations of respective samples using spectrophotometer. Chlorophyll can easily be quantified with a spectrophotometer based on the Beer-Lambert Law and the extinction coefficient for chlorophyll. The equations of Arnon (1949) were used for quantification of the total chlorophyll, chlorophyll **a** and chlorophyll **b** content in an 80% acetone extract:

$$\begin{aligned} \text{Total chlorophyll } (\mu\text{g/ml}) &= 20.2 (A_{645}) + 8.02(A_{663}) \\ \text{Chlorophyll a } (\mu\text{g/ml}) &= 12.7 (A_{663}) - 2.69 (A_{645}) \\ \text{Chlorophyll b } (\mu\text{g/ml}) &= 22.9 (A_{645}) - 4.68 (A_{663}) \end{aligned}$$

where A_{663} is the solution absorbance at 663 nm and A_{645} is the absorption at 645.

The amount of chlorophyll present in the leaf tissue is expressed mg/g of the sample based on the formula $V \times (1000 \times W) = 100 \times (1000 \times 1) = 0.1$ and substituting with the equation.

$$\begin{aligned} \text{Total chlorophyll (mg/g)} &= 20.2 (A_{645}) + 8.02 \\ & (A_{663}) \times 0.1 \\ \text{Chlorophyll a (mg/g)} &= 12.7 (A_{663}) - 2.69 \\ & (A_{645}) \times 0.1 \\ \text{Chlorophyll b (mg/g)} &= 22.9 (A_{645}) - 4.68 \\ & (A_{663}) \times 0.1 \end{aligned}$$

V= final volume of chlorophyll extract in 80% acetone which in this case is 100 ml and W=fresh weight of tissue extracted which is 1 g.

Results and Discussion

Greens are important sources of protective food which are highly beneficial for the maintenance of good health and prevention of diseases. In this study estimating the chlorophyll content in the under-utilized commonly available edible greens helped us to understand their nutritive and medicinal benefits in a better way. A total of ten edible greens belonging to eight different species were selected for this study these include *Amaranthus spinosus*, *A. viridis*, *Barringtonia acutangula*, *Cardiospermum halicacabum*, *Chenopodium album*, *Oxalis corniculata*, *Portulaca oleracea*, *Salvadora persica*, *Sauropus androgynous* and *Trianthema portulacastrum*. Greens used in this study belong to the herbs except for *Barringtonia* and *Salvadora* that are tree species. *Cardiospermum halicacabum* is a well-known medicinal green used in this study. A survey conducted in the local vegetable market (Tambaram, Chennai, India) has revealed that only three greens *Cardiospermum halicacabum*, *Oxalis corniculata* and *Portulaca oleracea* are occasionally sold in the market whereas other greens are not available in the market and they are found distributed in the disturbed areas & wastelands. *Sauropus androgynosis* rarely found in the cultivation.

Chlorophyll estimation was done on the green fresh leaf samples extracted with the acetone solvent the absorbancy readings of chlorophyll extracts were measured in two different wavelengths 645nm and 663nm

respectively using Spectrophotometer (Table 1). Based on the absorbancy values calculations were made using Arnon's (1949) equation and the amount of chlorophyll a, chlorophyll b and total chlorophyll were estimated and tabulated (Table 2).

Table 1. Spectrophotometric absorbancy values of leaf samples

| Binomial of the Green | 645nm | 663nm |
|----------------------------------|-------|-------|
| <i>Amaranthus spinosus</i> | 0.468 | 1.128 |
| <i>Amaranthus viridis</i> | 0.356 | 0.979 |
| <i>Barringtonia acutangula</i> | 0.305 | 0.772 |
| <i>Cardiospermum halicacabum</i> | 0.686 | 1.643 |
| <i>Chenopodium album</i> | 0.374 | 0.934 |
| <i>Oxalis corniculata</i> | 0.092 | 0.260 |
| <i>Portulaca oleracea</i> | 0.221 | 0.622 |
| <i>Salvadora persica</i> | 0.118 | 0.404 |
| <i>Sauropus androgynous</i> | 0.582 | 0.602 |
| <i>Trianthema portulacastrum</i> | 0.205 | 0.555 |

The results of the chlorophyll estimation shows that *Cardiospermum halicacabum* has the maximum chlorophyll content of chlorophyll a (1.902 mg/g), chlorophyll b (0.802 mg/g) and total chlorophyll (2.703 mg/g) among the ten green samples followed by *Amaranthus spinosus* and *Chenopodium album*. The least amount of chlorophyll content is found in the *Oxalis corniculata* showing chlorophyll a (0.305 mg/g), chlorophyll b (0.089 mg/g) and total chlorophyll (0.394 mg/g). Among the leaves of two tree species analyzed *Barringtonia acutangula* showed high content of chlorophyll a (0.898 mg/g), chlorophyll b (0.337 mg/g) and total chlorophyll (1.235 mg/g).

The result signifies that most of the benefits of various green foods seem to be

related to their chlorophyll content. Chlorophyll has the power to regenerate our bodies at the molecular and cellular level. It has been found that average consumption of 300-400 grams of green vegetables and fruits per day will supplement enough chlorophyll. So only chlorophyll is called as "green blood". Recent studies have indicated that diet rich in chlorophylls help to prevent diseases but the method of extraction only results in the either denature or loss of this chlorophyll pigment (Ferruzzi and Blakeslee, 2007; Kizhedath and Suneetha, 2011 and

Levent İnanç, 2011). Hence, this study suggest that instead of going on for chlorophyll supplements or chlorophyll liquid solutions or chlorophyll extracts or chlorophyll tablets for availing the nutritive and health benefits, greens can be directly consumed in cooked state or sometimes eaten as fresh. These underutilized greens can be used effectively in human dietary consumption as a source of nutrient supplement (Chlorophyll) for availing health and nutrient benefits of chlorophyll at relatively affordable cost.

Table 2. Estimation of Chlorophyll a, Chlorophyll b and Total Chlorophyll in leaf samples

| Binomial of the Green | Chlorophyll a (mg/g) | Chlorophyll b (mg/g) | Total chlorophyll (mg/g) |
|----------------------------------|---------------------------------|---------------------------------|-------------------------------------|
| <i>Amaranthus spinosus</i> | 1.307 | 0.543 | 1.850 |
| <i>Amaranthus viridis</i> | 1.147 | 0.357 | 1.504 |
| <i>Barringtonia acutangula</i> | 0.898 | 0.337 | 1.235 |
| <i>Cardiospermum halicacabum</i> | 1.902 | 0.802 | 2.703 |
| <i>Chenopodium album</i> | 1.085 | 0.419 | 1.564 |
| <i>Oxalis corniculata</i> | 0.305 | 0.089 | 0.394 |
| <i>Portulaca oleracea</i> | 0.730 | 0.214 | 0.945 |
| <i>Salvadora persica</i> | 0.481 | 0.081 | 0.562 |
| <i>Sauropus androgynous</i> | 0.607 | 1.051 | 1.658 |
| <i>Trianthema portulacastrum</i> | 0.649 | 0.209 | 0.859 |

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