



# The Effect of Eco-Enzyme Spraying on Chlorophyll Content of Hydroponic Spinach (*Amaranthus sp.*)

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**Abstract.** The green pigment chlorophyll gives a plant's leaves and stems their color. The process of photosynthesis depends heavily on chlorophyll, which also contains significant amounts of antioxidant, anti-inflammatory, and wound-healing compounds. Many green vegetables, including spinach, contain chlorophyll. The vitamins, proteins, minerals, calcium, phosphorus, sitosterol, and minerals found in spinach leaves, notably iron, are essential for the growth and wellness of the human body. Both hydroponically and non-hydroponically, spinach can be grown. A preliminary investigation carried out in December 2020 discovered that the chlorophyll concentration of spinach grown hydroponically was lower than that of spinach grown conventionally. This study aims to determine how spraying ecoenzym affects the amount of chlorophyll in hydroponically produced spinach. Ecoenzymes are created when organic kitchen waste, sugar, and water are fermented in a 3:1:10 ratio. This process can speed up biochemical activities in nature and create enzymes that are helpful for processing leftover fruit or vegetables. From April to August 2021, this study was carried out at the Wire House and Plant Physiology Laboratory, Department of Biology, Universitas Negeri Padang. This study used an experimental design with four treatments (addition of 1 ml, 2 ml, 3 ml, and 4 ml of ecoenzymes in 1 L of water) and two replications. The design was entirely randomized. Spectrophotometric methods were used to analyze the chlorophyll content. Following the completion of the investigation, it was discovered that lettuce grown hydroponically with ecoenzymes has an average total chlorophyll content in mg/L of P1 (control), 3,181, P2, 5, 739, P3, 6,151, and P4, 6,755. The study's findings showed that adding chlorophyll enhanced the overall chlorophyll content of spinach grown hydroponically numerically, while adding ecoenzymes had no statistically significant impact at the 5 percent level.

**Keywords:** Chlorophyll · Hydroponics · Ecoenzyme · Spectrophotometry · Spinach

## 1 Introduction

The pigment chlorophyll provides a plant's leaves and stems their green hue. In addition to that, chlorophyll is crucial to the process of photosynthesis. Additionally, chlorophyll has large levels of anti-inflammatory, antioxidant, and wound-healing compounds. Many green vegetables, including spinach, contain chlorophyll. The vitamins, proteins, minerals, calcium, phosphorus, sitosterol, and minerals found in spinach leaves, notably iron, are essential for the growth and wellness of the human body. Both hydroponically and non-hydroponically, spinach can be grown.

Hydroponics is an alternative that can be used to increase crop productivity, especially on small holdings. Mechanical cultivated hydroponics using media other than soil and using water as a nutrient. As explained by [1], that hydroponics is a way of cultivating plants without using soil media. Hygiene for hydroponic cultivation is usually not only determined by the medium used, it is also determined by the nutrient solution given because the nutrients provided greatly affect the growth of plants that are cultivated hydroponically [2]. Nutrient solutions commonly used in hydroponic systems are inorganic fertilizers, namely *AB mix*. *AB mix* is a nutrient solution consisting of inorganic chemical compounds that have macronutrients and micronutrients combined as nutrients for plants [3]. In addition to the high price, the use of inorganic fertilizers can pollute the environment. This is what makes researchers interested in spraying coenzymes in hydroponic spinach cultivation. Since a preliminary study was completed in December 2020, the comparison shows that spinach grown hydroponically has a lower chlorophyll concentration than spinach grown conventionally.

*Ecoenzyme* is a refined organic waste product made from household trash that is fermented with sugar, water, and organic waste. The liquid ecoenzyme is dark brown in color and smells strongly of sour/freshness. [4]. Although the fermentation process is quite long, namely for 3 months. However, the resulting solution has very many properties. In the fermentation process alone, it has produced a lot of O<sub>3</sub> (ozone) which the earth's atmosphere need. When water is added to the solution ecoenzym, a reaction occurs that allows for the usage of organic fertilizer, a mixture with water when used to water plants will give better flower, fruit, or harvest yields.

This study aims to determine how spraying ecoenzym affects the amount of chlorophyll in hydroponically produced spinach.

## 2 Material and Methods

### 2.1 Tools and Materials

This research uses tools such as NFT system, tray, plastic cup, 250 mL measuring cup, 1000 mL beaker glass, stirring rod, digital scale, TDS (Total Dissolved Solid) meter, pH meter, oven, millimeter paper, glass funnel, bottles. Spray, knife, scissors, nails, ruler, camera.

The material used is ecoenzyme taken from UNP biology lecturer which is produced at home wire department of biology, lettuce seeds obtained from hydroponic agriculture in West Sumatra, hydroponic nutrition (*AB mix*), water, black plastic, toothpick, rockwool, plastic, label paper.

## 2.2 Experimental Design

The design used in this study was a completely randomized design (CRD) with 5 treatments and 4 replications. Consists of control (K), 1mL eco enzyme + 1 L water, 2mL eco enzyme + 1 L water, 3mL eco enzyme + 1 L water and 4mL eco enzyme + 1 L water.

### 2.2.1 Making Container Planting Medium

West Sumatra's hydroponic community provided the spinach seeds. Seeding is done for 7 days until the seedlings have 3–4 perfect leaves. The spinach seeds were sown on rockwool that had been cut to a size of 2 x 2 x 2 cm. Then rockwool doused with water until moist, give a hole. Place it in a place exposed to sufficient sunlight.

### 2.2.2 Preparation and Seeding of Spinach Seeds (*Amaranthus Sp.*)

The West Sumatra hydroponic community provided the spinach seeds. Seeding is done for 7 days until the seedlings have 3–4 perfect leaves. The spinach seeds were sown on rockwool that had been cut to a size of 2 x 2 x 2 cm. Then rockwool doused with water until moist, give a hole. Place it in a place exposed to sufficient sunlight.

### 2.2.3 Nutrition Making

Make a hydroponic system parent solution, namely AB *mix* nutrition. Each stock A and stock B was dissolved in 500 mL of water. The recommendation for using AB Mix for 1 dose is 5 mL of stock A and 5 mL of stock B with 1 L of water (Sharif, 2019).

### 2.2.4 Application

Taking 1 mL of eco-enzyme is put into a glass beaker that already contains 1 L of water, then stirred until smooth. Repeat the same for the control, 2 mL eco enzyme, 3 mL eco enzyme, and 4 mL eco enzyme.

### 2.2.5 Seed Transfer

Take the seeds that are 7 days old or those that already have 3–4 leaves and then transferred to the NFT system planting media container that has been prepared.

### 2.2.6 Observation

Observations were made on the growth of Spinach plants.

## 2.3 Plant Height Observation

Observations of plant height were carried out every 6 days, until harvest, which is 5 weeks after planting.

## 2.4 Measurement of Chlorophyll Level

Chlorophyll content is evaluated using a spectrophotometric method. The new spinach leaves from hydroponic plants were weighed as much as 1 g, then cut into small pieces. The pieces were then extracted using 95% alcohol by grinding using a mortar until all the chlorophyll dissolved. Then the solution is filtered using gauze, making sure the kailan dregs are white. Then the chlorophyll solution was put into a 100 ml volumetric flask. Add 95% alcohol until the volume becomes 100ml. Then the solution was put into a cuvette and its absorbance was calculated at wavelengths of 649 and 665 nm.

## 2.5 Data Analysis

The calculation of chlorophyll content refers to Wintermans and De Mots formulas [5] can be calculated using the:

$$\text{Total chlorophyll (mg/L)} = 20.0 \text{ OD}_{649} + 6.1 \text{ OD}_{665}.$$

$$\text{Chlorophyll a (mg/L)} = 13.7 \text{ OD}_{665} - 5.76 \text{ OD}_{649}.$$

$$\text{Chlorophyll b (mg/L)} = 25.8 \text{ OD}_{649} - 7.7 \text{ OD}_{665}.$$

## 3 Result and Discussion

Based on the research that has been done, the following results are obtained.

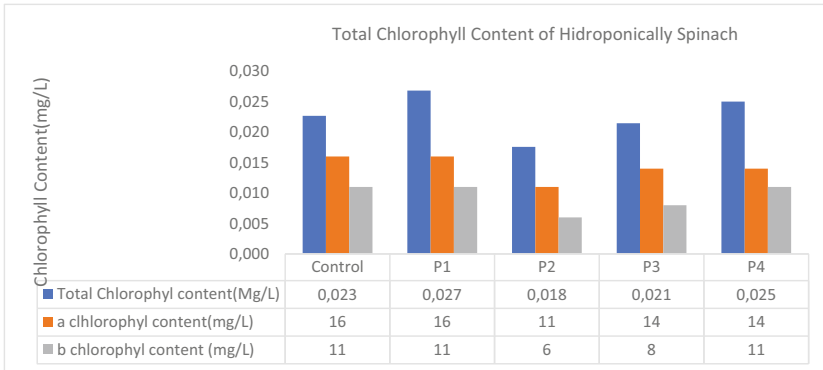
### 3.1 Measurement of Chlorophyll Levels

The formation of chlorophyll is influenced by several factors, namely plant genetic factors, oxygen, carbohydrates, water, light intensity, temperature, and nutrients. The availability of nutrients in the soil varies depending on the habitats [6].

All green plants contain chlorophyll a and chlorophyll b. Chlorophyll makes up 75% of the total chlorophyll. The chlorophyll content in plants is about 1% dry weight [7]. While chlorophyll b is an auxiliary pigment that serves to absorb light, chlorophyll plays a role in the light reaction, which transforms solar energy into chemical energy and transferring energy to chlorophyll in the light reaction. Chlorophyll a is blue-green while chlorophyll b is yellow-green.

The chlorophyll levels can be seen in Fig. 1. The results of When hydroponically grown ecoenzymes are added, the average total chlorophyll concentration in spinach is measured in mg/l as a control., 2,678 mg/l p1, 26,822 mg/l, p2, 17,584 mg/l, p3; 21,463 mg/l and p4, 25,011 mg/l. The study found that spinach grown hydroponically had an increase in total chlorophyll concentration.at p1 and decreases at p2, then increase again until p4. The highest clorophyll content is p1; 26,822 mg/l and the lower clorophyll content is p2; 17,584 mg/l. that meant the content of chlorophyll was numerically different from the spraying ecoenzyme, However, based on statistics, the ecoenzyme spraying had no discernible impact at the 5% level.

A mixture of complex organic molecules known as ecoenzyme is created through the fermentation of organic waste, sugar, and water. Ecoenzyme has a strong sour/fresh scent and dark brown color. Ecoenzymes can be used as plant fertilizers, as repellents



**Fig. 1.** Spinach chlorophyll content in various treatments.

for various plant pests, and as conservationists for the surrounding environment because eco-enzymes can neutralize various pollutants that pollute the surrounding environment [8].

Chlorophyll in plants plays a role in photosynthesis, the process of absorbing and converting light energy into chemical energy. The more chlorophyll content in the leaves, the higher the photosynthesis process in plants. Photosynthesis is an important process to maintain the growth and development of a plant [9]. The higher the chlorophyll content, the more maximal the photosynthetic reaction will be, and vice versa. If the chlorophyll content is low or low, there is a possibility that the photosynthetic reaction will not be optimal [10].

In addition to affecting photosynthesis, chlorophyll also has many health benefits including as an antioxidant, promoting anticancer detoxification, and antiaging. Therefore, by consuming vegetables that contain high chlorophyll, it will be better for the health.

## 4 Conclusion

According to studies, hydroponically grown spinach saw an increase in total chlorophyll content at P1 and decreases at P2, then increase again until P4. The highest chlorophyll content is P1; 26,822 Mg/L and the lower chlorophyll content is P2; 17,584 Mg/L. That meant the content of chlorophyll numerically different with the spraying ecoenzyme. However, based on statistics, the ecoenzyme spraying had no discernible impact at the 5% level.

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## References

1. Y.Chadirin, K. Hidaka, Y. Sago, T. Wajima, M. Kitano, Application of temperature stress to root zone of spinach II. Effect of the high-temperature pre-treatment on quality, *Environmental Control in Biology* 49, no. 4, 2011, pp. 157–164.
2. D.Z. Vidiyanto, S. Fatimah, C. Wasonowati. Penerapan panjang talang dan jarak tanam dengan sistem hidroponik NFT (nutrient film technique) pada tanaman kailan (*Brassica oleraceae* var. alboglabra, Agrovigor: Jurnal Agroekoteknologi 6, no. 2, 2013, pp. 128–135.
3. H. Mas'ud, L. Widhianto, Pertumbuhan Dan Hasil Tanaman Tomat (*Lycopersicum esculentum* Mill) Pada Media Substrat Hidroponik Dengan Konsentrasi Nutrisi ABmix Yang Berbeda, *Agrotekbis: E-Jurnal Ilmu Pertanian* 9, no. 2, 2021, pp. 495–503.
4. M. Hemalatha, P. Visantini, Potential use of eco-enzyme for the treatment of metal-based effluent, *IOP Conference Series: Materials Science and Engineering*, vol. 716, no. 1, p. 012016. IOP Publishing, 2020.
5. L. Sumenda, Analisis kandungan klorofil daun mangga (*Mangifera indica* L.) pada tingkat perkembangan daun yang berbeda, *Jurnal Bios Logos* 1, no. 1, 2011.
6. B.L. Mpapa, Analisis kesuburan tanah tempat tumbuh pohon Jati (*Tectona grandis* L.) pada ketinggian yang berbeda, *Jurnal Agrista* 20, no. 3, 2016, pp. 135–139.
7. A.J. Pratama, A. N. Laily, Analysis of chlorophyll content of gandasuli leaves (*Hedychium gardnerianum* Shepherd ex Ker-Gawl) at three different development areas, *Proc. SNKP SDA* 216, no. 1, 2015.
8. R.L. Utpalasar, I. Dahliana, Analisis hasil konversi eco enzyme menggunakan nenas (*Ananas comosus*) dan pepaya (*Carica papaya* L.), *Jurnal Redoks* 5, no. 2, 2020, pp. 135–140.
9. J. Yu, Y. Zhang, C. Niu, J. Li, Effects of two kinds of allelochemicals on photosynthesis and chlorophyll fluorescence parameters of *Solanum melongena* L. seedlings, *Ying Yong sheng Tai Xue bao = The journal of applied ecology* 17, no. 9, 2006, pp.1629–1632.
10. G. Rizal, S. Karki, V. Thakur, S. Wanchana, H. Alonso-Cantabrana, J. Dionora, J. E. Sheehy, R. Furbank, S. von Caemmerer, W. P. Quick, A sorghum (*Sorghum bicolor*) mutant with altered carbon isotope ratio, *Plos one* 12, no. 6, 2017, p. e0179567.

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