Analysis of ascorbic acid content (vitamin C) of purslane (*Portulaca oleracea* L.) at various altitudes in East Java, Indonesia

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Abstract. Purslane (*Portulaca oleracea* L.) is a weed that can be used as a source of natural antioxidants. One of the ingredients in purslane is ascorbic acid (vitamin C). One of the preventive measures related to infections caused by the coronavirus is taking vitamin C to support the body's immunity, which is proven to have a positive effect on the body's immunity. Vitamin C can prevent susceptibility to reduce respiratory infections in certain conditions. The objective of this study was to get purslane plants (*Portulaca oleracea* L.) as a source of quality bioactive components of ascorbic acid (vitamin C) from various altitudes in East Java, Indonesia. The study conducted by taking purslane planting material in the lowlands 3 m above sea level (Rungkut, Surabaya); medium plains 593 m above sea level (DAU, Malang), and highlands 945 m above sea level (Bumi Aji, Batu Malang). Vitamin C analysis was carried out using LCMS (Liquid chromatography-mass spectrometry). The results of this study were the analysis of the content of purslane (*Portulaca oleracea* L.) ascorbic acid (Vitamin C) from three altitudes was at highlands 9.24 mg/kg, medium plains 2.40 mg/kg, and lowlands 9.73 mg/kg.

1. Introduction

Public demand for natural foods that have health functions is increasing, along with the increasing patient with degenerative diseases, such as cancer, coronary heart disease, diabetes mellitus, liver, kidney failure, and so on. One of the plants that have multiple benefits, both as a food that has high nutritional value and also has medicinal properties (functional food), is the purslane plant (*Portulaca oleracea* L.).

Purslane plants are a weed that has nutritional value [1] cause its content. Purslane used as a source of food that has excellent benefits. Purslane is a food material from local resources, easy to obtain and cultivated, cheap, apart from being a food ingredient, it also has several advantages as medicinal properties. Purslane plants contain many components of active compounds. All parts of this plant contain l-norepinephrine, carbohydrates, fructose, vitamin A, vitamin B1, vitamin B2, vitamin E, and are rich in ascorbic acid [2].

Vitamin C is a water-soluble vitamin, also called ascorbic acid [3]. Vitamin C is well known for its role in collagen synthesis [4] in connective tissue and acts as an antioxidant. Vitamin C supports...
immune function [5] and protects the body against infections caused by viruses. Vitamin C can act as a weak anti-histamine agent to relieve flu-like symptoms such as sneezing, nasal congestion, and swollen sinuses. Three controlled trials in humans have reported a significantly lower incidence of pneumonia in the group taking vitamin C supplements. This study indicates that vitamin C can prevent susceptibility to lower respiratory tract infections under certain conditions. COVID-19 has been reported to cause higher respiratory tract infections, so that vitamin C can be an effective option for preventing COVID-19 [6].

Purslane can grow at various heights. The increasing of altitude causes the temperature to drop. The rate of temperature reduction is generally around 0.6 °C for each additional 100 m asl. However, this varies depending on the place, season, time of day, water vapor content in the air, and other environmental factors [7]. The difference in temperature for each height range causes the metabolic processes in a plant to be different, so the production of secondary metabolites is different.

The purpose of this study was to analyze absorbic acid content (vitamin C) of purslane (Portulaca oleracea L.) as a source of quality bioactive components from various altitudes in East Java, Indonesia.

2. Materials and methods
The study conducted by taking purslane plants which was planted in the lowlands 3 m above sea level (Rungkut, Surabaya); medium plains 593 m above sea level (DAU, Malang); and highlands 945 m above sea level (Bumi Aji, Batu Malang). The samples used were 2 purslane plants at each altitude. Sampling by dismantling the purslane plants. Coordinate point of lowlands (Rungkut, Surabaya) is LAT 7°09'17.89S Lng 112°47'22; medium plains (DAU, Malang) is LAT 7°41'78.9 S Lng 112°58'24.5 E; highlands is LAT 7°54'14.41S Lng 122°52'03.84 E.

Method: Analysis of vitamin C was carried out by means of: Weigh ± 0.2 grams of sample and put in a 12 mL conocele. Add 10.0 mobile phase A, vortex for 2 minutes. Sonification for 60 minutes, vortex for 2 minutes. Centrifuge with 8000 rpm for 10 minutes. Filter with 0.22 µM millex.Inject 2 µL into LCMSMS
HPLC condition:
Columns : ACQUITY UPLC @BEH C18 1.7 µm
Eluent : A = 10 mM ammonium formate 0.1% formic acid in Aquabidest
        B = 10 mM ammonium formate 0.1% formic acid in MeOH
Flow : 0.6 ml / min
Grad : 0.00 min 99% A
      2.00 min 99% A.
      3.00 min 45% A.
      3.10 min 99% A.
      3.5 min 99% A.

Vitamin C 177> 141 24 8

3. Results and discussion
Purslane was extracted and analyzed to determine its antioxidant content. The antioxidant activity of the fraction was evaluated to combat free radicals. Based on the results of the analysis, two polysaccharide fractions of purslane can be developed as natural antioxidants for the treatment of diseases.

| No. | Analysis     | Lowland (3 m asl) | Middle Plains (593 m asl) | Highland (945 m asl) |
|-----|--------------|-------------------|---------------------------|---------------------|
| 1   | Vitamin C (mg / Kg) | 9.73              | 2.40                      | 9.24                |
The results of the analysis of vitamin C (Table 1) based on the altitude are in the highlands of 9.24 mg/kg, medium plains 9.40 mg/kg, and lowlands 9.73 mg/kg. The antioxidant function of purslane is also related to the presence of ascorbic acid (vitamin C) [8]. Ascorbic acid found in 100 grams of purslane leaves is around 26.6 mg [9]. Vitamin C can act as a free radical scavenger and can react with superoxide anions, hydroxyl radicals, and lipid peroxides. Vitamin C can inhibit the formation of superoxide radicals, hydroxyl radicals, peroxy radicals, singlet oxygen, and hydrogen peroxide. Other endogenous antioxidant compounds in purslane are alpha-tocopherol, beta carotene, and glutathione. The results showed ascorbic acid content in purslane is about 26.6 mg and 506 mg (per 100 g fresh and dry weight, resp.) [10].

Table 2. The microclimate character of purslane (Portulaca oleracea L.)

| Altitudes     | Location          | Average temperature (°C) | Rainfall (mmr / yr) | Moisture (%) | Type of soil |
|---------------|-------------------|--------------------------|---------------------|--------------|--------------|
| Lowland       | Rungkut Surabaya  | 28.90                    | 180.60              | 74           | Vertisol     |
| 3 m asl       |                   |                          |                     |              |              |
| Medium plains | Dau Malang        | 24.20                    | 434                 | 66           | Andisol      |
| 593 m asl     |                   |                          |                     |              |              |
| Highland      | Bumi Aji Batu Malang | 22.80                | 517                 | 82           | Andisol      |
| 945 m asl     |                   |                          |                     |              |              |

Source: Observation results

Table 3. Physical and chemical properties of soils in various purslane habitats (Portulaca oleracea L.)

| Physical and chemical of soils | Lowland Value | Medium plains Value | Highland Value |
|-------------------------------|---------------|---------------------|----------------|
| pH                            | 6.78 High     | 7.24 High           | 7.09 High      |
| Organic C (%)                 | 0.77 Very low | 1.21 Low            | 1.12 Low       |
| Organic matter (%)            | 1.52 Very low | 2.09 Low            | 1.93 Very low  |
| Total N (%)                   | 0.07 Very low | 0.1 Low             | 0.09 Very low  |
| Available P (ppm)             | 7 Very low    | 16.92 Average       | 18.97 Average  |
| Exchanged K (me%)             | 0.15 Very low | 0.32 Very low       | 0.22 Very low  |
| Mg (me%)                      | 2.76 High     | 5.53 High           | 2.44 High      |
| Ca (me%)                      | 1.65 Very low | 17.92 High          | 15.48 High     |
| Texture:                      |               |                     |                |
| Dust (%)                      | 62            | 50.66               | 70.67          |
| Clay (%)                      | 36            | 46.99               | 27.77          |
| Sand (%)                      | 2             | 2.34                | 1.59           |

Source: Observation result

The results showed that the content of vitamin C in purslane showed a tendency to decrease with increasing altitude (Table 1). These results are in accordance with the opinion that the influence of altitude is mainly related to plant metabolic processes [11], such as biochemical processes and the synthesis of secondary metabolite compounds. This will affect growth, morphological characters, and the content of active compounds in a plant. Getting higher the altitude, getting higher the environmental stress (Table 3), for example, getting lower the temperature, getting higher the humidity, the smaller the sunlight intensity, getting shorter the exposure time. The stress of temperature, light, humidity, etc. can affect the production of secondary metabolites of plants. When plants are under pressure, then the production of secondary metabolites, including the production of vitamin C has decreased. This is a plant effort to fight against these environmental stresses.

Getting lower altitude, the sunshine intensity and the temperature is higher, so that vitamin C is more easily oxidized [12], especially if there is a catalyst for Fe, Cu, ascorbate oxidase enzymes, light, and high temperatures. It makes vitamin C levels at an altitude >800 m asl are lower than at an altitude
<200 m asl. Vitamin C dilute solution at pH <7.5 is still stable if there is no catalyst which can oxidized vitamin C. Oxidation of vitamin C will form dehydroascorbic acid [13]. The lower the altitude, the higher the antioxidant content. Purslane grown at different heights gives different antioxidant content. Purslane grown at different altitudes gives different antioxidant content.

Every plant has an optimum temperature for metabolism [14]. Likewise, the purslane plant has the optimum temperature for the formation of vitamin C. From the research data above, it can be stated that the optimum temperature of the purslane for vitamin C metabolism is temperature 28.9 °C at an altitude 3 m above sea level (Surabaya). Changes in environmental temperature affect plant growth and metabolism. The climate and light of a place depend on the length of exposure, the intensity, and the quality of the sunshine received [15], however, the period of plants can be changed by drought or cold. It can be said that the difference in plant response lies in the difference in environmental stress obtained by the plant.

The characteristic response of plant growth to temperature arises because high temperatures affect biochemical processes [16]. As the temperature of the plant cell increases, the speed of movement (vibration, rotation, and translation of the reacting molecules increases), causing more frequent collisions between molecules and a faster rate of reaction. All reactions that occur in the cell are accelerated by enzymes whose activity depends on the proper maintenance of the tertiary structure to which the reagents must properly adhere to react.

4. Conclusion

Based on the research, it appears that the results of the vitamin C content analysis purslane (Portulaca oleracea L.) in the highlands is 9.24 mg/kg, in the medium plains is 2.40 mg/kg, and in the lowland is 9.73 mg/kg. Getting higher altitudes, vitamin C content of purslane are lower.

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