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The growth patterns and *eleutherine* content of dayak onion (*Eleutherine palmifolia* Merr.) in sandy mineral soil and peat soil

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Abstract

This study aims to determine the growth pattern of Dayak onion *(Eleutherine palmifolia* Merr.) as well as to get the right harvest time with maximum tubers yield and high bioactive compound content in peat and sandy mineral soil media. The study was carried out in *Screen house,* Kalampangan Village, Sebangau Sub-District, Palangkaraya. The experimental design used is completely randomized design. The parameters observed by measuring the plant variables which consist of plant height, leaf number per panicle, the number of tillers per hill, tubers wet weight, tubers dry weight, and the content of bioactive compounds. All data observations were collaborated, processed, and presented in graphs and images so as to give an idea of the growth pattern of Dayak onion. The results showed that the best growth pattern is occured in sandy mineral soil with 20 weeks of DAS (days after sowing). On the other hand, the highest content of bioactive compounds was found in the *Eleutherine* of Dayakonion at the age of 18 DAS. For the growth of the onion crop dayak is recommended using sandy mineral soil media types and content of bioactive compounds was found in the *Eleutherine* of Dayakonion at the age of 18 DAS.

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Introduction

Central Kalimantan has such plant biodiversity that has medicinal potential within. Dayak onion *(Eleutherine palmifolia* Merr.) is one of the specific medicinal plants in Central Kalimantan that is used as a source of biopharmaceutical and is cultivated in order to not extinct from its natural habitat in the forest (Galingging, 2006). Dayak onion crop comes from division Spermatophyta, sub division Angiosperms, class, Monocotyledoneae order Liliales, family Iridaceae, genus *Eleutherine* and species *Eleutherine palmitoleic* Merr. (Megawati, 2005). In accordance with its name, this plant has the suitability in Borneo and is generally used as a medicinal plant for Dayak people (Pusat Studi Biofarmaka LPPM IPB, 2013).

Empirically, Dayak onion has been used by the local people (generation to generation) as a cure for various kinds of diseases such as breast cancer, colon cancer, high blood pressure (hypertension), diabetes (diabetes mellitus), ulcers, high cholesterol levels, andstroke (Galingging, 2009). The bioactive compounds of Dayakonion consist of *alkaloids, glycosides, flavonoids, phenolics, saponins, triterpenoids, and tannins (*Saptowahyono, 2007), *quinones* and *steroids* (Firdaus, 2006).

While clinically, the potential of Dayak onion as a multifunctional medicinal plant is very big. The bulb of *Eleutherine balbosa* and *Eleutherine americana* is known to contain secondary metabolic compounds and is classified as naphthoquinone (*Elecanacine, eleutherine, Eleutherol, Eleutherinone* (Hara *et al.*,1997; Alves *et al*., 2003; Nielsen dan Wege, 2006; Han *et al*., 2008).Naphthoquinone is known as antimicrobial, antifungal, antiviral, and antiparasitic which also have a bioactivity component that acts as anti-cancer and antioxidant. This compound is usually found in the vacuole cell in the form of glycosides (Robinson, 1995; Babula *et al.*, 2005).

There are two major types of soil in Central Kalimantan, among others, sandy mineral soil and peat soil.

However, the soil structure and soil fertility of this both types of soil are different. If it is viewed from the level of soil fertility, both types of soil have the equal opportunity to be used as a growing medium for Dayakonion.

Lingga and Marsono (2002) found that a sandy soil has some nutrient issues which are prone to rapid leaching. Soil porosity is very easy to infiltrate water nutrients into the deep ground so that plant nutrients are not covered by the roots. Meanwhile, according to Limin *et al.,* (2000), peat soil has low availability of macro and micronutrients and high level of soil acidity, high Cation Exchange Capacity (CEC), as well as low Base Saturation (BS).

Phytochemical screening results dayak onion plants contain alkaloids from Central Kalimantan (++++/strong positive once), steroids (++++/strong positive once), glycosides (++/positive), flavonoids (++/positive), phenolic (++/positive), tannins (++/positive) and saponins negative (-) (Galingging, 2006.) The ethanol extract bulbous wild onions growing Dayak origin Banjarbaru (South Kalimantan) have antioxidant activity with IC50 values of 25, 33 ppm (Kuntorini and Astuti, 2010) and antioxidant activity at age 6 mst stronger (IC50 = 93 ppm) compared to age 12 mst (IC50 = 50.42 ppm) but weaker than vitamin C (IC50 = 3.03 ppm) and BHT (IC50 = 5.52 ppm) as a control (Kuntorini, 2013).

This study is essential to be concerned because of both types of soil; there has been no research on the growth of Dayak onion as well as on the sandy mineral soil and peat soil in a specific location. To ensure that both types of land are suitable for the growth of Dayak onion, we need a study that determines the growth pattern of Dayak onion and the right time to harvest maximum tubers yield and to achieve high content of bioactive compounds.

Materials and methods

The research was conducted for 6months from March to September 2016 in Kalampangan Village, Sebangau Sub-District, Palangkaraya, Central Kalimantan Province Indonesia.

Tools and Materials

The materials used in this study include Dayak onion bulbs, sandy mineral soil, peat soil, and chicken manure fertilizer. Nevertheless, the tools used to support the study consist of *Screen house* with paranet, polybag, scales and analytics tools, camera, oven, GPS map 60 CSX, Delta-T.D Cambridge-England, soil thermometer, air thermometer, office stationery, and other equipment to support research activities.

Research design

The experimental design used is *completely randomized design* (CRD). The study was carried out in *Screen house,* Kalampangan Village, Sebangau Sub-District, Palangkaraya. Measurement of plant variables such as plant height, leaf number per panicle, the number of tillers per hill, tuberswet weight, and tubers dry weight. Otherwise, the analysis of bioactive compounds was done by qualitative test using Liquid Chromatograpy-Mass Spectroscopy (LC-MS) through1) Separation (extraction) that is based on the nature of *Naphthoquinone/Eleutherine* compound, 2) an approach (molecular weight, molecular structure) of compound identification which was done by ESI (Electro Spray Ionization) mode ±. All data from the observations were collaborated, processed, and displayed in graphs and images. The presented graphsare expected to give an idea of the growth patterns. The results of this research are also a part of the research steps such as the preparation of soil media (sandy mineral soil and peat soil), soil sifting process, soil transference into a polybag, the provision of basic fertilizer, maintenance, plants observation, and the analysis of bioactive compound.

Step-by-step preparation (qualitative test) using LC-MS are:

1) Sample weighted ± 0.5 grams of
2) added methanol + H20 (80: 20)
3) Added Buffer Ammonium 20 m mol format
4) Disonifikasi for 30 minutes
5) centrifuged at 4500 rpm for 10 minutes
6) Filtered with 0.2 µm
7) Diluted 10 times
8) analyzed using LC-MS

LC-MS are two tools that are combined into one, which serves to separate multiple compounds or mixtures of compounds based on polarity (the working principles of chromatography), after which a mixture of these compounds separately, then the pure compounds will be identified molecular weight. The data obtained is the molecular weight plus some of the charge and the molecular weight of the solvent.

As for how the liquid chromatography is the same as the HPLC or liquid chromatography another, are:

a. Analyte along with an eluent of a syringe pump or LC enter into capillary.in the Capillary contained anode (negative pole) on the Taylor cone and the cathode (negative pole) near the input of the analyte and eluent. Kutup function so that the charge gathered on Taylor cone is positively charged so that later on during a spraying and formed droplets (drops-drops) did not join - join into larger droplets.

b. Analyte and solvent (eluent) are sprayed through a Taylor cone.

Will form droplets - droplets which droplets - droplets that will undergo phase solvent evaporation to reduce solvent attached to the analyte. For a time, in the case of evaporation continuously then the solvent which includes the analyte trapped in positive charge excess, in English this stage is called the 'rayleigh' limit is Reached, there will be an explosion called coulomb explosion which will occur breakdown droplet (droplets) earlier. There are several possibilities on droplet - droplet, namely:

1) analyte will add one positive charge
2) analyte will add some positive charge
3) analyte will add one positive charge and one molecule of solvent
4) analyte will add one positive charge and some solvent molecules
5) analyte will add some positive charge and some solvent molecules.

c. Droplets experience explosion coulombs will be entered into the cone where the left and right already flowing nitrogen gas (N2).Gas function so that the analyte happened the last stable in shape and is not disturbed by the influence of oxygen gas. Droplet into the capillary transfer then is analyzed by the mass spectrometer.

The positive charge on the solvent derived from the ions Na+, Li+, K+, NH4+, and other actions. Because of the area Taylor cone in the capillary needle negatively charged, then the analyte in a solvent that has a positive charge will gather area. Taylorcone as a result, at the time of spraying droplets (droplet) a surface has a positive charge, and each droplet (droplet) is no longer stuck together (to form larger droplets). In the spectra of molecular weight gain is often the case these ions or molecules in addition to weight gain usually written with [M + molecular ions]. Possible molecular ions were detected in the mass spectroscopy is an [M + H+],[M - H+], as well as the analyte with extras such as Na+, K+, H3O+, NH4+, and the molecules of the mobile phase (Audrey, 2003).

Results and discussion

From this research, we can seethe growth pattern of Dayak onion on its characteristic and media both on sandy mineral soil and peat soil. The observation of the growth pattern starts at 8 weeks of DAS (days after sowing) where at this point the leaves and roots begin to appear. Then, a subsequent observation was carried out on 12, 16, 20, and 24 weeks of DAS.

Fig. 1. Dayak Onion (*Eleutherine palmifolia* Merr.)

Thus, the growth pattern of plant height, number of leaves, number of tillers, tubers wet weight, and tubers dry weight on sandy mineral soil also peat soil is presented in the following Figure 2.

The height growth pattern of Dayak onion that is planted in sandy mineral soil has experienced an increased from 8 weeks of DAS (33,4 cm), 12 weeks of DAS (40,7 cm), 16 weeks of DAS (47,7 cm), 20 weeks of DAS (52,6 cm), and 24 weeks of DAS (52,8 cm) with an increase of 7,3 cm, 7 cm, 4,9 cm, and 0,2 cm.

Moreover, an increase of plant height also occurred in the peat soil media from 8 weeks of DAS (3,0 cm), 12 weeks of DAS (6,3 cm), 16 weeks of DAS (9,5 cm), 20 weeks of DAS (13,5 cm), and 24 weeks of DAS (14,25 cm) by 3,3 cm, 3,2 cm, 4 cm, and 0,75 cm. From here, we can see that the height growth in sandy mineral soil is greater than the peat soil media. Since the beginning of 8 weeks of DAS, the growth pattern in sandy mineral soil is higher than the plants in peat soil media (Figure 2.).

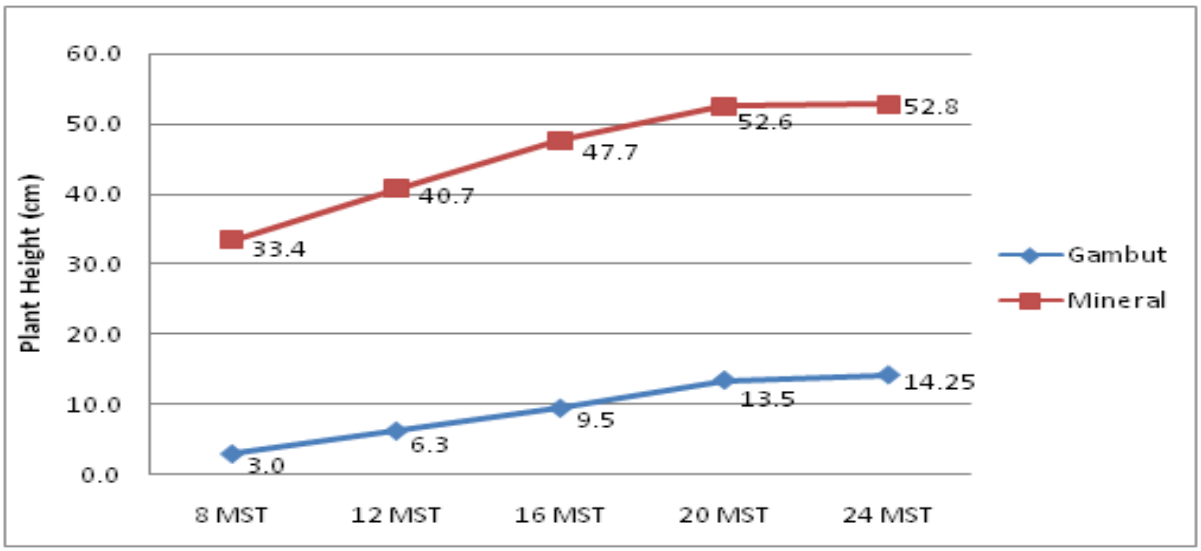

Fig. 2. The height growth pattern of Dayak onionat the age of 8, 12, 16, 20, and 24 weeks of DAS planted in mineral soil media and peat soil media.

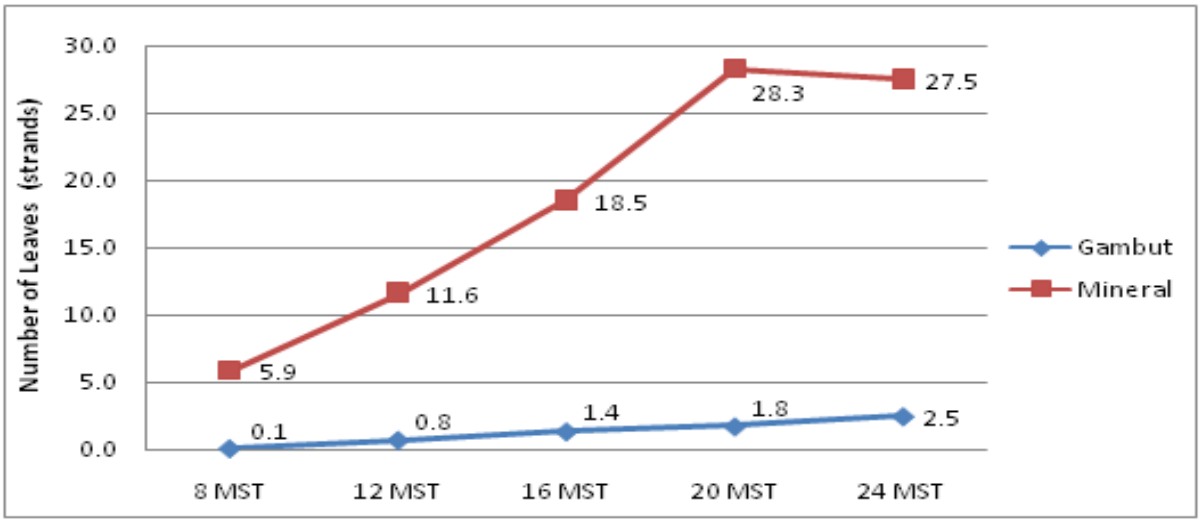

Fig. 3. The growth pattern in the number of leaves of Dayak onion at the age of 8, 12, 16, 20, and 24 weeks of DAS in sandy mineral soil media and peat soilmedia.

Likewise, in the number of leaves, the Dayak onion that is grown in sandy mineralsoil has more numbers than in peat soil (Graph 2). The growth pattern in the number of leaves on sandy mineral soil media is increased from the age of 8 weeks of DAS (5,9 leaves), 12 weeks of DAS (11,6 leaves), 16 weeks of DAS (18,5 leaves), and 20 weeks of DAS (28,3 leaves) and a decrease in the age of 24 weeks of DAS (27,5 leaves) with an increase of 5,7 leaves, 6,9 leaves, 9,8 leaves, and a decrease of 0,8 leaves on each period. While on the other hand, the growth pattern in peat soil media has increased at the age of 8 weeks of DAS (0,1 leaves), 12 weeks of DAS (0,8 leaves), 16 weeks of DAS (1,4 leaves), 20 weeks of DAS (1,8 leaves), and 24 weeks of DAS (2,5leaves) with an increase of 0,7 leaves, 0,6 leaves, 0,4 leaves, and 0,7 leaves.

The growth pattern in the number of tillers on sandy mineral soil media has experienced an increase since the age of 8 weeks of DAS (1,8 tillers), 12 weeks of DAS (5,3 tillers), 16 weeks of DAS (7,6 tillers), and 20 weeks of DAS (12,0 tillers), but not in the age of 24 weeks of DAS (12,0 tillers) with an increase of 3,5 tillers, 2,5 tillers, and 4,2 tillers.

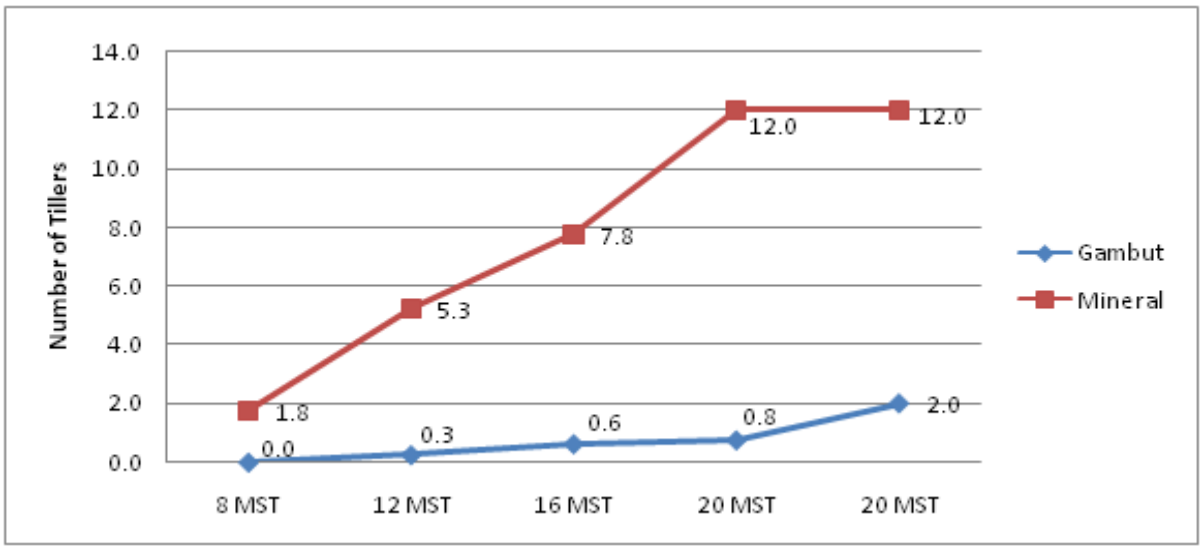

Fig. 4. The growth pattern in the number of tillers of Dayak onion at the age of 8, 12, 16, 20, and 24 weeks of DAS in sandy mineral soil media and peat soil media.

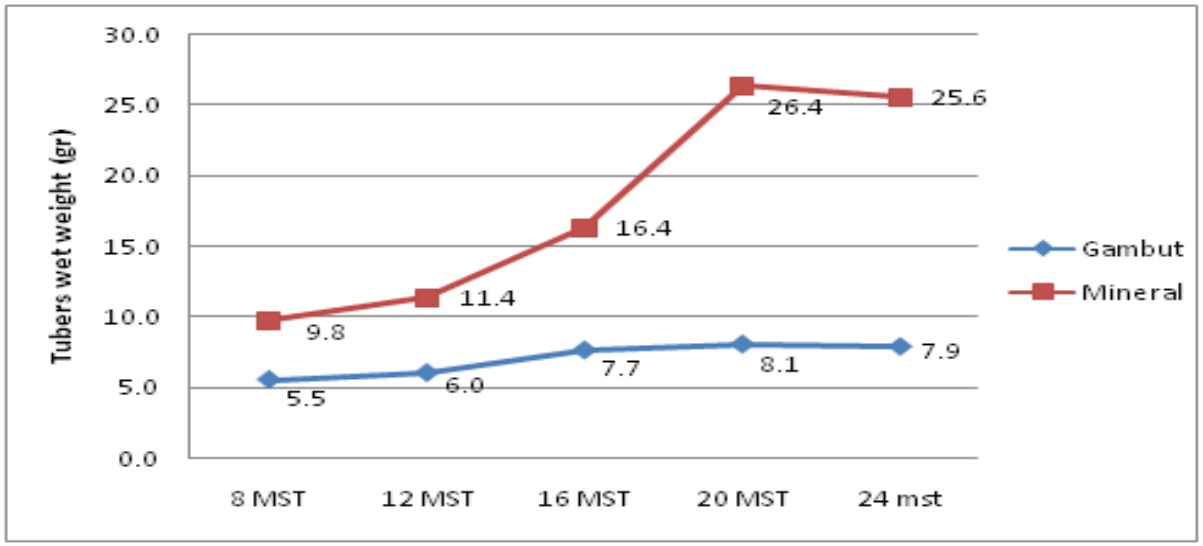

Fig. 5. The growth pattern in the tubers wet weight of Dayak onion at the age of 8, 12, 16, 20, and 24 weeks of DAS in sandy mineral soil media and peat soil media.

Different results occur on peat soil media that only encountered a low enhancement from 8 weeks of DAS (0,0 tillers), 12 weeks of DAS (0,3 tillers), 16 weeks of DAS (0,6 tillers), 20 weeks of DAS (0,8 tillers), and 24 weeks of DAS (2,0 tillers) with an increase of 0,3 tillers, 0,3 tillers, 0,2 tillers, and 1,2 tillers.

The growth pattern in the tubers wet weight on sandy mineral soil media has experienced a sigmoid increase from the age of 8 weeks of DAS (9,8 g), 12 weeks of DAS (11,4 g), 16 weeks of DAS (16,4 g), 20 weeks of DAS (26,4 g) while there is a slight decrease at the age of 24 weeks of DAS (25,5 g); each period has the increase of 1,6 g, 5 g, 10 g and also a decrease of 0,8 g. Whereas, the results from peat soil media show a weak number which are 8 weeks of DAS (5,5 g), 12 weeks of DAS (6,0g), 16 weeks of DAS (7,7 g), 20 weeks of DAS (8,1 g), and a decrease at the age of 24 weeks of DAS (7,9 g) with a respective increase of 0,5 g, 1,7 g, 0,4 g, and a decrease of 0,2 g.

The growth pattern in the tubers dry weight of Dayak onion in sandy mineral soil media is increased greatly from the age of 8 weeks of DAS (3,2 g), 12 weeks of

DAS (5,0 g), 16 weeks of DAS (7,2 g), 20 weeks of DAS (17,4 g), while there is a slight decrease at the age of 24 weeks of DAS (16,9 g) with each increase by 1,8 g, 2,2 g, 10,2 g and a decrease of 0,5 g.

There is also an increase in peat soil media at the age of 8 weeks of DAS (2,4 g),12 weeks of DAS (2,7 g), 16 weeks of DAS (3,3 g), 20 weeks of DAS (3,7 g), and a decrease at the age of 24 weeks of DAS (3,5 g) with such a consecutive increase of 0,3 g, 0,6 g, 0,4 g, and a decrease of 0,2 g.

As we can see from the Graph 6 above, the highest *Eleutherine* compound occurs at the age of 18 weeks of DAS and is higher on the sandy mineral soil than on the peat soil.

Fig. 6. The growth of tubers wet weight: a= peat soil media and b= sandy mineral soil media.

Discussion

Apparently, based on the presented graphs of plant height, the number of leaves, the number of tillers, tubers wet weight, tubers dry weight, and bioactive content, both soil types have equally met an increase but there is a huge difference. On the sandy mineral soil, the observation of all variables has generated much higher results than in peat soil in which this happens due to the diverse soil characteristics.

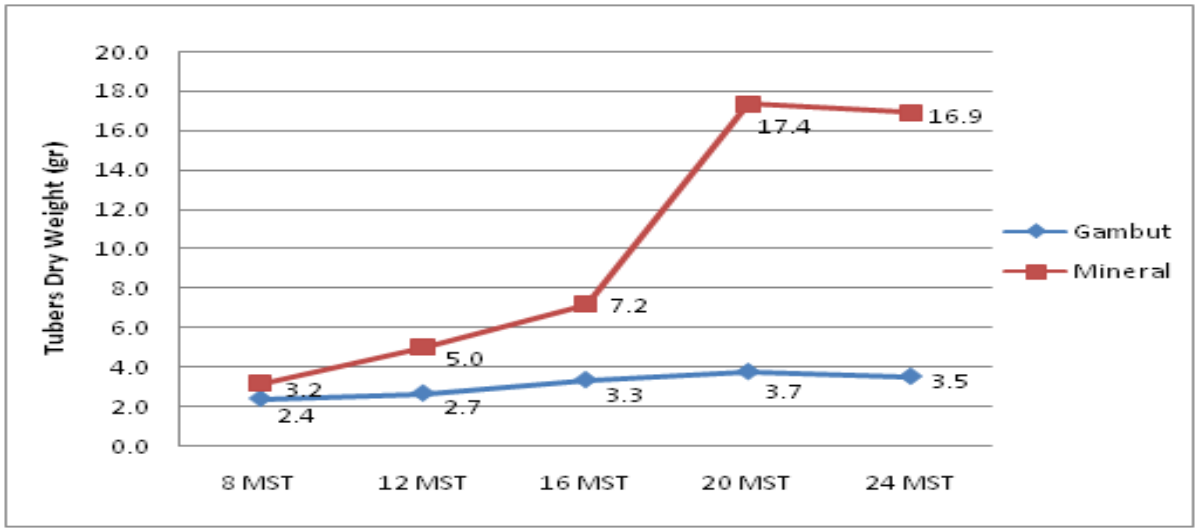

Fig. 7. The growth pattern in the tubers dries weight of Dayak onion at the age of 8, 12, 16, 20, and 24 weeks of DAS in sandy mineral soil media and peat soil media.

The differences in soil quality as a growing medium will affect the growth and development of the plants.

The growth and development of the plants are a process of life and propagation in which it depends on the results of assimilation, hormones, and growth substance and environment which support and generate growth pattern (Gardner, Pearce and Mitchell, 1991).

The results of the soil analysis show that the character of peat--dusty, clay, and loam textured soil contains the pH of H_2O by 4,1, Cation Exchange Capacity (CEC) by 74,56 me/100 g, Base Saturation (BS) by 11%, C-organic by 34,23%, C/N by 44, N-total by 0,78%, and P-Bray by 29,58 mg kg-1. Besides that, the loamy-sandy soil has the pH of H_2O by 4,8, CEC by 21,07 me/100 g, BS 22%, C-organic by 4,73%, C/N by 14, N-total by 0,33%, and P-Bray by 13,94 mg kg-1.

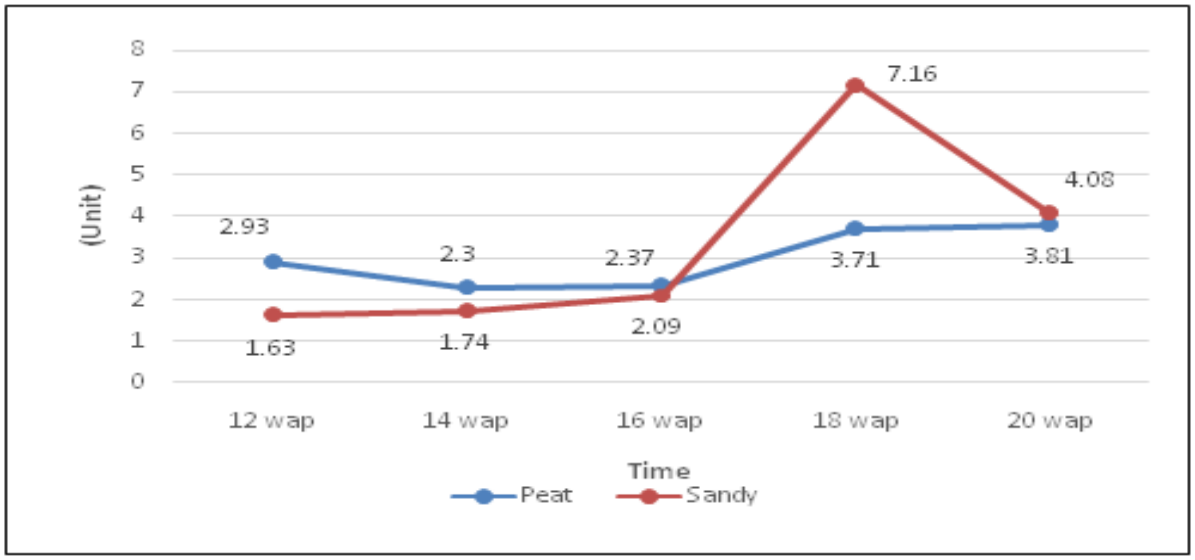

Fig. 8. The content of *Eleutherine* Compound of Dayak Onion.

The growth pattern in peat soil media points out that the growth and development of the plants are hampered in all variables.

This is most likely due to the low availability of macro and micro nutrients and the compounds that are toxic to the peat soil. High level of soil acidity, high CEC, and low BSare also an obstacle in this type of soil media (Limin *et al.,* 2000). By that, the plants could not grow optimally in which this can be seen from the stunted plant, a small number of leaves and tillers as well as low tubers wet weight and dry weight. Instead, the growth pattern of sandy mineral soil tends to be better than the peat soil because the sandy mineral soil has a pretty good aeration and drainage system. Plants will grow higher and the number of leaves and tillers will be optimal to capture the sunlight in order to get more energy supply (Hardjowigeno, 1995). More sunlight will enhance the growth of plant height, number of leaves, number of tillers, tubers wet weight, tubers dry weight as well as the production of Dayak onion bulbs. In addition, higher content of pH and better aeration system on the sandy mineral soil will generate a greater nutrient uptake.

Similarly, the bioactive compound on sandy mineral soil has a higher *Eleutherine*-compared to the peat soil which it mainly occurs at the age of 18 weeks of DAS. This is happened due to the differences in the characteristics and fertility rates of these soil types.

The synthesis of bioactive compounds in the plants is basically influenced by 1) the hereditary factors (genetic component), 2) the ontogeny factors (developmental stages), and 3) the environmental factors. Therefore, the environmental factors which affect the production of secondary metabolism include climate, growing area, plants environment, and cultivation methods (Robbers *et al.,* 1996).

Conclusion

Central Kalimantan has diverse soil characteristics and fertility rates. It is important to note that both sandy mineral soil and peat soil are likely to be used as a medium for Dayak onion cultivation. Sandy-mineral soil gives a high growth pattern of plant height, number of leaves, number of tillers, tubers wet weight, tubers dry weight, and better bioactive compounds than peat soil. Whereas, the highest growth pattern of tubers wet weight and tubers dry weight in this Dayak onion has happened at the age of 20 weeks of DAS while the highest bioactive compounds can be found in the *Eleutherine* content at the age of 18 weeks of DAS.

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