Activity of valerian catalase in Ultisol growing media by adding humic acid

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Abstract. Ultisols are soil in marginal lands which contain high Al and Fe, allowing the growing plants to have self-defense strategies by increasing catalase enzyme activity. The purpose of this study was to examine the response of valerian to Ultisol growing media by giving humic acid to the activity of the catalase enzyme, and to determine the potential planting medium to increase the activity of the catalase enzyme. The research method used was experimental with 2 factors of factorial completely randomized design (CRD). The first factor is the planting medium which consists of two media types namely (1) Ultisols and (2) Ultisols + organic matter. The second factor is the concentration of humic acid which consists of 4 treatment levels namely 0 g / kg; 4 g / kg; 8 g / kg and 12 g / kg. Of the two factors observed, 8 treatment combinations were produced, each with 3 replications. Parameters observed were catalase enzyme activity and growth parameters. As supporting data, the content of N, P, K, Ca, Mg, S, Mn, Fe, and Al in Ultisols have been analyzed. The results showed that the growing medium had an effect on the catalase enzyme activity. Catalase activity is influenced by the planting medium, plants that experience stress increase the catalase activity. Valerian grown on Ultisol growing media increased catalase activity, whereas those in Ultisol growing media with the addition of organic matter decreased catalase activity.

1. Introduction

Valerian (Valeriana officinalis L.) can be a source of antioxidants [1] because it contains several active compounds. Active compounds are products of secondary metabolites, including polyphenols and alkaloids which function as antioxidants. The content of secondary metabolites in medicinal plants reflects the quality of these plants. Therefore, studying the active compound content in valerian can contribute to information about these plants. Valerian has been shown to have sedative properties, treat insomnia [2] and contains essential oils as a raw material for perfume [3]. With the main content of valereic acid, which is an active compound belonging to the flavonoid group [4]. Antioxidants are molecules that can slow down or prevent the oxidation process of other molecules. The oxidation process can produce free radicals, thus triggering a chain reaction that can damage cells.

Studying the ability of valerian as a producer of antioxidants and a producer of medicinal raw materials, it is necessary to study its development in marginal soils such as ultisols which are acid soils with high Al and Fe content. This is a limiting factor for plant growth. The initial symptom of Al poisoning is inhibited root elongation, resulting in decreased absorption of water and minerals [5]. The physiological sign in response to Al stress is an imbalance in the production of reactive oxygen species (ROS) during metabolic processes in cells that induces antioxidant defenses. The antioxidant system in

plant cells provides protection against the toxic effects of ROS [6]. An important component of the protection system is enzymatic defense, such as catalase which can avoid O2- and H2O2 [7]. Al can produce superoxide radicals, resulting in an increase in lipids in the experiment using Allium cepa var. agrogarum L. [8].

Catalase enzymes include antioxidants that prevent oxidation which is a reaction process that can create free radicals. The enzyme catalase eliminates H2O2 by converting it to water molecules. An increase in stress levels in plants will usually be followed by an increase in antioxidant enzymes so that the analysis of a plant's tolerance to stress can be seen from the amount of the enzyme production. The purpose of this study was to examine the response of valerian to Ultisol growing media by giving humic acid to the activity of the catalase enzyme, and to determine the potential planting medium to increase the activity of the catalase enzyme.

2. Methodology

The research was conducted experimentally using polybags, with a height of 30 cm and a diameter of 25 cm. Planting the seeds is carried out directly in the media in polybags after being treated. Each polybag is planted with 1 plant. The experiment was carried out in the Unsoed Faculty of Biology greenhouse. The research materials used were valerian seeds, ultisols, organic fertilizers and humic acid. The tools used are: anlytic scales and a spectrophotometer. The parameters studied were plant growth and catalase activity.

The experiment was carried out in polybags containing 5 kg of planting medium. The planting media used were Ultisols, organic fertilizers and humic acid. The design used was a completely randomized design (CRD) with a two-factor factorial pattern. The first factor is the type of growing media which consists of 2 types, namely: Ultisols and Ultisols + Organic Materials (BO). The second factor is the concentration of humic acid which consists of 4 types, namely: 0 g / kg; 4 g / kg; 8 g / kg and 12 g / kg. From the two factors that were tested, there were 8 kinds of treatment combinations, each with 3 replications. Quantitative data analysis using Duncan's Multiple Range Test (DMRT)

2.1 Determination of catalase activity

Samples were pounded at 4°C in a porcelain mortar with a buffer (100 mM K2HPO4, pH = 7.0, 1 mM EDTA, 0.5 mM phenylmethylsulfonyl fluoride (PMSF), and 0.3% ethanol). Homogenization was carried out using 5 mL of buffer per g of fresh sample weight. The homogenate was centrifuged at 3300 rpm for 5 minutes at 4°C. The supernatant produced was used for the catalase activity test. As standard catalase consists of a mixture of 0.85 mL of sample; 0.05 mL homogenization buffer, and 0.1 mL 0.09 M H2O2. The absorbance value was measured at 253 nm in a Shimadzu UV-1601 spectrophotometer. The rate of reduction in H2O2 was converted to catalase units (U) from the standard catalase curve [9].

3. Result and Discussion

The root morphology of valerian as a response to growing media showed a different branching system. The ultisol planting medium caused the development of the root system to be imperfect compared to the development of the roots in the ultisol planting medium with the addition of organic matter (Figure 1). The same thing was stated that Al treatment on hydroponic medium inhibited root growth [10].

Figure 1. Valerian on ultisol (A) growing media and ultisol + organic matter (B) growing media

Morphologically, it appears that valerian grown on growing media with the addition of organic matter shows a more complete root system. It is assumed that the toxicity of Al in the form of Al3 + will be chelated by organic compounds so that it becomes neutral. The addition of organic matter also increases the growth of plant organs. In accordance with the statement [11], the combination of humic acid and nitrogen significantly increased root dry weight, increased total root length, root diameter, root surface area, and root volume.

The results of this study showed that the growing medium had an effect on catalase activity. Catalase is an enzyme involved in the degradation of hydrogen peroxide to water and oxygen and is the most effective antioxidant enzyme in the prevention of oxidative damage. The results of the analysis of the catalase enzyme activity on valerian grown on ultisols, due to Al stress in Table 1, show high catalase activity in ultisol growing media, and decreased activity in ultisol growing media with the addition of humic acid and organic matter.

Table 1. Valerian catalase activity in various growing media

Planting Media	Leaf Catalase (u/mg)	Root catalase (u/mg)
Ultisol + humic acid 0	0,81 a	2,18 a
Ultisol + humic acid 4g/kg	0,61 bc	0,81 bc
Ultisol + humic acid 8g/kg	0,54 de	0,66 cd
Ultisol + humic acid 12g/kg	0,51 e	0,66 cd
Ultisol BO + humic acid 0	0,66 b	0,93 d
Ultisol BO + humic acid 4g/kg	0,64 bc	0,61 cd
Ultisol BO + humic acid 8g/kg	0,59 cd	0,59 d
Ultisol BO + humic acid 12g/kg	0,49 e	0,52 d
10 P	0,00	0,00

Note: different letters in the same column indicate differences between treatments

The increase in catalase activity with the same pattern was also found in a study [12], which stated that the higher the Al concentration in the hydroponic medium could increase the catalase activity of Nigella arvensis. Furthermore, the activity of catalase and ascorbate peroxidase, glutathione reductase showed an increase due to stress conditions [13]. Catalase activity in planting media with the addition of organic matter is lower, because in the absence of stress, catalase activity occurs due to the differentiation process and se development. This is supported by research [14], which states that catalase activity is an indication of root cell differentiation and growth in experiments using Zea mays.

4. Conclusion

Catalase activity is influenced by the planting medium, plants that experience stress increase the catalase activity. Valerian grown on ultisol growing media increased catalase activity, while in ultisol growing media with the addition of organic matter it decreased catalase activity.

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