

# Ratio of Yeast Extract Administration on Induction in vitro planlet black orchid (*Coelogyne pandurata* lindl) From Kalimantan

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**Abstract.** Black Orchid (*Coelogyne pandurata*) is a flora from Kalimantan that has uniqueness and beauty on the tongue of its flowers. Plants multiply naturally, non-natural propagation has many obstacles, difficult crossing. Plant breeding efforts are required. Tissue culture techniques are expected to be carried out, several endogenous, exogenous factors and the addition of growth regulators (ZPT) are expected to be research efforts for the propagation of this orchid. Efforts to add yeast extract are expected to be a source of growth regulators, because yeast is rich in proteins, free amino acids, peptides, nucleotides, and complex vitamins. This study used the addition of Yeast Extract to the Induction of in Vitro Planlet Black Orchid (*Coelogyne pandurata* Lindl) 0.5 g / l, 1 g / l and 1.5 g / l. test analysis using SPSS with Kruskal-Wallis non-parametric test analysis, the results showed that the administration of yeast extract had a significant influence on the number of shoots and the number of roots. A concentration of 1 g/l has an influence on the number of shoots (4.33) and the number of leaves (8.00). A concentration of 1.5 g/l of yeast extract affects the number of roots (3.00).

Keywords : Yeast Extract, Black Orchid (Coelogyne pandurata), In Vitro culture.

## **1.Introduction**

The labellum on the black orchid flower which is black, is a uniqueness possessed by this type of Orchid. Flowers bloom around April-July in their natural habitat and smell good when blooming, the duration of blooming is about 5-6 days (Sasongko, 2019).

The uniqueness causes this orchid to be in great demand, however, it has a short flowering time and flowers are difficult to cross. The business of propagating black orchids has been carried out through conventional methods, but this method has its drawbacks. According to Saputri (2015) in Serliana et al., (2017) Conventional propagation of orchids takes a long time to acquire saplings and is limited. Conventional propagation efforts using seeds cannot be carried out, since orchid seeds do not have an endosperm and mutualism symbosis with mycorrhizal fungi is required. In the in vitro propagation of black orchids, growth regulators are expected to help, it is necessary to add hormones that can play an active role. ZPT can add synthetic and organic compounds as needed. Important components in the culture medium are vitamins, amino acids and complex organic substances. Complex organic substances play an important role in increasing plant growth, complex organic substances that have a relatively affordable price are yeast extracts (Nurilmala, 2018).

Yeast extract contains amino acids, vitamins and peptides. Vitamin yang terdapat dalam ekstrak ragi adalah thiamin, riboflavin, piridoksin, asam pantotenat dan niasin. as used as a complementary ingredient to in vitro culture media. Thiamin plays a very important role in in vitro culture, giving thiamin in small amounts can stimulate the growth of explants as well as increase root growth. Yeast extract also acts as a source of nitrogen which is influential in physiological processes, namely the formation of proteins, nucleic acids and coenzymes. Nitrogen is a macronutrient that is indispensable for plants, namely for the formation of



protoplasm and improving growth vegetative plants (Widiastoety & Kartikaningrum, 2003).

## 2. Ingredients and Methods

#### 2.1. Plant Material

Explants of black orchid planlets (C. pandurata) with a height of 1-1.5 cm, the number of leaves 2 strands and without roots, Baker's yeast, MS medium (Murashige & Skoog

#### 2.2 Media creation

The manufacture of the substrate is carried out by heating sterile aquades 500 mL, to which sugar is added as much as 30 grams homogeneous add instant MS media of 4.43 grams, and yeast extract according to treatment. Suffice it with the addition of sterile aquadest up to 1000 mL, take optimal pH measurements of 5.6-5.8.

Warm up using add agar 8 grams to the medium solution. The medium is poured into the culture bottle, carry out the autoclave sterilization process at a temperature of 121oC with a pressure of 1 atm, 30?. After the sterilization process is complete cool the culture bottles in the culture chamber

#### 2.3 Yeast Extract Solution

To obtain yeast extract is carried out dilution as much as 0.5 g, 1 g and 1.5 g in 5 mL of sterile aquadest. Each of these solutions is homogenized with a magnetic stirrer for 30? with a temperature of 70oC so that the yeast powder becomes homogeneous. The solution will form 2 layers (precipitate layer and supernatant solution). Next, a solution is taken supernatan and mixed in murashige & Skoog medium

#### **2.4 Initiation Treatment**

The explant is separated from the clump and from the planting medium, the planlet is then carried out the initiation of the planlet in the treatment medium. Each culture bottle contains one black orchid planlet. Storage of culture in an incubation planting room.

#### 2.5. Data Analysis

Using a Complete Randomized Design (RAL) on the administration of yeast extract 4 levels of concentration as follows: 0 g / L, 0.5 g / L, 1 g / L, 1.5 g / L with three repeats there were 12 experimental units, the SPSS test of normality and homogeneity and the analysis of the Kruskal-Wallis non-parametric test at a significance level of 5% to determine the influence between treatments

#### 3. Results

Concentration ratio of Yeast Extract at In Vitro Initiation of Black Orchid (C. pandurata)

The application of yeast extract has a significant effect on the average number of buds. The average number of shoots is highest in the treatment medium 1 g / l of yeast extract with an average number of shoots of 4.33 buds. The average number of black orchid leaves does not have a significant effect from the administration of yeast extract. The highest number of leaves is found at a concentration of 1 g / l with an average number of leaves. The best concentration of yeast extract

The average number of black orchid roots shows that the administration of yeast extract has a significant



effect, with the best concentration of yeast extract being found at a concentration of 1.5 g / 1, namely in the treatment of the average number of roots a total of 3 roots.

### **4.Discussion**

#### Number of shoots

The most bud growth is found in the yeast extract treatment with a concentration of 1 g / 1

**Table 1.** Average increase in the number of shoots of planlet culture Black Orchid (C. pandurata)

Concentration Yeast extract	Mean ±Standard error (SE)
0 (g/l)	$2,00 \pm 0,00^{a}$
0,5 (g/l)	$3,33\pm0,33^{\text{b}}$
1 (g/l)	$4,33 \pm 0,33^{b}$
1,5 (g/l)	$3,\!67\pm1,\!45^b$

**Caption** : Images followed by the same letter in the same column and row do not differ significantly based on the Mann-Whitney test at 5% significance (P?0.05).

Yeast contains amino acids, vitamins and proteins that are high in The mechanism of cell differentiation is that protein synthesis becomes faster and more optimal. Gansau et al., (2015) stated that the content of amino acids and vitamins contained in yeast extract also increases levels of endogenous hormones in plants such as IAA and GA3 by increasing hormone levels The endogenous then stimulates cell enlargement and differentiation, causing more shoots.

The results of this study are in accordance with the research of Safitri et al., (2013), namely the administration of yeast with a concentration of 8% gives the best results on the number of buds on the multiplication of mangosteen buds in vitro. In the treatment of 0.5 g / l of yeast extract) had the lowest average number of shoots, which was 3.33 buds. This indicates that a concentration of 0.5 g/l of yeast extract has not been able to induce the number of shoots properly. According to Tuhuteru et al., (2012) the response to explants depends on the explant's ability to absorb exogenous ZPT and balance with endogenous ZPT.

Number of leaves

The highest average number of leaves was found in the treatment of 1 g / l of yeast extract with 8 leaves

**Table 2**. Average increase in the number of leaves of planlet culture black orchid (c. pandurata)

Concentration Yeast extract	Mean ±Standard error (SE)
0 (g/l)	$4,67 \pm 0,67^{a}$
0,5 (g/l)	$7,67 \pm 0,33^{a}$
1 (g/l)	$8,00 \pm 0,5^{a}$
1,5 (g/l)	$7,67 \pm 0,33^{a}$

**Caption** : Images followed by the same letter in the same column and row do not differ significantly based on the Mann-Whitney test at 5% significance (P?0.05).



followed by a treatment of 0.5 g/l of yeast extract and a treatment of 1.5 g/l of yeast extract with an average leaf count of 7.67 strands. The best treatment according to research that has been carried out by Widiastoety & Kartikaningrum, (2003), Zulwanis et al., (2015) that the application of yeast extract in in vitro culture media with a concentration range of 1.00-1.75 g / l can produce the growth of the largest number of leaves in Dendrobium orchid planlets, and the application of yeast extract as much as 1.3 g / l in in in vitro culture media can increase the number of leaves in cattleya spp. orchid plants. Zunafika, (2019) in his research also stated that the addition of Yeast extract has a noticeable effect on the number of buds and leaves of banana explants with an average number of leaves of 6-17 strands, compared to other treatments without the addition of yeast extract.

Yeast extracts are rich in phytohormones such as cytokinins, amino acids, vitamins, enzymes and minerals that can stimulate cleavage and cell enlargement. Yeast excavation also contains nitrogen which plays a role in cell plasticity, so it can stimulate meristem cell division and cell enlargement. The activity of division of meristem cells will cause the occurrence of the formation of intervenous tissue on the leaves so that it can cause the area and number of leaves to increase (Widiastoety & Nurmalinda, 2010). Saepudin et al., (2020) stated that the parameter of the number of buds is closely related to the number of leaves, the more shoots that appear, the number of leaves will increase. The greater number of leaves on the plant can be interpreted that it is due to the good growth of shoots. As the number of leaves increases, the amount of chlorophyll levels will increase and the photosynthesis process will become larger.

#### Number of roots

The treatment on the best root number parameter is found in the treatment of 1.5 g / 1 of yeast extract with an average number of roots, namely 3 roots, in the treatment of 0.5 g / 1 of yeast extract) has the lowest average number of roots, namely 1.33 roots.

Concentration Yeast extract	Mean ±Standard error (SE)
0 (g/l)	$2,00 \pm 0,00^{a}$
0,5 (g/l)	$3,33 \pm 0,33^{b}$
1 (g/l)	$4,33 \pm 0,33^{b}$
1,5 (g/l)	$3,67 \pm 1,45^{b}$

Table 3. Average increase in the number of roots of planlet culture Black Orchid (C. pandurata)

**Caption** : Images followed by the same letter in the same column and row do not differ significantly based on the Mann-Whitney test at 5% significance (P?0.05).

This suggests that the higher the concentration of yeast extract given, the more it will increase the average number of black orchid roots. Santi et al., (2011) that in the yeast extract contained various nutrients such as proteins and vitamins. A source of vitamins that can stimulate root growth is vitamin B1. The study also mentioned that the administration of yeast extract to Vanda orchids had a noticeable influence on the parameters of the number and length of roots at a concentration of 1.5 g / 1. The physiological conditions of plants as well as different types of species can give different responses to each treatment. According to Hartati (2009) in Rahayu et al., (2021) states that root growth is supported by the supply of organic matter necessary to stimulate the increase in the number of roots. Giving yeast extract can also increase the content of endogenous IAA hormone in plants, high IAA levels can build resistance and increase root suction power



for the better. This further increases the nutrients that will be absorbed by the roots and causes the root growth to increase. Root growth occurs due to the cooperation between the hormone auxin and also cytokinins. Yeast extract has an important role, namely increasing the content of endogenous cytokinins and auxins (Rupawan et al., 2014). In this study, yeast extract added to in vitro culture media has succeeded in providing the highest average number of roots, it is because endogenous auxin in plants has been able to stimulate root growth. Rupawan et al., (2014) stated that the mechanism of auxin hormone can increase cell permeability by increasing pressure, developing cell walls and increasing the diffusion process so that water can enter the cells, and making it easier for cytokinins to carry out cell division.

Administration of ZPT in in vitro cultures in certain doses can stimulate growth, however, is an inhibitor if used in excess of the optimum dose. The parameter of the number of roots in the growth of in vitro cultures indicates that the explants are in healthy condition and are able to optimally absorb nutrients from the medium. The more roots that grow, the wider the range of plants and nutrients absorbed. According to Saepudin et al., (2020) root growth is also also t root growth also depends on nutrients found in media such as phosphorus, calcium, manganese and iron. The thiamin element contained in MS media can play a role in accelerating cell division activities in the root meristem and act as a coenzyme in reactions that produce energy and carbohydrates..

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