

# THE EFFECTS FERMENTED NATURAL PLANT GROWTH REGULATOR ON PRANCAK 95 TOBACCO (*NICOTIANA TABACUM* L. VAR PRANCAK 95) ACCLIMATIZATION

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**Abstract.** Prancak 95 tobacco has the potential to be developed in Indonesia because it has a distinctive aroma and a nicotine content of 2.13%. The quality of this tobacco raw material must be improved and started from the use of quality seeds through in vitro culture techniques. However, several things that must be considered are the acclimatization stages. One of the efforts to increase the growth of seedlings during the acclimation can be done by providing natural plant growth regulators (PGR) from extracts of organic materials such as coconut water, young corn, sprouts, and banana weevil that has been fermented in advance using EM-4 bioactivators. Based on this, the purpose of this study was to determine the optimal concentration of natural growth regulators in the acclimatization of Prancak -95 tobacco. This study uses a non factorial randomized block design with four treatments concentrated solution of natural growth regulators, which include: Control (no treatment), 10; 25; and 50 ml/L. The observed variables included the number of leaves, leaf area, plant height, and root length. Data analysis using Analysis of Variance (ANOVA) Test and continued with Duncan's Multiple Range Test (DMRT) Test at 95% confidence level to find out the best treatment. Based on the results of research that has been carried out for 60 days of observation, it can be concluded that the application of natural PGR is able to spur the growth of seedlings during the acclimatization period. Spraying application at a concentration of 10 ml / L can increase the number of leaves, leaf area, number of roots and height of Prancak 95 tobacco seedlings.

## 1. Introduction

The development of Indonesian tobacco is currently focused on local tobacco, and one of them is Prancak 95 variety. This tobacco originating from Madura has a distinctive aroma and is in demand by several large cigarette companies in Indonesia. Efforts to improve the quality of tobacco raw materials begin with the use of quality seeds. At present, tobacco tissue culture techniques have the potential to be developed because they can produce uniform seeds in a relatively short time [1].

The acclimation stage is the final stage which is relatively difficult because the seeds from the bottle must be adapted to the outside environment. While in vitro conditions, plantlets are accustomed to optimal conditions and the influence of growth regulators which can cause some changes in plant morphology, anatomy and physiology. Therefore, the most important thing to consider in the acclimation stage is adaptation to growth regulators, microclimates, and nutrients so that plantlets can grow and develop in ex vitro conditions [2].

One effort to increase the growth of seedlings can be done by providing growth regulators. However, growth regulators in the market have relatively expensive prices and are difficult to obtain. An alternative to replacing growth regulators is to use a fermentation solution of organic ingredients such as coconut water, young corn, sprouts, and banana weevil. These ingredients contain natural hormones such as auxin, cytokinins, and gibberellins. Based on several studies that have been done, Anwarudin (2017) states that banana weevil contains 1-Naphthaleneacetic Acid (NAA) compounds. Coconut water contains cytokinin and auxin hormones auksin [4]. Related to testing on plant growth, Admaja,

Sulistyowati, & Sarbino, (2015), stated that the use of natural PGR of 15 ml / L was able to stimulate the growth of rubber plants originating from sleeping eye stumps. In addition, Kaffi (2017) also stated that the administration of young corn extract spurred the growth of Nuda Indah plant cuttings. The use of banana weevil extract and coconut water has also been proven to be able to spur the growth of Kemiri Sunan seedlings [7]. Based on this, research is needed to find out some optimal natural PGR concentrations in the acclimatization of Prancak -95 tobacco.

## 2. Materials and Methods

This study was conducted in the greenhouse of tissue culture laboratory Politeknik Negeri Jember for 60 days. Plant materials using tobacco plants acclimatized 1 month old. Organic materials such as coconut water, young corn, bean sprouts and banana weevil are used as a source of natural PGR. The material is fermented using EM4 decomposers for 1 month. This study uses a non factorial randomized block design with four treatments of the concentration of natural PGR solution, which includes: Control (without natural PGR), 10 ml / L; 25 ml / L; 50 ml / L. The solution was applied once a week through the leaking method (200ml / plant). The observed variables included the number of leaves, leaf area, plant height, and root length. Data analysis using ANOVA Test and continued with DMRT Test at 95% confidence level to find out the best treatment.

## 3. Results and discussion

### 3.1 Number and Leaf Area

The results of analysis of variance in the parameters of the number and area of leaves showed that the application of natural PGR was very significant (Table 1). In addition, according to the data also shows that the application of organic PGR at different concentrations will produce different numbers of leaves. Seedlings that are not applied with natural PGR have the fewest number of leaves compared to seeds that are applied.

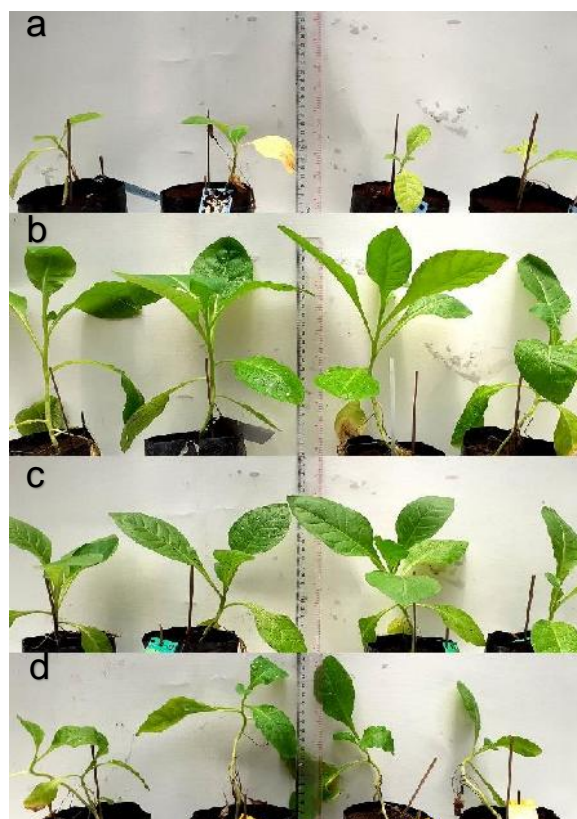
**Table 1.** The effect of some natural PGR concentrations on the number and extent of Prancak 95 tobacco seedlings aged (60 DAP).

Concentrations of natural PGR	Leaf Number	Leaf Area (cm <sup>2</sup> )
0ml/L	2,75 a	9,79 a
10ml/L	8,25 b	42,91 c
25ml/L	6,75 b	37,51 c
50ml/L	4,25 a	19,84 b

<sup>a</sup> The number followed by the same letter are not significantly different at ( $p < 0.05$ ) level of Duncan's test.

Based on result, concentration of 10 ml / L showed the best results in increasing the number of leaves with an average of 8.25 leaves per seed. The higher the concentration of natural PGR was seen to suppress leaf formation, as in the concentration of 50 ml / L the average leaves formed were 4.25. The results of leaf area in **Table 1.** show that the widest leaves, 42.91 cm<sup>2</sup>, were produced from seedlings that were applied with 10 ml / L natural ZPT. The results were also not significantly different from the concentration of 25 ml / L which had an average leaf area of 37.51 cm<sup>2</sup>. The leaf area decreases at a concentration of 50 ml / L.

Natural PGR gives a very significant effect on seedling growth when compared to control treatment or without application (**Figure 1**). This shows that natural PGR is able to influence the growth of tobacco seedling leaves during the acclimatization period. Natural PGR given contains cytokines in the form of kinetin and zeatin. Cytokinins play an important role in cell division, spur the formation of shoots, leaf enlargement and delay senescence in leaves [8], [9] [10]. The level of concentration affects the ability of hormones to regulate the growth of tobacco seed leaves as in the results of the data in Table 1. The lower the natural PGR concentration shows the best results on the number and extent of leaves.



**Figure 1** Growth response of Tobacco-95 Seedlings on 60 DAP a) No natural PGR (Control); b) Treatment of 10 ml / L; c) 25 ml / L; d) 50 ml / L

### 3.2 Plant Height and Length of Root

The results of analysis of variance on plant height and root length parameters have a very significant effect (**Table 2**). This shows that the treatment of several natural ZPT concentrations stimulates stem height. Application of 10 ml / L was proven to stimulate the growth of seedling stems to be the highest at 15.93 cm. The concentration was not significantly different from the treatment of 25 ml / L (14.15 cm). The 50ml / L treatment had shorter shoots of 8.9 cm and was not significantly different from seedling height without natural ZPT treatment which was 6.7 cm.

**Table 2.** Effect of some natural PGR concentrations on plant height and root length on Prancak 95 tobacco seedlings (60 DAP)

Concentrations of natural PGR	Plant height (cm)	Root Length (cm)
0ml/L	6,70 a	13,58 a
10ml/L	15,93 b	21,00 b
25ml/L	14,15 b	18,90 ab
50ml/L	8,90 a	18,27 ab

<sup>a</sup> The number followed by the same letter are not significantly different at ( $p < 0.05$ ) level of Duncan's test.

The balance of natural PGR concentration in the application of 10 ml / L is thought to spur the growth of Prancak 95. Seedlings at a concentration of 10 ml / L proven to be more able to spur stem height and root length than other concentrations. According to Tanimoto & Hirano (2013) gibberellins will play a role in root growth if at low concentrations and at high concentrations it will suppress root lengthening. As the results of observations on the root length in **Table 2**. shows that the treatment of 10 ml / L gives the best results on the root length that is 21,00 cm. But these results were not significantly

different from the treatment of 25ml / L and 50 ml / L. Whereas the shortest root (13,58 cm) was found in the control treatment or without the application of natural PGR.

Natural PGR is made containing auxin hormone (IAA) and gibberellins (GA3) which play a role in the elongation of stems and roots [12]. Auxin plays an important role in apical dominance and root formation. The length of the stem is also affected by the enlargement and elongation of cells by auxin. In influencing cell enlargement and elongation, auxin is able to increase osmotic pressure, and increase cell permeability to water, thus spurring the diffusion of water into cells so that cells will increase in size [13].

#### 4. Conclusions

Based on the results of research that has been done, it can be concluded that the application of natural PGR is able to spur the growth of seedlings during the acclimatization period. Spraying application at a concentration of 10 ml / L can increase the number of leaves, leaf area, number of roots and height of Prancak-95 tobacco seedlings.

#### 5. Acknowledgements

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#### 6. References

- [1] N. Kumar and M. Reddy, "In vitro Plant Propagation: A Review," *J. For. Environ. Sci.*, vol. 27, 2011.
- [2] J. Pospíšilová, I. Tichá, P. Kadleček, D. Haisel, and Š. Plzáková, "Acclimatization of micropropagated plants to ex vitro conditions," *Biol. Plant.*, vol. 42, no. 4, pp. 481–497, 1999.
- [3] S. Anwarudin, "Uji Keberadaan Zat Pengatur Tumbuh (ZPT) Organik Auksin dari Tauge dan Bonggol Pisang yang telah Difermentasi Menggunakan MOL, EM-4, dan PGPR dengan Metode HPLC," Universitas Islam Negeri Sunan Kalijaga Yogyakarta, 2017.
- [4] J. W. H. Yong, L. Ge, Y. F. Ng, and S. N. Tan, "The chemical composition and biological properties of coconut (*Cocos nucifera* L.) water," *Molecules*, vol. 14, no. 12, pp. 5144–5164, 2009.
- [5] W. Admaja, H. Sulistyowati, and S. Sarbino, "Pengaruh Campuran Hormon Organik dan Pupuk Organik Cair terhadap Peningkatan Daya Tumbuh Bibit Stum Mata Tidur Tanaman Karet," *Perkeb. dan Lahan Trop.*, vol. 4, no. 2, pp. 18–21, 2015.
- [6] U. Kaffi, "Uji Efektifitas Pertumbuhan Vegetatif Bunga Nusa Indah (*Mussaenda Pubescens*) Terhadap Pemberian ZPT Organik Jagung Muda Pada Berbagai Sumber Setek," *Agrovital*, vol. 2, no. 2, 2017.
- [7] F. Kurniati, T. Sudartini, and D. Hidayat, "Aplikasi berbagai bahan ZPT alami untuk meningkatkan pertumbuhan bibit kemiri sunan (*Reutealis trisperma* (Blanco) Airy Shaw)," *J. Agro*, vol. 4, no. 1, pp. 40–49, 2017.
- [8] P. J. Davies, "The plant hormones: their nature, occurrence, and functions," in *Plant hormones*, Springer, 2010, pp. 1–15.
- [9] J.-M. Davière and P. Achard, "Organ communication: Cytokinins on the move," *Nat. plants*, vol. 3, no. 8, p. 17116, 2017.
- [10] J. Skalák *et al.*, "Multifaceted activity of cytokinin in leaf development shapes its size and structure in *Arabidopsis*," *Plant J.*, vol. 97, no. 5, pp. 805–824, 2019.
- [11] E. Tanimoto and K. Hirano, "Role of Gibberellins in Root Growth," in *Plant Roots: The Hidden Half, Fourth Edition*, 2013, pp. 11–13.
- [12] E. Tanimoto, "Regulation of Root Growth by Plant Hormones—Roles for Auxin and Gibberellin," *CRC. Crit. Rev. Plant Sci.*, vol. 24, no. 4, pp. 249–265, 2005.
- [13] K. Takahashi and T. Kinoshita, "The regulation of plant cell expansion: auxin-induced turgor-driven cell elongation," *Mol. cell Biol. growth Differ. plant cells*, pp. 156–173, 2016.