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Original research article

Adaptive Responses and Differences in Morphophysiological Resistance of *Dendrobium* sp., *Phalaenopsis* sp., and *Vanda* sp. Orchids to Abiotic Stress

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Abstract

Orchids are an important horticultural commodity in Indonesia with high economic and conservation value; however, their sustainability is threatened by abiotic stress, such as salinity. This study aims to evaluate the resistance of Dendrobium sp., Phalaenopsis sp., and Vanda sp. to NaCl-induced salinity stress using a completely randomized design (CRD) with three replications. The treatments (0%, 1%, 5%, and 10% NaCl) were applied every three days for five weeks under greenhouse conditions. Morphological responses were observed visually, while chlorophyll content (mg/L) was measured spectrophotometrically at 649 and 665 nm. The results showed that chlorophyll content decreased significantly with increasing NaCl concentration. Total chlorophyll of Phalaenopsis sp. decreased sharply with increasing NaCl concentration from 5.25 mg/L (control), 3.78 mg/L (1%), 1.74 mg/L (5%) and 0.54 mg/L (10%); In contrast, Dendrobium sp. showed fluctuations from 5.02 mg/L (control), 8.20 mg/L (1%), 3.74 mg/L (5%) and 4.05 mg/L (10%). While Vanda sp. experienced a moderate decrease from 9.42 mg/L (control), 9.41 mg/L (1%), 4.53 mg/L (5%), and 4.26 mg/L (10%). Vanda sp. showed the highest salinity tolerance, followed by Dendrobium sp., while Phalaenopsis sp. was the most susceptible. NaCl stress significantly reduced chlorophyll content in all orchids.

1. INTRODUCTION

Orchids are a horticultural commodity with high economic value [1]. Market demand for orchids continues to increase year after year [2]. This situation has driven massive exploitation of orchids and their rapid propagation through intensive cultivation [3]. However, in practice, orchid cultivation often faces challenges including drought [4], salinity [5], and changes in temperature and climate [6]. These factors can inhibit plant growth and development, resulting in serious economic losses.

Abiotic stress has been shown to affect plant growth and the morphological, physiological, and genomic characteristics of orchids both *in vitro* and *in vivo* [3; 7; 8; 9]. Among abiotic stressors, salinity has emerged as a major constraint on plant productivity worldwide. Salinity stress, primarily caused by the accumulation of sodium chloride (NaCl), disrupts plant physiological and metabolic processes through osmotic stress, ion toxicity, and oxidative damage [10]. These effects lead to reduced water uptake, impaired photosynthesis, including photosynthetic pigment levels, particularly chlorophyll, and ultimately decreased plant growth and yield [11], [12], [13]. Therefore, resistance to abiotic stress (salinity stressors) is key to developing superior orchid varieties [14].

In addition to physiological factors, exposure to abiotic stress affects plant morphology. Morphological responses to abiotic stress are associated with the accumulation of phenolic compounds, which act as secondary metabolites involved in plant defence mechanisms. These compounds play a role in mitigating oxidative stress and maintaining cellular integrity, thereby influencing visible morphological changes such as leaf chlorosis, necrosis, and growth inhibition [9]. The ability to produce secondary metabolites varies among species [15] and can serve as a natural plant defense [16].

Identifying morphological and physiological responses to abiotic stress is crucial as a basis for developing conservation and plant breeding strategies. Studies on the effects of stress on orchid varieties are still very limited [17], particularly the comparison of morphological and physiological responses of three native Indonesian orchid varieties to abiotic stress. The selection of *Dendrobium*, *Phalaenopsis*, and *Vanda* in this study is based on their ecological diversity, physiological variation, and economic importance. Collectively, these three genera represent a gradient of adaptive capacity from tolerant, and intermediate to sensitive. This variation provides a strong scientific basis for comparative analysis aimed at identifying key traits associated with salinity tolerance. This study aimed to determine the morphological and physiological responses of *Dendrobium* sp., *Phalaenopsis* sp., and *Vanda* sp. orchid varieties to NaCl abiotic stress and to determine the orchid varieties with the best resistance characteristics to NaCl abiotic stress.

2. MATERIALS AND METHODS

This study used an experimental approach with a completely randomized design (CRD) to test orchid resistance to abiotic stress. The study was conducted in a greenhouse and biology laboratory. Samples included *Dendrobium* sp., *Phalaenopsis* sp., and *Vanda* sp., with the independent variable being NaCl abiotic stress and the dependent variable being the plant's resistance level based on morphological and physiological data.

Abiotic Stress Injection

The abiotic stress treatment consisted of injections of NaCl solutions (0%, 1%, 5%, and 10%) every three days for five weeks using a root wounding method using a syringe. A fixed volume of NaCl solution (1 mL per plant) was injected into the root zone to ensure uniform exposure. The root-wounding injection method was selected to facilitate direct uptake of the saline solution and to simulate localized salinity stress conditions, thereby enhancing the consistency and effectiveness of stress induction.

Morphological Data Collection and Chlorophyll Testing

Research documentation was conducted to obtain morphological data and compare the NaCl injection treatment with the control. The chlorophyll test was conducted by grinding 0.5 g of leaves with 10 ml of 96% ethanol until the leaves were pale and the solvent green. The extract solution was then filtered through filter paper and placed in a dark vial. The solution was transferred to a cuvette up to the mark and then read using the absorbance at 649 nm and 665 nm. The chlorophyll a and b levels, as well as total chlorophyll (mg/L) were calculated using the following formula [18]:

$$Chl\ a\ (mg/L) = (13.7 \times A_{665}) - (5.76 \times A_{649})$$

$$Chl\ b\ (mg/L) = (25.80 \times A_{649}) - (7.60 \times A_{665})$$

$$Total\ chlorophyll\ (Chl\ a + Chl\ b)\ (mg/L) = (6.10 \times A_{665} + 20.04 \times A_{649})$$

Data Analysis

The measurement data were analyzed quantitatively using Microsoft Excel software, including the calculation of mean values and standard deviation to evaluate data variability, and were subsequently presented in tables and graphical forms. The standard deviation presented reflects variability among treatment levels.

3. RESULTS

The responses of Dendrobium sp., Phalaenopsis sp., and Vanda sp. to NaCl salinity stress varied significantly, both in terms of chlorophyll content and morphological changes (Table 1; Figure 1). The total chlorophyll content of Phalaenopsis sp. decreased drastically with increasing NaCl concentration. In the control, the chlorophyll content was 5.25 mg/L, which decreased progressively to 3.78 mg/L (1% NaCl), 1.74 mg/L (5% NaCl), and 0.54 mg/L (10% NaCl). This reduction in chlorophyll content was accompanied by observable morphological changes, including leaf chlorosis, necrotic lesions along the leaf margins, and root browning.

A slightly different trend was observed for Dendrobium sp. Chlorophyll content increased at 1% NaCl (8.20 mg/L) compared to the control (5.02 mg/L), indicating a stimulatory effect at low salinity levels. However, at higher salinity levels, chlorophyll decreased to 3.74 mg/L (5% NaCl) and 4.05 mg/L (10% NaCl). Morphological observations revealed only mild necrotic spots on the leaves and reduced shoot elongation. These results indicate that Dendrobium sp. has a moderate level of salinity tolerance, maintaining pigment stability at lower salt concentrations but being susceptible at higher levels.

Table 1. Data on chlorophyll a and b levels and total chlorophyll (mg/L)

Sample Name	A_{649} *(Sample absorbance)*	A_{665} *(Sample absorbance)*	Chl. a content (mg/L)	Chl. b content (mg/L)	Total Chlorophyll (mg/L)
Dendrobium sp. Kontrol	0,16	0,31	3,33	1,69	$5{,}02 \pm 2{,}04^b$
Dendrobium sp. - NaCl 1%	0,25	0,52	5,75	2,45	$8{,}20 \pm 2{,}04^a$
Dendrobium sp. - NaCl 5%	0,12	0,24	2,58	1,16	$3{,}74 \pm 2{,}04^b$
Dendrobium sp. - NaCl 10%	0,12	0,26	2,89	1,15	$4{,}05 \pm 2{,}04^b$
Phalaenopsis sp. Kontrol	0,17	0,32	3,37	1,88	$5{,}25 \pm 2{,}10^a$
Phalaenopsis sp. - NaCl 1%	0,12	0,22	2,30	1,48	$3{,}78 \pm 2{,}10^b$
Phalaenopsis sp. - NaCl 5%	0,05	0,12	1,36	0,38	$1{,}74 \pm 2{,}10^c$
Phalaenopsis sp. - NaCl 10%	0,02	0,02	0,18	0,36	$0{,}54 \pm 2{,}10^d$
Vanda sp. Kontrol	0,29	0,58	6,32	3,09	$9{,}42 \pm 2{,}90^a$
Vanda sp. - NaCl 1%	0,29	0,58	6,28	3,13	$9{,}41 \pm 2{,}90^a$
Vanda sp. - NaCl 5%	0,14	0,29	3,22	1,31	$4{,}53 \pm 2{,}90^b$
Vanda sp. - NaCl 10%	0,13	0,27	2,94	1,31	$4{,}26 \pm 2{,}90^b$

Note: Data are presented as mean ± standard deviation. Different superscript letters in the same column indicate significant differences at $p < 0.05$

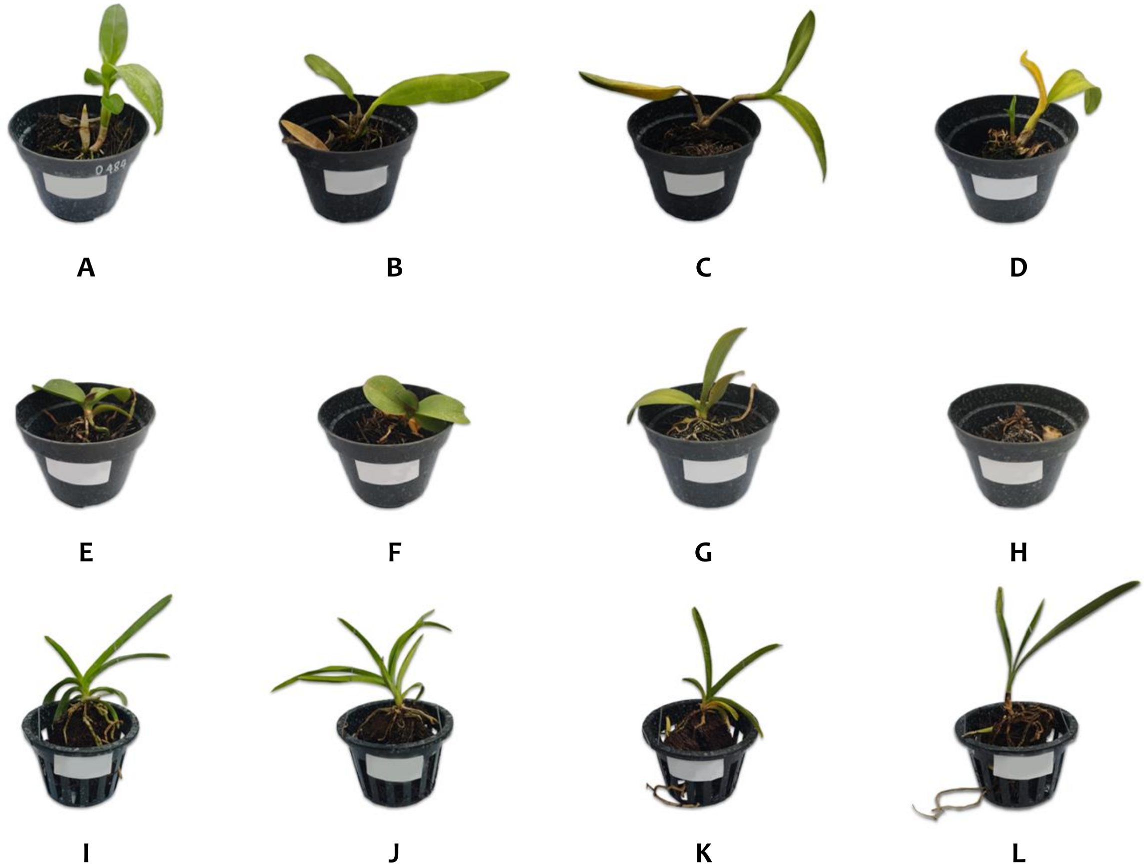

Figure 1. Morphology of the orchid after salinity stressors. *Dendrobium* sp. showed moderate tolerance. (A-D), *Phalaenopsis* sp. was the most sensitive (E-H), and *Vanda* sp. showed the highest tolerance (I-L) treated with 1% NaCl (B,F,J), 5% NaCl (C,G,K), 10% NaCl (D,H,L) compared with the control (A,E,I)

The chlorophyll content of *Vanda* sp. remained virtually unchanged between the control (9.42 mg/L) and 1% NaCl (9.41 mg/L), indicating no negative impact at low salinity. At 5% and 10% NaCl, chlorophyll content decreased to 4.53 mg/L and 4.26 mg/L, respectively, indicating moderate but manageable pigment reduction. Morphological symptoms were relatively mild, consisting of leaf yellowing limited to older leaves without severe necrosis. Compared with other genera, *Vanda* sp. showed the strongest tolerance to salinity. Based on chlorophyll content and morphological response, the order of relative tolerance to salinity was *Vanda* sp. > *Dendrobium* sp. > *Phalaenopsis* sp

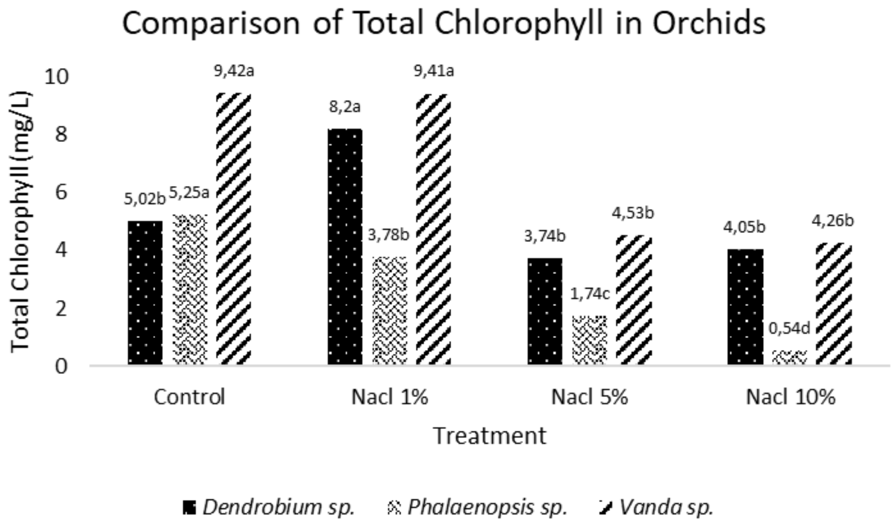

Figure 2. Comparison of total chlorophyll in *Dendrobium* sp., *Phalaenopsis* sp., and *Vanda* sp. orchids treated with several different NaCl concentrations

4. Discussion

The results clearly show that salinity stress, caused by NaCl, significantly reduces chlorophyll content and causes morphological damage in orchids, but the tolerance level varies across genera. The drastic decrease in chlorophyll in *Phalaenopsis* sp. indicates its high sensitivity to salt stress. A decrease from 5.25 mg/L (control) to 0.54 mg/L (10% NaCl) is associated with severe chlorosis and necrosis. This indicates that *Phalaenopsis* sp. is unable to regulate ion uptake or maintain osmotic balance under salinity stress. Salinity is known to disrupt chloroplast ultrastructure, increase the accumulation of reactive oxygen species (ROS), and inhibit the chlorophyll biosynthesis pathway [3; 8]. *Phalaenopsis* sp. showed greater sensitivity to salinity stress, which may be associated with less effective physiological adaptation mechanisms compared to more tolerant orchid genera.

In contrast, *Dendrobium* sp. showed an increase in chlorophyll content under 1% NaCl. This response is likely due to hormesis, where low levels of stress stimulate metabolic activity, including increased chlorophyll biosynthesis and antioxidant defenses. Several studies have reported similar findings in epiphytic and halophytic orchids, where mild stress can activate the production of osmolytes (e.g., proline, glycine betaine) and ROS-scavenging enzymes, thereby enhancing short-term photosynthetic performance [19; 20]. However, at higher salt concentrations (5–10%), the protective mechanisms of *Dendrobium* sp. appear inadequate, leading to chlorophyll degradation and mild necrotic symptoms.

Vanda sp. exhibits the highest tolerance level, maintaining stable chlorophyll levels at low salinities and showing only moderate declines at higher concentrations. Morphological damage is minimal, limited to leaf yellowing in older tissues. This suggests the presence of efficient protective mechanisms, such as compartmentalization of toxic ions within vacuoles, maintenance of membrane integrity, and activation of stress-responsive genes. Recent studies have highlighted that *Vanda* orchids exhibit greater metabolic plasticity, including efficient stomatal regulation and ROS detoxification, compared to other genera [9]. This makes *Vanda* sp. a strong candidate for breeding salt-tolerant orchid cultivars.

The salinity tolerance ranking (*Vanda* sp. > *Dendrobium* sp. > *Phalaenopsis* sp.) highlights the importance of genotypic variation among orchids. It also supports the idea that orchids, as tropical epiphytes, employ diverse adaptive strategies that reflect their ecological niches. From a practical perspective, identifying tolerant orchids like *Vanda* provides valuable genetic resources for germplasm conservation and breeding programs. Furthermore, understanding the morphophysiological basis of salinity tolerance can inform in vitro selection, genetic engineering, and genome editing approaches aimed at improving abiotic stress tolerance in orchids. Overall, these findings confirm that NaCl salinity is a major limiting factor in orchid cultivation due to its strong inhibitory effect on chlorophyll content and photosynthesis [21]. However, the variation in tolerance between genera highlights opportunities for conservation and improvement. The development of salinity-tolerant orchid varieties will be crucial to ensure sustainable orchid production, especially in areas affected by soil salinization and climate change.

5. Conclusion

This study demonstrates differences in morphophysiological responses among *Dendrobium* sp., *Phalaenopsis* sp., and *Vanda* sp. under NaCl-induced salinity stress. *Phalaenopsis* sp. showed the highest sensitivity, *Dendrobium* sp. exhibited moderate tolerance, and Vanda sp. showed the greatest tolerance by maintaining relatively stable chlorophyll levels and minimal morphological damage. These findings highlight variation in salinity resistance among orchid genera and identify *Vanda* sp. as a potential candidate for developing salinity-tolerant varieties, with implications for orchid cultivation and conservation under increasing salinity stress.

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