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Palynological Studies and Genetic Compatibility Levels of Orchid through Intergeneric Crossing *Vanda* sp., *Phalaenopsis* sp., and *Dendrobium* sp.

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Abstrak

Penelitian ini bertujuan untuk mengkaji viabilitas dan daya perkecambahan polen serta tingkat kompatibilitas genetik pada persilangan intergenerik antara *Vanda*, *Phalaenopsis*, dan *Dendrobium*. Sebanyak 14 spesies anggrek digunakan sebagai tetua jantan dan betina dengan 29 skema persilangan. Uji viabilitas dilakukan menggunakan larutan tetrazolium, sedangkan daya perkecambahan polen diuji secara *in vitro* pada media Brewbaker & Kwack. Hasil menunjukkan *Vanda tricolor* memiliki viabilitas polen tertinggi (82,5%) dan *Vanda* Dr Anek × Gordon Dillon daya perkecambahan tertinggi (67,2%). Viabilitas dan perkecambahan polen berkorelasi positif dengan keberhasilan persilangan. Kombinasi *Phalaenopsis* (jantan) × *Vanda* (betina) menunjukkan tingkat keberhasilan tertinggi (88,89%), sedangkan persilangan *Dendrobium* × *Vanda* menunjukkan kompatibilitas sedang (50%). Keberhasilan pembentukan buah menunjukkan adanya kompatibilitas genetik dan fisiologis yang memungkinkan pembuahan lintas genus dalam famili Orchidaceae. Hasil ini menegaskan bahwa hibridisasi intergenerik antar ketiga genus tersebut berpotensi menghasilkan varietas baru dengan karakter ornamental unggul, meskipun diperlukan uji viabilitas biji dan konfirmasi molekuler untuk memastikan kestabilan genetik hibrida.

Kata kunci: Anggrek, Intergenerik, Kompatibilitas genetik, Persilangan, Polen

Abstract

This study examined pollen viability, germination, and genetic compatibility in intergeneric hybridization among Vanda, Phalaenopsis, *and* Dendrobium. *Fourteen orchid species were used as male and female parents in 29 crossing schemes. Pollen viability was tested using tetrazolium solution, and germination was evaluated in vitro on Brewbaker & Kwack medium. Vanda tricolor showed the highest pollen viability (82.5%), while Vanda Dr Anek × Gordon Dillon had the highest germination rate (67.2%). Pollen viability and germination were positively correlated with crossing success. The highest compatibility occurred in the Phalaenopsis (male) × Vanda (female) combination (88.89%), while Dendrobium × Vanda crosses showed moderate compatibility (50%). Successful fruit formation indicates genetic and physiological compatibility that allows cross-genus fertilization within Orchidaceae. These findings highlight the potential of intergeneric hybridization among the three genera to develop new hybrids with superior ornamental traits, though further seed viability and molecular analyses are needed to confirm hybrid stability.*

Keywords: Crossbreeding, Genetic compatibility, Intergeneric, Orchid, Pollen

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INTRODUCTION

Horticultural commodities are a major commodity that plays a variety of complex and important roles, particularly from an economic perspective (Yanuan Putra et al., 2025). One interesting horticultural commodity that is still being widely developed is ornamental plants. Ornamental plants have high economic and aesthetic value, making them popular among various groups. One example of a prospective ornamental plant commodity with high economic value is the orchid. Orchid flowers come in a variety of attractive shapes, colors, patterns, and aromas (Dormont et al., 2019), and they also have a long lasting bloom. With their stunning uniqueness, orchids have attracted the attention of ornamental plant enthusiasts and collectors. Market demand for orchids has also tended to increase year to year (Ardiyanti & Nuraini, 2024), with uses as ornamental cut flowers, whole ornamental plants, raw materials for the perfume and vanilla flavoring industries, traditional medicine, and for ritual purposes in several religious beliefs (Pant, 2013).

In terms of biodiversity and genetic resources, there are nearly ±50,000 wild orchid species in ±1,200 genera recognized worldwide (Fatmah Hiola et al., 2019), this is a diverse and important genetic resource for research purposes in the field of genetics and plant breeding. In Indonesia, there are around 5,000 orchid species spread across several islands (Handoyo & Prasetya, 2006). Orchids are included in the Orchidaceae family, which is one of the largest families of flowering plants in the world. The increasing number of new species and variations can be obtained through the natural cross-pollination process of various types in nature, natural mutations, and plant breeding processes (Mondragón-Palomino & Theißen, 2009). The activity of increasing variation in orchids is very important because it aims to meet market demand for orchids with certain characteristics and improve the desired properties of orchid plants.

There are three large genera of orchids with various species that are popular/widely cultivated by collectors and ornamental plant hobbyists, namely the genera *Vanda*, *Phalaenopsis*, and *Dendrobium*, all three orchid genera have unique flowers that are different each other. According to Hartati et al. (2017) orchids from the *Vanda* genera are more sought after by collectors, because they have the characteristics of large, round flowers and bold colors. However, the availability of Vanda orchids on the market is not as much as orchids from other genera, so the selling price is more expensive. The prospects for developing new orchid varieties are still wide open, so through plant breeding activities, orchid varieties with high market value and unique characteristics can be obtained.

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Increasing orchid diversity can be achieved through cross-breeding between genera (Han et al., 2025), especially from the genera *Vanda*, *Phalaenopsis*, and *Dendrobium*, which have many species and variations, making them suitable for use as parent plants in hybridization.

There are several considerations before carrying out orchid breeding through hybridization. Crossbreeding in plants requires genetic compatibility. Genetic compatibility is the ability of two organisms to produce normal offspring, free from congenital diseases, and genetic harmony in carrying out reproductive functions to produce offspring (Andersson & Simmons, 2006). In the concept of plant breeding, genetic compatibility can be understood as the ability to carry out pollination/cross-pollination and demonstrate successful seed formation. Not all parent combinations can fertilize each other. Genetic incompatibility mechanisms (both self-incompatibility and cross-incompatibility) prevent fertilization even if pollination occurs.

Apart from genetic compatibility, the viability and germination of orchid pollen also determine the success of hybridization between genera in orchids (Bellusci et al., 2010). Pollen viability is the ability of pollen grains to remain alive, physiologically active, and able to fertilize egg cells in the ovules to produce embryos (Althiab-Almasaud et al., 2024). while pollen germination is the ability of viable pollen to germinate and form a pollen tube after falling on the surface of the stigma (W. Zhao et al., 2023). In orchids, pollen viability and germination are essential for the pollen tube to reach the ovule and form an embryo. Without this, even mechanical pollination will fail to produce normal seeds.

Based on previous research by Hartati et al. (2017) which only tested the compatibility of intergeneric crosses between *Vanda* sp. and *Phalaenopsis* sp., it appears that there are still limitations in scope to only two genera. In fact, a broader genetic compatibility study involving more genera is very necessary to open up opportunities for the formation of new hybrids with higher genetic diversity. Therefore, this study not only focuses on compatibility but also focuses on palynology studies and the level of genetic compatibility through intergeneric crosses of three orchid genera, namely *Vanda* sp., *Phalaenopsis* sp., and *Dendrobium* sp. Information regarding the level of compatibility between orchid genera, pollen viability, and pollen germination is important in orchid breeding to make it more efficient and precise in planning the determination of progeny.

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so that it is expected to provide more comprehensive information and become a basis for more innovative orchid breeding efforts.

RESEARCH METHODS

The research was conducted in 2 locations, the first location is located in the orchid screenhouse located in Ngabeyan, Tegowanuh Village, Kaloran District, Temanggung, the second location is in the screenhouse and tissue culture room located in Tlogorejo, Temanggung. The research was conducted from September 6, 2025 to October 20, 2025. The materials used in this study were 14 orchid species.

Two orchids are from the genus *Vanda*, namely *Vanda* Dr Anek x Gordon Dillon and *Vanda tricolor* (species). Five orchids are from the genus *Phalaenopsis*, namely *P. amboinensis* var. common, *P*. Lime Maroon, *P*. Tropic Wonderland, *P*. Tying Shin new, and *P. violacea*. Seven orchids are from the genus *Dendrobium*, namely *D. anosmum* var. *huttoni*, *D. aphyllum*, *D*. Blue Helga, *D*. Red Butterfly, *D*. Superbiens, *D*. Tanida Pink Stripe x Hawaii Stripe, and *D*. Thongchai Gold. Also pollen from each orchid species, 5ml tubes, labels, string ropes, Gandasil foliar fertilizer (B and D), tween solution, tetrazolium solution, Brewbaker & Kwack media (Sucrose, $Ca(NO_3).2H_2O$, $MgSO_4.2H_2O$, H_3BO_2, and KNO_3), and distilled water (H_2O) were also used. For tools, bent splinter tweezers, magnifying glass, markers, vortex, object glass, 90mm plastic pertrisid, pipettes, water sprayers, ocular microscopes, cameras with macro lenses, and counters were used.

Research was conducted by a survey to select the parent orchids that would be used in the crossbreeding. Then, after obtaining the orchids to be used, pollen samples were first taken to be tested for viability and germination. Pollen collection was carried out at 7:00 a.m., and was only taken from flowers that were 3 days old after blooming. This condition kept the pollen in optimal condition and function because it was estimated that it was still below the mid-flowering age and too close to the wilting age, so it was still optimal (Yuan et al., 2018). There were 10 samples/replications from 10 flowers taken from each species. The observations were pollen viability (%) and pollen germination (%). The amount of pollen viability can be calculated using the formula (number of viable pollen/observed pollen) x 100%, while the pollen germination power is calculated using (number of germinated pollen/total number of pollen) x 100%.

Crossings were performed in the morning, from 7:00 a.m. to 10:00 a.m. in both screenhouses. There were 29 crossbreeding schemes, each with three replicates, so that the total becomes 87 crosses. Crossings were performed by first collecting pollen and then

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transferring it to the stigma of an orchid designated as the female parent/pollen recipient. Crossbreeding is only done on orchid plants that are already flowering, below is a list of crossbreeding codes and schemes:

Table 1. Crossing Codes and Parent Species of Orchids

No.	Code	Crossbreeding parents
1.	PPG X DKLR	♂ *P*. Tropic Wonderland X ♀ *D*. Thongchai Gold
2.	DKLR X PPG	♂ *D*. Thongchai Gold X ♀ *P*. Tropic Wonderland
3.	DALU X PPG	♂ *D. anosmum* var. *huttoni* X ♀ *P*. Tropic Wonderland
4.	DALU X DKLR	♂ *D. anosmum* var. *huttoni* X ♀ *D*. Thongchai Gold
5.	PPG X DALU	♂ *P*. Tropic Wonderland X ♀ *D. anosmum* var. *huttoni*
6.	DKLR X DALU	♂ *D*. Thongchai Gold X ♀ *D. anosmum* var. *huttoni*
7.	PPG X PVI	♂ *P*. Tropic Wonderland X ♀ *P. violacea*
8.	PAM X PVI	♂ *P. amboinensis* var. common X ♀ *P. violacea*
9.	DKLR X DAA	♂ *D*. Thongchai Gold X ♀ *D. aphyllum*
10.	PVI X PAM	♂ *P. violacea* X ♀ *P. amboinensis var. common*
11.	PPG X DAA	♂ *P*. Tropic Wonderland X ♀ *D. aphyllum*
12.	PVI X V.Ung	♂ *P. violacea* X ♀ *V*. Dr Anek x Gordon Dillon
13.	PPG X V.Ung	♂ *P*. Tropic Wonderland X ♀ *V*. Dr Anek x Gordon Dillon
14.	DAA X V.Ung	♂ *D. aphyllum* X ♀ *V*. Dr Anek x Gordon Dillon
15.	DUP X V.Ung	♂ *D*. Blue Helga X ♀ *V*. Dr Anek x Gordon Dillon
16.	V.Ung X DUP	♂ *V*. Dr Anek x Gordon Dillon X ♀ *D*. Blue Helga
17.	VT X PMU	♂ *V. tricolor* X ♀ *P*. Lime Maroon
18.	VT X DAA	♂ *V. tricolor* X ♀ *D. aphyllum*
19.	V.Ung X PMU	♂ *V*. Dr Anek x Gordon Dillon X ♀ *P*. Lime Maroon
20.	DUP X DUK	♂ *D*. Blue Helga X ♀ *D*. Superbiens
21.	DUK X DUP	♂ *D*. Superbiens X ♀ *D*. Blue Helga
22.	DUP X DMA	♂ *D*. Blue Helga X ♀ *D*. Red Butterfly
23.	DUP X DPG	♂ *D*. Blue Helga X ♀ *D*. Tanida Pink Stripe x Hawaii Stripe
24.	PMU X VT	♂ *P*. Lime Maroon X ♀ *V. tricolor*
25.	VT X PUPB	♂ *V. tricolor* X ♀ *P*. Tying Shin new
26.	DAA X VT	♂ *D. aphyllum* X ♀ *V. tricolor*
27.	DMA X VT	♂ *D*. Red Butterfly X ♀ *V*. tricolor
28.	VT X V.Ung	♂ *V. tricolor* X ♀ *V*. Dr Anek x Gordon Dillon
29.	V.Ung X VT	♂ *V*. Dr Anek x Gordon Dillon X ♀ *V. tricolor*

Notes: Each code/scheme is repeated 3 times, by adding a number at the end of the code as a repeat description

The variables observed in the cross were the age of flower bloom (weeks after blooming/WAB), success of the cross (SC), duration of flower wilting after pollination (days/FW), time of fruit formation (days after pollination/TFF), resistance of fruit to falling (days after success/FF), fruit condition (ready to harvest/falling/FC), and fruit formation (formed/not/FFR).

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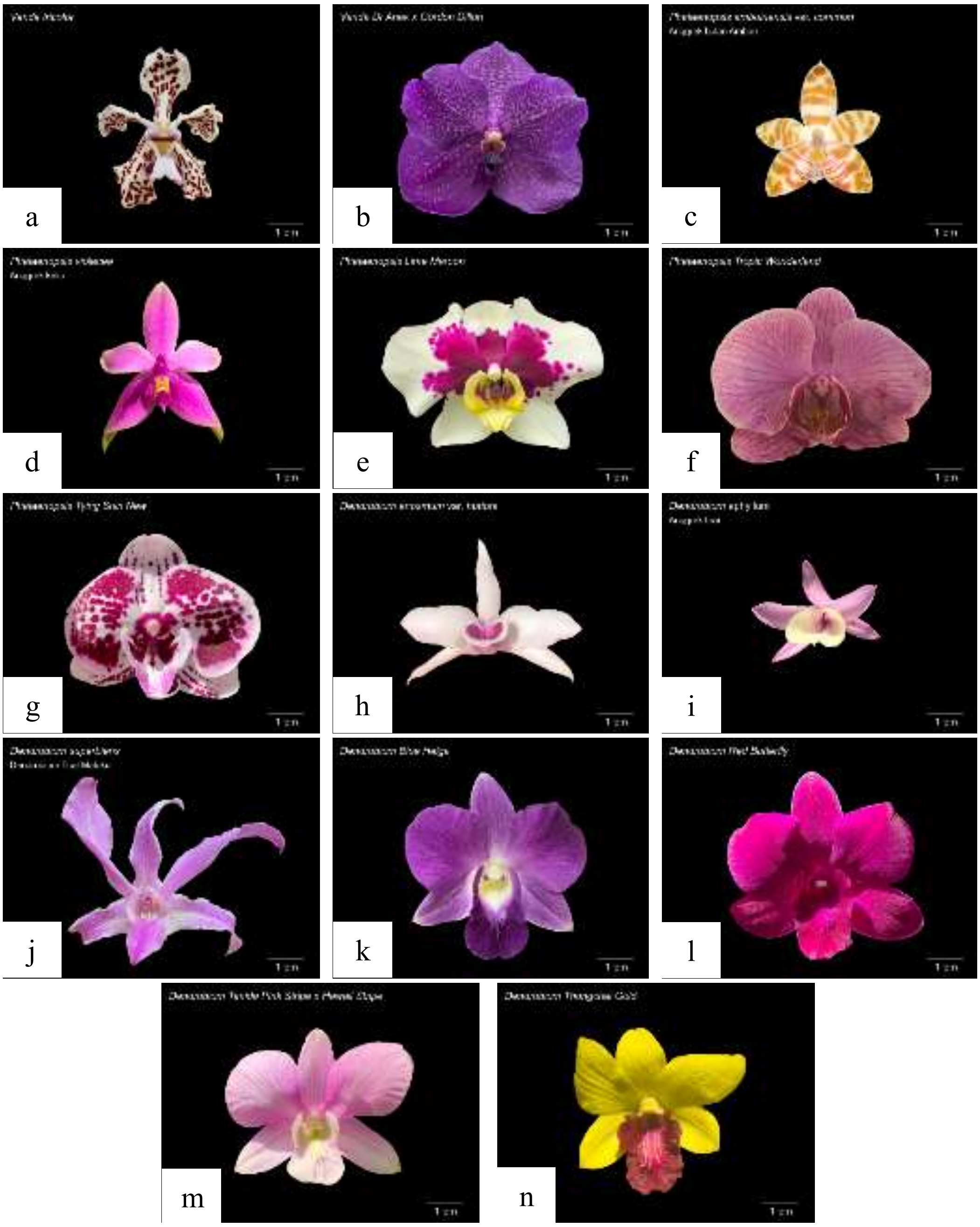

Figure 1. The appearance of each parent orchid flower, a : *Vanda tricolor*; b : *Vanda* Dr Anek x Gordon Dillon; c : *P. amboinensis* var. common, d : *P. violacea*; e : *P.* Lime Maroon, f : *P.* Tropic Wonderland, g : *P.* Tying Shin new; h : *D. anosmum* var. *huttoni*; i : *D. aphyllum*; j : *D.* Superbiens; k : *D.* Blue Helga; l : *D.* Red Butterfly; m : *D.* Tanida Pink Stripe x Hawaii Stripe; and n : *D.* Thongchai Gold

This research is exploratory using a mixed methods approach. This approach was chosen because the research aims not only to obtain quantitative data but also to explore

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and describe the phenomenon of crosspollination in orchids. In the qualitative section, the research was conducted descriptively by observing the success of cross-pollination between species and between genera, as indicated by the formation of capsules or fruit. The results of these crosses were then analyzed descriptively to illustrate the level of compatibility between cross-pollination combinations. If the crossing scheme is viewed based on genus and crosses with the same genus as a comparison, the combination will be as in the table below.

Table 2. Number of Crosses by Genus

No.	Crossing scheme	Number of crosses	Number with replication
1.	♂ *Vanda* X ♀*Phalaenopsis*	3	9
2.	♂ *Phalenopsis* X ♀*Vanda*	3	9
3.	♂ *Vanda* X ♀ *Dendrobium*	2	6
4.	♂ *Dendrobium* X ♀ *Vanda*	4	12
5.	♂ *Phalenopsis* X ♀ *Dendrobium*	3	9
6.	♂ *Dendrobium* X ♀ *Phalaenopsis*	2	6

Table 3. Number of Crosses Based on the Same Genus as A Comparison

No.	Crossing scheme (same genera)	Number of crosses	Number with replication
1.	♂ *Vanda* X ♀ *Vanda*	2	6
2.	♂ *Phalenopsis* X ♀ *Phalaenopsis*	3	9
3.	♂ *Dendrobium* X ♀ *Dendrobium*	7	21

Meanwhile, in the quantitative part, the research focused on experimental testing of pollen viability and pollen germination. Pollen viability was measured using a tetrazolium solution to calculate the percentage of viable pollen grains, while pollen germination was tested through an in vitro method on Brewbaker & Kwack media by calculating the percentage of pollen that successfully germinated. The experiment was designed using a one-factor Completely Randomized Design (CRD), namely orchid species consisting of 14 species from three main genera (*Vanda*, *Phalaenopsis*, and *Dendrobium*). The data were then analyzed using analysis of variance with a test level of 1% ($\alpha = 0.01$) and then post hoc tests were carried out using Duncan's Multiple Range Test. data from intergeneric crossing (success rate, capsule formation, fruit set duration, and classification of compatibility) were analyzed descriptively by calculating percentages and comparing them with the classification of compatibility levels as proposed by Hartati (2017). Fully compatible if the cross produces more than 60% of the fruit, partially compatible if the fruit produced is between 30 and 60%, and fully incompatible if the cross yields less than 30%.

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RESULTS AND DISCUSSION

Based on the research that has been conducted, several important data and information have been obtained that can be used as considerations for carrying out or just planning orchid plant breeding, especially through simple methods such as artificial crossing by transferring pollen from one orchid to another. Before reviewing the success of crosses between genera, a review of the quality of orchid pollen is first carried out, including pollen viability and pollen germination power. Pollen viability is important to know, especially to assess whether the plant used as a male is truly fertile or not, in other words it has the ability to fertilize the ovules of the female flower/parent (Impe et al., 2020), Low pollen viability will increase the chance of failure in the pollination process (Pascual et al., 2021), especially in the process of crossing between genera which takes into account genetic compatibility.

Pollen germination also determines the success of pollination (Tran et al., 2023), this aspect is important for understanding the ability of pollen to grow and fertilize the ovules/ovaries found in female flowers. The higher germination rate of pollen, the greater chance of the pollen forming a pollen tube capable of reaching ovule and fertilizing female gamete, thus producing viable seeds. Its correlation with pollen viability is that viability indicates whether the pollen is still alive and functional, while germination rate indicates the ability of the living pollen to grow actively and form a pollen tube. Therefore, pollen must be viable before it can germinate, and generally the two have a positive correlation (Sulusoglu & Cavusoglu, 2014). However, in several male orchid parents studied (Table 4.), the species with the highest pollen viability, *Vanda tricolor*, did not show the highest germination results either, but the highest pollen germination was in *Vanda* Dr Anek x Gordon Dillon, this could be due to physiological conditions, pollen maturity levels, and different genotypes.

If we review the data in Table 4, we can understand that in most species that have high viability also have quite high pollen germination, even in *D.* superbiens, the lowest viability also has an impact on the lowest pollen germination. This shows that pollen viability is correlated with pollen germination, although in a small number of orchid species in this study this did not occur. Although there is no direct standard that states what percentage of pollen viability is considered good in orchids, because it is very dependent on the species and genus, in many cases of orchid crossing, pollen viability of more than 50% is an average that indicates successful pollination (Bellusci et al., 2010), although this

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is independent of genetic compatibility. In this study, there were only two orchid parents that had pollen viability below 50%, namely *D. anosmum* var. *huttoni* and *D.* Superbiens. Previous research conducted by Kahagalla et al. (2019) stated that orchids from Dendrobium are included in the orchid genus that has high pollen viability ranging from 77-94%. Low pollen viability can be caused by pollen maturity and genetic degradation in hybrid species that have been breeding for a long time.

Table 4. Results of Viability and Germination Tests of Orchid Pollen

Male parent species	Pollen viability (%)	Pollen germination (%)
DAA (*D. aphyllum*)	75,7±3,03[a]	39,0±5,59[ef]
DALU (*D. anosmum* var. *huttoni*)	46,9±6,83[c]	36,5±6,70[f]
DKLR (*D.* Thongchai Gold)	61,0±3,07[b]	46,2±3,87[cde]
DMA (*D.* Red Butterfly)	62,4±8,05[b]	35,9±8,20[f]
DPG (*D.* Tanida Pink Stripe x Hawaii Stripe)	63,5±6,00[b]	34,6±9,07[f]
DUK (*D.* Superbiens)	36,3±9,18[d]	9,3±3,39[g]
DUP (*D.* Blue Helga)	57,5±7,61[b]	33,2±6,73[f]
PAM (*P. amboinensis* var. common)	81,1±5,09[a]	50,1±6,02[bcd]
PMU (*P.* Lime Maroon)	77,6±7,06[a]	48,7±4,00[bcd]
PPG (*P.* Tropic Wonderland)	79,3±5,31[a]	57,9±7,96[b]
PUPB (*P.* Tying Shin new)	75,4±8,21[a]	41,6±7,89[def]
PVI (*P. violacea*)	81,2±3,63[a]	54,3±9,96[bc]
V.Ung (*V.* Dr Anek x Gordon Dillon)	78,5±4,52[a]	67,2±8,49[a]
VT (*V. tricolor*)	82,5±6,14[a]	56,7±4,94[a]
Mean	68,49	43,66
F	46,76**	37,67**
C/V	6,90%	11,97%

In orchids, normally, the pollen germination rate will be lower than the pollen viability percentage, this also occurs in other plants (Sudha et al., 2022), the reason is because viability is the ability to remain alive, while germination requires certain environmental conditions that are not always met especially in medium, and is also influenced by other factors such as genetic defects or inadequate vigor (R. Zhao et al., 2022). However, this number will really determine whether the pollination carried out can successfully form fruit and seeds, because pollen germination is the initial stage of fertilization in orchids. In this research, the parent with the lowest pollen germination power was *D.* Superbiens and the highest was *V.* Dr Anek x Gordon Dillon.

Table 6. Table of Crossing and Fruit Formation Success

No.	WAB ♂/♀ (week after blooming)	SC	FW (days)	TFF (days after pollination)	FF (days after success)	FC	FFR
1.	5/6	-	3	-	-	-	-
2.	6/5	-	2	-	-	-	-

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No.	WAB ♂/♀ (week after blooming)	SC	FW (days)	TFF (days after pollination)	FF (days after success)	FC	FFR
3.	1/5	-	2	-	-	-	-
4.	1/6	-	3	-	-	-	-
5.	5/1	-	2	-	-	-	-
6.	6/1	-	3	-	-	-	-
7.	5/3	✓	2	7	✓	✓	✓
8.	4/3	✓	4	10	✓	✓	✓
9.	6/1	-	2	-	-	-	-
10.	3/4	✓	3	6	✓	✓	✓
11.	5/1	-	1	-	-	-	-
12.	3/2	✓	2	13	✓	✓	✓
13.	5/5	✓	2	12	✓	✓	✓
14.	1/2	-	2	10	(-) 17	-	✓
15.	5/2	-	2	9	(-) 13	-	✓
16.	2/5	✓	3	12	✓	✓	✓
17.	3/1	-	3	13	(-) 17	-	✓
18.	3/3	-	2	13	(-) 15	-	✓
19.	2/7	✓	1	10	✓	✓	✓
20.	5/6	-	2	-	-	-	-
21.	6/5	-	3	-	-	-	-
22.	5/4	-	3	-	-	-	-
23.	5/3	-	6	15	(-) 23	-	✓
24.	3/3	✓	2	10	✓	✓	✓
25.	3/2	-	4	12	(-) 20	-	✓
26.	1/3	✓	5	8	✓	✓	✓
27.	4/3	✓	3	6	✓	✓	✓
28.	3/2	✓	3	9	✓	✓	✓
29.	2/3	✓	3	6	✓	✓	✓

Note: The symbol (-) means no or not experiencing the process at all, while the symbol (✓) means yes or experiencing the process

From the crosses carried out, in Table 6, it can be understood that there are only 12 cross schemes that successfully produce fruit and do not fall off, this shows that in the 12 cross schemes there is suitability for crosses, while there are 18 crosses that successfully form fruit, but in the process of development and filling of the fruit, 6 cross schemes experienced fruit loss, namely schemes number 14, 15, 17, 18, 23, and 25, while the rest did not experience any formation at all. However, all crossing schemes indicated wilting in females after pollen transfer from another flower, with an average time of 2 and 3 days, and the longest 6 days in scheme number 23, It is thought that each species has a different response and cross breeding ability, therefore the wilting response in flowers also varies. Orchids will require more nutrients and energy to form fruit (Zhang et al., 2022), therefore

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in this study, orchids/parent plants were still given fertilizer to ensure that fruit formation was not too affected by nutrient dynamics.

Orchids will wilt when their pollen is taken and pollination is carried out because the purpose of waiting and attracting pollinators is felt to be finished, this mechanism is also triggered by hormonal activity which signals the flower to divert all its energy and resources to developing a seed pod (Arditti et al., 1971). However, wilting is not always a sign that pollination was successful. This was proven in this study that in all crossing schemes, all female parents experienced wilting, but only 12 schemes showed success.

The phenomenon of fruit formation that occurs in schemes 14, 15, 17, 18, 23, and 25 which then fall off can be caused by the failure of the perfect fertilization process or physiological imbalance after pollination, as well as several causes such as imperfect fertilization, incompatibility, disturbed hormonal balance, unsuitable environment, and also physiological factors of the parent plant. However, in this study, the failure, the cause that is more highlighted is the existence of incompatibility, where this incompatibility does not only occur in different genus crossing schemes but also occurs within the same genus of different species. This happens because there is a genetic incompatibility between the two parents, so that the hybrid is formed imperfectly or even fails, so that the female parent will reject the fruit formation process until it is perfect and it will fall off (Szymajda et al., 2022).

The success rate of a cross-pollination is determined by counting the number of fruit formed from the total number of flowers crossed. Successful pollination occurs when the pollinarium can be inserted into the rostellum. The cross is considered successful if the orchid flowers show fresh, green ovaries, and the base of the flower stalk appears enlarged. A few days later, the perianth will appear to wilt (Hartati et al., 2017). If we look at the results of crosses based on genus (Table. 7), then it can be understood that, only the crossing scheme of *Phalaenopsis* as male with *Dendrobium* as female as well as *Dendrobium* as male with *Phalaenopsis* as female, showed 0% success in crossing.

Table 7. Success Rate of Intergeneric Crossing

No.	Crossing scheme	Number of crosses	Number with replication	Number of successful	Success rate (%)
1.	♂V X ♀P	3	9	3	33,33%
2.	♂P X ♀V	3	9	8	88,89%
3.	♂V X ♀D	2	6	3	50%
4.	♂D X ♀V	4	12	6	50%
5.	♂P X ♀D	3	9	0	0%
6.	♂D X ♀P	2	6	0	0%

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Table 8. Success Rate of Crossing in Same Genus

No.	Crossing scheme	Number of crosses	Number with replication	Number of successful	Success rate (%)
1.	♂V X ♀V	2	6	6	100%
2.	♂P X ♀P	3	9	7	77,78%
3.	♂D X ♀D	7	21	0	0%

The highest success rate of intergenera crosses was in the *Phalaenopsis* scheme as a male with *Vanda* as a female at 88.89% and vice versa only 33.33%, which means that the highest compatibility in this study was only achieved when *Phalaenopsis* was used as a male and *Vanda* as a female in the cross. However, what is interesting in this study is the success of crosses between the *Vanda* and *Dendrobium* genera, both of which reciprocally showed a compatibility percentage of 50%. In previous studies, cross-breeding experiments between the *Vanda* and *Dendrobium* genera have almost always failed. These two orchids are not closely related and have many biological differences, for example, the *Vanda* is monopodial and the *Dendrobium* is sympodial. Large differences in many aspects, especially genetics, will further increase the chance of failure in cross-breeding because it will clearly be incompatible and cannot produce a clear hybrid. Ploidy and chromosomes also determine the success of crosses between genera. Incompatibility in both parents, especially in the number of chromosomes, will result in sterile offspring and fertilization will not occur perfectly (Sattler et al., 2016).

The successful cross between the *Dendrobium* and *Vanda* demonstrates the genetic and physiological compatibility that allows for cross-genus fertilization within the Orchidaceae family. This indicates that the mechanisms of pollen recognition, pollen tube growth, and embryo development can occur even though the two parents are from different genera and are distantly related. This success is significant for orchid breeding programs, as it opens up the possibility of developing intergeneric hybrids with a combination of superior traits, such as the resistance of *Dendrobium* and the attractive flower color or shape of *Vanda*. However, the success of fruit formation requires further study through viability and seed germination tests to ensure that fertilization occurs successfully and produces viable offspring. These findings also have scientific implications for understanding the kinship relationships and mechanisms of reproductive isolation between genera within the Orchidaceae.

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Figure 2. Orchid crossbreeding that shows success with the formation of seed pods, a : ♂ *P. Tropic Wonderland X* ♀ *P. violacea*; b : ♂ *P. amboinensis var. common X* ♀ *P. violacea*; c : ♂ *P. violacea X* ♀ *P. amboinensis var. common*, d : ♂ *P. violacea X* ♀ *V. Dr Anek x Gordon Dillon*; e : ♂ *P. Tropic Wonderland X* ♀ *V. Dr Anek x Gordon Dillon*, f : ♂ *V. Dr Anek x Gordon Dillon X* ♀ *D. Blue Helga*, g : ♂ *V. Dr Anek x Gordon Dillon X* ♀ *P. Lime Maroon*; h : ♂ *P. Lime Maroon X* ♀ *V. tricolor*; i : ♂ *D.aphyllum X* ♀ *V. tricolor*; j : ♂ *D. Red Butterfly X* ♀ *V. tricolor*; k : ♂ *V. tricolor X* ♀ *V. Dr Anek x Gordon Dillon*; and l : ♂ *V. Dr Anek x Gordon Dillon X* ♀ *V. tricolor*

Overall, if we refer to the standards set by Hartati et al. (2017), the cross between V X P in this study is classified as partially compatible because the percentage of crosses is between 30-60%, while P X V is classified as fully compatible because it is above 60%, namely 88.89%. Then, the crosses between *Vanda* and *Dendrobium* are each in the partially

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compatible category because they are between 30-60%. For comparison, as can be seen in Table 8, interspecific crosses between several species in the genera Vanda and Phalaenopsis both showed high levels of compatibility, each above 60%, except for the genus Dendrobium which showed 0% success. This proves that the level of incompatibility in orchid crosses will be higher in intergenera crosses than in interspecific crosses. This is because the genetic distance will be greater when they are in different genera, thus creating a more complex genetic barrier than when they are in different species but in the same genus (Kaneko & Bang, 2014).

Incompatibility occurs due to the inability of male and female organs to produce sexual seeds due to physiological factors, and the inability to produce pollen tubes quickly enough to reach the ovules. Incompatibility in cross-breeding is caused by incompatibilities between male and female organs. This incompatibility is controlled by environmental, genetic, and physiological factors. The success of crossing is indicated by the appearance of fruit that can survive until harvest and has a normal physical/appearance as in Figure 2. Each genus that is used as a female parent has a different fruit appearance, The fruit will later be used as planting material which will produce hybrid offspring from both parents. This intergeneric crossing is ideal for creating hybrids with more diverse characters, especially to bring out characters that do not exist in a genus.

The successful intergeneric hybridization between *Dendrobium* and *Vanda* indicates that certain species from these genera possess sufficient genetic and physiological compatibility to overcome reproductive barriers commonly observed in Orchidaceae. This finding highlights the potential for generating novel hybrids with desirable traits and provides an important reference for future orchid breeding strategies. However, further analyses, including seed viability tests and molecular confirmation of hybrid status, are necessary to ensure that fertilization was truly successful and that the offspring are genetically stable.

CONCLUSIONS AND SUGGESTIONS

The study revealed that pollen viability and germination significantly influenced the success of intergeneric hybridization among *Vanda*, *Phalaenopsis*, and *Dendrobium*. High pollen viability and germination were generally associated with greater crossing success, emphasizing their importance in determining male fertility and compatibility potential. The highest compatibility was observed when *Phalaenopsis* acted as the male parent and *Vanda* as the female, while crosses between *Dendrobium* and *Vanda* showed

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moderate compatibility, indicating that both genera still share functional reproductive mechanisms despite their taxonomic differences. These findings confirm that intergeneric hybridization among these orchids is feasible and valuable for breeding programs aimed at developing novel hybrids with superior ornamental and adaptive traits. Further studies on seed viability and molecular verification are needed to confirm true hybrid formation and genetic stability.

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