

# Performance evaluation of new Papaya Ring Spot Virus-tolerant Papaya (*Carica papaya* L.) F1 Hybrids in three selected locations

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## ABSTRACT

Papaya production is consistently challenged by various diseases such as Papaya Ring Spot Virus (PRSV). To mitigate the impact of diseases, PRSV tolerant inbred lines with good horticultural characteristics were selected and used to develop novel PRSV-tolerant Papaya F<sub>1</sub> hybrids. Subsequently, the researchers conducted field trials across three locations in Southern Tagalog to evaluate the field performance of the three newly developed hybrids, namely Hirang, Liyag, and Timyas. These hybrids exhibited semi-dwarf growth, possessed robust stems at the onset of flowering, and displayed few nodes at first flowering. Notably, hybrids showcased remarkable fruit-bearing habits when compared to control varieties. The hybrids exhibited varying characteristics, including fruit weight, length, and width. They had thick flesh with high sweetness level, low titratable acidity, and high edible portion. Liyag displayed maize yellow flesh, Timyas had saffron yellow flesh, while Hirang had vermilion red flesh. Furthermore, hybrids exhibited

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moderate tolerance to the virus, with mild symptoms of PRSV. Although these tested positive for PRSV via ELISA, the hybrids consistently yielded marketable quantities (44.95 MT/ha for Liyag, 44.78 MT/ha for Hirang, and 41.23 MT/ha for Timyas). The new hybrids exhibited consistent characteristics across locations and thus are recommended for potential release throughout Luzon, Philippines.

*Keywords:* Hirang, Liyag, Papaya, PRSV, Timyas

## INTRODUCTION

Papaya (*Carica papaya* L.) (2n=18) is a high-value crop cultivated in the tropics and sub-tropics such as the Philippines (Magdalita and Signabon 2017). In the Philippines, papaya ranks 5<sup>th</sup> among the major fruit crops in terms of production volume (PSA 2016) and its production is a highly profitable industry. Philippine papayas are exported to USA, Japan, Hong Kong, UAE, Canary Islands, New Zealand, Saudi Arabia, Singapore, among others (Dela Cueva et al. 2017). However, the papaya industry faces significant challenges, particularly from diseases and insect pests such as Papaya Ring Spot Virus (PRSV), Bacterial Crown Rot disease (*Erwinia mallotivora*), root rot (*Phytophthora*) (Rivarez 2017), anthracnose, damping-off, and fruit rot. Emerging diseases such as Papaya crown yellows, detected in Mindanao (Billones 2001) and Papaya *Maleira* virus, found in Mexico but not yet detected in the Philippines, (Brito-Perez et al. 2012) may pose serious threats in the future. PRSV infection affects all growth stages of papaya and is transmitted by aphid vectors in a non-persistent manner and spreads like a wildfire (Magdalita and San Pascual 2019).

The PRSV infestation in Cavite and Laguna has inflicted significant damage on the industry, prompting the implementation of mitigation measures. One critical approach involved the development of PRSV-tolerant papaya varieties. During the 1990s, breeding of PRSV-tolerant hybrids began with the development of the first Papaya F<sub>1</sub> hybrid, Sinta, using tolerant inbred lines. Given evolving market preferences and the persistent challenges of PRSV resurgence and new diseases, ongoing efforts aim to create F<sub>1</sub> hybrids with similar or superior PRSV tolerance and fruit quality to previously released varieties such as Sinta, Carinosa, Red Lady, and Red Royale.

To achieve this, continuous purification process of selected inbred lines is essential, ensuring that inbred lines exhibit consistent performance after approximately eight inbreeding cycles. Selected parental lines, distinguished by exceptional fruit quality and PRSV tolerance, were crossed to produce F<sub>1</sub> hybrids. From numerous crosses, a select few F<sub>1</sub> hybrids were chosen, pretested in the field in 2013 in Laguna, and

demonstrated moderate PRSV tolerance, high yields, and superior fruit quality. Once their productivity was established in one location, the next step was to conduct on-farm trials in at least two other locations to confirm performance of these hybrids before potential release.

This study aims to assess the performance of three PRSV-tolerant Papaya F<sub>1</sub> hybrids. Specifically, it seeks to evaluate their performance in terms of yield, fruit morphology, quality traits, and PRSV reaction in three locations in Southern Tagalog, namely Laguna, Batangas, and Cavite.

## **MATERIALS AND METHODS**

### **Plant materials and their preparation**

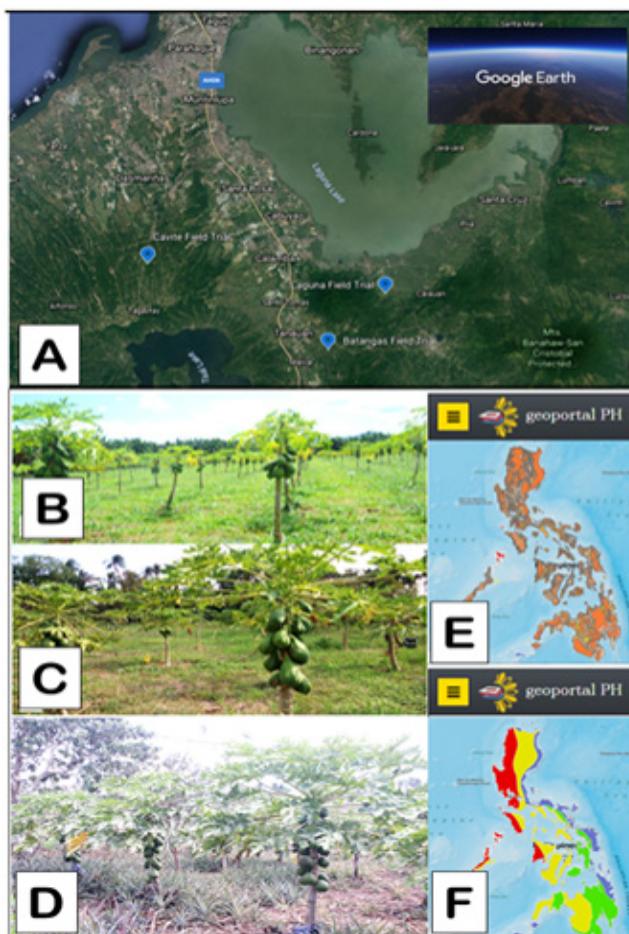
In this study, the performance of three new PRSV-tolerant Papaya F<sub>1</sub> hybrids was tested under field conditions in three locations. The three hybrids are: Liyag (5648 x 336), Hirang (097 x 4172) and Timyas (4173 x 5648). Two other lines were used in the study as inbred control, 4172 (Cavite Special) and the F<sub>1</sub> hybrid Sinta (5648 x 4172) that served as the PRSV-tolerant control.

The hybrid seeds, which were produced and maintained by the Fruit and Ornamental Crops Breeding Section of the Institute of Plant Breeding (IPB), College of Agriculture and Food Science (CAFS), University of the Philippines Los Baños, were sown in a sterile potting medium composed of a mixture of garden soil, coir dust and compost (1:1:1). The seedling bags were placed in the screen house with partial shade using a black net with a light intensity of 585-790 lux measured by the light meter (TL-600 USB Lux Meter, DongGuan XinRui Instruments, Co., LTD, China). The medium was watered accordingly when the moisture content of the medium fell to the rating scale of 2-3 using the Soil Moisture meter (Tekamura Electric Works Ltd. Japan). The seeds germinated within seven days.

Seedlings were initially cultivated in plastic bags arranged in steel benches inside a screen house, maintaining a temperature range of 25-30°C. After seven days, seedlings received fertigation with a complete water-soluble fertilizer and were nurtured for two months. To enhance their resilience, plants underwent a one-week hardening process by gradually exposing them to sunlight. Subsequently, seedlings were transplanted, with each planting hole receiving a 50 g basal fertilizer mix (urea + complete fertilizer, 2:1 ratio). Regular watering was provided in the absence of rainfall. Furthermore, plants were subjected to periodic applications of appropriate insecticides and fungicides, ensuring their health and protection.

## Locale of the Study

The field trials were conducted in the UPLB experimental station in i) Tranca, Bay, Laguna, ii) ACS Industrial Farms in Sto. Tomas Batangas, and iii) Montealegre's Farm in Silang, Cavite (Figure 1). The study was conducted from January 2018 to December 2020.



**Figure 1.** The location of the field trials (pinned in blue) (A), the fruiting papaya plants in (B) Laguna, (C) Batangas, and (D) Cavite, the (E) soil type map of BSWM and DA, and the (F) climate map of PAGASA, classifying the soil and climate type of each location.

## Evaluation of horticultural traits and PRSV reaction of promising Papaya F<sub>1</sub> hybrids

Evaluation of different horticultural characteristics and PRSV reaction of the three promising papaya F<sub>1</sub> hybrids was conducted. Horticultural traits observed at flowering stage and the fruits of the three F<sub>1</sub> papaya hybrids and the controls

were evaluated in three locations. The experimental layout spanned three distinct locations (Figure 1). In each of these locations, replications were bordered with Sinta hybrid. The experiment took place within a 2,250-square-meter area. The experimental plants were spaced 3 m between rows and 2 m between plants, mulched to conserve moisture and provided with an irrigation facility.

The pictures and layout of field trials done in Batangas, Cavite, and Laguna from 2018 to 2019 are presented in Figures 1A-D and 2, respectively. The plants were bordered with Sinta. At the flowering stage, horticultural characteristics of the three promising  $F_1$  hybrids and the controls, including plant height (cm), stem diameter (cm), number of nodes, and number of fruits/tree/cycle, were evaluated.

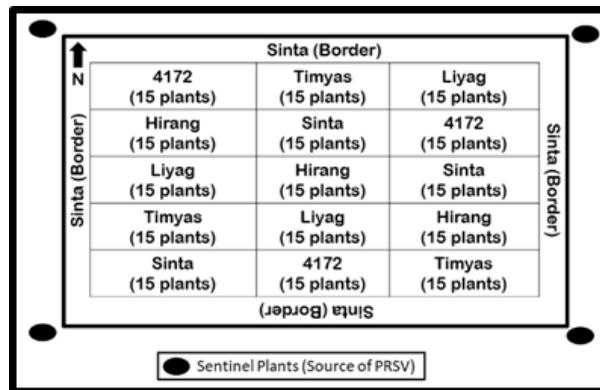


Figure 2. Replicated field trial of candidate papaya hybrids.

Mature fruits per tree were counted at maturity stage to determine yield. Marketable and non-marketable fruits were also counted. Physiologically mature fruits with Peel Color Index 2 (PCI 2), i.e., tinge of yellow on the apex of fruit, were harvested. Five fruit samples were taken from each tree and allowed to ripen on benches. At fully ripe stage, fruits were evaluated for morphological characteristics, namely weight, length, and width; peel and flesh thickness, peel and seed weight, and fruit quality traits such as total soluble solids, titratable acidity, flesh firmness, and edible portion. The reaction to PRSV of three papaya  $F_1$  hybrids was monitored. Final reaction was assessed at the fruiting stage in three locations. The typical symptoms of PRSV are ringspot in the fruit, mottling and chlorosis on the leaves, and water-soaked lesions in the stem and petioles. Alviar et al. (2012) scored severity symptoms as 1- No PRSV-like symptoms; 3- Mottling; 5- Mosaic; 7- Mosaic and leaf deformation; 9- Mosaic, leaf deformation, and shoestring. In addition, virus titer was measured using an Enzyme-linked Immunosorbent Assay (ELISA) using standard procedure and then quantified. Both severity scoring and ELISA were done in collaboration with pathologists from the same institute.

## Statistical Analysis

Field trial experiments were conducted using a randomized complete block design (RCBD) with three replications, each consisting of 15 trees. Five fruit samples were collected per tree. Locations served as blocks in the experimental design. Data on horticultural characteristics were subjected to analysis of variance (ANOVA) using the F-test. When significant differences among treatment means were found, the least significant difference (LSD) test at  $\alpha = 0.05$  was used for mean separation. All statistical analyses were performed using the Statistical Tool for Agricultural Research (STAR) software (IRRI 2014).

## RESULTS AND DISCUSSION

A field trial was conducted in three different locations whose characteristics in terms of climate, soil type and their geographic coordinates are presented in Table 1.

**Table 1. Characteristics of three locations in terms of climate, soil type, and GPS coordinates.**

Location		Climate type*	Soil Type***	GPS
Montealegre's Farm	Silang, Cavite**	Type 1	Tagaytay Loam	14o10'43"N, 121o58'00"E
ACS Industrial Farm	Santo Tomas, Batangas	Type 1	Lipa Loam	14o03'29"N, 121o11'36"E
UPLB Experimental Field	Los Baños, Laguna	Type 3	Lipa Clay Loam	14o08'12"N, 121o15'46"E

\*Based on PAGASA, \*\* Has high inoculum of PRSV, \*\*\* Based on BSWM, DA

### Tree characteristics of Papaya F<sub>1</sub> hybrids planted in Laguna, Batangas and Cavite trials.

Tree characteristics of three PRSV tolerant Papaya F<sub>1</sub> hybrids planted in three locations are presented in Table 2. For plant height, significant differences were observed in Laguna field trial, where Hiran and Liyag had the highest mean plant height compared to Timyas, Sinta, and 4172. The hybrids were semi-dwarf, but 4172 was dwarf in Batangas and Cavite (0.98 m and 0.86 m). Dwarf papaya has plant height below 1 m, while semi-dwarf has a height of 1-1.5 m (IBPGR 1988). Philippine Papaya semi-dwarf varieties include Red Royale and Cariñosa (EWSC nd). Furthermore, stem diameter (2.3-5.11 cm) of hybrids were the controls across locations. Semi-dwarf characteristics and thicker stems are essential parameters, as it is suitable in typhoon-stricken areas (Magdalita and San Pascual, 2019).

Sakamoto et al. (2003) observed that in rice, semi-dwarf cultivars are more resistant to wind and rain damage and thus yield more grain when fertilized. Moreover, comparable mean number of nodes to first flower was observed in all hybrids of Laguna and Cavite field trials. However, significant differences were observed on in the average number of nodes to the first flower in the Batangas trial.

**Table 2. Plant height, stem diameter, and no. of nodes of the three Papaya F<sub>1</sub> hybrids in Laguna, Batangas, and Cavite**

Hybrid	Plant Height (m)			Stem Diameter (cm)			No. of Nodes		
	Lag (p=0.000)	Bat (p=0.058)	Cav (p=0.185)	Lag (p=0.090)	Bat (p=0.853)	Cav (p=0.817)	Lag (p=0.200)	Bat (p=0.035)	Cav (p=0.229)
Hirang	1.40 <sup>a</sup> ± 0.05	1.16 ± 0.12	1.18 ± 0.11	5.11 ± 0.44	2.40 ± 0.44	3.13 ± 0.49	14.67 ± 1.53	29.00 <sup>a</sup> ± 0.10	24.33 ± 3.79
Liyag	1.41 <sup>a</sup> ± 0.08	1.28 ± 0.10	1.24 ± 0.07	4.93 ± 0.49	2.63 ± 0.42	3.13 ± 0.38	13.33 ± 0.58	29.67 <sup>a</sup> ± 1.53	26.00 ± 2.65
Timyas	1.28 <sup>b</sup> ± 0.06	1.36 ± 0.21	1.03 ± 0.15	4.75 ± 0.63	2.93 ± 0.61	3.03 ± 0.80	14.00 ± 2.00	28.33 <sup>a</sup> ± 1.15	27.33 ± 4.73
Sinta	1.25 <sup>b</sup> ± 0.04	1.18 ± 0.08	1.08 ± 0.23	4.97 ± 0.15	2.43 ± 0.25	2.90 ± 0.53	17.88 ± 3.36	24.67 <sup>b</sup> ± 2.31	27.00 ± 3.61
4172	1.27 <sup>b</sup> ± 0.04	0.98 ± 0.15	0.96 ± 0.04	4.07 ± 0.25	2.73 ± 1.19	2.30 ± 0.20	17.00 ± 2.65	26.67 <sup>ab</sup> ± 2.52	31.67 ± 3.06

\* Values are means ± standard deviation. Superscript letters indicate significant differences based on ANOVA and post-hoc tests ( $\alpha = 0.05$ ); means with the same letters within each column are not significantly different. Lag = Laguna, Bat = Batangas, Cav = Cavite.

Table 3 shows number of fruits and yield of three papaya F<sub>1</sub> hybrids in Laguna, Batangas, and Cavite. Significant differences were observed in terms of number of fruits per tree across hybrids in Laguna and Cavite trials. In Laguna, three hybrids had comparable mean number of fruits but were significantly different from the control, 4172. Further, in Cavite, significant differences were observed in the mean number of fruits because the three hybrids, Liyag, Hirang, and Timyas, differed from the controls.

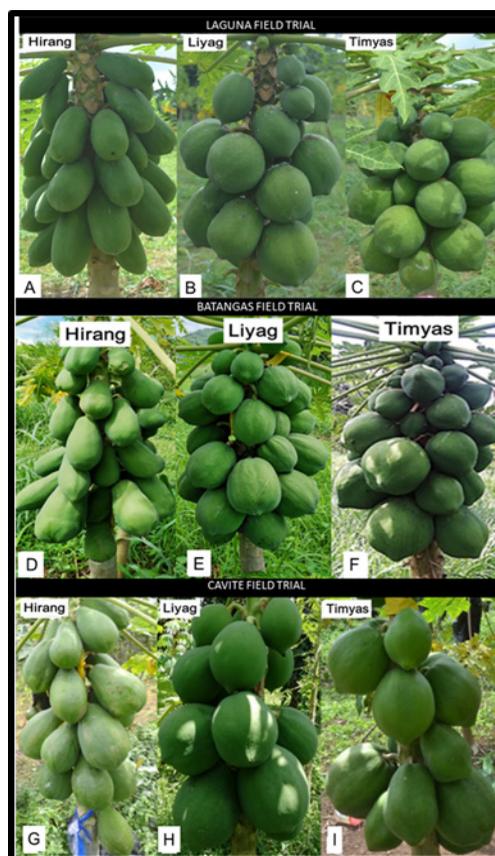
**Table 3. The number of fruits and yield of the three Papaya F<sub>1</sub> hybrids in Laguna, Batangas, and Cavite**

Hybrid	No. of fruits			Yield (MT/ha)		
	Lag (p=0.013)	Bat (p=0.182)	Cav (p=0.000)	Lag (p=0.038)	Bat (p=0.669)	Cav (p=0.000)
Hirang	19.34 <sup>a</sup> ± 2.04	20.00 ± 8.33	11.63 <sup>a</sup> ± 2.72	50.84 <sup>a</sup> ± 5.36	49.64 ± 22.00	33.86 <sup>b</sup> ± 6.51
Liyag	14.46 <sup>ab</sup> ± 0.41	17.15 ± 3.45	12.83 <sup>a</sup> ± 1.76	48.33 <sup>a</sup> ± 2.10	44.66 ± 9.19	41.85 <sup>a</sup> ± 4.44
Timyas	19.15 <sup>a</sup> ± 4.59	16.36 ± 3.21	13.22 <sup>a</sup> ± 1.07	45.42 <sup>a</sup> ± 13.96	42.97 ± 14.69	35.30 <sup>a</sup> ± 2.70

Sinta	17.82 <sup>a</sup> ± 3.36	16.20 ± 1.91	6.19 <sup>b</sup> ± 2.72	46.36 <sup>a</sup> ± 12.91	40.16 ± 12.84	17.09 <sup>b</sup> ± 9.43
4172	9.92 <sup>b</sup> ± 2.57	10.00 ± 2.96	3.75 <sup>b</sup> ± 0.25	24.21 <sup>b</sup> ± 7.32	30.79 ± 16.17	7.43 <sup>c</sup> ± 0.60

\* Values are means ± standard deviation. Superscript letters indicate significant differences based on ANOVA and post-hoc tests ( $\alpha = 0.05$ ); means with the same letters within each column are not significantly different. Lag = Laguna, Bat = Batangas, Cav = Cavite.

Regarding yield, significant differences were observed across hybrids in Laguna and Cavite field trials. In Laguna, three hybrids had similar average yield to Sinta but significantly different with 4172. In Cavite trial, Liyag and Timyas produced the highest average yields and followed by Hirang. Representative prolific bearing trees of Hirang, Liyag, and Timyas are presented in Figure 3. In a similar study by Janthasri and Chaiyaboon (2015), yellow Krang, a hybrid of Red Krang x Sai Nampeung, had more flowers and fruits compared to control inbred lines. Furthermore, Yellow Krang had higher yield compared to Khaeg Dum Gosym and Khaeg Nuan, two of released Papaya Thailand varieties.



**Figure 3. Prolific fruit bearing trees of Hirang (A, D and G), Liyag (B, E and H), and Timyas (C, F, I) in Laguna (A-C), Batangas (D-F), and Cavite (G-I) field trials.**

Significant differences on mean weight of fruits were observed on the different hybrids planted in Laguna and Cavite trials ( $p=0.000$ ). Liyag produced the heaviest and longest fruits. Furthermore, significant differences in fruit width were only observed in the Cavite trial, where Hirang, Liyag, Timyas, and Sinta all had wider fruits than 4172. Liyag was observed to have thicker flesh like Sinta and 4172 but was significantly different from Timyas and Hirang in the Laguna field trial. While in terms of mean seed weight, significant differences were observed in Laguna and Cavite field trials, where Liyag, Timyas, and Sinta had the heaviest seed weight in the Laguna trial, while in the Cavite trial, Liyag had the heaviest seed weight. Aside from quantitative fruit traits, organoleptic properties must be determined since papaya is generally eaten fresh. The organoleptic traits such as sweetness and firmness should be considered. Fruits should have sweet and firm flesh. Moreover, Figure 4 shows the different representative fruits found in different locations while their internal appearance is shown in Figure 5.

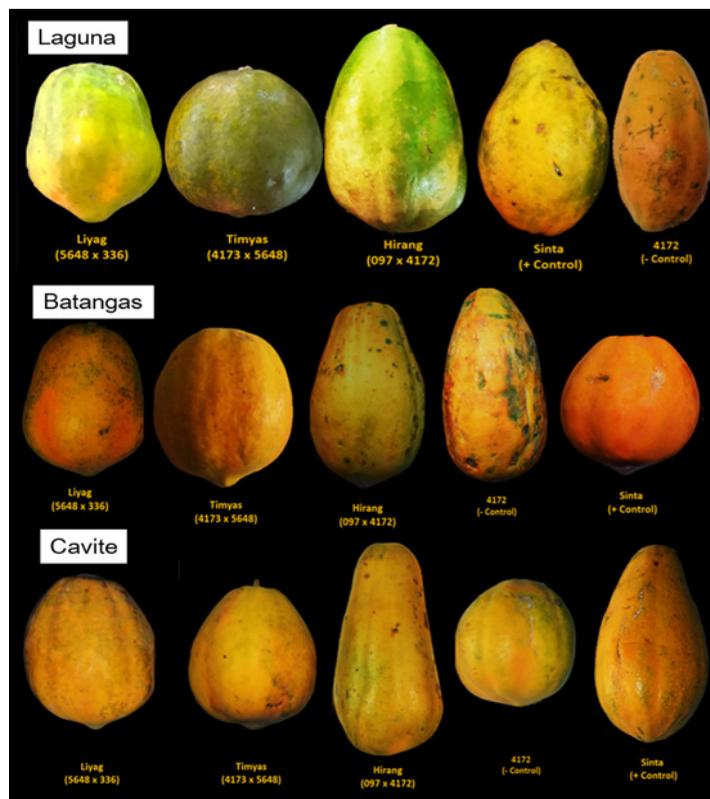


Figure 4. External appearance of mature fruits of Hirang, Liyag, Timyas, 4172, and Sinta across Laguna, Batangas and Cavite field trials.

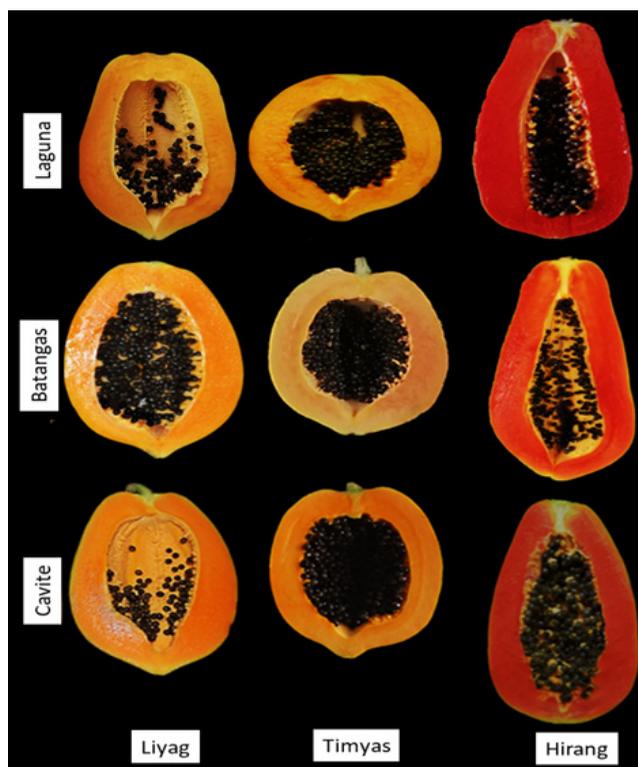


Figure 5. Internal appearance of mature fruits of HIRANG, LIYAG, TIMYAS, and 4172 across Laguna, Batangas and Cavite field trials.

Table 4. Fruit weight and fruit dimensions (length and width) of the three Papaya F1 hybrids in Laguna, Batangas, and Cavite

Hybrid	Fruit Weight (g)			Fruit length (mm)			Fruit width (mm)		
	Lag (p=0.000)	Bat (p=0.867)	Cav (p=0.000)	Lag (p=0.090)	Bat (p=0.026)	Cav (p=0.077)	Lag (p=0.056)	Bat (p=0.987)	Cav (p=0.044)
Hirang	1051.82 <sup>b</sup> ± 11.62	980.40 ± 162.63	1173.48 <sup>ab</sup> ± 58.55	180.86 <sup>b</sup> ± 1.71	178.26 ± 8.44	181.62 ± 7.42	108.80 ± 1.71	109.19 ± 8.86	116.10 <sup>a</sup> ± 7.43
Liyag	1338.74 <sup>a</sup> ± 58.86	1076.45 ± 320.66	1309.43 <sup>a</sup> ± 80.98	209.61 <sup>a</sup> ± 8.58	199.35 ± 26.08	201.04 ± 23.50	118.14 ± 5.35	109.93 ± 11.76	123.51 <sup>a</sup> ± 3.15
Timyas	936.48 <sup>b</sup> ± 77.53	1033.47 ± 162.07	1068.74 <sup>b</sup> ± 20.16	181.88 <sup>b</sup> ± 22.02	182.17 ± 11.91	164.71 ± 14.82	108.46 ± 4.04	113.61 ± 8.18	119.14 <sup>a</sup> ± 6.84
Sinta	1030.62 <sup>b</sup> ± 103.58	975.61 ± 217.65	1071.42 <sup>b</sup> ± 135.60	174.21 <sup>b</sup> ± 6.90	166.07 ± 15.58	163.93 ± 3.34	111.18 ± 7.83	108.55 ± 9.08	119.33 <sup>a</sup> ± 12.61
4172	968.06 <sup>b</sup> ± 48.35	1169.22 ± 330.21	846.42 <sup>c</sup> ± 47.17	190.80 <sup>ab</sup> ± 3.52	190.83 ± 15.67	186.17 ± 20.39	102.56 ± 5.85	111.33 ± 19.71	100.53 <sup>b</sup> ± 7.32

\* Values are means ± standard deviation. Superscript letters indicate significant differences based on ANOVA and post-hoc tests ( $\alpha = 0.05$ ); means with the same letters within each column are not significantly different. Lag = Laguna, Bat = Batangas, Cav = Cavite.

Total Soluble Solid (TSS) is the most critical parameter for the selection of this crop since it is consumed fresh (Magdalita and Signabon 2017). High TSS is an indicator that fruit flesh is sweet. TSS ranges from 6-17°Brix. High TSS is desirable for papaya, ranging from 8-12°Brix. Minuye and Zerihune (2020) reported that desirable papaya TSS in Ethiopia could range from 10-16°Brix. The three hybrids, including the controls, had high TSS. Comparable TSS was observed across hybrids in Laguna. However, significant differences were observed across hybrids in Cavite and Batangas trials. In Batangas and Cavite trials, Liyag had the highest TSS. However, all the hybrids and inbred 4172 have acceptable sweetness as they had high TSS (8-12°Brix). Firmness is another measured characteristic that refers to the texture of the flesh. Papaya flesh should neither be too soggy nor too hard. All four hybrids had firm flesh. The mean firmness was found not significantly different across varieties in all three locations and had mean firmness that ranged from 1.16-1.65 kg/cm<sup>2</sup>.

**Table 5. Total soluble solids and firmness of the three Papaya F1 hybrids in Laguna, Batangas, and Cavite**

Hybrid	Total Soluble Solids (°Brix)			Firmness (kg/cm <sup>2</sup> )		
	Lag (p=0.460)	Bat (p=0.038)	Cav (p=0.006)	Lag (p=0.094)	Bat (p=0.980)	Cav (p=0.370)
Hirang	10.70 ± 0.53	11.51 <sup>c</sup> ± 0.45	11.13 <sup>a</sup> ± 0.53	1.33 ± 0.03	1.31 ± 0.022	1.44 ± 0.044
Liyag	11.04 ± 0.52	13.00 <sup>a</sup> ± 0.45	11.68 <sup>a</sup> ± 0.38	1.27 ± 0.06	1.46 ± 0.31	1.44 ± 0.1858
Timyas	12.08 ± 2.11	11.68 <sup>bc</sup> ± 0.11	11.19 <sup>b</sup> ± 0.50	1.29 ± 0.12	1.48 ± 0.12	1.41 ± 0.33
Sinta	11.84 ± 0.31	12.64 <sup>ab</sup> ± 0.34	10.50 <sup>b</sup> ± 0.72	1.38 ± 0.22	1.51 ± 0.43	1.30 ± 0.17
4172	11.67 ± 0.21	11.58 <sup>bc</sup> ± 0.46	10.11 <sup>b</sup> ± 0.17	1.07 ± 0.11	1.65 ± 0.64	1.16 ± 0.043

\* Values are means ± standard deviation. Superscript letters indicate significant differences based on ANOVA and post-hoc tests ( $\alpha = 0.05$ ); means with the same letters within each column are not significantly different. Lag = Laguna, Bat = Batangas, Cav = Cavite.

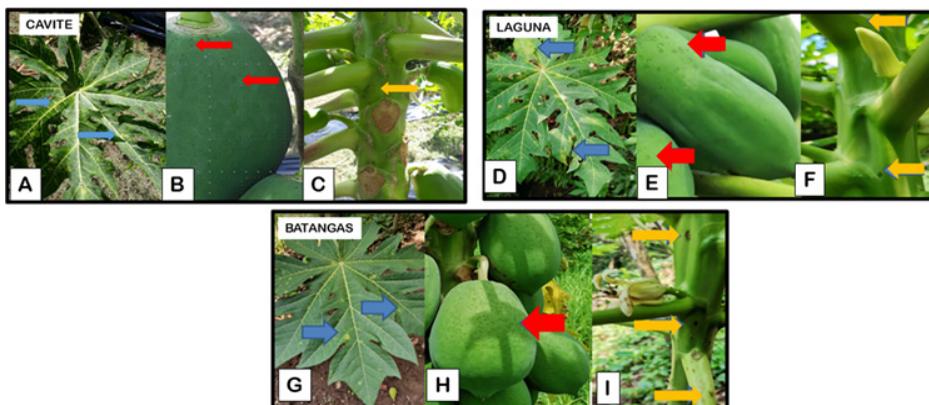
Comparable titratable acidities (TA) were observed among hybrids across locations. Mean TA ranges from 1.83-2.20 meq/10 mL juice. Minuye and Zerihune (2020) reported that low TA was acceptable to consumers which were in the range of TA in different cultivars of Papaya in Ethiopia (1.45-1.98 meq). Similarly, comparable high edible portion among varieties were observed in all three locations. Mean edible portions ranged from 78.30-87.21%.

**Table 6. Titratable acidity and edible portion of the three Papaya F1 hybrids in Laguna, Batangas, and Cavite**

Hybrid	Titratable Acidity (meq/10 mL juice)			Edible Portion (% EP)		
	Lag (p=0.089)	Bat (p=0.274)	Cav (p=0.081)	Lag (p=0.102)	Bat (p=0.790)	Cav (p=0.554)
Hirang	2.13 ± 0.20	2.20 ± 0.076	2.13 ± 0.21	83.31 ± 0.98	82.83 ± 5.36	86.89 ± 1.28
Liyag	2.10 ± 0.11	2.08 ± 0.189	2.00 ± 0.20	84.97 ± 0.70	82.81 ± 0.70	88.07 ± 0.75
Timyas	2.17 ± 0.12	2.08 ± 0.126	2.20 ± 0.19	83.01 ± 0.77	82.98 ± 2.09	86.76 ± 1.36
Sinta	1.93 ± 0.06	2.02 ± 0.076	2.07 ± 0.16	83.41 ± 1.08	80.79 ± 2.17	84.71 ± 2.89
4172	1.90 ± 0.09	1.92 ± 20.075	1.83 ± 0.1	84.62 ± 1.08	85.20 ± 0.76	85.74 ± 4.24

\* Values are means ± standard deviation. Lag = Laguna, Bat = Batangas, Cav = Cavite.

Regarding flesh color, Timyas had saffron yellow flesh (RHCC 22B). In addition, Liyag had maize yellow flesh (RHCC 21B) (Figure 6). Consumers prefer yellow-fleshed papayas in Southern Tagalog and Central Luzon areas. Since the introduction of Solo papaya, which had yellow flesh, the consumers have become used to eating yellow-fleshed papayas. Hirang had vermillion red (RHCC 41B) flesh. Red-fleshed papayas are preferred in Bicol region and by the Chinese communities. The canning and food-and-beverage industries also has preference for red-fleshed papayas.



**Figure 6. Symptoms observed in F1 hybrids in Laguna, Batangas, and Cavite trials. Hybrids show moderate tolerance to PRSV such as mild chlorosis (blue arrows in A, D, G), ringspots on fruit (red arrows in B, E, H), and water-soaked lesions in stem (yellow arrows in C, F, I).**

## Reaction to PRSV

The reaction of the hybrids to PRSV was assessed. Visual symptomatology rating indicated that hybrids were all moderately tolerant to PRSV. Visual symptoms of PRSV are presented in Table 7 and Figure 6.

**Table 7. Virus rating, symptoms observed and virus titer of the three Papaya F1 hybrids in Laguna, Batangas, and Cavite**

Hybrid	Virus Rating			Symptoms			Virus Titer (A450 nm)			
	Lag	Bat	Cav	Lag	Bat	Cav	Lag	Bat	Cav	
Hirang	MT	MT	MT	mRS	mMo, VB	mMo	0.54	0.875	0.39	
Liyag	MT	MT	MT	mC, mRS	mMo, mM	mMo	1.53	1.00	0.53	
Timyas	MT	MT	MT	mMo	mMo, mM	mM	0.86	0.75	0.12	
Sinta	MT	MT	MT	mMo	Mo, mM	mM, mVB	0.81	1.25	1.42	
4172	MT	MT	MT	Mo,VB	mC, mM	Mo, mM	0.61	1.375	0.76	
							-	0.02	0	0
							+	0.502	1	1

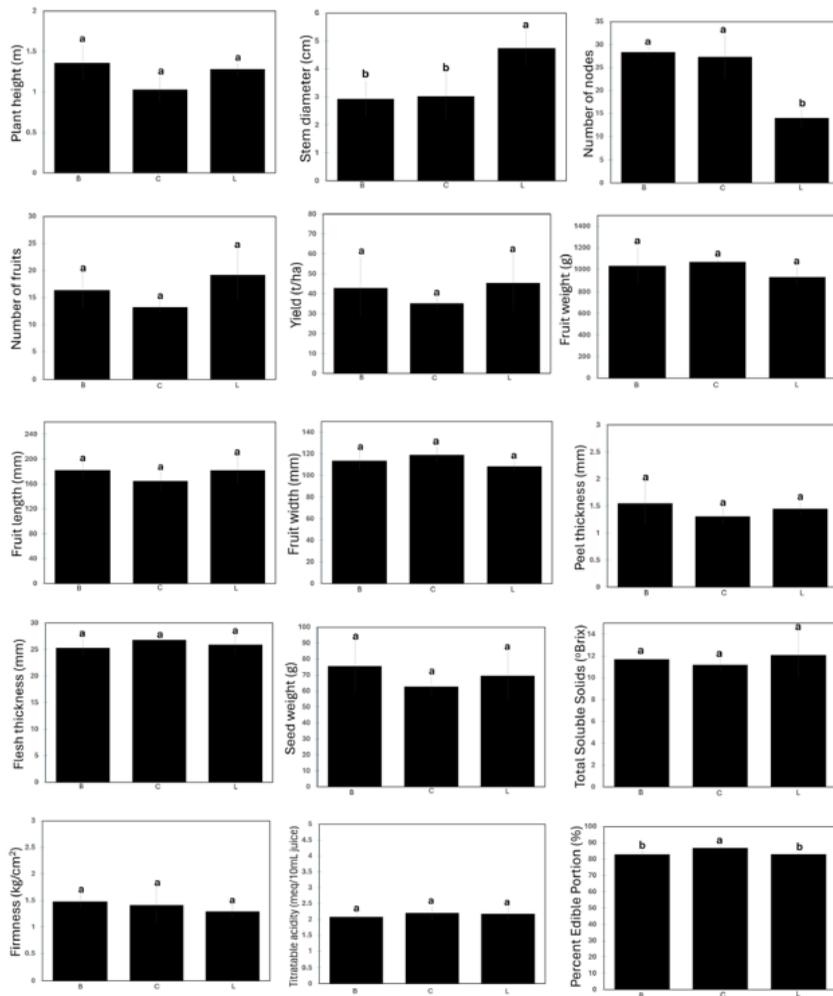
\* MT = moderately tolerant, mRS = mild ringspots, mC = mild chlorosis, Mo = mottling, mVB= mild vein banding, mM = mild mosaic, mMo = mild mottling, - = negative control, + positive control, Lag = Laguna, Bat = Batangas, Cav = Cavite.

Table 7 shows the virus titer of hybrids planted in Laguna, Batangas, and Cavite field trials. Moderate tolerance to PRSV was observed in all the hybrids and control. However, varying mild symptoms were observed across hybrids with common symptoms such as mild mottling, mild mosaic, and mild chlorosis. Some of the symptoms are shown in Figure 6. In Laguna, all hybrids had a mean visual rating of 3, indicating moderate tolerance to the virus.

This observation is similar to previous reports by Alviar et al. (2012), where Sinta and Cariflora had moderate tolerance to PRSV. High virus titer at A450 nm was observed in all hybrids and both positive and negative controls planted in Laguna. Liyag exhibited the highest virus titer, followed by Timyas, then Sinta, and finally Hirang. The data shows that all the hybrids were positive for PRSV. The virus titer results showed that the hybrids can tolerate the presence of the virus in Laguna and these can still produce marketable yield despite infection.

In Batangas, hybrids and controls were positive for PRSV but are rated 3, observing moderately tolerance since they produced marketable yield despite the presence of the virus. Furthermore, the results of the ELISA test for PRSV detection on collected samples from the hybrids planted in the Batangas field trial showed a higher mean score in Liyag and in the two controls than the positive control for ELISA. At the same time, the lower score for ELISA for PRSV was observed in Hirang and Timyas.

Lastly, in Cavite, a positive inoculum of PRSV was observed in Liyag, Sinta, and 4172. Liyag, when compared to ‘Sinta’ and 4172, showed tolerance to PRSV as seen in higher virus titer compared to the positive control. Timyas and Hirang showed lower virus titer. Even though there was a high inoculum, a low virus titer was observed in the two hybrids. The low virus titer indicates that the hybrids are not affected by the high inoculum of PRSV, hence the low virus titer. The different symptoms are shown in Figure 7 observed in Laguna, Batangas, and Cavite field trials.



**Figure 7. Size, yield, and fruit characteristics of papaya hybrid ‘Timyas’ grown in three locations (Batangas (B), Cavite (C), and Laguna (L)). Bars represent means and vertical lines indicate standard deviations. Different letters above bars denote significant differences among locations based on ANOVA and post-hoc tests ( $\alpha = 0.05$ ); means sharing the same letter are not significantly different.**

## Performance of individual hybrids across three locations

In addition to the influence of papaya genotypes environmental conditions contribute to variations expressed in these fruits (Morton and Miami 1987). Superior genotypes therefore, should meet minimum criteria first before recommendation for cultivation and release (da Luz et al. 2018).

### Timyas

When characteristics of Timyas were analyzed, majority of parameters measured for hybrid Timyas showed no significant differences as observed on mean plant height, number of fruits, and most importantly, yield (Figure 7). Only the average number of nodes to first flower and stem diameter showed statistical significance. Higher mean number of nodes was observed in allocations for Timyas, whereas the highest mean number of nodes to first flower was observed in Batangas and Cavite trials. Earlier flowering was observed in Laguna, and much later in Cavite and Batangas. For papaya, it is desirable to have a shorter stature with smaller number of nodes to first flower. In all three locations, semi-dwarf characteristics and prolific bearing of Timyas were observed, revealing that Timyas is moderately tolerant to PRSV.

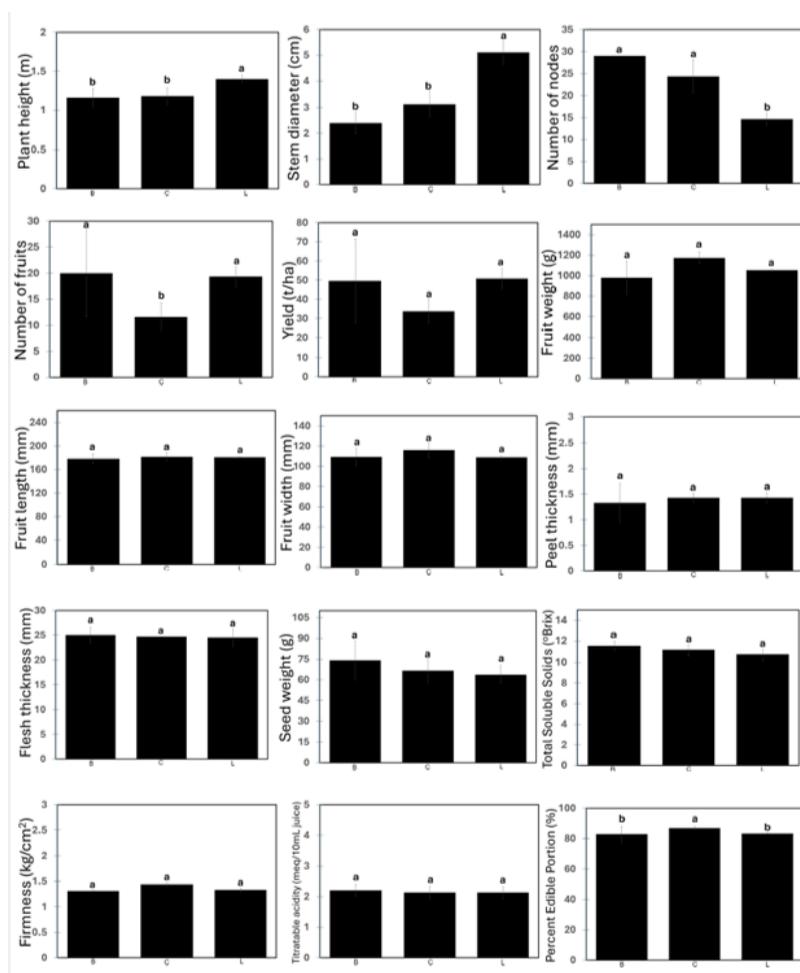
Timyas, in all three locations, had comparable mean fruit weight, fruit length, fruit width, peel thickness, flesh thickness and seed weight. Figure 6 shows that in all locations, fruits had uniform yellow orange flesh. Lastly, comparable total soluble solids, firmness, and titratable acidity were observed. However, percent edible portion was observed to be significantly different for hybrid Timyas in all locations. Timyas fruits harvested in Cavite had the highest (86.76%) edible portion. Analysis of performance of hybrid Timyas in all three locations showed that in most characteristics measured, fruit and fruit quality traits were all comparable except percent edible portion. However, Timyas had high (>80%) edible portion.

### Hirang

When tree, yield, and fruit characteristics of Hirang were analyzed, significant differences were observed in mean plant height, stem diameter, number of nodes, and number of fruits. In terms of plant height, trees were shortest in Cavite (1.18 m) and differed significantly in Batangas (1.16 m) and Laguna (1.40 m). Similarly, stem diameter was thickest in Laguna (5.11 cm) and different significantly with Batangas (2.4 cm) and Cavite (3.13 cm). Moreover, the highest number of nodes was observed in Batangas (29) and in Cavite (24.33). In terms of mean number of fruits, comparable mean number of fruits were observed in Batangas (19.99) and in Laguna (19.34) but was significantly different to the mean number of fruits of

Hirang in Laguna (11.63). Plant height, an important trait for resisting wind and rain damage from typhoons, showed that Hirang was semi-dwarf with a thick stem (>2.5 cm). In addition, high yields were observed for Hirang.

In all locations, comparable fruit characteristics of Hirang were observed (Figure 8), particularly its mean fruit weight, fruit length, fruit width, peel thickness, flesh thickness, seed weight, total soluble solids, firmness, and titratable acidity. Only percent Edible Portion (%EP) of fruits harvested was significantly different in all three locations. Highest %EP was observed in Cavite (86.89%), followed by Laguna (84.62%) and Batangas (82.81%).



**Figure 8.** Size, yield, and fruit characteristics of papaya hybrid ‘Hirang’ grown in three locations (Batangas (B), Cavite (C), and Laguna (L)). Bars represent means and vertical lines indicate standard deviations. Different letters above bars denote significant differences among locations based on ANOVA and post-hoc tests ( $\alpha = 0.05$ ); means sharing the same letter are not significantly different.

## Liyag

In Liyag, only stem diameter and number of nodes were found to be significantly different across locations for tree characteristics and yield. Liyag in Laguna (4.93 cm) field trial had thicker stems compared to those planted in Batangas (2.63 cm) and Cavite (3.13 cm). However, in terms of number of nodes to first flower, the least number of nodes to first flower was observed for Liyag planted in Laguna. Mean plant height ranged from 1.24-1.41 m while the mean number of fruits for Liyag ranged from 12.83-17.15 and the mean yield ranged from 41.85-48.33 MT (Figure 9).

In terms of fruit characteristics, only flesh thickness was observed to be significantly different across locations for Liyag. Mean fruit weight, fruit length, fruit width, peel thickness, and seed weight were comparable for Liyag fruits. Mean flesh thickness was observed to be highest in Laguna (27.32 mm) compared to Liyag fruits in Cavite (24 mm). Figure 5 shows that the yellow orange fruit color of Liyag is uniform across locations. For fruit quality traits, only TSS was significantly different across locations where the highest TSS was observed in fruits of Liyag grown in Batangas (13°Brix). The range of TSS of the hybrid was found to be desirable for the three locations.

In all locations tested for individual varieties, there is evidence of variation in plant characteristics. However, only few fruit characteristics varied in all locations. Despite these observed variations, plants were generally semi-dwarf, with high yield, produced sweet fruits and had high edible portion. In terms of qualitative characteristics such as flesh and peel colors, these expressions were uniform in all 3 locations.

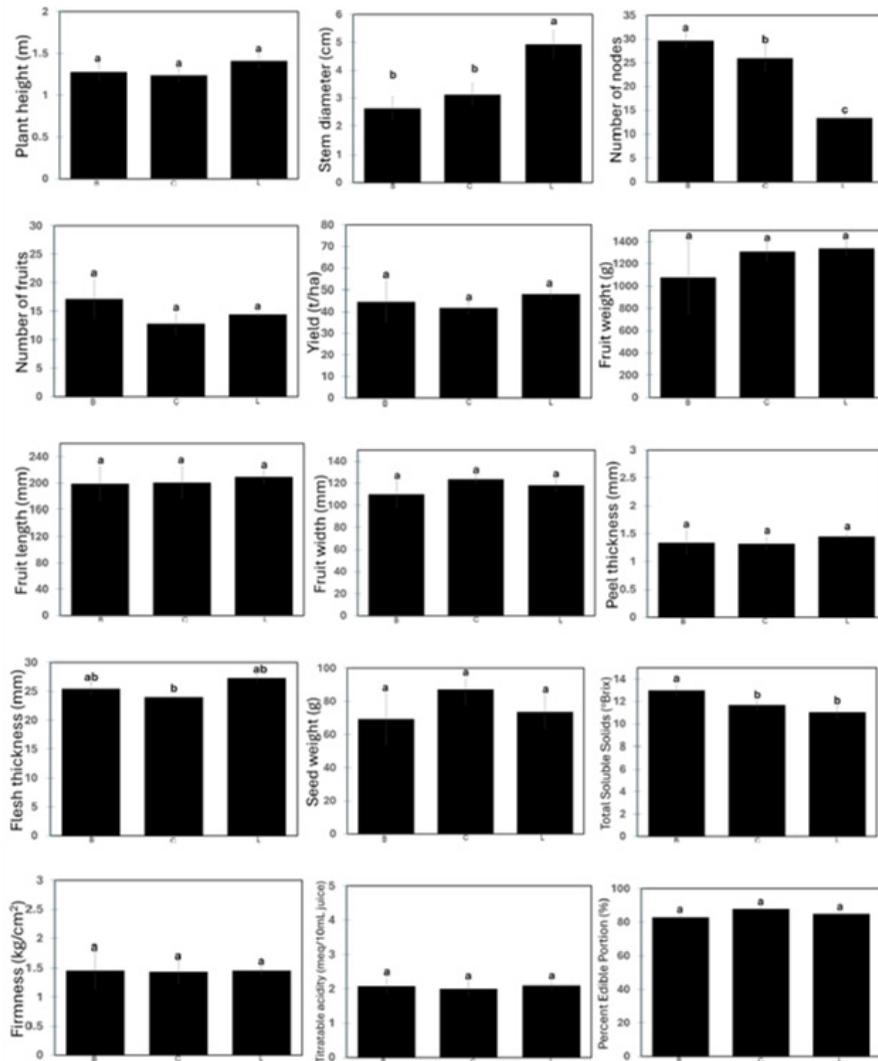


Figure 9. Size, yield, and fruit characteristics of papaya hybrid 'Liyag' grown in three locations (Batangas (B), Cavite (C), and Laguna (L)). Bars represent means and vertical lines indicate standard deviations. Different letters above bars denote significant differences among locations based on ANOVA and post-hoc tests ( $\alpha = 0.05$ ); means sharing the same letter are not significantly different.

## SUMMARY AND CONCLUSION

Papaya with PRSV tolerance has been developed in the Philippines to counter losses due to the Papaya Ringspot Virus. One effective approach to counter those losses is the development of PRSV tolerant  $F_1$  hybrids by crossing PRSV tolerant inbred lines, the first of which to be developed in this manner was Sinta Papaya. New  $F_1$  hybrids with PRSV tolerance, namely Hirang, Liyag, and Timyas are being developed, maintained, characterized, evaluated, and genetically advanced in IPB, UPLB. This study aimed to assess the performance of the hybrids in three locations: Laguna, Batangas, and Cavite.

Hirang (097 x 4172) produces vermillion red (RHCC 41B) fleshed fruits while Liyag (5648 x 336) produces maize yellow (RHCC 21B) fleshed fruits and Timyas (4173 x 5648) produces saffron yellow (RHCC 22B) fleshed fruits. All hybrids are moderately tolerant to PRSV, semi-dwarf and thick stemmed, has sweet, firm fleshed fruits with high edible portions.

In the Laguna field trial, all varieties have semi-dwarf characteristics with comparable stem diameter and number of nodes to the first flower. Further, more harvestable fruits were observed in the three hybrids and Sinta. Similarly, yield was higher in the three hybrids and Sinta than in 4172. Moreover, Liyag had the heaviest and longest fruits. All hybrids are moderately tolerant to PRSV with only mild symptoms such as chlorosis, ringspot and water-soaked lesions recorded. In terms of virus titer, Hirang, Liyag, Timyas and Sinta plants were all positive for PRSV but still produced marketable yield indicating moderate tolerance to PRSV.

In Batangas trial, comparable plant height, stem diameter, number of fruits, and yield were observed among the different varieties planted in the trial. Significant differences were observed in average number of nodes to the first flower. Further, similar fruit characteristics were observed. However, only TSS differed significantly among hybrids in fruit quality traits. The hybrids had high edible portion and low titratable acidity. Liyag was observed to have a similar mean TSS with Sinta, followed by Timyas and then Hirang. All hybrids planted in Batangas were sweet as indicated by high TSS. While in terms of ELISA, Liyag, Sinta, and 4172 showed an equally higher amount of virus than the positive control. Hirang and Timyas showed the presence of virus higher than the negative control but a little lower than the positive control. Liyag had the highest virus titer among the three hybrids, equal to the positive control. However, Liyag also showed prolific bearing with good quality fruits as observed with high TSS, low TA, and high edible portions.

Lastly, in Cavite field trial, comparable tree characteristics were observed. However, significant difference was observed in terms of average number of fruits and yield. Higher mean number of fruits was observed in the three hybrids compared to controls. Liyag and Timyas had higher yield, followed by Hirang and then Sinta. Regarding fruit characteristics, heavier fruits were observed in Liyag. Regarding fruit width, hybrids and Sinta had similar average fruit widths. While in terms of other fruit characteristics, hybrids showed comparable attributes. Regarding fruit quality traits, similar means of titratable acidity, % edible portion, and firmness were observed except TSS where Hirang and Liyag were sweeter than Timyas and Sinta. Furthermore, in terms of virus titer, Liyag had high virus titer compared to the positive control. Low virus titer intermediate of the positive and negative control was observed in Hirang and in Timyas. Liyag showed tolerance to PRSV.

In conclusion, hybrids Hirang, Liyag and Timyas is recommended for release in Luzon, Philippines because of their consistent performance across locations as observed in individual hybrid analyses. These hybrids are high yielders, moderately tolerant to PRSV, and have good quality flesh and fruit.

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