Stability of Yield and Yield Components of Pepper (*Capsicum annuum*), and Evaluation of Publicly Available Predictive Meteorological Data in East and Southeast Asia

Derek W. Barchenger²

The World Vegetable Center, Shanhua, Tainan 74151, Taiwan

Robert A. Clark, III

University of Oklahoma, Cooperative Institute for Mesoscale Meteorological Studies and National Oceanic and Atmospheric Administration, National Weather Service, National Centers for Environmental Prediction, Storm Prediction Center, Norman, OK 73072

Paul A. Gniffke¹, Dolores R. Ledesma, Shih-wen Lin, Peter Hanson, and Sanjeet Kumar

The World Vegetable Center, Shanhua, Tainan 74151, Taiwan

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Abstract. Multilocation trials are important for breeding programs to identify high-yielding, adapted lines for a wide range of environments. In this study, we evaluated yield and yield components (fruit weight, fruit length, and fruit width) as well as days to 50% anthesis and fruit maturity of the 10 chili pepper lines in the International Chili Pepper Nursery 15 (ICPN15) distributed by the World Vegetable Center to interested cooperators worldwide. Performance data of the ICPN15 entries were received from collaborators evaluating the set in seven different environments in five countries (Indonesia, South Korea, Thailand, Taiwan, and Vietnam). Significant genotype-by-environment ($G \times E$) interactions were detected for all traits evaluated. Additive main effect and multiplicative interaction analyses indicated high environmental influence on yield, days to 50% anthesis, and maturity, whereas genotype was the greatest contributor to variability in the market-driven yield components of fruit length, width, and weight. Four lines (ICPN15-4, -5, -7, and -10) were identified as highly stable and could serve as sources of yield and yield component stability in either short fruit market segments (ICPN15-4) or long fruit market segments (ICPN15-5, -7, and -10). We attempted to use publicly available weather data to help in explaining the source of the environmental variability; however, differences between analyzed and observed weather were too different to be useful. This is evidence that weather data should be collected at each testing environment in future studies. This study provides a basis for future studies in the stability of important horticultural traits in pepper, and highlights the need for further work in this area.

Pepper (*Capsicum annuum*) is an important crop worldwide, with an estimated 25% of people consuming some form (vegetable,

spice, or food colorant) of pepper daily (Smith, 2015). Originating in the Americas, peppers have been widely adopted into the cuisines of Africa, Asia, and Europe (Guzman et al., 2011; Sherman and Billing, 1999; Smith, 2015). In 2014, global harvested area of pepper was $\approx 3,600,000$ ha, with the vast majority ($\approx 65\%$) of production occurring in Asia (Food and Agriculture Organization of the United Nations, 2015). Peppers are a high-value crop (DeWitt and Bosland, 1993) and have short-term economic benefits for smallholder farmers (Kahane et al., 2013). Peppers are also important sources of provitamin A compounds (Guzman et al., 2011; Kantar et al., 2016) and vitamin C; thus, they have long-term nutritional benefits.

Pepper production occurs across a broad range of agroecological conditions, including

the humid tropics, deserts, and cool temperate climates (Bosland and Votava, 2012), and different systems ranging from open field to protected cultivation (sweet pepper). Productivity of pepper is often reduced by both biotic and abiotic stresses, the types of which can vary greatly among region. High-yield, disease-resistant cultivars are one of the most effective ways for farmers to increase productivity. Although no cultivar of a given crop is adapted everywhere, cultivars differ in the extent of their adaptation. A major objective of most breeding programs is high and stable yield and yield components. Breeders must examine whether a given cultivar is better adapted to a specific type of environment, and whether its performance is stable relative to that of other cultivars. Predictable performance over a broad range of conditions benefits farmers and seed producers by expanding the range of adaptation, and increased uniformity and potential sales. Consequently, potential cultivars are evaluated over locations and/or years to assess cultivar adaptation. $G \times E$ interaction is a major concern in cultivar development, testing, and release. This interaction occurs when the relative performance of genotypes changes in different environments. Partitioning of environments to reduce $G \times E$ interaction is challenging, especially in areas where there is extensive climatic variation.

Consumer preference of pepper is region specific and can vary greatly within a country (Bosland and Votava, 2012). Although studies in this area are limited, it has been observed that regional-specific preferences for pepper fruit shape, size, and color, and capsaicinoid content, as well as yield, are major driving forces for farmer cultivar selection. Similarly, for pepper, yield components include fruit length, fruit width, and fruit weight, among other traits. Therefore, evaluating the stability of yield and yield components of multiple market types in diverse environments is essential for successful pepper breeding. Unfortunately, studies to evaluate $G \times E$ interaction in *Capsicum* generally have focused on accumulation of capsaicinoids (Butcher et al., 2012; Gurung et al., 2011; Lee et al., 2005; Zewdie and Bosland, 2000) and not on the stability of yield or yield components (Gurung et al., 2012; Zewdie and Poulos, 1996). The World Vegetable Center (WorldVeg) pepper breeding program seeks to combine superior traits into broadly useful backgrounds of hot peppers for use in diverse production regions around the world. Adaptation to warm, humid climates; high yield potential; yield stability; and multiple disease resistance are the key breeding goals. Acceptable fruit shape, size, heat, and flavors are also critically important for adoption. Breeding activities and line development are carried out in Taiwan.

The ICPN at WorldVeg was initiated during the early 1990s as a platform to distribute sets of improved pepper lines to interested cooperators around the world, and as an opportunity to gather performance data

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²Corresponding author. E-mail: Derek.barchenger@worldveg.org.

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from diverse ecological settings. Nursery participants volunteer to conduct the trial and have the opportunity to evaluate new pepper lines, and to identify those that have potential as new cultivars or as parents in localized breeding programs. The multilocation data of ICPN can provide valuable information on $G \times E$ interactions that can be used in improving pepper cultivar development and testing programs. The additive main effect and multiplicative interaction (AMMI) analysis can be an effective model to assess stability of genotypes because it captures a large portion of the $G \times E$ sum of squares and allows for the separation of main and interaction effects, providing meaningful interpretation of the data (Pacheco et al., 2016).

Our objectives were to characterize the presence and relative magnitude of genotype, environment, and $G \times E$ interaction in the ICPN15 and to evaluate the performance and stability of entries in the ICPN15, while determining the utility of publicly available meteorological data as a tool to determine the effects temperature and precipitation play on the stability of yield in pepper.

Materials and Methods

The ICPN15 was composed of 10 highly inbred lines (Table 1), including ICPN15-4 and ICPN15-7, long-term checks representing medium (10 ± 2 cm) and long (15 ± 2 cm) fruit lengths, respectively. Selection of lines for inclusion in ICPN15 was based on resistance to bacterial wilt pathogen (*Ralstonia solanacearum*), *Potato virus Y* (PVY), and *Cucumber mosaic virus* (CMV) (Table 1), and overall performance (yield, plant vigor, etc.) in combination with diverse fruit characteristics (size, pungency, and color) during preliminary evaluation (data not shown). Although all breeding and selection were done in Taiwan, the parental material for the ICPN15 lines originated from diverse regions such as India (Pant C1), Malaysia (Kulim and MC-12), France (HDA 248 and HDA 295), and the Philippines (Kawit). Co-operators were requested to adhere to a uniform protocol for plot size, row length,

plant spacing, data collection, and experimental design (Lin et al., 2014). A total of 103 collaborators in 36 different countries received ICPN15, but only 21 provided feedback. Due to inconsistencies in experimental design, incomplete data, or other problems such as high disease incidence (resulting in missing data points), on seven locations were sufficient to analyze traits of interest for $G \times E$ interaction and stability analysis.

For this study, two locations in Taiwan and Vietnam and one location each in Indonesia, South Korea, and Thailand were selected. In Vietnam, the ICPN15 experiment was conducted at the Southern Horticulture Research Institute (Vietnam-1) (lat. 10.4°N , long. 106.3°E) in a clay loam soil as well as at a farm in the Phu Giao District, Binh Duong Province (Vietnam-2) (lat. 11.3°N , long. 106.8°E) in a well-drained sandy loam soil following a crop of bitter melon (*Momordica charantia* L.) and bottle gourd [*Lagenaria siceraria* (Molina) Standl.]. The ICPN15 lines were planted in a red-yellow podzolic soil with a pH of 6.5 in Campaka, Purwakarta, West Java, Indonesia (lat. 6.5°S , long. 107.7°E ; elevation, 50 m) following a pepper (*C. annuum*) crop. In South Korea, the experiment was conducted at the National Horticultural Research Institute in the city of Suwon, Kyonggi Province (lat. 37.3°N , long. 126.9°E ; elevation, 50 m) following a Chinese cabbage (*Brassica rapa* ssp. *pekinensis*) crop in a silt sandy loam (pH 6.8). The experiment was conducted in Suphanburi, Thailand (lat. 14.5°N , long. 100.0°E) in a loamy clay soil with a pH of 6.5 to 7.0 following a cucumber (*Cucumis sativus*) crop. The experiment was also conducted at the WorldVeg in Shanhua, Tainan, Taiwan (lat. 23.1°N , long. 120.3°E ; elevation, 50 m) over two seasons (summer and fall) in two different sandy loam fields that were previously planted in *Sesbania grandiflora* (Taiwan-1) and rice (*Oryza sativa*) (Taiwan-2) with a pH of 7.5 and 7.7, respectively.

Most cooperators did not or were not able to provide weather data (air temperature and daily rainfall accumulation) during the trial,

and such information is important in understanding possible reasons for differences in overall performance among locations, and underlying causes of $G \times E$ interactions. To fill this gap, daily maximum and minimum temperatures during the experiment in each of the trial environments were obtained from the National Oceanic and Atmospheric Administration's Global Forecast System (GFS) (www.ncdc.noaa.gov; Global Climate & Weather Modeling Branch, 2016) for analysis for each experimental site. The GFS predicts global weather conditions by assimilating millions of weather observations (from surface observation sites, weather balloons, aircraft, satellites, weather radar, and other sources) into an "analysis," or best guess, of the state of Earth's atmosphere at model runtime. Then, the equations governing the behavior of the atmosphere are integrated forward in time to produce a "forecast" of the state of the atmosphere at several forecast hours. We used outputs from the analysis step in this study so that errors resulting from forecast uncertainty are minimized. The horizontal grid spacing of the analyses is $1^\circ \times 1^\circ$. New GFS analysis and forecast cycles begin every 6 h, so daily maximum and minimum temperatures were calculated for each experimental site based upon these 6-h data. We obtained estimates of precipitation from the National Aeronautics and Space Administration and the Japan Aerospace Exploration Agency's joint Tropical Rainfall Measuring Mission (TRMM). One of the products from TRMM is the 3-h TRMM 3B42 V7 Multisatellite Precipitation Analysis (<https://trmm.gsfc.nasa.gov/>; Huffman et al., 2007). This precipitation analysis uses information from space-borne microwave and radar instruments, which are then adjusted with in situ information from rain gauges to create quasi-global (50°N to 50°S) analyses of accumulated rainfall every 3 h at a resolution of $0.25^\circ \times 0.25^\circ$. The inclusion of rain gauge reports is intended to correct for errors inherent in the process of estimating precipitation from space-borne instruments. The 3-h accumulated rainfall analyses were summed by month to yield total estimated monthly accumulations of rainfall at each experimental

Table 1. Summary of International Chili Pepper Nursery 15 (ICPN15) lines evaluated in seven locations in East and Southeast Asia.

ICPN code	Name	Parentage ^a	Disease resistance ^b				
			BW	CVMV	Pc3	CMV	PVY
ICPN15-1	VI059328	Pant C1	R ^c	S	S	R	R
ICPN15-2	AVPP0302	Long Chili/TM-888	R	S	R	R	R
ICPN15-3	AVPP0303	Shiang Yen No. 3/HyHot3	S	S	S	S	S
ICPN15-4	AVPP9813	Kulim/HDA 295	R	S	S	R	R
ICPN15-5	AVPP0304	Field62-selection-1/MC-12	R	S	MR	MR	R
ICPN15-6	AVPP0305	Kawit/MC-12	R	S	MR	R	MR
ICPN15-7	AVPP9905	Susan's Joy	R	R	S	MR	R
ICPN15-8	AVPP0306	VC232/HiHot3 selection	MR	S	S	R	R
ICPN15-9	AVPP0105	Kulim/HDA248	R	R	S	R	R
ICPN15-10	AVPP0307	LongThick/9852-173	R	S	S	MR	MR

^aAll the ICPN15 lines evaluated in this study were highly inbred. The lines are either the result of a single cross followed by seven to nine generations of selection and self-pollination (two parents listed) or a selection made within a segregating population of an accession or landrace followed by seven to nine generations of pure line selection and self-pollination.

^bDisease screening was conducted in the screenhouse at the World Vegetable Center, Shanhua, Tainan, Taiwan, using pathogen isolates from Taiwan. The diseases are: BW = bacterial wilt (*Ralstonia solanacearum*); CVMV = *Chili vein mottle virus* (aphid-transmitted potyvirus); Pc3 = *Phytophthora capsici* race 3; CMV = *Cucumber mosaic virus* (aphid-transmitted cucumovirus); PVY = *Potato virus Y* (aphid-transmitted potyvirus).

^cResistance scores are: R = resistant (50% to 100% resistant); MR = moderately resistant (10% to 49%); S = susceptible (<10%).

site. Daily maximum and minimum temperature as well as precipitation were recorded at the Taiwan-1 and Taiwan-2 locations. Because of the differences in weather conditions as analyzed in a global weather model and measured in situ, these data were not included in the $G \times E$ or correlations analyses.

The ICPN15 lines were planted on 30-cm-high raised beds with two rows. The plot size was 5.4 m long and 1.5 m wide between furrows, with between- and within-row spacing of 0.5 m and ≈ 45 cm, respectively (total of 24 plants/plot with 12 plants/row). The total plant density was 29,630 plants/ha. The plots were managed as uniformly as possible according to best recommended management practices (Berke et al., 2005), which included supplemental irrigation at each location to prevent drought stress.

The collaborators collected data on important traits associated with yield and yield components. Yield was calculated based on the weight of the marketable fruit (those free from damage associated with biotic or abiotic stress) recorded weekly for 10 weeks. The individual weekly yields were summed to calculate total plot yield. Total plot yield was converted into tons per hectare using the following formula: Yield ($\text{ton}\cdot\text{ha}^{-1}$) = {[total plot yield (kg) / 1,000 ($\text{kg}\cdot\text{ton}^{-1}$)] / [harvested area (m^2) / 10,000 ($\text{m}^2\cdot\text{ha}^{-1}$)]}. Average fruit length (measured in centimeters), average fruit width (measured in centimeters), and average fruit weight (measured in grams) were calculated using 10 randomly selected marketable fruits from the second harvest. Collaborators were also asked to record the number of days after transplanting (DAT) to 50% anthesis and

50% maturity, which was when 50% of the plants in a plot have open flowers at the second node and have fully ripe fruit, respectively.

Plots were arranged in a randomized complete block design with three replications randomized at each location. The combined analysis of variance (ANOVA) and correlations using Pearson's correlation coefficient for yield and yield components, as well as for days to 50% anthesis and maturity across all environments were calculated using R (version 3.4.1). The $G \times E$ analysis with R for Windows (version 4.0) (Pacheco et al., 2016) was used for AMMI analysis.

Results

F tests from the combined ANOVA indicated significant $G \times E$ interactions for yield ($P \leq 2 \times 10^{-16}$) and the yield components of fruit length ($P = 3.1 \times 10^{-14}$), fruit width ($P = 4.92 \times 10^{-8}$), and fruit weight ($P \leq 2 \times 10^{-16}$), as well as days to 50% anthesis ($P \leq 2 \times 10^{-16}$) and maturity ($P = 0.0409$). As a result of the highly significant $G \times E$ mean squares for all traits (Table 2), AMMI analysis was conducted to assess the interaction component more fully.

The AMMI analysis revealed that environments, genotypes, and $G \times E$ interactions were highly significant for yield, fruit width, fruit weight, fruit length, and days to 50% anthesis ($P \leq 0.0001$) (Table 3). For yield, the environment, genotype, and $G \times E$ interaction accounted for 77%, 2%, and 21% of the treatment combinations of sums of squares, respectively (Table 3). For days to 50% anthesis, environment was by the far the greatest contributor to the percentage of the treatment combination sum of squares (99%). Fruit width, fruit length, fruit weight, and genotype accounted for most of the treatment combination sums of squares (64%, 64%, and 56%, respectively), followed by environment (25%, 25%, and 28%, respectively), and $G \times E$ (11%, 11%, and 16%, respectively). For days to 50% maturity, the main effects of environment ($P \leq 0.0001$) and genotype ($P = 0.03$) were significant and accounted for 98% and 0.7% of the treatment combination sums of squares, respectively (Table 3).

The AMMI biplots with main effects on the abscissa and the PCA scores for both genotypes and environments as the ordinates are presented in Fig. 1. For the traits evaluated here, environments generally had high variability in both main effects and interactions (Figs. 1 and 2, and Table 3). The South Korea, Taiwan-1, and Vietnam-2 sites exerted the least interactive forces on yield (Fig. 1A). The ICPN15 lines 9, 7, and 10 were the greatest yielding lines (13.9, 13.0, and 12.7 $\text{t}\cdot\text{ha}^{-1}$, respectively), whereas ICPN15-6, -8, -9, and -10 had the most stable yield across the growing environments, although ICPN15-8 had less yield than the grand mean (Fig. 1A). However, the majority of the ICPN15 lines performed differently in each environment, illustrating the large $G \times E$ interaction (Fig. 2). The ICPN15 lines had the greatest average yield when grown in the Taiwan-2 site (24.7 $\text{t}\cdot\text{ha}^{-1}$); they had the least yield when grown at the Vietnam-1 and Thailand sites (2.7 $\text{t}\cdot\text{ha}^{-1}$ and 1.4 $\text{t}\cdot\text{ha}^{-1}$, respectively) (Figs. 1A and 2A). South

Table 2. Means squares in the combined analysis of variance across seven locations in East and Southeast Asia for the 10 International Chili Pepper Nursery 15 lines.

	df ²	Yield ($\text{t}\cdot\text{ha}^{-1}$)	Fruit wt (g)	Fruit width (cm)	Fruit length (cm)	50% Anthesis (DAT) ³	50% Maturity (DAT)
Genotype (G)	9	43.0***	447.5***	1.98***	90.5***	68***	169***
Environment (E)	6	2,172.7***	336.9***	1.16***	52.9***	17,499***	37,782***
$G \times E$	54	64.6***	21.4***	0.06***	2.6***	12***	58*
Replication (E) ^x	14	29.5***	1.3 NS	0.01 NS	0.9 NS	8**	441***
Residuals	126	4.3	1.6	0.02	0.5	4	39
Total	209						

²Degrees of freedom.

³Days after transplanting.

^xReplication as a function of environment.

NS, *, **, ***Nonsignificant or significant at $P < 0.05$, 0.01, or 0.001, respectively.

Table 3. Additive main effects and multiplicative interaction analysis of variance including the first six principal component analysis (PCA) axes.

	df ²	Yield ($\text{t}\cdot\text{ha}^{-1}$)		Fruit wt (g)		Fruit width (cm)		Fruit length (cm)		50% Anthesis (DAT) ^x		50% Maturity (DAT)	
		Mean sq	Var % ^y	Mean sq	Var %	Mean sq	Var %	Mean sq	Var %	Mean sq	Var %	Mean sq	Var %
E ^w	6	2,172.7***	77.1	336.9***	28.1	1.16***	25.1	52.9***	24.9	17,499.2***	98.8	37,782.4***	98.0
G	9	43.0***	2.3	447.5***	55.9	1.98***	64.0	90.5***	64.0	67.8***	0.6	169.2*	0.7
$G \times E$	54	64.6***	20.6	21.4***	16.0	0.06***	10.9	2.6***	11.1	12.2***	0.6	57.7 NS	1.3
PCA1	14	107.7***	43.2	55.4***	67.2	0.10***	45.9	5.4***	53.1	23.3***	49.5	183.5**	82.4
PCA2	12	93.6***	32.2	21.2***	22.1	0.07***	27.7	3.3***	27.8	16.0***	29.1	22.8 NS	8.8
PCA3	10	46.1***	13.2	6.2**	5.3	0.04**	14.3	1.2*	8.7	9.4*	14.2	19.2 NS	6.2
PCA4	8	45.8***	10.5	5.2**	3.6	0.02 NS	5.7	1.2*	6.5	4.5 NS	5.4	8.6 NS	2.2
PCA5	6	3.4 NS	0.6	3.0 NS	1.6	0.02 NS	4.6	0.6 NS	2.4	1.5 NS	1.4	2.1 NS	0.4
PCA6	4	2.8 NS	0.3	0.6 NS	0.2	0.01 NS	1.7	0.5 NS	1.4	0.6 NS	0.3	0.2 NS	0.0
Res	140	6.8		1.5		0.02		0.5		4.2		79.5	

²Degrees of freedom.

³Variance proportion for analysis of variance and principal component analysis.

^xDays after transplanting.

^wE = environment, G = genotype, $E \times G$ = environment $\times$ genotype, PCA = principal component analysis, Res = residuals.

NS, *, **, ***Nonsignificant or significant at $P < 0.05$, 0.01, or 0.001, respectively.

Fig. 1. Additive main effects and multiplicative interaction analysis biplots of the yield and yield component, and days to 50% anthesis and maturity means, and the first principal component axis score of 10 International Chili Pepper Nursery 15 lines (1–10) (○) and seven environments (●) (1 = Indonesia; 2 = South Korea; 3 = Thailand; 4 = Taiwan-1; 5 = Taiwan-2; 6 = Vietnam-1; and 7 = Vietnam-2). PCA = principal component analysis; DAT = day after transplanting.

Korea, Taiwan-1, and Vietnam-2 had negative and smaller interactions compared with other locations.

Lines ICPN15-7, -3, and -5 had the greatest fruit weight (19.5, 18.3, and 15.6 g, respectively), whereas ICPN15-1, -5, and

-10 had the most stable fruit weight (Fig. 1B). ICPN15-2 and -8 had the least fruit weight (6.9 and 6.9 g, respectively) (Fig. 1B). The least interactive forces on fruit weight was observed when the ICPN15 lines were grown in South Korea; the strongest interac-

tion occurred in Taiwan-2 and Thailand, and average fruit weight was greatest in Indonesia and Taiwan-2 (15.0 and 14.9 g, respectively) (Figs. 1B and 2B).

ICPN15-3 and -7 had the longest average fruit length (14.3 and 14.9 cm, respectively)

Fig. 2. Average yield, yield components, and days to 50% anthesis for the 10 International Chili Pepper Nursery 15 (ICPN15) lines averaged within each environment. DAT = day after transplanting.

ICPN15-1, -4, and -8 had the shortest (9.3, 9.5, and 8.7 cm, respectively), and ICPN15-1, -7, -8, and -10 generally had among the lowest environmental interactions (Fig. 1C). Most locations had strong interactive forces on fruit length, with South Korea and Vietnam-1 having the least (Fig. 1C). For fruit width, the PCA1 scores for most ICPN15 lines were generally low, with ICPN15-3 and -10 having the greatest environmental interaction (Fig. 1D). ICPN15-7 as well as ICPN15-5 and -3 had the widest fruit (2.0, 2.0, and 1.9 cm, respectively), whereas ICPN15-2, -8, and -6 had the narrowest fruit (1.2, 1.2, and 1.5 cm, respectively) (Fig. 1D).

Environment strongly influenced days to 50% anthesis (Fig. 1E). ICPN15-1, -3, and -10 showed the most stability for date to 50% anthesis (Fig. 1E). ICPN15-1 and -8 were latest for days to 50% anthesis (57.8 and 59.1 DAT, respectively), and ICPN15-2, -6, and -4 had the earliest (53.2, 54.0, and 54.6 DAT, respectively) (Fig. 1E). Similar to days to 50% anthesis, ICPN15-1 and -8 had the latest days to 50% maturity (105.5 and 103.3 DAT, respectively), whereas ICPN15-5, -7, and -10 had the shortest (95.4, 98.4, and 98.5 DAT, respectively) (Fig. 1F). The lines with the most stable days to 50% maturity were ICPN15-2, -4, and -8 (Fig. 1F). When grown in Indonesia, Vietnam-1, and Vietnam-2, the ICPN15 lines had the earliest days to 50% anthesis (32.4, 51.2, and 25.0 DAT, respectively) and maturity (63.1, 77.6, and 61.4 DAT, respectively) (Figs. 1E and 1F), whereas average days to 50% anthesis (96.1 and 74.6 DAT, respectively) and maturity (145.9 and 142.9 DAT, respectively) was greatest South Korea and Taiwan-1 (Figs. 1E and 1F).

The typical diurnal temperature range during the production period at the sites in Indonesia, Thailand, and Vietnam was relatively large compared with the ranges seen in South Korea and Taiwan (Fig. 3). On the other hand, in South Korea and Taiwan, the effects of season upon average daily temperature during the production period are readily apparent. Observed differences at the Taiwan location in both environments were considerably greater than the predicted model, but still less than the analyzed differences in Thailand (Fig. 3). The average analyzed temperatures in South Korea and Indonesia were relatively similar to one another (21.6 and 21.9 °C, respectively) as were the average analyzed temperatures at the Taiwan-1, Taiwan-2, Thailand, Vietnam-1, and Vietnam-2 sites (27.6, 25.6, 26.8, 27.3, and 27.6 °C, respectively) during the production period (Fig. 3). The average observed temperatures in the Taiwan-1 and Taiwan-2 environments were 27.3 and 23.3 °C, respectively.

Estimated monthly rainfall was relatively stable for the locations in South Korea, Vietnam-1, and Vietnam-2, excluding the partial months of May in South Korea and June in Vietnam-1 (Table 4). The estimated monthly rainfall at Taiwan-1, Taiwan-2, Thailand, and Indonesia was much more variable (Table 4). Taiwan-2 and Thailand had greater precipitation early during the production season, which diminished to nearly 0 mm toward the end of the season (Table 4). Precipitation was greatest in the middle of the production season and was least early and late in the season at Taiwan-1 and Indonesia (Table 4). The observed precipitation for the Taiwan-1 and Taiwan-2 locations

followed a similar pattern as the multisatellite estimates. However, the precipitation was greatly underestimated for nearly every month during the season at Taiwan-1. Precipitation was overestimated during September at Taiwan-1, but was underestimated in other months. The exception to the difference in estimated and observed precipitation is when there is no precipitation, such as in August and December (Table 4).

As expected, there were significant positive correlations between fruit length ($r = 0.75$) and fruit width ($r = 0.94$) with fruit weight, as well as days to 50% anthesis and maturity ($r = 0.8$) (Table 5). However, no other significant correlations among the yield components evaluated in this study were identified (Table 5).

Discussion

As a result of the significant interaction of $G \times E$, the usefulness of genotype mean as a single parameter to measure stability is limited (Pritts and Luby, 1990). Therefore, a hybrid model that incorporates additive (combined ANOVA) and multiplicative components (PCA), such as AMMI, is preferred (Shaffi and Price, 1992). In the AMMI model, the additive portion of the variance is separated from the multiplicative variance (interaction) by ANOVA, and PCA analysis is then applied to the interaction component from the ANOVA to extract a new set of coordinate axes that account more effectively for the interaction patterns (Shaffi and Price, 1992). Estimation of the PCA axes is accomplished according to the least-squares principle (Bradu and Gabriel, 1978). For the AMMI biplot display, points on a perpendicular line

Fig. 3. Analyzed daily maximum (●) and minimum (●) temperatures of the seven growing environments during the International Chili Pepper Nursery 15 (ICPN15) experiment. These data were obtained from the Global Forecast System (GFS) (www.ncdc.noaa.gov) using the GPS coordinates at each experimental location.

Table 4. Total/average daily estimated precipitation (measured in millimeters) per month at seven growing environments and observed precipitation for two Taiwan environments in International Chili Pepper Nursery 15. These data were collected from the Tropical Rainfall Measuring Mission 3B42 V7 multisatellite precipitation analysis using the GPS coordinates at each location.

Month	South Korea	Vietnam-1	Vietnam-2	Taiwan-1	Taiwan-1 observed	Taiwan-2	Taiwan-2 observed	Indonesia	Thailand
April				24.5/0.8	32.0/0.8				
May	10.5/0.7			87.6/2.8	228.0/7.4				
June	81.0/2.7	4.7/0.6		319.1/10.6	1679.0/56.0				
July	89.5/2.9	74.7/2.4	149.4/5.0	200.9/6.5	656.0/21.2				
August	82.1/2.6	73.6/2.4	129.1/4.2	0.0/0.0	0.0/0.0				1.3/0.1
September	88.9/3.3	64.6/2.2	150.1/5.0			44.4/2.3	20.0/1.1		119.1/4.0
October		86.8/9.6	96.3/7.4			21.1/0.7	94.0/3.0		40.9/1.3
November						3.7/0.1	8.0/0.3	47.0/4.7	43.9/1.5
December						7.9/0.2	10.0/0.3	83.8/2.7	6.4/0.2
January						0.0/0.0	0.0/0.0	161.2/5.2	0.0/0.0
February								61.7/2.2	8.9/0.3
March								45.0/5.6	0.0/0.0
Total	352.0/2.6	304.4/2.8	524.9/5.1	632.1/5.2	2595.0/21.3	77.1/0.6	132.0/1.1	398.7/3.7	220.5/1.1
Average	70.4	60.9	131.2	126.4	519.0	15.4	26.4	79.7	27.6

Table 5. Pearson correlation coefficients (*r*) for yield and yield components, and days to 50% anthesis and maturity.

	Yield (t·ha ⁻¹)	Fruit wt (g)	Fruit length (cm)	Fruit width (cm)	50% Anthesis (DAT ^a)	50% Maturity (DAT)
Yield (t·ha ⁻¹)	1.00					
Fruit weight (g)	0.22	1.00				
Fruit length (cm)	0.28	0.75*	1.00			
Fruit width (cm)	0.30	0.94**	0.56	1.00		
50% Anthesis (DAT)	0.13	-0.08	0.29	-0.22	1.00	
50% Maturity (DAT)	0.05	-0.06	0.14	-0.25	0.80*	1.00

^aDays after transplanting.

*, **Correlation coefficient values significant at $P \leq 0.05$ or 0.01, respectively.

have similar means; those that fall almost on a horizontal line have similar interaction patterns. Genotypes or environments with large PCA1 scores (either positive or negative) have high interactions, whereas genotypes or environments with PCA1 scores near zero have small interactions. The genotypes with low PCA1 values can be considered

more stable; environments with low PCA1 values are considered to have less of an influence on genotype means compared with those with greater PCA1 values.

We found significant $G \times E$ interaction effects for the yield and yield components evaluated here, indicating that the ICPN15 lines responded differently in different grow-

ing environments. For yield of the ICPN15 lines, environment was the largest contributor to variability observed in this study, which supports the findings of Gurung et al. (2012). Environment also played a major role in the observed variability for days to 50% anthesis and maturity. However, individual yield components such as fruit length, width, and

weight were influenced more strongly by the main effect of genotype (Table 3), which differs from the findings of Cabral et al. (2017). The high influence of environment on yield indicates that more testing is needed in diverse environments in the breeding program. It is also possible that the environments in this study were too diverse, resulting in high environmental effects. It might be useful to analyze the data in terms of broad macroenvironments in future studies; however, as a result of the limited number of environments tested here (seven), such an analysis was not possible.

Stommel and Griesbach (2008) found that fruit shape (fruit length/fruit diameter at midpoint) had high broad-sense heritability ($H = 0.97$) and relatively low narrow-sense heritability ($h^2 = 0.25$), indicating a large genetic component and smaller environment component, with predominantly dominant and epistatic gene action. They also reported fruit shape was simply inherited with several minor effect-modifying genes. The high genetic and lower environmental component associated with the variability in fruit shape (Stommel and Griesbach, 2008) is supported by our results.

Varietal preferences for pepper fruit length and width are market driven; therefore, it was important to parse yield into yield components for evaluation. For example, ICPN15-7 was a high-yielding line across all the environments in our study, but ICPN15-7 also produced long fruit. If the preferred market type in a given region is short fruit, ICPN15-7 will not be a good choice for the producers in that region. Although fruit length and width correlated highly with fruit weight, fruit weight did not correlate significantly with yield. This suggests that yield might be more influenced by traits such as fruit number per plant, although this was not measured in this study. Likely resulting from the large genotype effect, the ICPN15 lines were generally stable for fruit length and width across growing environments. Therefore, the target market should be selected early on in the breeding program because these traits are easily fixed, and should focus on improving other traits such as early maturity and high yield within specific market types.

In general, the ICPN15 lines had the greatest yields and produced the greatest fruit length, width, and weight when grown in the Taiwan-2 environment. This could be because Taiwan was also the selection environment. The ICPN15 lines were likely better adapted to Taiwan and therefore had lower performance in diverse climates, especially in the Vietnam-1 and Thailand environments, despite diverse genetic backgrounds (Table 1). Taiwan-2 was the fall season, which is more favorable for pepper production because it is generally less hot and rainy compared with the summer season (Taiwan-1).

Yield was generally lower in the Indonesia, Thailand, and Vietnam environments, and greater in the South Korea and Taiwan environments. Interestingly, the analyzed di-

urnal temperature range was greatest in Indonesia, Thailand, and the Vietnam environments, and lowest in South Korea and Taiwan. However, observed daily maximum and minimum temperatures in the Taiwan environments were considerably different from those in the GFS analyses. The analyzed daily maximum and minimum temperatures were generally greater in Thailand and Vietnam than in the other environments (Fig. 3). Furthermore, days to 50% anthesis was often greater in the South Korea and Taiwan environments compared with the Indonesia, Thailand, and Vietnam locations (Fig. 1). Temperature strongly influences flower and fruit development in pepper (Polowick and Sawhney, 1985). In addition, flower number and fruit set percentage are greatly influenced by the diurnal temperature range and mean daily temperature (Bakker, 1989). In general, pepper flower number and fruit set are greatest at 28 to 32 °C and 18 to 26 °C day/night temperatures, and are reduced at temperatures more than or less than these ranges (Aloni et al., 2001; Erikson and Markhart, 2002; Polowick and Sawhney, 1985; Pressman et al., 1998; Wubs et al., 2009). However, the majority of these studies were conducted on sweet pepper, which is generally considered more sensitive to temperature than chili pepper. The analyzed temperatures in Taiwan and South Korea were more similar to those found to be ideal for flower and fruit set compared with the Indonesia, Thailand, and Vietnam environments. The large differences in analyzed daily maximum and minimum temperatures might contribute to the overall lower performance of the ICPN15 lines in Indonesia, Thailand, and Vietnam, although other factors also played important roles.

It was difficult to evaluate the influence of estimated rainfall on performance of the ICPN15 lines. Although gauge-adjusted satellite precipitation estimates are generally useful for large-scale or long-lasting precipitation events, space-borne precipitation estimation is considerably more difficult when rainfall occurs as the result of small or highly transient convection. In South Korea, the rainfall was stable across the production season, and the lines performed well at this location. Similarly, Vietnam-1 and Vietnam-2 also had stable estimated monthly precipitation; however, the lines had among the lowest performance these locations. Both the estimated and observed precipitation was greatest at Taiwan-1 and least at Taiwan-2, and was variable in both environments, although these were among the best locations for ICPN15 performance. Therefore, we hypothesize that an external factor outside of precipitation was the major contributor the variability observed for location. This is supported by the fact that the trials at every location received supplemental irrigation when it was needed. However, high rainfall would likely result in greater disease incidence and lower performance.

One major challenge of multilocation testing through voluntary collaborators was

inconsistent collaborator adherence to requested protocols in experimental design and measurement of different traits, resulting in low proportions of usable data in this experiment. ICPN15 was successful in that it served as a means to distribute widely improved lines developed by WorldVeg; however, feedback on performance data were generally poor and inconsistent. One possible reason for this is the lack of incentive for recipients to provide data back to the breeding program. Regardless of whether useful feedback is provided, recipients of the ICPN lines are given germplasm and can continue to receive improved germplasm in the future. These issues need to be addressed in the future to improve the ICPN program as well as the WorldVeg pepper breeding program. Research in improved feedback and incentive programs in a participatory setting is limited (Sperling et al., 2001), and more studies in the area are required to address these issues better.

Another possible reason for poor feedback could be attributed to the use of ICPN lines by both public- and private-sector recipients directly for breeding, commercial release, and associated intellectual property concerns. For example, after further independent evaluation in national-level trials in respective countries, ICPN15-03 (AVPP0303) in Kazakhstan and Uzbekistan, ICPN15-04 (AVPP9813) in Armenia and Ghana, ICPN15-6 (AVPP0305) in Armenia, ICPN15-7 (AVPP9905) in Mali and Uzbekistan, and ICPN15-9 (AVPP0105) in Kazakhstan and Mali were released as cultivars by different local names (Lin et al., 2013). Furthermore, between 2005 and 2012, more than 30 ICPN lines have been released as open-pollinated cultivars in seven countries (Lin et al., 2013), and ICPN lines have been used as inbred parental lines in 34 hybrids, most of which ($n = 21$) were commercialized by Indian seed companies (Reddy et al., 2015). Although we did not receive usable data for ICPN15 from any of the Indian recipients, an empirical study suggested that about 13% of hybrid hot pepper seeds sold in India contain WorldVeg pepper germplasm, and an estimated 229,000 small-holder pepper growers planted cultivars with pedigrees containing WorldVeg germplasm in 2014 (Schreinemachers et al., 2016).

The long fruit check ICPN15-7 as well as ICPN15-5 and -10 were generally among the best-performing lines evaluated, combining high yield, large fruit (length, width, and weight), and early maturity with relatively high stability for these traits across growing environments. The short fruit check ICPN15-4 also had high yield and high stability for yield components across growing environments. Further testing in diverse growing environments is required to assess stability in these lines more fully. Although stability of yield and yield components across multiple environments is the goal of the breeding program, we found significant $G \times E$ interactions for most of the traits evaluated. Therefore, additional breeding efforts with local environment focus might be required

before commercializing these lines. More information is needed to assess the factors truly associated with the environmental variability identified in this study. In addition, it appears that the best publicly available prediction models for temperature and precipitation are not accurate enough and are likely not useful for this type of study, at least in Taiwan during the period of this experiment. This work provides a basis for assessing stability of important yield component traits in horticultural crops, which can greatly differ from those in agronomic crops, given the diversity in regional market preferences.

Literature Cited

- Aloni, B., M. Peet, M. Pharr, and L. Karni. 2001. The effect of high temperature and high atmospheric CO₂ on carbohydrate changes in bell pepper (*Capsicum annuum*) pollen in relation to its germination. *Physiol. Plant.* 112:505–512.
- Bakker, J.C. 1989. The effects of temperature on flowering, fruit set, and fruit development of glasshouse sweet pepper (*Capsicum annuum* L.). *J. Hort. Sci.* 64:313–320.
- Berke, T., L.L. Black, N.S. Talekar, J.F. Wang, P. Gniffke, S.K. Green, T.C. Wang, and R. Morris. 2005. Suggested cultural practices for chili pepper. AVRDC publication 05-620.
- Bosland, P.W. and E.J. Votava. 2012. Peppers: Vegetable and spice capsicums. 2nd ed. CAB International, Oxfordshire, UK.
- Bradu, D. and K.R. Gabriel. 1978. The biplot as a diagnostic tool for models of two-way tables. *Technometrics* 20:47–68.
- Butcher, J.D., K.M. Crosby, K.S. Yoo, B.S. Patil, A.M.H. Ibrahim, D.I. Leskovar, and J.L. Jifon. 2012. Environmental and genotypic variation of capsaicinoids and flavonoid concentrations in habanero (*Capsicum chinense*) peppers. *HortScience* 47:574–579.
- Cabral, N.S.S., A.M. Medeiros, L.G. Neves, C.P. Sudre, S. Pimenta, V.J. Coelho, M.E. Seafim, and R. Rodrigues. 2017. Genotype × environment interaction on experimental hybrids of chili pepper. *Genet. Mol. Res.* 16: doi: 10.4238/gmr16029551.
- DeWitt, D. and P.W. Bosland. 1993. The pepper garden. Ten Speed Press, Berkeley, CA.
- Erikson, A.N. and A.H. Markhart. 2002. Flower development stage and organ sensitivity of bell pepper (*Capsicum annuum* L.) to elevate temperature. *Plant Cell Environ.* 25:123–130.
- Food and Agriculture Organization of the United Nations (FAO). 2015. FAOSTAT statistics database. FAO, Rome, Italy.
- Global Climate & Weather Modeling Branch. 2016. The Global Forecast System (GFS)–Global Spectral Model (GSM) (GSM Version 13.0.2.). NOAA/NWS/NCEP/EMC. 21 Nov. 2017. <www.emc.ncep.noaa.gov/GFS/doc.php>.
- Gurung, T., S. Techawongstein, B. Suriham, and S. Techawongstein. 2011. Impact of environments on the accumulation of capsaicinoids in *Capsicum* spp. *HortScience* 46:1576–1581.
- Gurung, T., S. Techawongstein, B. Suriham, and S. Techawongstein. 2012. Stability of yield and capsaicinoids content in chili (*Capsicum* spp.) grown across six environments. *Euphytica* 187:11–18.
- Guzman, I., P.W. Bosland, and M.A. O'Connell. 2011. Heat, color, and flavor compounds in *Capsicum* fruit, p. 109–126. In: D.R. Gang (ed.). The biological activity of phytochemicals. Springer, New York, NY.
- Huffman, G.J., R.F. Adler, D.T. Bolvin, G. Gu, E.J. Nelkin, K.P. Bowman, Y. Hong, E.F. Stocker, and D.B. Wolff. 2007. The TRMM Multisatellite Precipitation Analysis (TMPA): Quasi-global, multiyear, combined-sensor precipitation estimates at fine scales. *J. Hydrometeorol.* 8:38–55.
- Kahane, R., T. Hodgkin, H. Jaenicke, C. Hoogendoorn, M. Hermann, J.D.H. Keatinge, J. d'Arros Hughes, S. Padulosi, and N. Looney. 2013. Agrobiodiversity for food security, health, and income. *Agron. Sustain. Dev.* 4:671–693.
- Kantar, M.B., J.E. Anderson, S.A. Lucht, K. Mercer, V. Bernau, K.A. Case, N.C. Le, M.K. Frederiksen, H.C. DeKeyser, Z.Z. Wong, J.C. Hastings, and D.J. Baumler. 2016. Vitamin variation in *Capsicum* spp. provides opportunities to improve nutritional value of human diets. *PLoS One* 11:e0161464.
- Lee, J.J., K.M. Crosby, L.M. Pike, K.S. Yoo, and D.I. Leskovar. 2005. Impact of genetic and environmental variant on development of flavonoids and carotenoids in pepper (*Capsicum* spp.). *Scientia Hort.* 106:341–352.
- Lin, S.W., Y.Y. Chou, H.C. Shieh, A.W. Ebert, S. Kumar, R. Mavlyanova, A. Rouamba, A. Tenkouano, V. Afari-Sefa, and P.A. Gniffke. 2013. Pepper (*Capsicum* spp.) germplasm dissemination by AVRDC–The World Vegetable Center: An overview and introspection. *Chronica Hort.* 53:21–27.
- Lin, S.W., H.C. Shieh, L.J. Kin, Z.M. Sheu, L. Kenyon, R. Srinivasan, and S. Kumar. 2014. Procedures for chili pepper variety field trials. AVRDC publication 14-784. Shanhu, Taiwan.
- Pacheco, A., M. Vargas, G. Alvarado, F. Rodríguez, M. López, J. Crossa, and J. Burguño. 2016. GEA-R (Genotype × Environment Analysis with R for Windows). Version 4.0. International Maize and Wheat Improvement Center. 2 Aug. 2017. <<http://hdl.handle.net/11529/10203>>.
- Polowick, P.L. and V.K. Sawhney. 1985. Temperature effects on male fertility and flower and fruit development in *Capsicum annuum* L. *Scientia Hort.* 25:117–127.
- Pressman, E., H. Moshkovitch, K. Rosenfeld, R. Shaked, B. Gamliel, and B. Aloni. 1998. Influence of low night temperatures on sweet pepper flower quality and the effect of repeated pollination, with viable pollen, on fruit setting. *J. Hort. Sci. Biotechnol.* 73:131–136.
- Pritts, M. and J. Luby. 1990. Stability indices for horticulture crops. *HortScience* 25:740–745.
- Reddy, M.K., A. Srivastava, S.W. Lin, R. Kumar, H.C. Shieh, A.W. Ebert, N. Chawda, and S. Kumar. 2015. Exploitation of AVRDC's chili pepper (*Capsicum* spp.) germplasm in India. *Taiwan Yuanyi* 61:1–9.
- Schreinemachers, P., K.P.C. Rao, W. Easdown, P. Hanson, and S. Kumar. 2016. The contribution of international vegetable breeding to private seed companies in India. *Gen. Resources Crop Evol.* doi: 10.1007/s10722-016-0423-y.
- Shaffi, B. and W.J. Price. 1992. Statistical analysis of genotype-by-environment using the AMMI model and stability estimates. *Proc. Conf. Appl. Stat. Agr.* 60–72.
- Sherman, P.W. and J. Billing. 1999. Darwinian gastronomy: Why we use spices: Spices taste good because they are good for us. *Bioscience* 49:453–463.
- Smith, S.H. 2015. In the shadow of a pepper-centric historiography: Understanding the global diffusion of *Capsicums* in the sixteenth and seventeenth centuries. *J. Ethnopharmacol.* 167:64–77.
- Sperling, L., J.A. Ashby, M.E. Smith, E. Weltzein, and S. McGuire. 2001. A framework for analyzing participatory plant breeding approaches and results. *Euphytica* 122:439–450.
- Stommel, J.R. and R.J. Griesbach. 2008. Inheritance of fruit, foliar, and plant habit attributes in *Capsicum*. *J. Amer. Soc. Hort. Sci.* 133:396–407.
- Wubs, A.M., E. Heuvelink, and L.F.M. Marcelis. 2009. Abortion of reproductive organs in sweet pepper (*Capsicum annuum* L.): A review. *J. Hort. Sci. Biotechnol.* 84:467–475.
- Zewdie, Y. and P.W. Bosland. 2000. Evaluation of genotype, environment, and genotype-by-environment interaction for capsaicinoids in *Capsicum annuum* L. *Euphytica* 111:185–190.
- Zewdie, Y. and J.M. Poulos. 1996. Stability analysis in hot pepper. *Capsicum Eggplant Nwsl.* 14:39–42.