Pepper Photosynthesis, Stomatal Conductance, Transpiration, and Water Use Efficiency Differ with Variety, Indigenous Habitat, and Species of Origin

John Erwin
Department of Horticultural Science, University of Minnesota, 1970 Folwell Avenue, St. Paul, MN 55108

Tanveer Hussein
Institute of Horticultural Sciences, University of Agriculture, Agriculture University Road, Faisalabad, Pakistan 38000

David J. Baumler
Department of Food Science and Nutrition, University of Minnesota, 1334 Eckles Avenue, St. Paul, MN 55108

Additional index words. humidity, Capsicum, photosynthesis, temperature

Abstract. The instantaneous photosynthetic rate ($P_n$), transpiration rate ($E$), and stomatal conductance ($g_s$) were measured for 33 outdoor-grown Capsicum varieties (varying in species of origin and indigenous habitat) between 29 July and 22 Aug. 2017 using a portable gas exchange meter. Cuvette leaf temperature ($T_{leaf}$) and relative humidity ($RH$) were recorded at that same time. $P_n$ differed from 3.6 to 3.7 for ‘Malawi Piquante’ and ‘Korean Long Green’ peppers to 16.3 μmol CO2/m2/s (fixed) for ‘Thai Hot’ peppers. The $g_s$ differed from 0.01 to 0.05 among 13 varieties to 0.28 mmol H2O/m2/s for ‘Thai Hot’ peppers. $E$ differed from 0.43 to 0.59 among three varieties to 4.14 to 4.20 mmol H2O/m2/s for ‘CGN 22091’ and ‘Peruvian Purple’ peppers. Water use efficiency (WUE; $P_n/E$) varied from 2.92 to 3.43 among three varieties to 5.10 to 7.20 for 16 other varieties. $C. annuum$ derived varieties had higher $P_n$ (9.4 μmol CO2/m2/s fixed) than varieties derived from other species (4.5–8.6 μmol CO2/m2/s fixed). Varieties originating from dry climates had higher $P_n$ (12.5 μmol CO2/m2/s fixed) than those originating from temperate or tropical climates (8.0–8.8 μmol CO2/m2/s fixed). $T_{leaf}$ (27 to 33 °C) and RH (38% to 39%) differed among varieties. $P_n$ was positively correlated with $g_s$, $E$, and RH and was negatively correlated with WUE. We found that Capsicum $P_n$, $E$, and $g_s$ varied more than has been previously reported, and our data suggested that $P_n$, $g_s$, and $E$ data of outdoor-grown peppers should be used only when selecting parents for a breeding program (unless progeny is intended for greenhouse production).

Peppers ($Capsicum$ sp.) were one of the first crops domesticated in the Western Hemisphere using several independent, geographically distinct, and regional domestication efforts (Bosland and Votava, 2012). Cultivated peppers are derived from five species ($C. annuum$ L., $C. baccatum$ L., $C. chinense$ Jacq., $C. frutescens$ L., and $C. pubescens$) from three indigenous habitats (dry, temperate, and tropical) were selected (Table 1). Seeds were sourced from different seed producers/suppliers, and the reported species of origin were confirmed by phenotyping of flowers, leaves, and fruits during the experiment (Table 1). Seeds were sown 0.6 cm deep in Master Garden Premium media (Premier Tech Horticulture, Ltd., Ontario, Canada) in 32-cell trays (one seed per cell; individual cell volume = 150 cm3) in January and February 2017. Trays were then covered with a clear plastic lid (10 cm above media) and were placed in a greenhouse (23 ± 2 °C air temperature). After seeds germinated and cotyledons unfolded, the lid was removed and plants were grown for 9 to 14 additional weeks under natural daylight plus 25 μmol·m−2·s−1 supplemental irradiance (0800–0200 hr; Sunblaze T5 fluorescent lights; Sunlight Supply, Inc., WA; +1.62 mol·m−2·d−1 daily light integral) in the same greenhouse. After five leaves unfolded, Howard and Wildman, 2007; Kantar et al., 2016; Palevitch and Craker, 1995; Russo and Howard, 2002; Topuz and Ozdemir, 2007; Wahyuni et al., 2011). Other pepper products are consumed as spices, including dried pepper flakes, processed “hot” sauces, and infused/pressed oils, among other products (Bosland, 1996; Zewdie and Bosland, 2001).

Abiotic factors such as low temperatures, high temperatures, salt stress, drought (Serrano et al., 2017), and waterlogging can limit pepper yield (Ou and Zou, 2012; Zhai et al., 2016). For instance, optimal temperature for pepper photosynthesis range from 25 to 35 °C, and temperatures outside this range can limit the yield; for example, $C. chinense$ flower abortion increased 2-fold and fruit set decreased 3-fold when greenhouse temperatures were increased from 30 to 40 °C (Garruna-Hernandez et al., 2014). Erickson and Markhart (2002) showed that Capsicum flower abortion after pollination was particularly sensitive to high temperatures (33 °C).

We determined whether photosynthetic and water use attributes differed among outdoor-grown pepper varieties that differed in parental species and indigenous climates. Previous studies of Capsicum focused on differences in photosynthetic and water use attributes among species only or varieties within a species. Past Capsicum research often used greenhouse-grown or controlled environment–grown plants. Our research objectives were to: 1) determine whether instantaneous $P_n$, $g_s$, $E$, WUE meter cuvette $T_{leaf}$ and RH of 33 outdoor-grown pepper varieties differed; 2) to determine whether variety photosynthetic and water use attributes differed based on indigenous parents or habitat; and 3) to determine whether pepper photosynthetic and/or water use attributes were correlated with each other.

Materials and Methods

Thirty-three pepper varieties derived from five species ($C. annuum$ L., $C. baccatum$ L., $C. chinense$ Jacq., $C. frutescens$ L., and $C. pubescens$) from three indigenous habitats (dry, temperate, and tropical) were selected (Table 1). Seeds were sourced from different seed producers/suppliers, and the reported species of origin were confirmed by phenotyping of flowers, leaves, and fruits during the experiment (Table 1). Seeds were sown 0.6 cm deep in Master Garden Premium media (Premier Tech Horticulture, Ltd., Ontario, Canada) in 32-cell trays (one seed per cell; individual cell volume = 150 cm3) in January and February 2017. Trays were then covered with a clear plastic lid (10 cm above media) and were placed in a greenhouse (23 ± 2 °C air temperature). After seeds germinated and cotyledons unfolded, the lid was removed and plants were grown for 9 to 14 additional weeks under natural daylight plus 25 μmol·m−2·s−1 supplemental irradiance (0800–0200 hr; Sunblaze T5 fluorescent lights; Sunlight Supply, Inc., WA; +1.62 mol·m−2·d−1 daily light integral) in the same greenhouse. After five leaves unfolded,
Table 1. Variation in the instantaneous photosynthetic rate ($P_n$; mmol m$^{-2}$ s$^{-1}$), stomatal conductance ($g_s$; mmol H$_2$O m$^{-2}$ s$^{-1}$), transpiration rate ($E$; mmol H$_2$O m$^{-2}$ s$^{-1}$), cuvette leaf temperature ($T_{leaf}$; $^\circ$C), and relative humidity after 5 min (RH; %) and water use efficiency (WUE; mmol H$_2$O mmol CO$_2$$^{-1}$) among 33 outdoor-grown pepper varieties that vary in indigenous habitat and species. Indigenous habitats used for analysis are shown as superscript numbers after the variety name (1: desert; 2: temperature; 3: tropical; ?: unknown).

| Variety                | Country of Origin | Species       | $P_n$ | $g_s$ | E    | $T_{leaf}$ | RH  | WUE  |
|------------------------|-------------------|---------------|-------|-------|------|------------|-----|------|
| Malawi Piquante$^1$    | Malawi            | baccatum      | 3.6 a | 0.02 a| 0.47 a| 31 ab      | 44 abcdef| 5.9 b|
| Korean Long$^2$        | Korea             | annuum        | 3.7 a | 0.01 a| 0.43 a| 32 ab      | 40 abc  | 6.8 b|
| Antoii Romanian$^3$    | Romania           | annuum        | 4.3 a | 0.02 a| 0.59 a| 33 b       | 39 ab  | 6.3 b|
| Pakistan$^1$           | Pakistan          | annuum        | 4.3 a | 0.02 a| 0.69 a| 32 ab      | 41 abc  | 6.5 b|
| Fatali$^1$             | Africa            | chinense      | 4.4 a | 0.02 a| 1.03 a| 32 ab      | 42 abc  | 4.6 ab|
| Tabasco Red$^1$        | Mexico            | frutescens    | 4.5 a | 0.03 a| 0.90 a| 32 ab      | 38 a    | 5.8 b|
| Hawaiian Sweet$^1$     | Hawaii            | annuum        | 4.8 a | 0.03 a| 1.75 a| 30 ab      | 44 abc  | 3.7 ab|
| Hungarian Sunshine$^1$ | Hungary           | annuum        | 4.8 a | 0.07 a| 1.75 a| 30 ab      | 44 abc  | 3.7 ab|
| II Sweet$^2$           | USA               | annuum        | 5.9 abc| 0.08 abc| 1.39 abc| 28 a | 51 abcdef| 4.7 ab|
| Red Rocoto$^1$         | Ecuador           | pubescens     | 6.0 abc| 0.04 a| 1.23 abc| 32 ab | 41 abc  | 4.9 ab|
| Shishitou$^2$          | Japan             | annuum        | 6.1 abc| 0.08 abc| 2.03 abc| 32 ab | 45 abc  | 3.1 a|
| Moses Orange$^1$       | Kenya             | annuum        | 6.3 abc| 0.04 a| 1.13 abc| 33 b | 39 ab  | 6.0 b|
| Trinidad Moruga        | Trinidad          | chinense      | 7.7 abc| 0.05 a| 1.27 ab | 32 ab | 42 abc  | 7.2 b|
| Jamaican Red Hot$^1$   | Jamaica           | chinense      | 7.7 abc| 0.03 a| 1.05 ab | 33 b | 40 abc  | 6.8 b|
| Apple Hungarian$^1$    | Hungary           | annuum        | 7.9 abc| 0.05 a| 1.50 ab | 32 ab | 42 abc  | 5.7 b|
| Brazilian Star Fruit   | Brazil            | baccatum      | 8.1 abc| 0.08 ab| 1.69 ab | 32 ab | 45 abc  | 6.3 b|
| Yellow$^1$             | Costa Rica        | chinense      | 11.5 cdef| 0.21 abc| 4.14 c | 31 ab | 49 abcdef| 2.92 a|
| Explosive Ember$^1$    | Italy             | annuum        | 12.6 defg| 0.19 abc| 3.09 abc| 27 a | 56 bcdef| 4.84 ab|
| Aji Lemon$^1$          | Costa Rica        | chinense      | 11.5 cdef| 0.21 abc| 4.14 c | 31 ab | 49 abcdef| 2.92 a|
| Makoko Akosnade$^1$    | Ghana             | annuum        | 9.3 abc| 0.12 abc| 2.19 abc| 28 a | 54 abc  | 5.9 b|
| Giant Mexican Rocoto   | Mexico            | pubescens     | 10.4 abc| 0.16 abc| 2.73 abc| 28 a | 54 abcdef| 5.5 b|
| Bebere Ethiopian Brown$^1$ | Ethiopia      | annuum        | 10.5 abc| 0.17 abc| 3.60 abc| 32 ab | 45 abcdef| 3.71 a|
| Czechoslovakian Black$^2$ | Czech Rep.        | annuum        | 10.7 bedfg| 0.10 abc| 2.21 abc| 28 ab | 54 abcdef| 5.17 ab|
| Zimbabwe Bird$^1$      | Zimbabwe          | annuum        | 10.8 bedfg| 0.14 abc| 3.32 abc| 32 ab | 45 abcdef| 3.43 a|
| Coronong$^1$           | Malaysia          | chinense      | 10.9 bedfg| 0.19 abc| 2.85 abc| 28 ab | 55 abcdef| 4.48 ab|
| Bolivian Rainbow$^1$   | Bolivia           | annuum        | 11.1 bedfg| 0.13 abc| 2.27 abc| 28 ab | 57 def  | 4.61 ab|
| CGN 22901$^3$          | Costa Rica        | chinense      | 11.5 cdef| 0.21 abc| 4.14 c | 31 ab | 49 abcdef| 2.92 a|
| Pepperoncini$^2$       | Italy             | annuum        | 12.6 defg| 0.19 abc| 3.09 abc| 27 a | 56 bcdef| 4.84 ab|
| Numex Sunset Chili$^2$ | USA               | annuum        | 13.1 cefg| 0.20 abc| 3.11 abc| 28 ab | 57 def  | 4.69 ab|
| Aleppo$^1$             | Syria             | annuum        | 13.3 fg  | 0.20 abc| 3.13 abc| 29 ab | 57 ef   | 4.53 ab|
| Bulgarian Carrot$^1$   | Bulgaria          | annuum        | 13.5 fg  | 0.16 abc| 3.13 abc| 29 ab | 56 cdef  | 4.76 ab|
| Hait Cluster$^1$       | Haiti             | annuum        | 14.1 fg  | 0.20 abc| 3.30 abc| 28 ab | 56 bcdef| 5.90 b|
| Peruvian Purple$^1$    | Peru              | annuum        | 14.6 fg  | 0.27 bc| 4.21 c | 29 ab | 55 abcdef| 4.83 ab|
| Thai Hot$^1$           | Thailand          | annuum        | 16.3 g  | 0.28 c| 3.87 bc| 27 a | 59 f    | 5.10 b|

ANOVA

| Species                  | Climate     | $P_n$ | ** | NS | NS | NS | NS |
|--------------------------|-------------|-------|----|----|----|----|----|
| Variety                  | $P_n$       | 6   | ***| ***| ***| ***| ***|
| Region                   | $P_n$       | 6   | ***| ***| ***| ***| ***|

Species: $P_n$, Climate, $P_n$, RH

annuum, Desert, 12.5 b, 56 b
baccatum, Temperate, 8.0 a, 47 ab
chinense, Tropical, 8.8 a, 46 a
frutescens, 4.5 a
pubescens, 8.2 ab

$^a$Means followed by different lowercase letters within each column are significantly different based on Tukey’s Honestly Significant Difference Test (P < 0.05) for mean separation based on the analysis of variance (ANOVA).

$^b$Statistical significance as determined by analysis.
ns, *, **, ***Nonsignificant or significant at P ≤ 0.05, 0.01, or 0.001, respectively.
were independent variables (Table 1). Indigenous habitats were subjectively categorized as dry/desert, temperate, and tropical based on the climate in the country of origin (Table 1). Data were collected at six different times from a different leaf each time (198 total values each for Pn, E, gs, and WUE). Data were analyzed using an analysis of variance followed by mean separation (Tukey’s honestly significant difference and least significant difference; α < 0.05) using the SPSS statistical software package (IBM SPSS Statistics, version 24; IBM Corp., Armonk, NY). Pearson correlations among dependent variables were also determined.

Results

Pn varied from 3.6 to 3.7 for ‘Malawi Piquante’ and ‘Korean Long Green’ peppers to 16.3 μmol CO2/m2/s (fixed) for ‘Thai Hot’ peppers (Table 1). The gs varied from 0.01 to 0.59 for most varieties (Malawi, Korean Long Green, Antohi Romanian, Pakistan, Fatali, Tabasco Red, Hlan Sweet Hot, Hungarian Sunshine, Red Roccoto, Moses Orange, Trinidad Moruga Scorpion, Jamaican Red Hot, and Apple Hungarian) to 0.28 mmol H2O/m2/s for ‘Thai Hot’ pepper (Table 1). E varied from 0.43 to 0.59 for ‘Malawi Piquante’, ‘Korean Long Green’, and ‘Pakistan peppers’ to 4.14 and 4.21 mmol H2O/m2/s for ‘CGN 22091’ and ‘Peruvian Purple’ peppers, respectively (Table 1). WUE differed from 2.92 to 3.43 among three varieties to 5.10 to 7.20 for 16 other pepper varieties (Table 1). Tleaf varied from 27°C for ‘Thai Hot’ and ‘Pepperoncini’ peppers to 33°C for ‘Pakistan’, ‘Moses Orange’ and ‘Jamaican Red Hot’ peppers (Table 1). Cuvette RH differed from 38% for ‘Hawaiian Sweet’ peppers to 59% for ‘Thai Hot’ peppers (Table 1).

C. frutescens, C. baccatum, C. chinense, and C. pubescens derived varieties had lower Pn (4.5–8.6 μmol CO2/m2/s fixed) than C. annuum derived varieties (9.4 μmol CO2/m2/s fixed) (Table 1). There were no differences in gs, E, Tleaf, RH, or WUE among varieties based on parental species (Table 1). Varieties originating from temperate climates had lower Pn (8.0 μmol CO2/m2/s fixed) than those originating from dry regions (12.5 μmol CO2/m2/s fixed) (Table 1). Varieties from tropical regions had a lower cuvette RH (46% to 47%) than those from dry regions (56%) (Table 1) when tested in our experiment.

Pn was positively correlated with gs (Pearson correlation = 0.88), E (0.86), and RH (0.62), and it was negatively correlated with WUE (–0.35) (Table 2). The gs was positively correlated with E (0.95) and RH (0.56), and it was negatively correlated with Tleaf (–0.18) and WUE (–0.56) (Table 2). E was positively correlated with RH (0.44) and negatively correlated with WUE (–0.67) (Table 2). Tleaf was negatively correlated with gs (–0.18) and RH (–0.21) (Table 2). RH was negatively correlated with Tleaf (–0.21) (Table 2).

Discussion

Capsicum Pn values varied more in our experiment (3.6–3.7 to 16.3 μmol CO2/m2/s fixed) than they did in other experiments reported by others; for example, Ou and Zou (2012) reported that the Pn of five Capsicum species varied from 15.8 to 21.8 μmol CO2/m2/s (fixed). Among pepper varieties within a species, Pn also varied more in our experiment than it did in others (Ghasemi et al., 2016; Rosado-Souza et al., 2015). Borisev et al. (2012) reported that the Pn of 10 C. annuum varieties differed from 14.5 to 16.6 μmol CO2/m2/s (fixed); the C. annuum variety Pn (22 varieties) in our work ranged from 3.7 to 16.3 μmol CO2/m2/s (fixed) (Table 1). Ridzuan et al. (2018) reported that the C. annuum variety/accession Pn differed from 11.5 to 19.1 μmol CO2/m2/s (fixed). In contrast, Hassan et al. (2014) reported that the C. annuum variety Pn varied from 2.5 to 5.3 μmol CO2/m2/s (fixed; irradiance not reported). Pérez-Grajales et al. (2004) reported similar Pn rates (at a lower irradiance of 500 μmol/m2/s) for C. pubescens. Rosado-Souza et al. (2015) reported that the C. chinense accession Pn varied from 17 to 25 μmol CO2/m2/s (fixed).

Differences between our Pn values and others may have been related to the irradiance levels when Pn was measured, where plants were grown, or the number of varieties studied. For instance, irradiance in our work was at saturating photosynthetic levels (1000 μmol·m−2·s−1), whereas irradiance in other experiments was undocumented or at 300 to 500 μmol·m−2·s−1. Because peppers are typically grown in the field, where they are routinely exposed to saturating irradiance levels, we believe that our data more accurately reflect actual field Pn. Furthermore, Pn can vary more for field-grown than for greenhouse-grown/controlled environment–grown Capsicum. Borisev et al. (2012) reported that the Pn of nine Capsicum varieties differed from 14.5 to 16.6 μmol CO2/m2/s (fixed), but that Pn differed from 12.8 to 18.7 μmol CO2/m2/s (fixed) when those same varieties were grown in the field. Furthermore, the greater variation in Pn in this study compared with that in some other studies may have occurred because we evaluated a greater number of varieties that were intentionally selected to represent a broad array of genetic and indigenous backgrounds.

Our data regarding the varieties derived from C. annuum included collectively higher Pn than the data regarding the varieties derived from other species. However, broad conclusions regarding which species have higher Pn should be made cautiously because abiotic factors can interact with species and affect Pn. For instance, Ou and Zou (2012) reported that the Pn of C. frutescens was higher than that of four other Capsicum species (21.8 vs. 15.8–17.7 μmol CO2/m2/s fixed) when plants were grown at 35 °C, but the Pn of C. pubescens was highest when plants were grown at 15 °C. In the same study, the Pn of C. baccatum and the Pn of C. pubescens were higher than that of three other Capsicum species when plants were drought-stressed. In contrast, Okunlola et al. (2017) reported that the C. chinense Pn was more drought-tolerant than the Pn of C. annuum or C. frutescens. The environment where a plant is grown or the water status of a plant when Pn data are collected can lead to different conclusions about which Capsicum species has higher Pn. Although varieties are reportedly derived from a species, many are interspecific hybrids (often unreported) (Petkova et al., 2014) and genetically related (Amaral-Junior et al., 2005). That hybridization alone may impact the photosynthetic or water use data. Petkova et al. (2014) showed that the Pn of four F1 pepper hybrids was more tolerant to high temperatures (33 to 35 °C) than that of parental species.

The range and gs values reported in our study (0.01–0.28 mmol H2O/m2/s) were less than those reported by others. Ridzuan et al. (2018) reported that the gs of the C. annuum variety (14) varied from 0.25 to 0.68 mmol H2O/m2/s. Hassan et al. (2014) reported that the gs of the C. annuum variety varied from 0.50 to 0.60 mmol H2O/m2/s. Differences between our gs data and that of others may have been related to the environments where plants were grown. For instance, plants grown at temperatures below 15 °C or above the optimal temperature for pepper Pn can decrease the Capsicum gs (Jaimez and Rada, 2016). Because the Tleaf data were collected on some days when the temperature was above the optimal temperature reported for

Table 2. Pearson correlation between instantaneous photosynthetic rate (Pn), stomatal conductance (gs), transpiration rate (E), cuvette leaf temperature (Tleaf), and humidity after 5 min (RH) and water use efficiency (WUE; WUE = Pn/E).

| Factor | Pn | gs | E | Tleaf | RH | WUE |
|--------|----|----|----|------|----|-----|
| (2-tailed) | 0.88** | 0.86** | -0.09 | 0.62** | -0.35** |
| | *** | *** | NS | *** | *** |
| (2-tailed) | 0.95** | -0.18* | 0.56** | -0.56** |
| | *** | * | *** | *** |
| (2-tailed) | -0.02 | 0.44** | -0.67** |
| | NS | *** | *** | *** |
| (2-tailed) | -0.21** | NS | -0.010 |
| | NS | NS | NS |

*Statistical significance as determined by the analysis of variance. NS, *, **, ***NSignificant or significant at P ≤ 0.05, 0.01, or 0.001, respectively.
Capsicum Pn (25 to 30 °C), the gs may have been impacted (Table 3).

Our data indicated that gs does not vary among domesticated Capsicum species (Table 1). Millá et al. (2013) also found that gs did not vary between wild-type and domesticated C. annuum or C. baccatum. Interestingly, we observed that gs differed among varieties derived from those species. Rosado-Souza et al. (2015) reported that the C. annuum variety gs differed. Yet, in both of those studies, the species gs did not differ. Similarly, Percy et al. (1996) showed that the gs of interspecific selections of cotton (Gossypium barbadense L) was higher than that of the parents or ancestral lines. Differences in the gs of varieties observed by us and others may have been due to the integration of a mutation into breeding programs over time and/or interspecific hybridization resulting in greater variations in gs than that observed for genetic parents. Furthermore, a recent study has shown a transgenerational effect on watercress (Lepidium sativum) gs; the maternal light environment effects on stomatal density and gs were expressed in seed-propagated progeny, suggesting that differences in gs in this experiment and in past experiments, to some degree, may be impacted by the environment in which seeds were produced (Vrablova et al., 2018). A similar relationship between the maternal water status and progeny WUE was also reported for Eucalyptus (Vivas et al., 2019).

The E values reported here also varied more than those reported by others, although the maximum E values were similar (3.21–4.05 mmol H2O/m2/s) (Ou and Zou, 2012). Ridzuan et al. (2018) reported that the E of the C. annuum variety differed from 4.61 to 7.11 mmol H2O/m2/s (indoor-grown). Furthermore, differences in E values in our work and that of others may have been related to where plants were grown. Borisev et al. (2012) reported that the E of Capsicum variety (nine) differed more for outdoor-grown than indoor-grown peppers (4.4–6.1 vs. 4.7–5.0 mmol H2O/m2/s, respectively). In contrast to the Pn, E, and gs data presented here, the WUE values we observed (3.1–7.2) were similar to those reported by others for peppers grown in a controlled environment (3.9–6.8) (Ou and Zou, 2012). However, our values were generally higher than those reported by Borisev et al. (2012) who also reported that the WUE of indoor-grown C. annuum variety (nine) varied less than that of field-grown plants (3.2–3.5 and 2.3–3.9, respectively). The WUE of Capsicum is reportedly impacted by irradiance; WUE was higher when plants were grown under high irradiance (400 μmol·m−2·s−1; 2.2) compared to low irradiance (200 μmol·m−2·s−1; 1.8) (Fu et al., 2010). However, irradiance in our study was 1000 μmol·m−2·s−1; therefore, it would be considered high. Moreover, the negative correlation between Capsicum WUE and Pn that we observed agreed with the observations of Antony and Singandhupe (2004).

Ridzuan et al. (2018) reported that the C. annuum variety E and gs were correlated with the E, as we observed (0.55 and 0.87, respectively). However, Pn is not always correlated with the gs. Yun and Ahn (2009) reported that increasing temperature and atmospheric CO2 concentrations increased the pepper Pn, but not the pepper gs. The strong correlation between pepper gs and Pn reported here (0.95) (Table 2) supports the assertion that gs limitations may reduce pepper Pn (Serrano et al., 2017). However, the observations of Yun and Ahn (2009) suggested that gs limitations on Pn may occur less if CO2 concentrations are increased, as is common for greenhouse-produced bell peppers (Doras, 2003; Erwin and Gesick, 2017).

A negative correlation between gs and Tleaf was previously reported (r2 = 0.76) for cotton (G. barbadense L–derived) (Radin et al., 1994), although that correlation was greater than what we observed here for pepper (Table 2). Interestingly, some have suggested that gs measurements obtained later in the afternoon (after 1300 hr) may be important when evaluating plants for higher yield based on gs, for instance. Rebetzke et al. (2002) recommended that water breeding projects should involve selected progeny with higher gs collected later in the day because gs collected later in the day correlated more with yield than gs collected earlier. Similarly, Radin et al. (1994) found that the cotton yield (related to Pn) was associated more with gs collected in the afternoon than in the morning, and that varieties with high yield under hot temperatures had higher gs in the afternoon, but not in the morning (Lü et al., 1997). We collected data when Pn and gs were highest during the day; whether Capsicum yield is associated more with gs collected at certain times of the day is not known.

Differences in outdoor environmental conditions for data collection dates during our experiment may have reduced the resolution of our experiment. Tleaf was higher on collection dates 3 and 4 (33.6 to 34 °C; reported Pn optima) than on other dates (27.7 to 29.6 °C) (Table 3). Furthermore, cvette RH was higher on collection dates 2, 3, and 4 (54% to 58%) than on collection dates 1 and 6 (40% to 43%) (Table 3). Because Tleaf and RH are correlated with Pn, gs, and E (Table 2), the environmental differences among collection dates may have impacted values. Care should be taken when determining conclusions about plant photosynthesis or yield based on instantaneous Pn or water use data based on the leaf area per unit. Collecting data based on the leaf area per unit does not account for differences in the individual leaf area, plant leaf number, whole-plant leaf area, or whole-plant photosynthesis. Furthermore, instantaneous Pn, data do not account for plant acclimation to changing environmental conditions (Hikosaka et al., 2006). Therefore, our study is of value because it compared photosynthetic and water use attributes of a larger number of pepper varieties at the same time of day and under the same environmental conditions simultaneously. Our study also demonstrated that outdoor-grown pepper variety photosynthetic and water use attributes vary more than previously reported, and that the reported parental species and indigenous habitat are associated with some photosynthetic and water use attributes.

**Literature Cited**

Amarul J'Junior, A.T., R. Rodrigues, C.P. Suder'e, E.M. Riva, and M. Karasawa. 2005. Genetic divergence between "chilli" and sweet pepper accessions using multivariate techniques. Hort. Bras. 23(1):22–27.

Antony, E. and R.B. Singandhupe. 2004. Impact of drip and surface irrigation on growth, yield and WUE of capsicum (Capsicum annuum L.). Agr. Water Mgt. 65(2):121–132.

Borisev, M., B. Krstic, D. Gvozdenova, and J. Gvozdanovic-Vargic. 2012. Photosynthesis and water use efficiency relations to yield of ten pepper varieties (Capsicum annuum). Bulg. J. Agr. Sci. 18(4):589–594.

Boslund, P.W. 1996. *Capsicums*: Innovative uses of an ancient crop, p. 479–487. In: J. Janick (ed.). Progress in new crops. ASHS Press, Arlington, VA.

Boslund, P.W. and E.J. Votava. 2012. Peppers: Vegetable and spice capsicums. CAB Intl., Wallingford, UK.

Carvalho, J.A., F.C. Rezende, R.F. Aquino, W.A. Freitas, and E.C. Oliveira. 2011. Productive and economic analysis of red peppers under different irrigation depths cultivated in greenhouse. Rev. Bras. Eng. Agr. Ambient. 15:569–574.
Dorais, M. 2003. The use of supplemental lighting for vegetable crop production: Light intensity, crop response, nutrition, crop management, and cultural practices. Can Greenhouse Conf. 2003:1–8.

Eriksen, A.N. and A.H. Markhart. 2002. Flower developmental stage and organ sensitivity of bell pepper (Capsicum annuum L.) to elevated temperature. Plant Cell Environ. 25:123–130.

Erwin, J. and E. Gesick. 2017. Photosynthetic responses of Swiss chard, kale, and spinach cultivars to irradiance and carbon dioxide concentration. HortScience 52:706–712.

Food and Agriculture organization (FAO). 2014. FAOSTAT–Agriculture <http://www.fao.org>.

Fu, Q.S., B. Zhao, Y.J. Wang, S. Ren, and Y.D. Guo. 2010. Stomatal development and associated photosynthetic performance of capsicum in response to different light availabilities.Photosynthetica 48(2):189–198.

Garruna-Hernandez, R., R. Orellana, A. Larque-Saavedra, and A. Canto. 2014. Understanding the physiological responses of a tropical crop (Capsicum chinense Jacq.) at high temperature. PLoS One 9(11):e111402.

Ghasemi, M., M. Modarresi, B. Jelodar, N. Bagheri, and A. Jamali. 2016. The evaluation of exogenous application of salicylic acid on physiological characteristics, proline and essential oil content of chamomile (Matricaria chamomilla L.) under normal and heat stress conditions. Agriculture 6(3):31.

Hassan, M., T. Haider, S.N. Chowdhury, F. Howlander, and A.F.M. Uddin. 2014. Study on morphophysiological and yield performance of four chilli (Capsicum spp.) lines. J. Biosci. and Agr. Research 2(1):1–12.

Hikosaka, K., K. Ishikawa, A. Borjigida, O. Muller, and Y. Onoda. 2006. Temperature acclimation of photosynthesis: Mechanisms involved in the changes in temperature dependence of photosynthetic rate. J. Expt. Bot. 57:291–302.

Howard, L.R., S.T. Talcott, C.H. Brenes, and B. Villalon. 2000. Changes in phytochemical and antioxidant activity of selected pepper cultivars (Capsicum species) as influenced by maturity. J. Agr. Food Chem. 48:1713–1720.

Howard, L.R. and R.E.C. Wildman. 2007. Antioxidant vitamin and phytochemical content of fresh and processed pepper fruit (Capsicum annuum), p. 165–191. In: R.E.C. Wildman (ed.). Handbook of Nutraceuticals and Functional Foods. CRC Press, Boca Raton.

Jaimez, R.E. and F. Rada. 2016. Gas exchange, growth, flowering and fruit production in sweet pepper (Capsicum chinense Jacq.) along a vertical gradient determined by altitudinal differences in a tropical region. Expt. Agr. 52(2):251–265.

Kang, S., L. Zhang, X. Hu, Z. Li, and P. Jerie. 2001. An improved water use efficiency for hot pepper grown under controlled alternate drip irrigation on partial roots. Sciento Hort. 89(4):257–267.

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(8):e0161464.

Lu, Z., J. Chen, R.G. Percy, and E. Zeiger. 1997. Photosynthetic rate, stomatal conductance and leaf area of two cotton species (Gossypium barbadense and Gossypium hirsutum) and their relation with heat resistance and yield. Austral. J. Plant Physiol. 24(5):693–700.

Milla, R., N. deDiego-Vico, and N. Martin-Robles. 2013. Shifts in stomatal traits following the domestication of plant species. J. Expt. Bot. 64(11):3137–3146.

Okunlola, G.O., O.A. Olatunji, R.O. Akinwale, A. Tariq, and A.A. Adelusi. 2017. Physiological response of the three most cultivated pepper species (Capsicum spp.) in Africa to drought stress imposed at three stages of growth and development. Scientia Hort. 224:198–205.

Ou, L.J. and X.X. Zou. 2012. The photosynthetic stress responses of five pepper species are consistent with their genetic variability. Photosynthetica 50(1):49–55.

Palevitch, D. and E.L. Craker. 1995. Nutritional and medicinal importance of red pepper (Capsicum spp.). J. Herbs Spices Med. Plants 3:55–83.

Percy, R.G., Z. Lu, J.W. Radin, E. Turcotte, and E. Zeiger. 1996. Inheritance of stomatal conductance in cotton (Gossypium barbadense). Physiol. Plant. 96(3):389–394.

Perez-Grajales, M., V.A. Gonzalez-Hernandez, M.C. Mendoza-Castillo, C. Peña Valdivia, A. Peña-Lomelli, and J. Sahagun-Castallanos. 2004. Physiological characterization of Capsicum annuum hot pepper (Capsicum pubescens R & P) landraces. J. Amer. Soc. Hort. Sci. 129:88–92.

Petkova, V., V. Todorova, and N. Tomlekova. 2014. Efficiency of photosynthetic apparatus of sweet pepper (Capsicum annuum L.), hybrids and their parental components in high temperature conditions. Plant Science (Bulgaria).

Qin, C., Y. Changshui, Y. Shen, X. Fang, L. Chen, J. Min, J. Cheng, S. Zhao, M. Xu, Y. Luo, Y. Yang, Z. Wu, L. Mao, H. Wu, C. Ling-Hu, H. Zhou, H. Lin, S. Gonzalez-Morales, D.L. Trejo-Saavedra, H. Tian, X. Tang, M. Zhao, H. Huang, A. Zhou, X. Yao, J. Cui, W. Li, Z. Chen, Y. Feng, Y. Niu, S. Bi, X. Yang, W. Li, H. Cai, X. Luo, S. Montes-Hernández, M.A. Leyva-González, Z. Xiong, X. He, L. Bai, S. Tan, X. Tang, D. Liu, J. Liu, S. Zhang, M. Chen, L. Zhang, L. Zhang, Y. Zhang, W. Liao, Y. Zhang, M. Wang, X. Lu, B. Wen, H. Liu, H. Luan, Y. Zhang, S. Yang, X. Wang, J. Xu, X. Li, S. Li, J. Wang, A. Falloix, P.W. Bosland, Y. Li, A. Krogh, R.F. Rivera-Bustamante, L. Herrera-Estrella, Y. Yin, J. Yu, K. Hu, and Z. Zhang. 2014. Whole-genome sequencing of cultivated and wild peppers provides insights into Capsicum domestication and specialization. Proc. Natl. Acad. Sci. USA 111(14):5135–5140.

Radin, J.W., Z. Lu, R.G. Percy, and E. Zeiger. 1994. Genetic variability for stomatal conduc-
tance in Pima cotton and its relation to improvements of heat adaptation. Proc. Natl. Acad. Sci. USA 91:7217–7221.

Rebetzke, G.J., A.G. Condon, R.A. Richards, and G.D. Farquhar. 2003. Gene action for leaf conductance in three wheat crosses. Austral. J. Agr. Res. 54(4):381–387.

Ridzuan, R., M.R. Yusic, M.M. Yusof, S.I. Ismail, G. Miah, and M. Usman. 2018. Genetic diversity analysis of selected Capsicum annuum genotypes based on morpho-physiological, yield characteristics and their biochemical properties. J. Sci. Food Agr., doi: 10.1002/ jfsa.9169.

Rosado-Souza, L., F. Scossa, I.S. Chaves, S. Kleessen, L.F. Salvador, J.C. Milagre, F. Finger, L.L. Bhering, R. Sulpice, W.L. Araújo, and Z. Nikoloski. 2015. Exploring natural variation of photosynthetic, primary metabolism and growth parameters in a large panel of Capsicum chine
tense accessions. Planta 242(3):677–691.

Russo, V.M. and L.R. Howard. 2002. Carotenoids in pungent and non-pungent peppers at various developmental stages grown in the field and glasshouse. J. Sci. Food Agr. 82:615–624.

Serrano, L.L., C. Penella, A.S. Bautista, S.L. Galarza, and A.C. Chover. 2017. Physiological changes of pepper accessions in response to salinity and water stress. Span. J. Agr. Res. 15(3):1–10.

Topaz, A. and F. Ozdemir. 2007. Assessment of carotenoids, capsaiacinoids and ascorbic acid composition of some selected pepper cultivars (Capsicum annuum L.) grown in Turkey. J. Food Compos. Anal. 20:596–602.

USDA NASS Report. 2016. http://usda.mannlib.cornell.edu/MannUsda/viewDocumentInfo.do?docum
tors for Capsicum species. Biochem. Syst. Environ. 156:316–324.

Wahyuni, Y., A.R. Ballester, E. Sudarmonowati, R.J. Bino, and A.G. Bovy. 2011. Metabolite biodiversity in pepper (Capsicum) fruits of thirty-two diverse accessions: Variation in health-related compounds and implications for breeding. Phytochemistry 72:1358–1370.

Yun, S. and M. Ahn. 2009. Effects on net photosynthesis in field-grown hot pepper responding to increased CO2 and temperature. Korean J. Environ. Agri. 28(2):106–112.

Zewdie, Y. and P.W. Bosland. 2001. Capsaicinoid profiles are not good chemotaxonomic indica-
tors for Capsicum species. Biochem. Syst. Ecol. 29:161–169.

Zhai, Y., M. Guo, H. Wang, J. Lu, J. Liu, C. Zhang, Z. Gong, and M. Lu. 2016. Auto-
phagy, a conserved mechanism for protein degradation, responds to heat, and other abiotic stresses in Capsicum annuum L. Front. Plant Sci. 7:131: doi: 10.3389/ fpls.2016.00131.