GAS EXCHANGE CHARACTERISTICS OF *VACCINIUM CORYMBOSUM* L. AND *VACCINIUM DARROWII* CAMP.

By

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Guidance Committee:

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ABSTRACT

GAS EXCHANGE CHARACTERISTICS OF *VACCINIUM CORYMBOSUM* L. AND *VACCINIUM DARROWII* CAMP.

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Comparisons were made between two highbush blueberry cultivars ('Jersey' and 'Bluecrop') and between 'Bluecrop' and Florida 4B, a selection of *Vaccinium darrowi*, with respect to influences of light, CO_2 concentration, temperature and vapor pressure deficit on net CO_2 assimilation, transpiration (E), leaf conductance to water vapor (g_1), and water use efficiency (WUE). The effect of temperature on the gas exchange characteristics of the interspecific hybrid (US75) between 'Bluecrop' and *V. darrowii* (F1a.4B) and two backcrosses to 'Bluecrop' (US239 and US245) were also determined

When measured under optimum conditions, non-significant differences were observed between 'Jersey' and 'Bluecrop' for net CO_2 assimilation, mesophyll conductance, CO_2 compensation point, E, g_1, and WUE. CO_2 assimilation, mesophyll conductance, E, and g_1 were significantly ($p=.05$) lower for *V. darrowii* compared to 'Bluecrop'.

CO_2 assimilation of leaves approached light saturation between 500-800 μmols s^{-1} m^{-2} for 'Bluecrop', 'Jersey' and *V. darrowii*. CO_2 assimilation increased

and g_1 decreased with increasing CO_2 up to 300-350 µl liter^{-1} and leveled off with further increases in CO_2 up to 800 µl liter^{-1}.

There was a 55-65% reduction in g_1 for 'Jersey' and 'Bluecrop' when vapor pressure deficit (VPD) was increased from 1 to 3 KPa but only a 20-25% reduction was observed in CO_2 assimilation. Significant reductions ($p=.05$) in CO_2 assimilation, g_1, and WUE, and significant increases in E were observed in V. darrowii in response to a VPD increase from 1 to 3 KPa. CO_2 assimilation for 'Bluecrop' was significantly higher than rates for V. darrowii at 1 KPa but not at 3KPa, while E and g_1 were significantly lower for V. darrowii at both 1 and 3 KPa. WUE was significantly higher for V. darrowii at 1 KPa but not at 3 KPa.

The temperature optimums for CO2 assimilation ranged between 18 to 26 degrees C for 'Jersey' and between 14 to 22 degrees C for 'Bluecrop'. The temperature optimums for V. darrowii and US75 were approximately 8 to 10 degrees higher than the optimum for 'Bluecrop'. The two backcrosses had contrasting temperature optimums with US239 having a optimum at 20 degrees C similar to 'Bluecrop' and US245 having a optimum at 30 degrees C similar to V. darrowii. V. darrowii had higher WUE's than 'Bluecrop' at both 20 and 30 degrees C, while US239 and US245 had significantly higher WUE's at 30 degrees C compared to 'Bluecrop'.

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INTRODUCTION

Genotypes which are native to differing environmental habitats may contain differences in CO_2 assimilation, transpiration (E), or water use efficiency (WUE) which contribute to their success in their respective environments. If useful adaptive mechanisms could be identified they might be incorporated into a plant breeding program or manipulated by cultural practices.

The tetraploid highbush blueberry (Vaccinium corymbosum L.) is native to swamps, bogs and stream margins from Michigan to Nova Scotia. It is well adapted to organic sandy soils of low pH and is poorly adapted to drought conditions and high temperature. In contrast, Vaccinium darrowii Camp. is a diploid lowbush blueberry species native to the Southeastern United States from Louisiana to Florida. Florida 4B, a selection of V. darrowii, is native to the sandy scrublands of the Ocala National Forest in central Florida, and survives under the high temperatures and the frequent drought conditions of its native habitat. However, there is no information on the mechanisms contributing to the survival of Fla. 4B under hot and dry conditions. Additionally, CO_2 assimilation, E, and leaf conductance to water vapor (g_l)

of V. darrowii and the highbush blueberry V. corymbosum have not been evaluated.

Fla. 4B frequently produces fertile hybrids when crossed with tetraploid Vaccinium species, presumably through the production of unreduced gametes. Fla. 4B was initially used in rabbiteye blueberry (V. ashei) breeding programs with major goals of developing cultivars requiring low chilling and possessing heat and drought resistance. Recently Fla. 4B has been used as a parent in an effort to accomplish these same goals in a highbush blueberry breeding program developed by A.D. Draper of the U.S.D.A. Fruit Lab at Beltsville, Md. Thus the tolerance of highbush blueberry to heat and drought might be improved through incorporation of genes from Fla. 4B.

The two major objectives of this study were to (1) compare the effects of light, CO2, vapor pressure deficit, and temperature on CO_2 assimilation, E, g_1 and WUE of two highbush blueberry cultivars ('Bluecrop' and 'Jersey') and Florida 4B and (2) to test the hypothesis that the tolerance of Fla. 4B to heat and drought results from WUE.

SECTION I

A BASIC COMPUTER PROGRAM FOR CALCULATION OF PHOTOSYNTHESIS, STOMATAL CONDUCTANCE AND RELATED PARAMETERS IN AN OPEN GAS EXCHANGE SYSTEM

Abstract

Computer programs written in BASICA (IMB'S VERSION OF BASIC) language were developed for the calculation of the gas exchange parameters of CO_2 assimilation, leaf conductance, stomatal conductance, residual conductance, intercellular CO_2 concentration, transpiration, water use efficiency and transpiration ratio in an open system. Formulas are discussed in both an algebraic and in a BASIC computer program form. Calculations based on mole fractions of CO_2 and water vapor are explained and both molar and mass fluxes are included in the program output to facilitate comparisons with data from the literature. Corrections are made in the program to accout for underestimation of CO_2 assimilation due to the increase in flow rates out of sample chambers caused by simultaneous transpiration. A sample output is included to illustrate the formatting capability of the program.

Introduction

Closed, semi-closed, and open systems have been used for the study of plant gas exchange (10,11). Of the three systems the open system provides the greatest flexibility for studying environmental effects on photosynthesis, transpiration, and stomatal conductance. The open system offers several convenient features useful in gas exchange studies, such as: 1) the ability to switch between serveral sample chambers to provide for replication; 2) manipu-

lation of CO_2, O_2, and water vapor concentration; and 3) simultaneous measurement of CO_2 and water vapor fluxes (8). Additionally, since the open system is a positive pressure system the technical difficulty of maintaining a totally leak-free system is avoided.

Frequent measurements are required to establish the effects that environmental variables such a CO_2 or vapor pressure concentration have on processes related to plant gas exchange. Measurements of plant gas exchange are often quite variable due to differences in plant material being measured, and interactions between complicated physiological processes, such as control of stomatal aperture, and the environment of the sample chamber. Thus when trying to characterize plant gas exchange, it is desirable to collect large quantities of data. For this reason computer programs were developed to facilitate the rapid calculation and analysis of net CO_2 assimilation, transpiration, leaf conductance, stomatal conductance, residual conductance, intercellular CO_2 concentration, transpiration ratio and water use efficiency in an open system. The programs were written in BASICA which can be used with IBM and IBM-compatible systems. BASIC is a simple and easily learned language which allows for easy manipulation of these programs to meet special needs.

The programs were written to calculate resistance in molar fluxes (m^2 s mol^{-1}), which are now preferred by many students of stomatal activity (3,4,6). The mass of water

vapor and CO_2 are affected by changes in temperature and pressure. Thus corrections for pressure and temperature are required when calculating mass flux. The use of mole fractions to calculate leaf and stomatal resistances removes the pressure and temperature dependence from the calculations (4). Additionally, molar fluxes (m^2 s mol^{-1} or mol m^{-2} s^{-1} for conductance) are more meaningful to the biologist than are the curious units arising form the calculation of mass fluxes (s cm^{-1} or cm s^{-1}).

Since the use of molar flux is relatively new in the literature, conversion factors are included which simultaneously calculate units in mass flux. The format of the program outputs both molar flux and mass flux of each parameter to facilitate comparison.

Calculations

The data that must be collected from the gas exchange system to use these programs is listed in Table 1. The fundamental body of calculations used in the programs is sixty-one lines in length as shown below.

```
 1 SAMPDEWPT=SAMPDPREADING * SAMPDPCORR
 2 AMBDEWPT=AMBDPREADING * AMBDPCORR
 3 SAMPDPCENT=(SAMPDEWPT-32)*5/9
 4 AMBDPCENT=(AMBDEWPT-32)*5/9
 5 T3=LEAFTEMP + 273
 6 T2=SAMPDPCENT + 273
 7 T1=AMBDPCENT + 273
 8 TS=373.16
 9 R1=TS/T1
10 R2=TS/T2
11 R3=TS/T3
12 PART1=-7.90298*(R1-1)+5.02808 * FNLGT(R1,10)
13 PART2=-1.3816*(10^(-7))*(10^(11.344*(1-T1/TS)-1))
14 PART3=8.1328*10^(-8)*(10^(-3.19149*(TS/(T1-1)))-1)
   +FNLGT(1013.246,10)
```

```
15 AMBIENTVAPORPRESSURE=10^(PART1+PART2+PART3)
16 AMBIENTVAPORPRESSURE(KPA)=AMBIENTVAPORPRESSURE/10
17 PART4=-7.90298*(R2-1)+5.02808*FNLGT(R2,10)
18 PART5=-1.3816*(10^(-7))*(10^(11.344*(1-T2/TS)-1))
19 PART6=8.1328*10^(-8)*(10^(-3.19149*(TS/T2-1)))-1)
   +FNLGT(1013.246,10)
20 SAMPLEVAPORPRESSURE=10^(PART4+PART5+PART6)
21 SAMPLEVAPORPRESSURE(KPA)=SAMPLEVAPORPRESSURE/10
22 PART7=-7.90298*(R3-1)+5.02808*FNLGT(R3,10)
23 PART8=-1.3816*(10^(-7))*(10^(11.344*(1-T3/TS)-1))
24 PART9=8.1328*10^(-8)*(10^(-3.19149*(TS/(T3-1)))-1)
   +FNLGT(1013.246,10)
25 LEAFVAPORPRESSURE=10^(PART7+PART8+PART9)
26 LEAFVAPORPRESSURE(KPA)=LEAFVAPORPRESSURE/10
27 VPD=LEAFVAPORPRESSURE(KPA)-
   SAMPLEVAPORPRESSURE(KPA)
28 WIN=AMBIENTVAPORPRESSURE*.001
29 WOUT=SAMPLEVAPORPRESSURE*.001
30 WLEAF=LEAFVAPORPRESSURE*.001
31 DELTACO2READING=IRGACF*DELTAIRGA
32 CALIRGA=.00148*CO2+.5498
33 CO2MICROMOLPERMOL=CALIRGA*DELTACO2READING
34 D=((P*29)/(.0821*T3))/29*1000
35 LEAFAREAM2=LEAFAREA*.01
36 FLOWM3PERS=FLOW*1.667E-05
37 FLOWIN=D*FLOWM3PERS
38 FLOWOUT=FLOWIN*(1-WIN)/(1-WOUT)
39 EMOLAR=FLOWOUT*(WOUT-WIN)/(1-WOUT)/LEAFAREAM2
40 E=EMOLAR*1000
41 WAVG=(WLEAF+WOUT)/2
42 TRANS=EMOLAR*1.8
43 RLMOLAR=(WLEAF-WOUT)/(EMOLAR*(1-WAVG))
44 RLMASS=RLMOLAR/2.5
45 RSMOLAR=RLMOLAR*1.6
46 RSMASS=RLMASS*1.6
47 GLMOLAR=1/RLMOLAR
48 GLMASS=1/RLMASS
49 GL=GLMOLAR * 1000
50 GSMOLAR=1/RSMOLAR
51 GSMASS=1/RSMASS
52 GS=GSMOLAR * 1000
53 A=FLOWOUT*CO2MICROMOLPERMOL*(1-WIN)/(1-WOUT)
   /LEAFAREAM2
54 CO2INTERCELLULAR=((CO2*(GSMOLAR-(EMOLAR/2)))-A/
   (GSMOLAR+(EMOLAR/2))
55 RM=CO2INTERCELLULAR/A
56 GM=1/RM * 1000
57 PNET=A*1.584
58 WUE=(A*.000001)/EMOLAR
59 TRATIO=1/WUE
60 RMMASS=RM/2.5
61 GMMASS=1/RMMASS
```

Table 1. Varibles that must be entered interactively or through the use of READ statements to make calculations of plant gas exchange.

Input Variable	Description
PLANTID$	Plant material identification
LEAFAREA	Leaf area in dm^2
SAMPLEDEWPT	Sample dew point[w]
AMBDEWPT	Ambient dew point[w]
LEAFTEMP	Leaf temperature in degrees C
CO2	Ambient CO_2 concentration
DELTAIRGA	Delta CO_2 reading from IRGA[x]
FLOW	Flow rate (liters min^{-1})[y]
LIGHT	PPFD ($\mu mol\ s^{-1}\ m^{-2}$)[z]

[w]The program will interactively ask for a correction factor to convert these variables to dew point. This allows for the use of a voltage reading from dew point hygrometers.

[x]The program will ask for a conversion factor to convert this reading to $\mu mol\ mol^{-1}$

[y]Flow rate is entered interactively into the program.

[z]This variable is entered interactively except in programs designed to analyze response to changing PPFD.

An algebraic description of the calculations used is listed below and referenced to the pertinent lines where these functions are performed in the program.

(1) Calculation of vapor pressure in KPa from dew point readings.

(a) Water vapor calculations form dew point readings (see references 1,7)

$$\text{Log}_{10}\text{ew} = -7.90298\ (\text{TS}/\text{T}-1)+5.02808\ \log_{10}(\text{TS}/\text{T}) -1.3816\text{x}10^{-7}(10^{11.344(1-\text{T}/\text{TS})}-1) +8.1328\text{x}10^{-8}(10^{-3.19149(\text{TS}/\text{T}-1)}-1) +\text{Log}_{10}\text{ews}$$

Where:

ew = saturation vapor pressure over a plane surface of pure liquid water in millibars (mb)

T = absolute (thermodynamic) temperature degrees K

TS = steam point temperature (373.16)

ews = saturation pressure of pure liquid water at steam-point temperature (1 standard atmosphere = 1013.246 mb)

In the program this calculation is made in lines 12-14 for the air dew point, lines 17-19 for the sample dew point, and lines 22-24 for the leaf temperature (the air spaces of the leaf are assumed to be saturated at the leaf temperature; see reference 8).

(b) Vapor pressure in mb = $10^{\text{Log}_{10}\text{ew}}$

(i) line 15 variable (AMBIENTVAPORPRESSURE) for air

(ii) line 20 variable (SAMPLEVAPORPRESSURE) for the sample

(iii) line 25 variable (LEAFVAPORPRESSURE) for the leaf

(c) Vapor pressure in KPa = vapor pressure in mb/10

(i) line 16 variable [AMBIENTVAPORPRESSURE(KPA)] for air

(ii) line 21 variable [SAMPLEVAPORPRESSURE(KPA)] for sample

(iii) line 26 variable [LEAFVAPORPRESSURE(KPA)] for leaf

Calculations of water vapor concentrations from dew point readings using the above equations have been compared with values obtained from the Smithsonian Tables and have been found to be accurate.

(2) Calculation of vapor pressure deficit (VPD) in KPa

VPD = Leaf vapor pressure minus sample chamber vapor pressure is calculated in line 27 variable (VPD)

(3) Calculation of flow rates into and out of the sample chamber

(a) Flow ($m^3 s^{-1}$) = Flow (liters min^{-1}) x 1.667×10^{-5} calculated in line 36 variable (FLOWM3PERS).

(b) Calculation of flow in mol s^{-1}

(i) calculate the density of air

$d=PM/RT$

Where: d is the density of air in grams

M is the molecular weight of air

R is the gas constant

T is the absolute thermodynamic temperature

(ii) Molar density of air in mol m^{-3}

$D = d/29 \times 1000$

and is calculated in line 34 variable (D)

(iii) Flow into the chamber in mol s^{-1}

Flow into chamber (mol s^{-1}) = D(mol m^{-3} x Flow $m^3 s^{-1}$) and is calculated in line 37 variable (FLOWIN).

(iv) The uptake of CO_2 in the sample chamber is balanced by the efflux of O_2 from the the photosynthetic reaction. However, the release of water vapor into the outgoing airstream through transpiration adds to the flow rate out of the chamber. If this additional flow is not accounted for a substantial underestimation in the calculation of CO_2 assimilation can occur (3,9,12). This error can be large under conditions of high vapor pressure deficit between the leaf and ambient air stream, which promotes rapid transpiration. This program makes a correction for additional flow out of the sample chamber due to

transpiration. For those who have systems where water vapor is scrubbed out of the exit stream before a measurement is made by the IRGA, this correction is unnecessary and this line should be removed (see references 3 and 9 for a full discussion).

(v) Calculation of flow out of chamber

$$f_o = \frac{f_i(1 - w_i)}{(1-w_o)}$$

Where: f_o and f_i are the molar flows of air on the outgoing and incoming air streams

w_o and w_i are the mole fractions of water vapor on the outgoing and incoming air streams.

(vi) w_i = ambient vapor pressure mmol mol^{-1}x.001 and is calculated in line 28 variable (WIN)

(vii) w_o = sample vapor pressure mmol mol^{-1}x.001 and is calculated in line 29 variable (WOUT)

(viii) Flow out is calculated in line 38 variable (FLOWOUT).

(4) Calculation of transpiration

$$E = \frac{f_o \times \frac{w_o - w_i}{1-w_o}}{LA}$$

Where: E is transpiration in mol $m^{-2}s^{-1}$

f_o is the flow out of the sample chamber mol s^{-1}

LA is the leaf area in m^2

w_i and w_o are the mole fractions of water vapor of the incoming and outgoing air streams

(a) Leaf area (m^2) = Leaf area (dm^2) * .01

Calculated in line 35, variable (LEAFAREAM2)

(b) Transpiration mol $m^{-2}s^{-1}$ is calculated in line 39 variable (EMOLAR)

(c) Transpiration in mmol $m^{-2}s^{-1}$ is calculated in line 40, variable (E)

(d) Transpiration in mg cm^{-2} s^{-1} is calculated with the use of a conversion factor in line 42, variable (TRANS)

(5) Calculation of CO_2 assimilation

$$A = \frac{f_o \times (c_i - c_o) \times \frac{(1-w_i)}{(1-w_o)}}{LA}$$

Where: $c_i - c_o$ is the delta CO_2 in the sample chamber

w_i and w_o are the mole fractions of water vapor on the incoming and outgoing air streams

LA is the leaf area in m^2

(a) CO_2 assimilation µmol $m^{-2}s^{-1}$ is calculated in line 54, variable (A)

(b) CO_2 assimilation mg $dm^{-2}hr^{-1}$ is calculated through the use of a conversion factor in line 57, variable (PNET)

(c) additional notes on the calculation of CO_2 assimilation:

(i) Delta CO_2 is calculated in line 31, variable (DELTACO2READING). The program allows for the use of a correction factor that is entered interactively. The use of this conversion factor can be useful in conserving expensive calibration standards.

(ii) Line 32, variable CALIRGA, is a regression equation to correct for change in IRGA response to changing CO_2 concentrations in the system. This correction is necessary when measuring CO_2 assimilation in response to large changes in ambient CO_2 concentration (see ref. 2 for further discussion and for an easy calibration procedure for generating this equation). This equation must be generated independently for each IRGA, and as such the users of these programs must calibrate their IRGA's using standard CO_2 tanks or appropriate CO_2 mixing systems to generate their own response equation. This equation should be substituted in the program at line 32.

(iii) Finally, delta CO_2 in $\mu mol\ mol^{-1}$ is calculated in line 33, variable (CO2MICROMOLPERMOL).

(6) Calculation of leaf resistance

(a) $$r_l = E \times \frac{w_l - w_o}{1 - w_{avg}}$$

Where: r_l is the leaf resistance in mol $m^{-2}s^{-1}$

w_{avg} is the average vapor pressure gradient from the leaf to air

w_l is the vapor pressure of the leaf, assuming that the leaf is saturated at its respective temperature.

w_o is the vapor pressure of the outgoing air stream.

(b) $$w_{avg} = \frac{w_l - w_o}{2}$$

w_{avg} is calculated in line 41, varable (WAVG)

(c) r_l is calculated in line 43, variable (RSMOLAR)

(d) r_l in s cm^{-1} is calculated through the use of a conversion factor (4) in line 44, variable (RLMASS)

Leaf resistance (r_l) estimates the total resistance to water vapor diffusion out of the leaf (8). Leaf resistance contains resistances of r_a (boundary layer resistance), r_s (resistance of the stomatal pore), and r_c (cuticular resistance). Evaluation of the path of these resistances shows that r_a and r_s are in series with each other and in parallel with r_c. Using the electrical analogy to evaluate conductances (1/resistance) gives the total conductance as: $1/r_l=(1/r_s+r_a)+1/r_c$. When the stomata are open r_c is extremely large compared to r_s and r_a and $1/r_c$

approaches zero. Even under stress conditions promoting stomatal closure, r_c is generally never responsible for more than 10% of total leaf resistance. Thus for all practical purposes the equation simplifies to $1/r_1 = 1/r_a + 1/r_s$ (see reference 8 for a more thorough discussion). Boundary layer must be determined independently in each sample chamber. This can be done using moistened filter paper replicas of leaves (8). In well stirred chambers r_a is often so low that it can be ignored. In our system r_a has been estimated to be less than 0.1 s cm^{-1} (2.5 m^2 s mol^{-1}). This value is only 2-5% of the total resistance (r_1) and has been ignored in the calculations (i.e. $r_1=r_s$). This value of r_a should be determined by the users of these programs for their chamber characteristics, leaf sizes, and leaf shapes. In these programs $r_1=r_s$ (line 43). If r_a is significant a line should be added to the calculations where $r_s=r_1-r_a$ (value determined).

(7) Estimating the CO_2 diffusion resistances (r'_s).

When estimating the resistance of CO_2 flux out of the leaf an additional resistance r'_r (residual resistance) is encountered due to the physical resistance of CO_2 diffusion through the mesophyll and the biochemical resistance related to the carboxylation reaction. Thus $r'_1=r'_a+r'_s+r'_r$. Calculations based on the physics of the diffusivities of air, CO_2, and H_2O vapor indicate that the best estimate of the resistance to CO_2 diffusion is equal to r_1 x 1.6 (3,4). Thus stomatal resistance to CO_2 diffusion

(r'_s) is given by: $r'_s = r_s \times 1.6$ which is calculated in line 45, variable (RSMOLAR). Stomatal resistance to CO_2 in mass flux is calculated through the use of a conversion factor in line 46, variable (RSMASS).

(8) Leaf conductance and stomatal conductance are calculated by taking the reciprocal of the respective resistance calculations.

(a) g_l in mol $m^{-2}s^{-1}$ is calculated in line 47, variable (GLMOLAL).

(b) g'_s in mol $m^{-2}s^{-1}$ is calculated in line 50, variable (GLMOLAR).

(c) g_l in cm s^{-1} is calculated in line 48, variable (GLMASS).

(d) g'_s in cm s^{-1} is calculated in line 51, variable (GSMASS).

(e) g_l in mmol $m^{-2}s^{-1}$ is calculated in line 49, variable (GL).

(f) g'_s mmol $m^{-2}s^{-1}$ is calculated in line 52, variable (GS).

(9) Intercellular CO_2

$$C_i = \frac{(g'_s - \frac{E}{2}) \times C_s - A}{(g'_s + \frac{E}{2})} \quad \text{(see ref.3)}$$

Where: C_i is the intercellular CO_2 concentration in μmol mol^{-1}

E is the transpiration rate in mol $m^{-2}s^{-1}$

C_s is the absolute CO_2 concentration in the sample chamber.

A is the CO_2 assimilation rate in μmol $m^{-2}s^{-1}$ Calculated in line 54, variable (CO2INTERCELLULAR).

(10) Residual resistance or mesophyll resistance

$r_r=C_i/A$

(a) Calculated in line 55, variable (RM).

(b) Residual conductance in mmol $m^{-2}s^{-1}$ is the recipocal of r_r and is calculated in line 56, variable (GM)

(c) r_r in s cm^{-1} is calculated in line 60, variable (RMMASS)

(d) g_r in cm s^{-1} is calculated in line 61, variable (GMMASS).

(11) Water use efficiency in moles CO_2 fixed per moles of H_2O lost in transpiration is calculated in line 58, variable (WUE).

(12) Transpiration ratio in moles of H_2O lost per moles of net CO_2 assimilation is calculated in line 59, variable (TRATIO).

Summary

The program was designed to facilitate the rapid calculation of most of the significant parameters related to plant gas exchange. These parameters are expressed both in mass flux and molar flux. The calculations can be incorporated into a computer program formated to generate output of the variables of interest to the researcher.

Programs have been designed to analyze response to changing temperature, CO_2 concentration, vapor pressure deficit gradient, and PPFD. Additional programs have been developed to analyze multiple replications with many sub-samples (over time) per replication per treatment. The output of these programs includes a list of the data, the calculation of all sub-sample values, and the means and standard deviations for each variable per treatment. These programs by their very nature are quite long and complicated and thus could not be used as examples in this publication. However, in order to illustrate the formatting capability possible, we have included a partial print out of the program for analysis of vapor pressure deficit response (Table 2).

Table 2. Sample output of a BASIC program to analyze plant gas exchange

LIST OF DATA

PLANT ONE	0.54	60.30	48.80	23.90	345.00	19.11
PLANT ONE	0.54	60.10	48.80	23.80	340.00	19.06
PLANT TWO	0.29	58.50	49.50	22.60	342.00	14.28
PLANT TWO	0.29	58.80	49.80	22.80	344.00	14.38

TREATMENT NUMBER 1

REPLICATION NUMBER 1

PLANT ID	V P DEFICIT KILOPASCALS	ASSIMILATION micromols PER m2-s	PNET mg CO2 PER dm2-hr	LIGHT micromols PER m2-s	L TEMP DEGREES CENTIGRADE
PLANT ONE	1.19	8.89	14.08	1000.00	23.90
PLANT ONE	1.18	8.80	13.95	1000.00	23.80
MEAN	1.19	8.85	14.01	1000.00	23.85
ST DEV	0.00	0.06	0.09	0.00	0.07

TREATMENT NUMBER 1

REPLICATION NUMBER 2

PLANT ID	V P DEFICIT KILOPASCALS	ASSIMILATION micromols PER m2-s	PNET mg CO2 PER dm2-hr	LIGHT micromols PER m2-s	L TEMP DEGREES CENTIGRADE
PLANT TWO	1.07	12.34	19.54	1000.00	22.60
PLANT TWO	1.09	12.45	19.72	1000.00	22.80
MEAN	1.08	12.39	19.63	1000.00	22.70
ST DEV	0.01	0.08	0.13	0.00	0.14

TREATMENT NUMBER 1

REPLICATION NUMBER 1

PLANT ID	V P DEFICIT KILOPASCALS	LEAF COND millimols PER m2-s	LEAF COND cm/s	S COND millimols PER m2-s	S COND cm/s
PLANT ONE	1.19	211.99	0.530	132.50	0.331
PLANT ONE	1.18	208.56	0.521	130.35	0.326
MEAN	1.19	210.28	0.526	131.42	0.329
ST DEV	0.00	2.43	0.006	1.52	0.004

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SECTION II

THE EFFECTS OF LIGHT, TEMPERATURE, CO_2, AND VAPOR PRESSURE DEFICIT ON PHOTOSYNTHETIC CO_2 ASSIMILATION, STOMATAL CONDUCTANCE, TRANSPIRATION AND WATER USE EFFICIENCY IN 'BLUECROP' AND 'JERSEY' HIGHBUSH BLUEBERRY, (*VACCINIUM CORYMBOSUM* L.)

Abstract

Gas exchange was compared in two cultivars of highbush blueberry (Vaccinium corymbosum L.) in order to characterize the response to light, CO_2, temperature, and vapor pressure deficit (VPD), and to evaluate water use efficiency (WUE) under high temperature and high VPD's. Cultivar differences in rates of CO_2 assimilation were not significant when expressed on a leaf area (11.86 µmols CO_2 m^{-2} s^{-1} for 'Bluecrop' and 11.51 for 'Jersey'), on a leaf dry weight (1.90 µmols CO_2 kg^{-1} dry wt. s^{-1} for 'Bluecrop' and 2.00 for 'Jersey') or on a total chlorophyll basis (19.03 µmols CO_2 g^{-1} chl s^{-1} for 'Bluecrop' and 19.39 for 'Jersey'). Under optimum conditions, differences between the two cultivars were also non-significant for mesophyll conductance (96.5 mmols CO_2 m^{-2} s^{-1} for 'Bluecrop' and 77.5 for 'Jersey'), transpiration (2.37 mmols H_2O m^{-2} s^{-1} for 'Bluecrop' and 2.20 for 'Jersey'), CO_2 compensation points (42.3 µl $liter^{-1}$ for 'Bluecrop' and 41.9 for 'Jersey'), water use efficiency (4.89 µmols CO_2/mmol H_2O for 'Bluecrop' and 5.43 for 'Jersey') and dark respiration (1.57 µmols CO_2 m^{-2} s^{-1} for 'Bluecrop' and 1.61 for 'Jersey'). The net CO_2 assimilation for leaves approached light saturation between 600-800 µmols s^{-1} m^{-2} photosynthetic photon flux density (PPFD) and the temperature optima for CO_2 assimilation ranged between 18 and 26 degrees C for 'Jersey' and between 14 and 22 degrees C for 'Bluecrop'. Leaf conductance to water vapor (g_1) dec-

reased and CO_2 assimilation increased with increasing CO_2 up to normal ambient levels (300-350 µl liter^{-1}) of CO_2 and increased at a slower rate with further increases in CO_2 up to 800 µl liter^{-1}. There was a 55-65% reduction in g_1 (236.4 to 104.8 mmols H_2O m^{-2} s^{-1} for 'Jersey' and 359.5 to 130.4 for 'Bluecrop') when vapor pressure deficit (VPD) was increased from 1 to 3 KPa but only a 20-25% reduction was observed in CO_2 assimilation (9.11 µmols CO_2 m^{-2} s^{-1} to 8.30 for 'Jersey' and 10.64 to 8.38 for 'Bluecrop') in response to VPD.

Stomata on leaves of terrestrial plants regulate gas exchange and therefore to a large extent control CO_2 assimilation and water loss. Thus control of stomatal aperture plays an essential role in determining whether an acceptable ratio of water loss to CO_2 intake is maintained by plants exposed to conditions favoring rapid transpiration (4, 8, 21, 22).

The effects that external environmental factors have on CO_2 assimilation and related gas exchange parameters have been widely investigated in fruit crops (7, 9, 11, 13, 25, 26). This information can be useful in evaluating environmental limitations to productivity in these crops, as well as providing insights into cultural or genetic means of improving their water and carbon efficiency.

Highbush blueberry is a fruit crop in which gas exchange has not been investigated. It is native to swamps, bogs, and stream margins from Michigan to Nova Scotia (3). The cultivated varieties are shallow rooted and devoid of root hairs (10). Highbush blueberry is well adapted to organic soils of low pH and performs poorly under drought conditions and high air temperatures (10).

The purposes of this study were to (a) characterize the gas exchange response of 'Jersey' and 'Bluecrop' highbush blueberry in relation to light, CO_2, temperature, and vapor pressure deficit and (b) evaluate the role of stomatal control over water use efficiency under conditions of high temperature and high vapor pressure deficits, environmental conditions often associated with drought.

Materials and Methods

Plant material. One-year-old 'Bluecrop' and 'Jersey' blueberry plants were grown in ten liter plastic pots in a mixture of 1 sand:1 peat (v/v). The plants were grown under 14 hour photoperiods in a glasshouse and supplemental light was provided by 1000 watt metal halide lamps (GE 1000W M47 BU/H36). Photosynthetic photon flux density (PPFD) at plant level during the growth and maturation of the vegetative flush used for measurements ranged from maxima of 650-1400 to minima of 85-225 µmols s^{-1} m^{-2}. The maximum day temperatures ranged from 18-40 and the minimum night temperatures from 18-27 degrees C. Fertilizer (200 ppm N, 100 ppm P, 100 ppm K, 50 ppm Mg, 100 ppm Fe w/w/w/w/w) was added to the

water used for irrigation as needed to maintain healthy leaf tissue. Phosphoric acid was used to adjust the pH of the water to 5.0.

Gas exchange measurements. Measurements were made on attached leaves of 8-12 week old terminal shoots of lateral branches. The terminal 1-3 leaves, which were fully expanded, were enclosed in environmentally controlled leaf chambers. Measurements were made in an open gas exchange system previously described by Sams and Flore (19), in which light (0-2000 μmols s^{-1} m^{-2} PPFD), ambient CO_2 (0-1000 μl $liter^{-1}$), temperature (10-45 degrees C), and vapor pressure deficit (0.5-3.5 KPa) could be monitored and controlled. Measurements were made on four different plants and twenty determinations were made per leaf per treatment level.

Unless otherwise indicated, gas exchange measurements were made at saturating light intensities (1000 μmols s^{-1} m^{-2}), a leaf temperature of 20 degrees C, ambient CO_2 concentration of 320-345 μl $liter^{-1}$, and at a leaf to air vapor pressure deficit of less than 1 KPa. Plant material was allowed to equilibrate for one hour under the respective initial treatment (light, CO_2, temperature, or vapor pressure deficit) level before measurements were made. Following a step change in treatment level, plant material was allowed to equilibrate for two hours to assure steady state conditions to allow for calculations of intercellular CO_2 concentrations.

Light response curves were determined by exposing leaves to saturating PPFD and subsequently decreasing the PPFD stepwise. The data were fitted by computer to an asymptotic curve of the form $y=a*e^{bx}$ (20). The temperature response curves were determined by measuring gas exchange after initially exposing the plant material to 10-15 degrees C. Leaf temperature was subsequently increased in steps of 4-5 degrees. Vapor pressure deficit was maintained at less than 1 KPa as temperature was increased. The data were fitted to quadratic equations using normal equations as described by Little and Hills (15).

The response to differing levels of ambient CO_2 was derived by exposing plant material to a low CO_2 concentration (90-120 µl liter^{-1}) and subsequently increasing CO_2 concentration stepwise to 800-900 µl liter^{-1}. A logarithmic curve was fitted to the data for CO_2 assimilation versus increasing ambient CO_2. Mesophyll conductance and CO_2 compensation points were determined from linear regression between intercellular CO_2 (0-250 µl liter^{-1}) and CO_2 assimilation.

Gas exchange response to vapor pressure deficit was determined by exposing plant material to low vapor pressure deficits (0.5-1 KPa) and subsequently increasing the vapor pressure deficit in step increments (0.75-1 KPa) up to 3 KPa.

Chlorophyll determinations. Chlorophyll was extracted from a 2.0 cm^2 leaf disk using n,n-dimethylformamide as

described by Moran and Porath (19), using 100 ml of solvent per 5 to 10 g fresh weight of tissue. Samples were kept in the dark at 5 degrees C for 48 hours, then absorbance was read at both 663 and 645 nm using a Beckmann spectrophotometer. Calculations of total chlorophyll were made using the equations of Arnon (1).

Data analysis. Gas exchange parameters were calculated as molar fluxes using the mole fractions of water vapor and CO_2 as suggested by Cowan (4). The calculations were made using computer programs described by Moon and Flore (18). Data were analyzed as a completely randomized design with each plant being a replication (n=4).

Results

Maximum rates of CO_2 assimilation under optimum conditions were the same for 'Bluecrop' and 'Jersey' (Table 1). Differences between the two cultivars were non-significant whether rates were expressed on a leaf area, on a dry weight, or on a total chlorophyll basis. Similar transpiration rates were observed for both cultivars (Table 1), and differences in water use efficiency (WUE), mesophyll conductance (g_m), CO_2 compensation point and dark respiration were also non-significant.

The initial slope of the curves of the response of CO_2 assimilation to PPFD was similar for 'Jersey' (Fig.1a) and 'Bluecrop' (Fig.1b). 'Jersey' approached light saturation at PPFD between 500 and 700 µmols s^{-1} m^{-2}, but 'Bluecrop' approached light saturation only at PPFD levels greater than

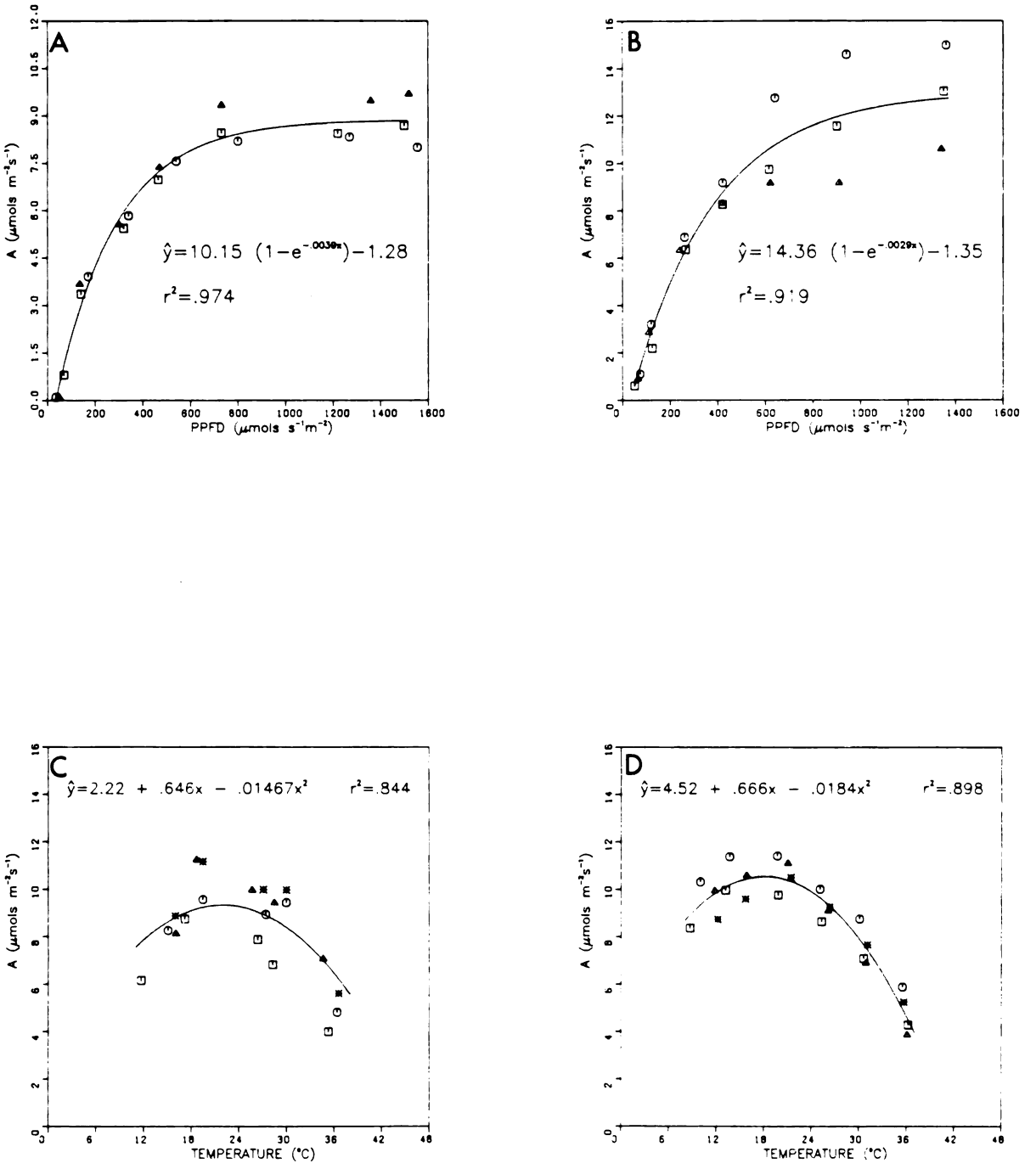
Table 1. Summary of gas exchange characteristics for 'Bluecrop' and 'Jersey'. CO_2 assimilation (A), transpiration (E), and water use efficiency (WUE) were measured at 21.2 degrees C (+/- 1.5), a leaf to air vapor pressure deficit of 1.06 KPa (+/- 0.26), saturating PPFD (1020.6 +/- 37.4 µmols s^{-1} m^{-2}) and at ambient CO_2 levels (328.5 +/- 5.1 µl $liter^{-1}$). Carboxylation efficiency (g_m) and CO_2 compensation points were estimated from the linear portion of the response of A to increasing intercellular CO_2.[a]

Gas exchange parameter	'Bluecrop'	'Jersey'
A (µmols CO_2 m^{-2} s^{-1})	11.86	11.51
A (µmols CO_2 Kg^{-1} dry wt. s^{-1})	1.90	2.00
A (µmols CO_2 g^{-1} chl. s^{-1})	19.03	19.39
E (mmols H_2O m^{-2} s^{-1})	2.37	2.20
WUE (µmols CO_2/mmol H_2O)	4.89	5.43
g_m (mmols CO_2 m^{-2} s^{-1})	96.5	77.5
CO_2 compensation point (µl $liter^{-1}$)	42.3	41.9
Dark respiration (µmols CO_2 m^{-2} s^{-1})	1.57	1.61

[a]Effect of cultivar on any gas exchange parameter was not significant at the 5% level.

Figure 1. Effects of photosynthetic photon flux density (PPFD) (A and B) and leaf temperature (C and D) on CO_2 assimilation (A) in 'Jersey' (A,C) and 'Bluecrop' (B,D). Measurements were made at ambient CO_2 levels (320-345 µl $liter^{-1}$). Response to PPFD was measured at 20°C and response to leaf temperature was made at 1000 µmol s^{-1} m^{-2} PPFD. Each value is the mean of 20 determinations and different symbols represent different plants.

FIGURE 1


A
ŷ=10.15 (1−e^−.0030x)−1.28
r²=.974
A (μmols m⁻²s⁻¹)
PPFD (μmols s⁻¹m⁻²)
B
ŷ=14.36 (1−e^−.0029x)−1.35
r²=.919
A (μmols m⁻²s⁻¹)
PPFD (μmols s⁻¹m⁻²)
C
ŷ=2.22 + .646x − .01467x²
r²=.844
A (μmols m⁻²s⁻¹)
TEMPERATURE (°C)
D
ŷ=4.52 + .666x − .0184x²
r²=.898
A (μmols m⁻²s⁻¹)
TEMPERATURE (°C)

700 µmols s^{-1} m^{-2} (Fig.1b). CO_2 assimilation increased in both cultivars (Figs. 2a and 2b) whereas leaf conductance to water vapor (g_l) decreased (Figs. 2c and 2d) with ambient CO_2. CO_2 compensation points and mesophyll conductance (g_m) were predicted from the linear portion of the response curve for CO_2 assimilation to intercellular CO_2 concentration (Table 1). CO_2 assimilation increased, then declined with increasing temperature in both cultivars (Figs. 1c and 1d). The optimum temperature for CO_2 assimilation ranged between 18-26 degrees C for 'Jersey' and between 14-22 degrees C for 'Bluecrop'.

Increasing the vapor pressure deficit (VPD), from 1 to 3 KPa significantly reduced CO_2 assimilation in 'Bluecrop' (Fig. 3b and Table 2) but not in 'Jersey' (Fig. 3a and Table 2). However, g_l was very sensitive to VPD and declined in both cultivars as VPD increased from 1 to 3 KPa (Figs. 3c,3d and Table 2). Increasing the VPD from 1 to 3 KPa significantly increased transpiration rates in both cultivars (Table 2). Increasing the VPD increased residual conductance significantly in 'Jersey' but not in 'Bluecrop', whereas it reduced WUE significantly for 'Bluecrop' but not for 'Jersey' (Table 2).

Discussion

The rates of CO_2 assimilation of 'Bluecrop'(11.86 µmols CO_2 m^{-2} s^{-1}) and 'Jersey' (11.51 µmols CO_2 m^{-2} s^{-1}) under optimum conditions are about twice those reported for rabbiteye blueberry (23), 50% lower than rates reported for

Figure 2. Effects of CO_2 on CO_2 assimilation (A) (A and B) and on leaf conductance (g_1) (C and D) in 'Jersey' (A,C) and 'Bluecrop' (B,D). Measurements were made at 20 °C and at saturating PPFD (1000 µmols s^{-1} m^{-2}). Each value is the mean of 20 determinations and different symbols represent different plants.

FIGURE 2

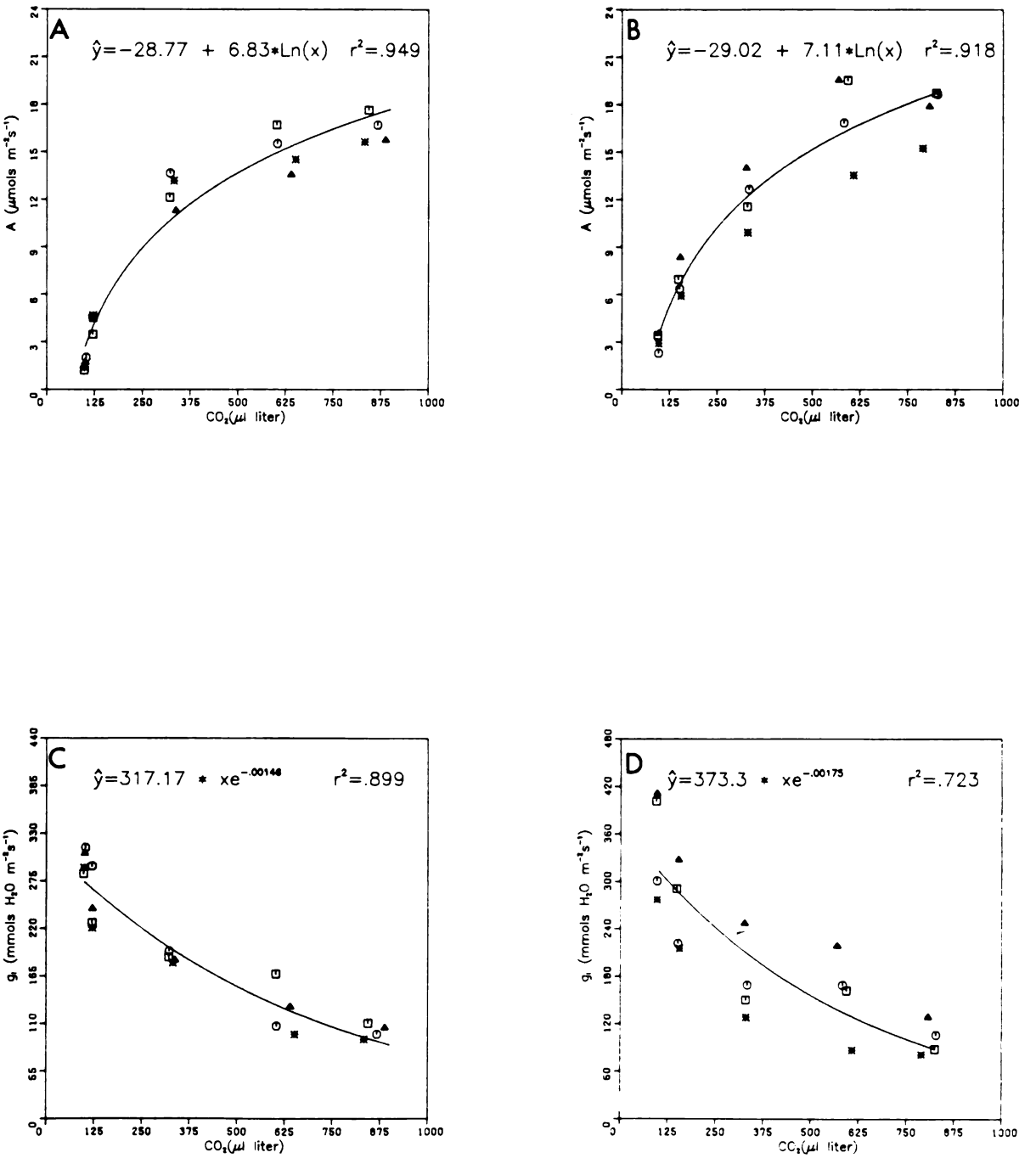

A
ŷ=−28.77 + 6.83*Ln(x) r²=.949
A (μmols m⁻²s⁻¹)
CO₂(μl liter)
B
ŷ=−29.02 + 7.11*Ln(x) r²=.918
A (μmols m⁻²s⁻¹)
CO₂(μl liter)
C
ŷ=317.17 * xe^-.00148 r²=.899
gₛ (mmols H₂O m⁻²s⁻¹)
CO₂(μl liter)
D
ŷ=373.3 * xe^-.00175 r²=.723
gₛ (mmols H₂O m⁻²s⁻¹)
CO₂(μl liter)

Figure 3. Effects of vapor pressure deficit (VPD) on CO_2 assimilation (A) (A and B) and on leaf conductance (g_l) (C and D) in 'Jersey' (A,C) and 'Bluecrop' (B,D). Measurements were made at 28 °C, at saturating PPFD (1000 μmols s^{-1} m^{-2}) and at ambient CO_2 concentrations (314-328 μl $liter^{-1}$). Each value is the mean of 20 determinations and different symbols represent different plants.

FIGURE 3

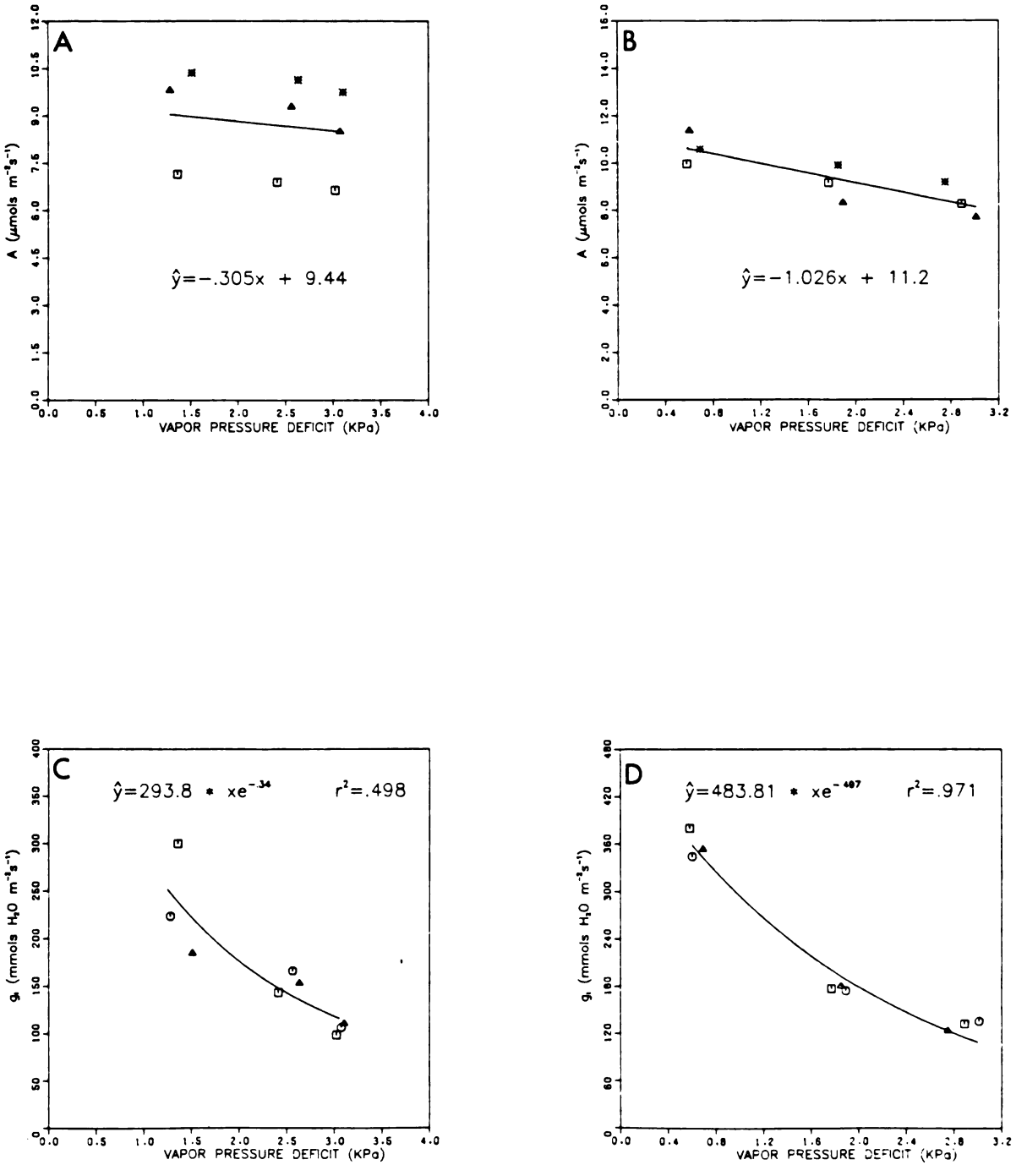

A
ŷ=-.305x + 9.44
A (μmols m⁻²s⁻¹)
VAPOR PRESSURE DEFICIT (KPa)
B
ŷ=-1.026x + 11.2
A (μmols m⁻²s⁻¹)
VAPOR PRESSURE DEFICIT (KPa)
C
ŷ=293.8 * xe^-.34
r²=.498
gs (mmols H2O m⁻²s⁻¹)
VAPOR PRESSURE DEFICIT (KPa)
D
ŷ=483.81 * xe^-.407
r²=.971
gs (mmols H2O m⁻²s⁻¹)
VAPOR PRESSURE DEFICIT (KPa)

Table 2. Effect of vapor pressure deficit on the gas exchange characteristics of 'Bluecrop' and 'Jersey'. CO_2 assimilation (A), leaf conductance to water vapor (g_l), transpiration (E), residual conductance to CO_2 (g_r), and water use efficiency (WUE) were measured at 28.6 degrees C (+/- 2.0), saturating PPFD (985 µmols s^{-1} m^{-2} +/- 37), and at ambient CO_2 concentrations (322.8 µl $liter^{-1}$ +/- 4.9).

Gas exchange parameter	'Jersey'		'Bluecrop'	
	1 KPa	3 KPa	1 KPa	3 KPa
A (µmol m^{-2} s^{-1})	9.11	8.30 n.s.	10.64	8.38*
E (mmol H_2O m^{-2} s^{-1})	1.90	3.44*	2.25	3.75*
g_l (mmol m^{-2} s^{-1})	236.4	104.8*	359.5	130.4*
WUE (µmol CO_2/mmol H_2O)	3.01	2.48 n.s.	4.78	2.60*
g_r (mmol CO_2 m^{-2} s^{-1})	39.2	51.9*	31.2	43.7 n.s.

* - significant at the 5% level.

n.s. - not significant at the 5% level.

apple (1,25), and 15% lower than maximum rates reported for peach, plum and cherry (6).

CO_2 assimilation increased in an asymptotic manner with increasing PPFD for both cultivars. The light saturation range of 500-800 μmols s^{-1} m^{-2} was similar to the saturation levels reported for other fruit crops (4, 5, 6, 9, 24) and for rabbiteye blueberry (23).

The mesophyll conductances of 'Bluecrop' (96.5 mmols CO_2 m^{-2} s^{-1}) and 'Jersey' (77.5 mmols CO_2 m^{-2} s^{-1}), were similar to values reported for almond, plum, peach and cherry (6) and the CO_2 compensation points were typical of those reported for C_3 species. CO_2 assimilation increased rapidly in both cultivars in response to increasing CO_2 concentration up to ambient levels, and then the rate of increase declined due to limitations imposed by the turnover of the carboxylase enzyme.

The general shape of the temperature response curves was parabolic with a decline in CO_2 assimilation at high temperature. The response to temperature was independent of humidity up to 30 degrees C, as VPD was controlled at less than 1 KPa. At temperatures above 30°C, VPD could not be stabilized at gradients less than 1 KPa because the laboratory temperature could not be raised sufficiently to prevent condensation of water vapor in the gas lines. VPD's ranged up to 1.7 KPa at high temperatures. The reduction in net CO_2 assimilation rate at high temperature was not as severe as some reported in many previous studies because VPD was

held constant as temperature increased. The broad temperature optimum range (18-26 degrees C) for 'Jersey' was similar to ranges reported for walnut (24) and pecan (5), whereas 'Bluecrop' had a lower temperature optimum (14-22 degrees C) than those observed for other fruit crops (5, 14, 20).

Stomatal closure in response to increasing VPD has been observed in most species of plants investigated (22). However, studies with several woody perennials have indicated that g_l effects on transpiration due to increasing VPD were small, and that substantial increases in transpiration occurred in these species at high VPD's (22). This was not the case with the highbush blueberry cultivars, as g_l is reduced by about 50% when VPD was increased from 1 to 1.5 KPa. Reduction of g_l in highbush blueberry due to increasing VPD imposed a greater restriction on transpiration than on CO_2 assimilation. When VPD was increased from 1 to 3 KPa there was only a modest (20-25%) reduction in CO_2 assimilation. Similar responses have been observed in Douglas fir (16, 17) and Sitka spruce (27). The fact that g_l was more sensitive to VPD than CO_2 assimilation suggests that highbush blueberry does not possess a strong feedback on stomatal aperture that maintains intercellular CO_2 constant. This contrasts with the strong coupling between g_l and CO_2 assimilation in apple (14). Residual conductance to CO_2 increased in highbush blueberry when VPD was increased from 1 to 3 KPa, which indicated that under the conditions of

this study CO_2 assimilation potential was not reduced by high VPD.

The range of g_1 is 90 to 320 mmol H_2O m^{-2} s^{-1} for deciduous fruit trees and 64 to 220 for other deciduous woody plants (12). Leaf conductance values for 'Bluecrop' (359.5) and 'Jersey' (236.4), at 28 degrees C and 1 KPa were above the upper limit of the range for woody perennials, but compared favorably with g_1 values reported for peach, plum, and cherry, which were measured under similar conditions (6). Thus, the stomatal control of water loss in highbush blueberry was no better or worse than that observed in other fruit crops. Transpiration rates and WUE for highbush blueberry were also similar to values reported for Prunus species (6). In contrast g_1 and transpiration rates were twice those reported for rabbiteye blueberry (23), but WUE's were similar, due to the higher CO_2 assimilation rates of highbush blueberry. The rabbiteye blueberry is reported to be drought tolerant (23) and restriction of water loss due to low rates of stomatal conductance may be a physiological adaptation that contributes to this drought tolerance. The rates of g_1 and CO_2 assimilation reported for rabbiteye blueberry are similar to those reported for apricot, some cultivars of which are adapted to desert conditions (6).

No evidence was obtained in this study, that inefficient stomatal control over water loss under high vapor pressure deficits is responsible for highbush blueberry's poor adaptation to drought. However, the WUE of highbush

blueberry might be improved through plant breeding if parents can be identified that can restrict transpirational losses through low values of g_l without imposing too large a restriction on CO_2 assimilation.

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SECTION III

A COMPARISON OF A LOWBUSH BLUEBERRY (*VACCINIUM DARROWII* CAMP.) VERSUS HIGHBUSH BLUEBERRY (*VACCINIUM CORYMBOSUM* L.): EFFECTS OF LIGHT, TEMPERATURE, CO_2 AND VAPOR PRESSURE DEFICIT ON PHOTOSYNTHETIC CO_2 ASSIMILATION, STOMATAL CONDUCTANCE, TRANSPIRATION AND WATER USE EFFICIENCY

Abstract

Gas exchange characteristics were determined for a wild diploid lowbush blueberry species (Vaccinium darrowii Camp.) and were compared with gas exchange characteristics previously determined for highbush blueberry (Vaccinium corymbosum L.), in order to determine if the greater tolerance of V. darrowii to high temperatures and drought conditions was due to stomatal control over water use efficiency. The maximum CO_2 assimilation rates for V. darrowii under optimum conditions were 8.55 µmols CO_2 m^{-2} s^{-1} on a leaf area, 1.04 µmols CO_2 kg^{-1} dry wt s^{-1} on a leaf dry weight and 13.45 µmols CO_2 g^{-1} chl s^{-1} on a total chlorophyll basis. Mesophyll conductance (55.3 mmols CO_2 m^{-2} s^{-1}), transpiration (E) (1.71 mmols H_2O m^{-2} s^{-1}), and leaf conductance (g_l) (140.4 mmols H_2O m^{-2} s^{-1}) were lower, while CO_2 compensation point (88.8 µl $liter^{-1}$) and water use efficiency (WUE) (5.00 µmols CO_2/mmol H_2O) were higher than values reported for 'Bluecrop'. CO_2 assimilation by leaves approached light saturation between 500 and 800 µmols s^{-1} m^{-2} photosynthetic photon flux density (PPFD). CO_2 assimilation increased and g_l decreased with increasing ambient CO_2. Increasing vapor pressure deficit (VPD) from 1 to 3 KPa significantly (p=.05) reduced CO_2 assimilation (8.87 µmols CO_2 m^{-2} s^{-1} to 7.43), g_l (168.1 mmols m^{-2} s^{-1} to 85.4) and WUE (6.45 µmols CO_2/mmol H_2O to 2.68), while E was significantly increased (1.38 mmols H_2O m^{-2} s^{-1} to 2.68). CO_2 assimilation for V. darrowii was significantly (p=.05) lower than rates for

'Bluecrop' at 1 KPa (10.63 μmols m^{-2} s^{-1} to 8.87) but not at 3 KPa, while E and g_1 were significantly lower at both 1 and 3 KPa. WUE was significantly (p=.05) higher for V. darrowii at 1 KPa (6.45 μmols CO_2/mmols H_2O) but not at 3 KPa. The temperature optimum for V. darrowii ranged between 25 to 30 degrees C, approximately 8 to 10 degrees higher than that reported for 'Bluecrop'. Transpiration and g_1 were lower and WUE was higher for V. darrowii than for 'Bluecrop' at both 20 and 30 degrees C. Residual conductance (g_r) to CO_2 increased for V. darrowii with a temperature increase from 20 to 30 degrees C, whereas it decreased in 'Bluecrop'.

Gas exchange characteristics differ in plants adapted to different environmental habitats (8), and their successful adaptation may depend upon differences in CO_2 assimilation, transpiration (E), or the ratio of the two which is called water use efficiency (WUE). Highbush blueberry (Vaccinium corymbosum L.) is well adapted to sandy organic soils of low pH. The root system is devoid of root hairs and the plants are reported to perform poorly under drought conditions and high temperature (10). In contrast, Vaccinium darrowii a wild diploid blueberry species, is well adapted to dry sites and high air temperatures in the Southeastern United States (3, 13, 16). However, the basis for the drought and heat tolerance of V. darrowii has not been investigated.

Previously (14) we reported on the effects that external environmental factors have on total gas exchange characteristics of highbush blueberry. This study was initiated to characterize the affect of light, CO_2, temperature and vapor pressure deficit on CO_2 assimilation and related gas exchange parameters for V. darrowii and to determine if there were differences in gas exchange between V. darrowii and highbush blueberry which could explain the differences in their heat tolerance and drought resistance. If useful adaptations in gas exchange properties occur and if they are heritable, they could be incorporated into highbush blueberry.

Materials and Methods

Plant material. One-year-old plants of Florida 4B, a selection of V. darrowii (supplied by A.Draper U.S.D.A. Fruit Lab, Beltsville, Md.) were grown in ten liter plastic pots in a mixture of 1 sand:1 peat (v/v). The methods used were identical to those employed for highbush blueberry (14), with the following exceptions: measurements were made on 10-20 leaves of an actively growing shoot; chlorophyll was determined on 2 to 4 whole leaves and measurements were made at 30 degrees C unless otherwise indicated.

Results

Maximum CO_2 assimilation for V. darrowii under optimum conditions was 8.55 µmols CO_2 m^{-2} s^{-1} on a leaf area basis, 1.04 µmols CO_2 kg^{-1} dry wt. s^{-1} on a leaf dry weight basis, and 13.45 µmols CO_2 g^{-1} chl s^{-1} on a total chlorophyll basis

(Table 1.). Mesophyll conductance (g_m) was estimated to be 53.3 mmols CO_2 m^{-2} s^{-1} and the CO_2 compensation point was 88.8 µl $liter^{-1}$ (Table 1). Transpiration (E) was 1.71 mmol H_2O m^{-2} s^{-1} and water use efficiency (WUE) was 5.00 µmol CO_2/mmol H_2O. Dark respiration (1.06 µmols CO_2 m^{-2} s^{-1}) was about 15% of CO_2 assimilation (Table 1).

CO_2 assimilation for leaves approached light saturation level at PPFD of 600-800 µmols s^{-1} m^{-2} (Fig. 1). The optimum temperature for CO_2 assimilation ranged between 25 to 30 degrees C (Fig. 1). CO_2 assimilation increased (Fig. 1) and leaf conductance to water vapor (g_l) decreased (Fig. 2) logarithmically as ambient CO_2 was increased. CO_2 assimilation decreased 15-20% as vapor pressure deficit (VPD) increased from 1 to 3 KPa (Fig. 1 and Table 2). Leaf conductance to water vapor decreased 50% as VPD increased to 1.5 KPa (Fig. 2) but did not decrease with further increases in VPD up to 3 KPa. Significant reductions (p=.05) were observed in CO_2 assimilation, g_l, and WUE as VPD increased from 1 to 3 KPa (Table 2), while E increased significantly.

CO_2 assimilation increased in V. darrowii as temperature rose from 20 to 30 degrees C, but decreased in highbush blueberry (Table 3). Transpiration was greater at the higher temperature in both V. darrowii and 'Bluecrop' but at either temperature E was much lower for V. darrowii (Table 3). WUE and g_l declined with temperature in both species, while calculated residual conductance to CO_2 (g_r) decreased in 'Bluecrop' but increased in V. darrowii (Table 3).

Table 1. Summary of gas exchange characteristics for *V. darrowii*. CO_2 assimilation (A), leaf conductance (g_l), transpiration (E), and water use efficiency (WUE) were measured at 30.7 degrees C (+/- 0.5), a leaf to air vapor pressure deficit of 1.08 KPa (+/- 0.37), saturating PPFD 1017 µmols s^{-1} m^{-2} (+/- 24.4), and ambient CO_2 levels 337.7 µl $liter^{-1}$ (+/- 11.4). Carboxylation efficiency (g_m) and CO_2 compensation point were estimated from the linear portion of the response of A to increasing intercellular CO_2.

Gas exchange parameter	Mean value
A (µmol CO_2 m^{-2} s^{-1})	8.55
A (µmol CO_2 Kg^{-1} dry wt s^{-1})	1.04
A (µmol CO_2 g^{-1} chl s^{-1})	13.45
E (mmol H_2O m^{-2} s^{-1})	1.71
g_l (mmol m^{-2} s^{-1})	140.2
WUE (µmol CO_2/mmol H_2O)	5.00
g_m (mmol CO_2 m^{-2} s^{-1})	53.3
CO_2 compensation point	88.8
Dark respiration (µmol CO_2 m^{-2} s^{-1})	1.06

Figure 1. Effect of photosynthetic photon flux density (PPFD) (A), leaf temperature (B), CO_2 (C), and leaf to air vapor pressue deficit (VPD) (D) on net CO_2 assimilation in V. darrowii. Measurements were made at 30 degrees C (A,C, and D), saturating PPFD (B,C, and D), and at ambient CO_2 levels (330-345 µl liter^{-1}) (A,B, and D). Each value is the mean of 20 determinations and different symbols represent different plants.

FIGURE 1

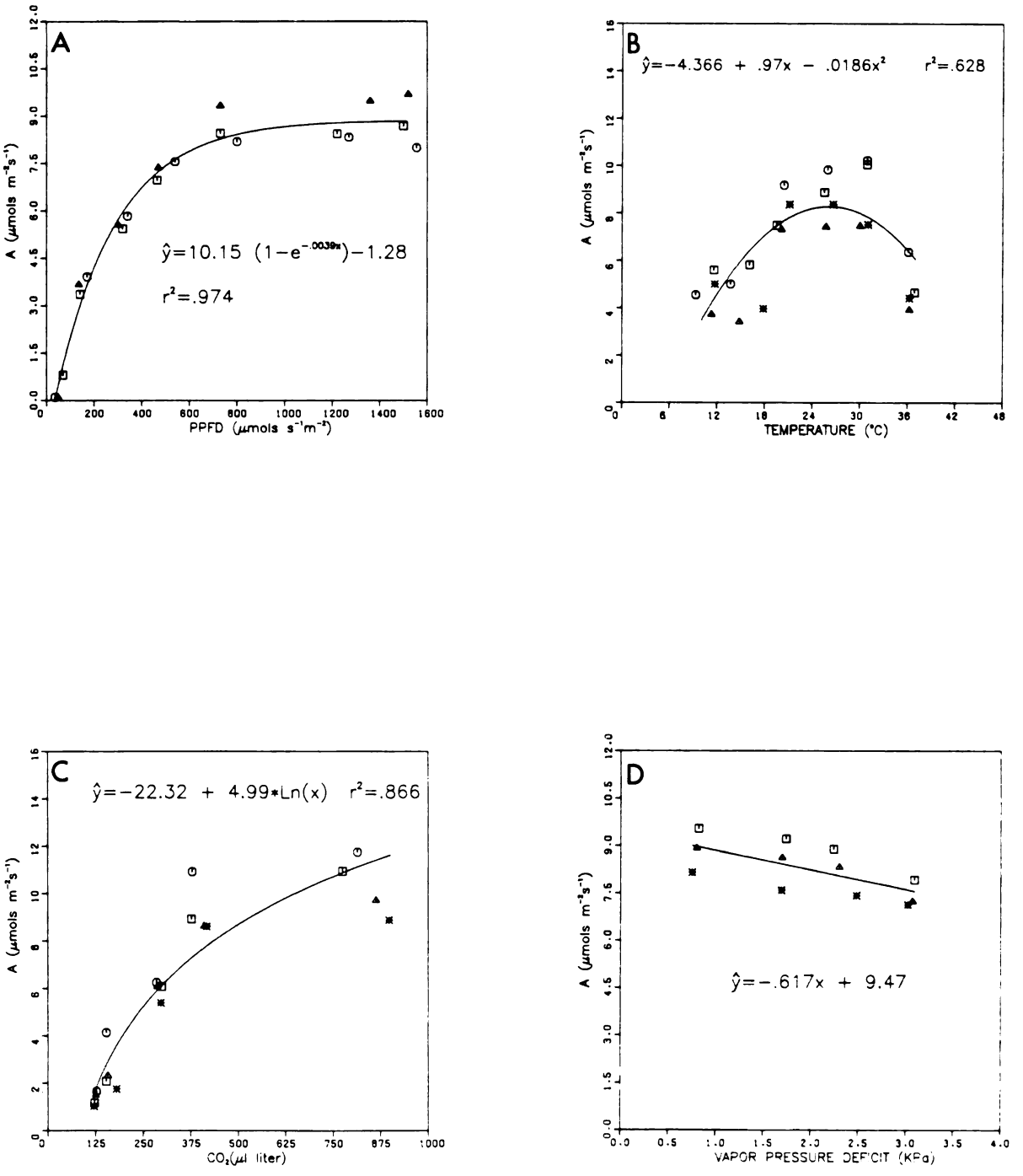
A
ŷ=10.15 (1−e^−.0039x)−1.28
r²=.974
A (μmols m⁻²s⁻¹)
PPFD (μmols s⁻¹m⁻²)
B
ŷ=−4.366 + .97x − .0186x²
r²=.628
A (μmols m⁻²s⁻¹)
TEMPERATURE (°C)
C
ŷ=−22.32 + 4.99*Ln(x)
r²=.866
A (μmols m⁻²s⁻¹)
CO₂(μl liter)
D
ŷ=−.617x + 9.47
A (μmols m⁻²s⁻¹)
VAPOR PRESSURE DEFICIT (KPa)

Figure 2. Effect of ambient CO_2 concentration (A) and leaf to air vapor pressure deficit (VPD) (B) on leaf conductance (g_l) for V. darrowii. Measurements were made at saturating PPFD (1000 µmol s^{-1} m^{-2}), and at 30 degrees C. Response to VPD (B) was measured at ambient CO_2 levels (330-335 µl $liter^{-1}$). Each value is the mean of 20 determinations and different symbols represent different plants.

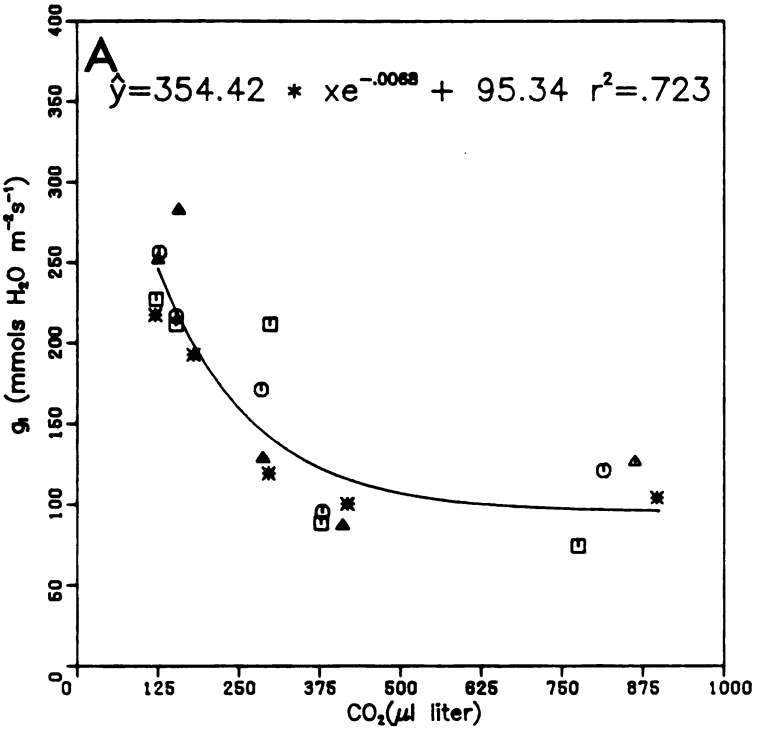
A
ŷ=354.42 * xe^-.0068 + 95.34 r²=.723
gₗ (mmols H₂O m⁻²s⁻¹)
CO₂(µl liter)
0
50
100
150
200
250
300
350
400
125
250
375
500
625
750
875
1000

FIGURE 2

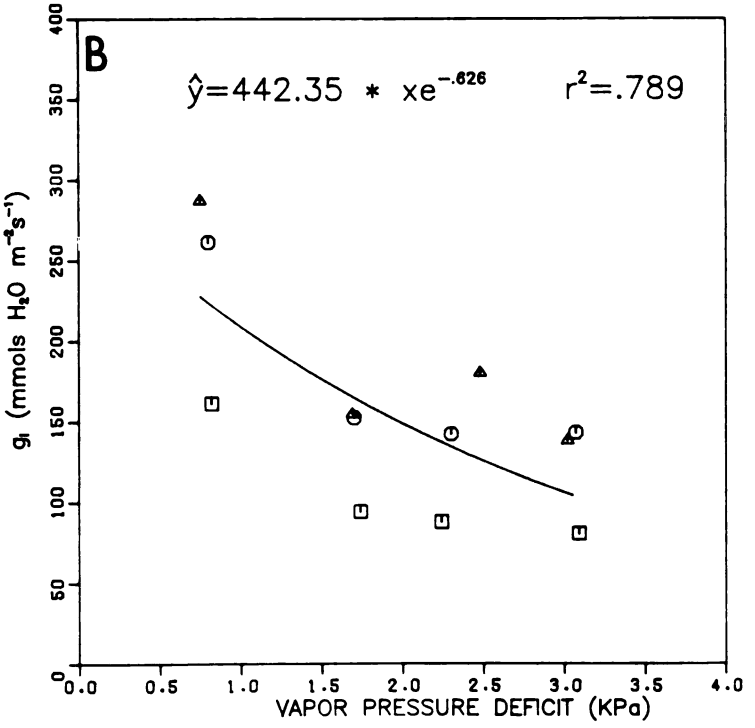
B
ŷ=442.35 * xe^-.626
r²=.789
gₗ (mmols H₂O m⁻²s⁻¹)
VAPOR PRESSURE DEFICIT (KPa)
0
50
100
150
200
250
300
350
400
0.0
0.5
1.0
1.5
2.0
2.5
3.0
3.5
4.0

Table 2. Summary of the effect of leaf to air vapor pressure deficit in kilopascals (KPa) on the gas exchange characteristics of *V. darrowii*. CO_2 assimilation (A), leaf conductance (g_l), transpiration (E), and water use efficiency (WUE) were measured at 30.6 degrees C (+/- 1.4), saturating PPFD (1033 µmol s^{-1} m^{-2} +/- 34), and at ambient CO_2 concentrations (334.7 +/- 3.1).

Gas exchange parameter	Vapor Pressure Deficit 1 KPa	3 KPa
A (µmol CO_2 m^{-2} s^{-1})	8.87	7.43*
E (mmol H_2O m^{-2} s^{-1})	1.38	2.68*
g_l (mmol m^{-2} s^{-1})	168.1	85.4*
WUE (µmol CO_2/mmol H_2O)	6.45	2.78*

* Significantly different from value for 1 KPa at the 5% level

Table 3. Comparison of the effects of leaf temperature on the gas exchange characteristics of highbush blueberry 'Bluecrop' and *V. darrowii*. Mean values for CO_2 assimilation (A), transpiration (E), leaf conductance (g_l), residual conductance to CO_2 (g_r), and water use efficiency (WUE) were obtained from temperature response curves measured at vapor pressure deficits less than 1.0 KPa, saturating PPFD (1000 µmol s^{-1} m^{-2}), and ambient CO_2 levels (330 µl $liter^{-1}$).

Gas exchange parameter	20 degrees C		30 degrees C	
	V. darrowii	'Bluecrop'	*V. darrowii*	'Bluecrop'
A (µmol CO_2 m^{-2} s^{-1})	8.10	10.70 *	8.83	7.63 n.s.
E (mmol H_2O m^{-2} s^{-1})	1.45	2.69 *	2.39	3.04 n.s.
g_l (mmol m^{-2} s^{-1})	117.5	237.3 *	93.1	155.8 *
WUE (µmol CO_2/mmol H_2O)	5.64	4.01 *	3.73	2.53 *
g_r (mmol CO_2 m^{-2} s^{-1})	34.1	42.3 *	49.9	32.0 *

* Significantly different at the 5% level.

while calculated residual conductance to CO_2 (g_r) decreased in 'Bluecrop' but increased in V. darrowii (Table 3).

Transpiration and g_1 were significantly lower (p=.05) in V. darrowii at VPD's of both 1 and 3 KPa (Table 4). CO_2 assimilation was significantly higher for 'Bluecrop' at 1 KPa but not at 3 KPa (Table 4). WUE of V. darrowii was 35-40% higher than 'Bluecrop' at 1 KPa but no significant difference in WUE was observed at 3 KPa (Table 4).

Discussion

Maximum CO_2 assimilation rates under optimum conditions for V. darrowii were 12-20% higher than rates reported for rabbiteye blueberries (19) and 25-35% lower than those reported for highbush blueberry (14).

The light response curve for V. darrowii was similar to those reported for rabbiteye (19) and highbush blueberry (14), as well as for other fruit crops (5, 6, 7, 9, 20) in that light saturation was approached between 500 and 800 μmols $s^{-1}m^{-2}$ PPFD. CO_2 assimilation increased and g_1 decreased with increasing CO_2 in a manner similar to that reported for highbush blueberry (14). Mesophyll conductance (g_m) which was estimated from the CO_2 response curve, was lower than in highbush blueberry (14). This lower capacity for CO_2 assimilation may reflect the lower range of g_1 observed, which restricts intercellular CO_2 concentration. A similar restriction in g_m due to stomatal aperture has been reported for apricot, some cultivars of which are adapted to desert conditions (7).

Table 4. Comparison of the effects of leaf to air vapor pressure deficit on the gas exchange characteristics of highbush blueberry 'Bluecrop' and *V. darrowii*. Mean values for CO_2 assimilation (A), transpiration (E), leaf conductance (g_l), residual conductance to CO_2 (g_r) and water use efficiency (WUE) were obtained from vapor pressure deficit response curves measured at 28 to 31 degrees C, ambient CO_2 concentrations 320-335 µl $liter^{-1}$ and at saturating PPFD 985-1035 µmol s^{-1} m^{-2}.

Gas exchange parameter	1 KPa		3 KPa	
	'Bluecrop'	*V. darrowii*	'Bluecrop'	*V. darrowii*
A (µmol CO_2 m^{-2} s^{-1})	10.63	8.87 *	8.50	7.43 n.s.
E (mmol H_2O m^{-2} s^{-1})	2.25	1.38 *	3.96	2.68 *
g_l (mmol m^{-2} s^{-1})	359.5	168.1 *	130.4	85.4 *
g_r (mmol CO_2 m^{-2} s^{-1})	39.2	37.6 n.s.	54.4	40.9 n.s.
WUE (µmol CO_2/mmol H_2O)	4.78	6.45 *	2.60	2.78 n.s.

* significant at the 5% level.

n.s. not significant at the 5% level.

VPD had a greater effect on g_l than on CO_2 assimilation, as reported for highbush blueberry (14).

CO_2 assimilation was optimum between 25 and 30 degrees C, as reported for peach (5), apple (11), and cherry (18); this is several degrees higher than the temperature optimum reported for highbush blueberry (14). Transpiration and g_l at 20 and 30 degrees C, were much lower for V. darrowii than for 'Bluecrop' highbush blueberry. Therefore one factor that determines the survival of V. darrowii under high temperatures and drought conditions may be an ability to restrict water loss by decreasing stomatal aperture. Such a response has also been suggested in rabbiteye blueberry which is reported to be drought resistant (10, 16, 19), and exhibits even lower ranges of g_l than V. darrowii (19). The restriction of water loss under conditions of high evaporative demand may more than compensate for the slightly lower rates of CO_2 assimilation in these drought resistant blueberries.

V. darrowii may possess a heritable component favoring survival at higher temperatures. Similar types of ecological differentiation have been reported in other species (2). Because crosses are possible between highbush blueberry and V. darrowii (16), the possibility that heat tolerance and drought resistance can be improved in highbush blueberry through the incorporation of genes from V. darrowii should be evaluated.

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SECTION IV

GENOTYPIC DIFFERENCES IN THE EFFECT OF TEMPERATURE ON CO_2 ASSIMILATION AND WATER USE EFFICIENCY IN BLUEBERRY

Abstract

In order to determine the feasibility of improving the net CO_2 assimilation and water use efficiency of highbush blueberry under high temperature, gas exchange determinations were made for a selection of Vaccinium darrowii (Florida 4B), a highbush cultivar 'Bluecrop' (V. corymbosum), their F1 hybrid (US75) and two backcrosses to 'Bluecrop' (US239 and US245). Maximum CO_2 assimilation of US75 (15 µmols CO_2 m^{-2} s^{-1}) was 30-40% higher than that of either parent. All genotypes responded parabolically to increasing temperature at vapor pressure deficits less than 1 KPa. CO_2 assimilation of US75 and F1a. 4B was optimum at 30 degrees C, that of 'Bluecrop' at 20 degrees. US239 had an optimum at 20 degrees C, similar to 'Bluecrop', and US245 had a higher temperature optimum (30 degrees C) similar to F1a 4B. F1a 4B had higher WUE's than 'Bluecrop' at both 20 (5.64 µmols CO_2/mmol H_2O to 4.01) and 30 degrees C (3.73 to 2.53). The backcrosses US239 and US245 had significantly ($p=.05$) higher WUE's at 30 degrees C than did 'Bluecrop'. Residual conductance to CO_2 decreased in 'Bluecrop' when temperature was raised from 20 to 30 degrees C but increased in all other genotypes. Due to the favorable gas exchange properties of US75 and US245 at 30 degrees C, we suggest that high temperature tolerance of V. darrowii may be heritable and that US245 may be a useful parent in a breeding program to improve the heat tolerance of highbush blueberry.

Vaccinium corymbosum L. grows well under relatively cool moist conditions; V. darrowii (Fla 4B) in contrast, occurs on hot dry sandy scrublands in central Florida (10). As such, they may possess physiological adaptations that improve their net CO_2 assimilation and water use efficiency under the temperature conditions of their respective habitats.

One selection of V. darrowii (Florida 4B) has a temperature optimum for net CO_2 assimilation approximately eight to ten degrees higher than that of 'Bluecrop', a cultivar of V. corymbosum (14). Although V. corymbosum and V. darrowii differ in ploidy, A. Draper (6) has developed a series of hybrids between these species presumably involving unreduced gametes. This study was undertaken to compare the photosynthetic performances of the parent genotypes with those of both an F1 hybrid (US75) and backcrosses between US75 and 'Bluecrop' (US239 and 245).

Material and Methods

Plant material. Dormant, one-year-old rooted cuttings of 'Bluecrop', Fla. 4B, US75, US239 and US245 (BC) blueberry were transplanted into ten liter plastic pots in a mixture of 1 sand:1 peat (v/v) in April of 1983. Plants were grown together for one year in a completely randomized design in a glasshouse under 14 hour photoperiods. Growth and cultural conditions were the same as those previously described (14). To induce dormancy plants were put into a growth chamber on January 26, 1984. Temperature in the growth chamber

was maintained at 3 degrees C and eight hour photoperiods were supplied with high pressure sodium lamps which provided light intensities between 200-350 μmols s^{-1} m^{-2} PPFD. Plants were removed from the growth chamber on April 16, 1984 and were again placed in the greenhouse in a completely randomized design. The maximum day temperature ranged from 22-40 degrees C and the minimum night temperatures from 22-28 degrees C. Maximum light intensities during the growth and maturation of the vegetative flush used in measurements ranged from 600-950 μmols s^{-1} m^{-2} PPFD. Gas exchange measurements were made during July of 1984 on 6-10 week old terminal shoots of lateral branches. The terminal 1-3 leaves of a mature flush of 'Bluecrop', US75, US239, and US245 and the terminal 10-20 leaves of an active vegetative terminal shoot of Fla 4B were enclosed in environmentally controlled leaf chambers. Measurements were made in an open gas exchange system previously described (14). Measurements were made on four different plants per genotype, and twenty determinations were made per leaf per treatment level.

Data analysis. Gas exchange parameters were calculated as molar fluxes using the mole fractions of water vapor and CO_2 as suggested by Cowan (5). The calculations were made using computer programs described by Moon and Flore (15). Data were analyzed for a completely randomized design with each plant being a replication. The effect of temperatures (20 and 30 degrees C) was analyzed statistically as a split

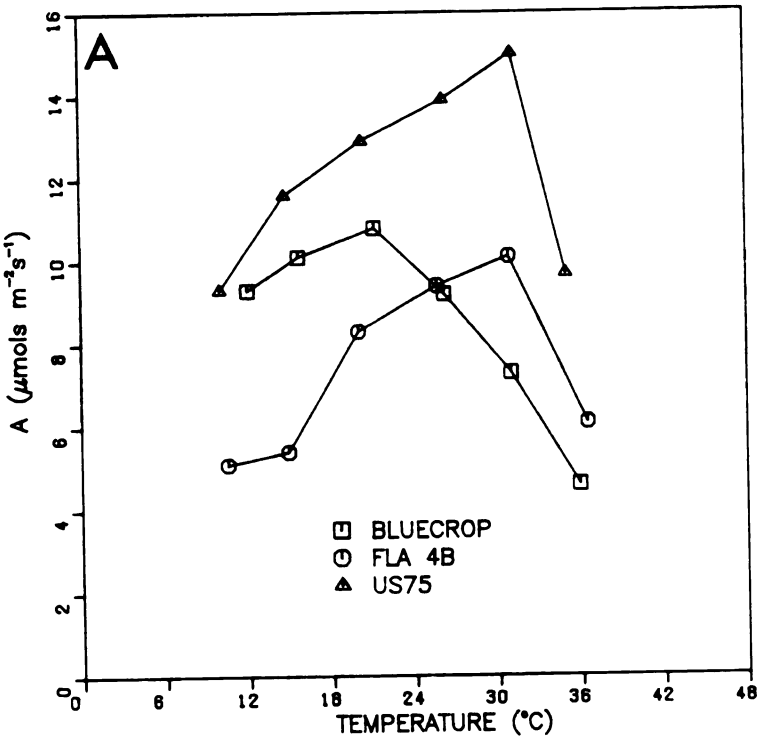
plot with four replications (plants) with genotype being the main effect.

Results

All genotypes responded to increasing temperature in a parobolic manner. CO_2 assimilation was greatest for 'Bluecrop' at about 20 degrees C (Fig. 1), whereas the optimum for F1a 4B was 28-30 degrees C. The interspecific hybrid (US75) had significantly ($p=.05$) higher rates of CO_2 assimilation than either parent (Fig 1,Table 1) and a broad temperature optimum 20 to 30 degrees C, that parallels F1a 4B. The two backcrosses appeared to be segregating for response of CO_2 assimilation to temperature with US239 resembling the 'Bluecrop' parent and US245 resembling F1a 4B (Fig. 1). Transpiration (E) increased and WUE decreased in all genotypes as temperature rose from 20 to 30 degrees C (Table 1). Leaf conductance to water vapor (g_l) declined by 35% in 'Bluecrop' and 21% in F1a 4B when temperature increased from 20 to 30 degrees C but g_l was unaffected in the two backcrosses (Table 2). There was a 91% increase in g_l in US75 as temperature increased (Table 2). High temperature reduced residual conductance to CO_2 (g_r) in 'Bluecrop' and 'US239', increased it in US245 and F1a 4B, but had no effect in US75 (Table 2).

The effects of genotype and temperature on all gas exchange parameters examined were significant with one exception (Table 3). Lack of significance in this case was probably due to the large differences in maximum rates of

Figure 1. Effect of leaf temperature on net CO_2 assimilation (A) for 'Bluecrop', V. darrowii (F1a 4B), and US75 (A), and US239 and US245 (B). Measurements were made at saturating PPFD (1000 µmol s^{-1} m^{-2}), at ambient CO_2 levels (340 µl $liter^{-1}$) and at leaf to air vapor pressure deficits less than 1 KPa. Each symbol represents the mean of 20 determinations and each curve is representative of a typical response for its respective genotype.


A
A (μmols m⁻²s⁻¹)
TEMPERATURE (°C)
BLUECROP
FLA 4B
US75
0
2
4
6
8
10
12
14
16
0
6
12
18
24
30
36
42
48

FIGRUE 1

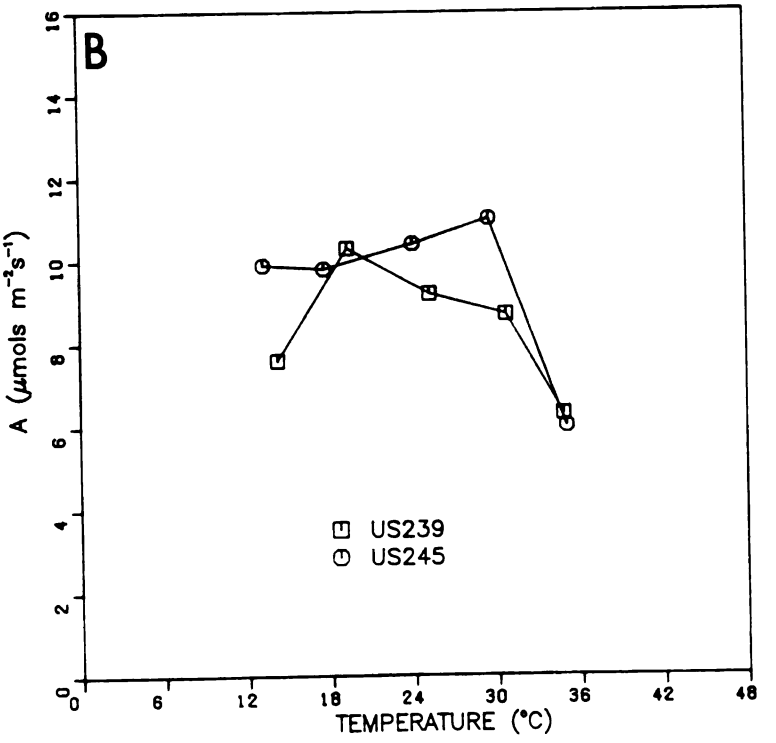

B
A (μmols m⁻²s⁻¹)
TEMPERATURE (°C)
US239
US245
0
2
4
6
8
10
12
14
16
0
6
12
18
24
30
36
42
48

Table 1. Summary of the effect of two levels of leaf temperature on the gas exchange characteristics of different blueberry genotypes. CO_2 assimilation (A), transpiration (E), and water use efficiency (WUE) were measured at leaf to air vapor pressure deficits less than 1 KPa, at saturating PPFD (1006 µmol s^{-1} m^{-2} +/- 14.7) and at ambient CO_2 concentrations (341.7 +/- 7.3 µl $liter^{-1}$).[z]

Genotype	A (µmol CO_2 m^{-2} s^{-1})			E (mmol H_2O m^{-2} s^{-1})			WUE (µmol CO_2/mmol H_2O)		
	20 °C	30 °C	Delta	20 °C	30 °C	Delta	20 °C	30 °C	Delta
'Bluecrop'	10.7^{ab}	7.6^{e}	-29%*	2.69^{a}	3.04^{ab}	+13%n.s.	4.01^{d}	2.53^{b}	-37%*
Fla. 4B	8.1^{c}	8.8^{de}	+9%n.s.	1.45^{d}	2.39^{a}	+65%*	5.64^{bcd}	3.73^{a}	-34%*
US75	12.9^{a}	15.0^{a}	+24%*	1.67^{cd}	3.86^{b}	+131%*	7.70^{a}	3.98^{a}	-49%*
US239	10.1^{bc}	9.2^{cde}	-9%n.s.	1.78^{bcd}	3.09^{ab}	+74%*	5.79^{abc}	3.02^{ab}	-48%*
US245	9.1^{bc}	11.0^{bcd}	+21%n.s.	2.16^{abc}	3.82^{b}	+77%*	4.20^{cd}	2.90^{ab}	-31%*

[z]Means within columns sharing the same letter are not significantly different at the 5% level using Tukey's test.

* change at 30 degrees is significantly different from the value at 20 degrees at the 5% level.

n.s. change at 30 degrees is not significantly different at the 5% level.

Table 2. Summary of the effect of two levels of leaf temperature on the gas exchange characteristics of different blueberry genotypes. Residual conductance to CO_2 (g_r), and leaf conductance (g_l) were measured at leaf to air vapor pressure deficits less than 1 KPa, at saturating PPFD (1006 µmol s^{-1} m^{-2} +/- 14.7) and at ambient CO_2 concentrations (341.7 +/- 7.3 µl $liter^{-1}$).[z]

Genotype	g_r (mmol CO_2 m^{-2} s^{-1}) 20 °C	30 °C	Delta	g_l (mmol H_2O m^{-2} s^{-1}) 20 °C	30 °C	Delta
'Bluecrop'	42.3^{abc}	32.0^{b}	-24.3% *	237.3^{a}	155.8^{c}	-34.3% *
Fla. 4B	34.1^{c}	49.9^{ab}	+46.3% *	117.5^{c}	93.1^{e}	-20.8% n.s.
US 75	57.2^{a}	59.1^{a}	+3.3% n.s.	217.2^{a}	415.2^{a}	+91.2% *
US 239	51.7^{ab}	44.9^{ab}	-13.2% n.s.	140.0^{bc}	134.7^{d}	-3.8% n.s.
US245	36.8^{bc}	49.0^{ab}	+33.2% n.s.	200.5^{a}	209.8^{b}	+4.6% n.s.

[z]Means within columns sharing the same letter are not significantly different at the 5% level using Tukey's test.
* change at 30 degrees is significantly different from the value at 20 degrees at the 5% level.
n.s. change at 30 degrees is not significantly different at the 5% level.

Table 3. Significance levels of F-tests of the analysis of variance of the effects of two levels of leaf temperature (20 and 30 degrees C) on CO_2 assimilation (A), transpiration (E), water use efficiency (WUE), leaf conductance (g_l), and residual conductance to CO_2 (g_r).[a]

Effect	Significance level				
	A	E	WUE	g_l	g_r
Genotype	*	**	**	**	**
Temperature	n.s.	**	**	*	*
Genotype x Temperature	**	**	**	**	**

** - significant at the 1% level.

* - significant at the 5% level.

n.s. - not significant at the 5% level.

[a]Measurement conditions were those described in Table 1 and Table 2.

CO_2 assimilation and in temperature optimums between the genotypes. However, interaction between genotype x temperature was significant in all cases (Table 3).

Discussion

CO_2 assimilation decreased 30% in 'Bluecrop' as temperature was increased from 20 to 30 degrees C and both g_l and g_r declined. The ratio of g_l/g_r was constant for 'Bluecrop' at 20 and 30 degrees C, which indicates that the reduction in CO_2 assimilation was due mainly to a reduction in stomatal aperture, but other non-stomatal aspects of photosynthesis and respiration may also be involved. In other studies, a decline in CO_2 assimilation at high temperature has been associated with an increase in respiration and a reduction in the efficiency of the carboxylase enzyme with increasing photorespiration (3, 7, 12, 13). Such a response could be the cause of the decline in the calculated g_r observed in 'Bluecrop'. Both CO_2 assimilation and g_r were decreased in US239 at 30 degrees C. In contrast, g_r increased with temperature in Fla 4B, the interspecific hybrid (US75) and US245. We suggest that the heat tolerance of Fla 4B is heritable and that it may be due to non-stomatal factors. Similar levels of heat tolerance due to non-stomatal factors have been reported for other species (2, 8, 9, 13) and heat tolerance has been associated in tomato with a heritable variation in the RuBP carboxylase-oxygenase enzyme (12).

The interspecific hybrid US75 had maximum photosynthetic rates that were 20 to 50% higher than the highbush parent and 30 to 80% higher than Fla 4B. This enhanced photosynthetic efficiency may be due to heterosis. Blueberry is sensitive to inbreeding depression and enhanced growth rates have been reported from crosses between unrelated Vaccinium genotypes (11).

The temperature plasticity and environmental range of highbush blueberry might be expanded through interspecific crosses with V. darrowii. Such crosses are possible due to the fact that V. darrowii frequently produces unreduced (diploid) gametes which form fertile hybrids when crossed with the tetraploid highbush blueberries (1, 6, 16). The backcrosses exhibited CO_2 assimilation values close to 'Bluecrop' at 20 degrees C, and that of US245 was 60% higher than 'Bluecrop' at 30 degrees C. In addition, US245 had only moderately higher rates of transpiration than 'Bluecrop' at 30 degrees C and a higher WUE. If these traits are heritable, as the variation in the temperature optimums of the two backcrosses indicates, the high temperature tolerance of V. darrowii can be transferred to highbush blueberry.

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