

FINAL PROJECT REPORT

WTFRC Project Number: AH-04-412

Project Title: Impact of cultural practices on apple canopy physiology

PI: M. Whiting

Organization: WSU-IAREC

Telephone/email: 5097869260 mdwhiting@wsu.edu

Address: 24106 N. Bunn

City: Prosser City:

State/Province/Zip WA

Cooperators: Jim McFerson, Horst Caspari, Troy Peters

Budget History:

Item	Year 1: 2004	Year 2: 2005	Year 3: 2006
Salaries			
Benefits			
Wages	14,000	14,000	14,000
Benefits	2,240	2,240	2,240
Equipment	4,000	4,000	4,000
Supplies	2,500	2,500	2,500
Travel	3,000	3,000	3,000
Miscellaneous			
Total	25,740	25,740	25,740

OBJECTIVES:

- Understand the impact of cultural practices on canopy carbon acquisition and water use
- Understand the horticultural significance of altered rates of gas exchange
- Continue to refine the design and efficiency of the whole-canopy gas exchange measurement system
- Begin initial development of whole-tree carbon budgets and models

Significant findings (2004-2006):

Chemical thinner trials

- Fish oil + lime sulphur (FOLS) and soy oil (SO) reduced significantly whole-canopy net CO₂ exchange rate
- FOLS and SO reduced canopy NCER similarly
- mean canopy NCER over 9 measurement days and 3 applications was ca. 76% and 77% of control for FOLS- and SO-treated trees, respectively
- a 4-d spray interval appeared effective for maintaining a deficit carbon balance
- trees recovered fully from thinning treatment
- trees recovered quicker from SO treatment than from FOLS treatment

Deficit irrigation trial

- no deficit irrigation strategy (DI or PRD) affected significantly whole-canopy or leaf gas exchange or stomatal conductance
- water use efficiency did not vary among irrigation treatments
- withholding irrigation for more than 3 weeks did not cause physiological stress to 14-year-old 'Fuji'/M.26 trees
- for many orchards, water resources may be conserved without reducing canopy carbon balance
- soil water content is not a good indicator of physiological status of canopy

Calcium trials

- both Mora-leaf® and calcium chloride at 6lbs/ac reduced leaf NCER and stomatal conductance compared to water-treated control
- both calcium treatments affected leaf physiology similarly
- leaf NCER was 21-27% lower and stomatal conductance was 34-36% lower from treated trees two days after application
- Mora-leaf® alone and combined with a cover-spray reduced slightly (ca. 10%) leaf gas exchange

METHODS:

Chemical thinner trials

- The effects of multiple applications of the chemical bloom thinners fish oil + lime sulphur (FOLS) and soy oil (SO) on 'Sun Fuji' whole-canopy gas exchange were studied in relation to water-sprayed 'control' trees. Thinning treatments consisted of 2% FO + 3% LS and 4% soy oil emulsion applied by PropTec sprayer at 100 gallons per acre. Treatments were applied at a 4-d interval beginning at 80% full bloom, three applications were made (e.g., 80% FB, 80% FB+4d,

and 80%FB+8d). Leaf and canopy gas exchange measurements were collected using a CIRAS-2 and CIRAS-1 gas analyzer, respectively, prior to, and following thinner applications. Whole-canopy measurements were taken before and after the initial application (80% full bloom) and again following the applications at 80%FB+4d and 80%FB+8d. Following the 80%FB+8 application, we collected gas exchange for three days. Three replications of each treatment were measured (i.e., total of 9 trees). System flow rates were calculated from 21 velocity measurements per inlet pipe.

Deficit irrigation trials

- The effects of irrigation regime on whole-canopy gas exchange and water relations were studied on 11-year-old ‘Fuji’ trees in Prosser. Two novel season-long irrigation strategies have been established in these orchards (both receiving approximately 50+% evapotranspiration replacement): deficit irrigation, and partial rootzone drying (see Caspari project #AH-04-413 for complete experimental details). Using whole-canopy chambers, gas exchange was monitored at key physiological growth stages. In 2005 data were collected between 7 August and 12 August and again between the 6th and 9th of September. Single leaf gas exchange data were also collected throughout the season using a TPS-1 (PP Systems) on a weekly basis (data not shown).
- A separate trial was conducted in 2006 to evaluate canopy gas exchange in relation to soil water content. Two treatments were imposed: an irrigated control, which received irrigation by drip emitters (four/tree) beneath the chambers, and an un-irrigated treatment which did not receive irrigation throughout the trial. From 22 August until 13 September, we measured whole-canopy gas exchange of six trees. In addition, almost every day we measured at noon, single leaf gas exchange (CIRAS-2, PP-Systems) of 3 sunlit and 3 shaded leaves per tree, and stem water potential (PMS Instruments pressure bomb) from 3 leaves acclimated within foil bags for a minimum of 3 hours. Soil water content was recorded daily from tensiometers (three per tree) at three depths (6", 12", and 18" below soil surface). Tensiometers were buried halfway from the trunk to the tree's dripline (ca. 2.5 feet from the trunk).

Calcium + cover spray trial

- The effects of calcium chloride, Mora-leaf®, and a prescribed codling moth cover spray on leaf gas exchange were compared in two separate trials in late summer 2005. In the first, trees were treated with water (control), Mora-leaf®, or Mora-leaf® + cover (5 lbs Imidan™, 8 fl. oz. Success™ per acre plus 1 pint Regulaid™ and 0.5 pint TriFol™ per 100 gallons) applied at 200 gallons/ac. Applications were made on 6 August (Day of year 218). In a separate trial, calcium chloride and Mora-leaf® were compared to untreated control. Applications of water, calcium chloride (6 lbs/ac), and Mora-leaf (6 lbs/ac) were made on 23 September to whole trees at 200 gallons per acre. For both trials, leaf gas exchange was measured on 3 sunlit leaves from 5 trees per treatment within 1 hr of solar noon. Data was collected the day before application and for several days following applications.

RESULTS & DISCUSSION

System for measuring canopy gas exchange

Over the course of this trial we have improved significantly our ability to monitor canopy gas exchange in remote orchard locations, and increase the number of trees (i.e., treatments or replications) we can evaluate. A 12kW continuous-duty diesel powered generator was mounted to a trailer and connected to an electrical service panel. This setup is supplied by a 40-gallon fuel tank and can provide electrical power for computers, gas analyzers, vacuum pumps, dataloggers, and blowers in remote locations. By using vacuum pumps to continuously withdraw gas samples from every chamber to the gas analyzer, and 12V solenoid valves and a manifold, we can switch among

chambers/treatments every minute. We made only subtle changes to the chamber design and still utilize mylar and hook-and-loop fastening. The plenum base allows high chamber volume exchange rates, important to minimize temperature buildup, at low air velocity. In short, we have developed and refined an efficient, reliable, and mobile system for studying fundamental canopy physiology.

Chemical thinner trials (2004 & 2005)

One likely mechanism of blossom/fruitlet thinning is via a reduction in net photosynthesis (or more specifically, carbon balance) and therefore, the supply of carbohydrate growth resources to developing fruit. Our lab has collaborated with others studying the horticultural benefits (i.e., thinning efficacy) of various blossom thinning agents (e.g., fish oil + lime sulphur (FOLS)) to better understand the tree's physiological response to this thinning agent and develop a successful organic thinning program. In 2004 and 2005 we compared the effects of several blossom thinning treatments on canopy gas exchange. In 2005, we studied the effects of applying FOLS and soy oil emulsion (SO) at a 4-day interval. Our data from research in 2004 had suggested that the reductions in net photosynthesis from a 7-day interval were insufficient to elicit an adequate thinning response (see our report in 2004 Apple Hort/Path book).

In 2005, due to poor weather conditions before 22 April, we were unable to collect gas exchange data prior to the first (e.g. 80% full bloom) thinner application. Within 24 hr of the initial application however we had recorded significant reductions in canopy NCER from both treatments (Table 1). Mean daily (0600 – 1730HR) NCER was 32% and 19% lower from SO- and FOLS-treated trees, respectively. High winds blew down our canopy chambers around 1800HR. As a result, we are missing data from late on the 23rd through to late morning on the 25th when conditions permitted chamber setup and data collection. By 3 days after the 80% FB application, trees had recovered from the initial reduction in NCER – mean NCER was only 13% lower than untreated trees, irrespective of treatment. It is not known, but unlikely that the greater initial reduction in NCER from the SO treatment is significant with respect to thinning efficacy, particularly because both treatments had similar NCERs two days later. We recorded greater reductions in NCER from the second application (e.g., 80% FB+4d) on 26 April. Both thinners reduced canopy NCER by 28% compared to the control. This reduction was recorded only over a 5-hr period in late afternoon because winds again blew down the chambers on the night of the 26th. By two days after the second application, canopy NCER was 42% and 35% lower from SO- and FOLS-treated trees, respectively. The greater reductions are not a result of a delay in thinner impact but due to differences in period of measurement of NCER between the 27th and 28th. We hypothesize that on the day after the second application (27th) we would have recorded similar or even greater reductions in NCER than those on the 28th had we recorded the full diurnal impact. Trees once again showed recovery from thinning treatments. By three days after the second treatment, SO-treated trees exhibited NCERs only 14% lower than those of untreated trees. FOLS-treated trees appear to recovery less rapidly – on the same day, canopy NCER had not appreciably recovered from the initial reduction. On 30 April, the third application was made (e.g. 80% FB+8d). We were able to record gas exchange late in the afternoon on the day of application and recorded slight reductions (15 – 18%) in NCER from both treatments. However, over the next day, both treatments again reduced canopy NCER similarly and by ca. 32%. Over the next two days SO-treated trees exhibited significant recovery – 3 days after the third application, NCERs were only 13% lower than those for the control. In contrast, on the same day, FOLS-treated trees had 22% lower NCER than control. Therefore, it appears that both FOLS and SO have similar immediate effects on canopy physiology but that FOLS is slightly more phytotoxic from characterized by a more persistent effect. It is not know whether the longer-lived reductions in NCER confer any greater thinning efficacy.

Overall, the reductions in canopy NCER we recorded match very closely those we reported previously from FOLS applications. What remains unknown is the relationship between thinner phytotoxicity and thinning efficacy. Future investigations should attempt to better define whole-tree carbon balance, taking in to account reproductive and vegetative growth rates as well as the impact of thinning agents on canopy carbon budgets. Moreover, further research must distinguish between

thinner effects on canopy carbon balance and interference with pollination/fruit set, and the relative roles of these modes of action for thinning.

Calcium + cover spray trial (2005)

In 2005 we also investigated the effects of commonly-applied micronutrient and insecticide cover sprays on apple tree gas exchange. In these preliminary investigations Mora-leaf® was tested alone and in combination with a standard codling moth cover spray. We recorded only slight negative effects of both treatments on leaf gas exchange (data not shown). Leaf NCER was reduced by 10 – 18%, irrespective of treatment. There was no additional effect of the cover spray. In addition, there was no consistent effect of either treatment on leaf transpiration or stomatal conductance. We hypothesize that the slight reductions in leaf NCER from Mora-leaf® are not horticulturally relevant – there is little chance that the reductions had any negative impact on fruit yield or quality though these parameters were not determined in this trial. However, in this first year we investigated only the response to a single application. We do not know whether trees would respond similarly to multiple applications or if chronic treatment with these products would impact yield or quality via reductions in carbon assimilation.

In 2005 we also compared the phytotoxicity of Mora-leaf® and calcium chloride by studying their effects on leaf gas exchange. Interestingly, there was little effect of either treatment the day after application but significant reductions in NCER two days after application. Approximately 24 hr following treatment, leaf NCER was ca. 10% lower from treated trees. However, leaf NCER from treated trees was 23% - 27% lower by 48 hr after application. Unfortunately we did not document the trend beyond 48 hr. Reductions in NCER were likely a result of lower stomatal conductance in treated trees. Again, there were only slight reductions 24 hr after treatment but significant reductions by 48 hr – both treatments exhibited ca. 34% lower stomatal conductance than untreated control. This delayed response is difficult to reconcile and merits further investigation. However, we discovered that both products are similar in their effect and that neither is particularly phytotoxic in single applications.

Deficit irrigation trials (2004 – 2006)

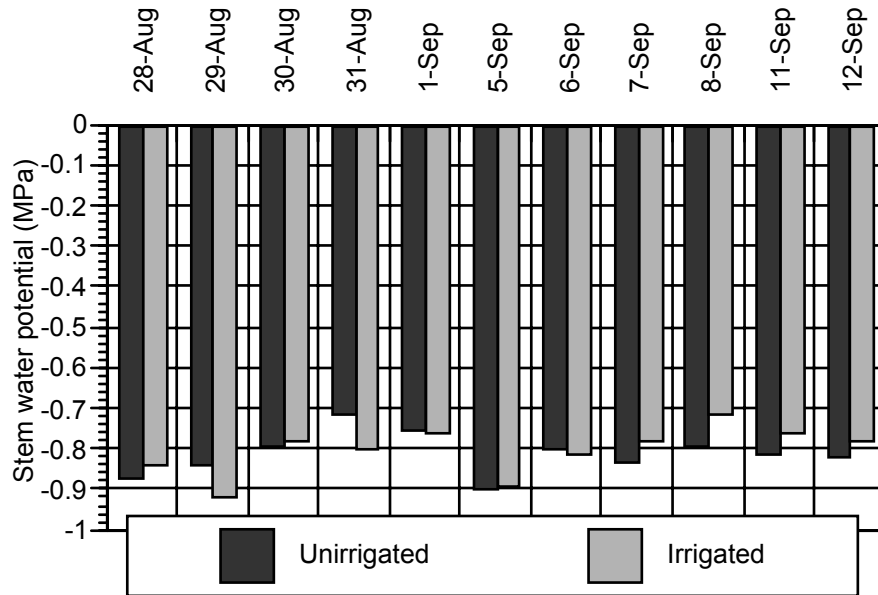
In 2004 and 2005, this project also documented the effects of two season-long irrigation regimes on whole-canopy and single leaf gas exchange in Prosser. For complete trial details including fruit yield, fruit quality, and soil water content see Caspari's report in the 2005 report (AH-04-413). Our results have been consistent across several years. In 2005 we recorded no effect of irrigation regime on whole-canopy gas exchange measure over a week in August and 3 days in September (data not shown). Similarly, in 2004 and 2003, neither deficit irrigation negatively impacted canopy net CO₂ exchange rates. Consistently we discovered that any differences among treatments were subtle and not statistically significant. Interestingly, this occurred despite significant differences in soil water content among treatments. In fact, across and within year, there was no clear or close relationship between soil water content and canopy NCER within the range studied in the current study. Soil water status has been often taken as a measure of drought stress in plants on the assumption that soil water status is proportional to plant water status. Clearly, this is an oversimplification of complex physiological phenomena and our data suggest that soil water content is not a good indicator of physiological stress. However, it is possible that we did not impose a physiologically significant water stress in this trial. Moreover, it may be more appropriate to examine only the soil profile where active root function exists. For the current analyses, we are comparing NCER to the water content of the entire 3.5 – 4' soil depth. Future trials should include measurements of leaf/stem water potential to improve our assessment of stress.

In 2006, we conducted our most detailed assessment of the relationship between soil water content and canopy/leaf physiology. We recorded whole-canopy gas exchange for ca. three weeks. Equipment malfunction spoiled the first week of data collection. Between the 3rd and 13th of September however, we were able to collect a tremendous amount of data. Soil water potential was monitored closely by tensiometers. At no point during the trial period did we record significantly

Figure 1. Midday stem water potential from leaves of irrigated and unirrigated 'Fuji'/'M.26' trees. Each bar is the mean of 9 leaves.

different soil water potential between the unirrigated and irrigated trees (data not shown). Similarities may have been due to the distance between the drip emitters and the tensiometers (i.e., tensiometers did not perceive the wetted soil). In addition, we did not apply large irrigation volumes to irrigated trees. Moreover, the tensiometers may not have been in proper contact with the soil and therefore, been generating false readings. If similar research were to be attempted, greater control over irrigation application and soil water content monitoring would be necessary to reconcile physiological data.

Our estimates of tree physiological stress support the contention that we did not impose a significant stress - we recorded no effect of irrigation treatment on stem water potential (SWP) (Fig.



1). In fact, despite not receiving any irrigation for 16 days, unirrigated trees exhibited no sign of water stress – there was no decline in midday SWP throughout the course of the trial. On 12 September, the penultimate day of this trial, SWP was not statistically different from the beginning of the trial (-0.87 vs. -0.82, respectively). This suggests that M.26 at maturity is reasonably drought resistant. This

may be due to its ability to access water resources deep in the soil profile, or to access a large soil volume. Very little research has investigated apple rootstock rooting patterns and the response of apple rootstocks to water deficits. Physiological stress (i.e., stomatal closure and decreased NCER) is believed to occur in apple at leaf and stem water potentials much lower than those we recorded (i.e., less than 2.0 MPa).

Single leaf gas exchange We recorded midday leaf gas exchange almost every day over the three week trial period. At no time did we record significant effects of irrigation treatment on any component of gas exchange (i.e., net CO₂ exchange rate, transpiration, stomatal conductance) (data not shown). However, we did record, throughout the trial, significantly lower leaf NCER and E and g_s from shaded leaves vs. sunlit leaves. Shaded leaves exhibited NCER of between 10% and 40% of sunlit leaves. Shaded leaf E was nearer to that of sunlit leaves, typically being only 15% to 30% lower. Therefore, water use efficiency of shaded leaves was much lower than that from sunlit leaves.

Whole canopy gas exchange We setup canopy chambers around 6 trees (3 irrigated, 3 unirrigated) on the 28th of August and collected data through to the 13th of September. Canopy transpiration from

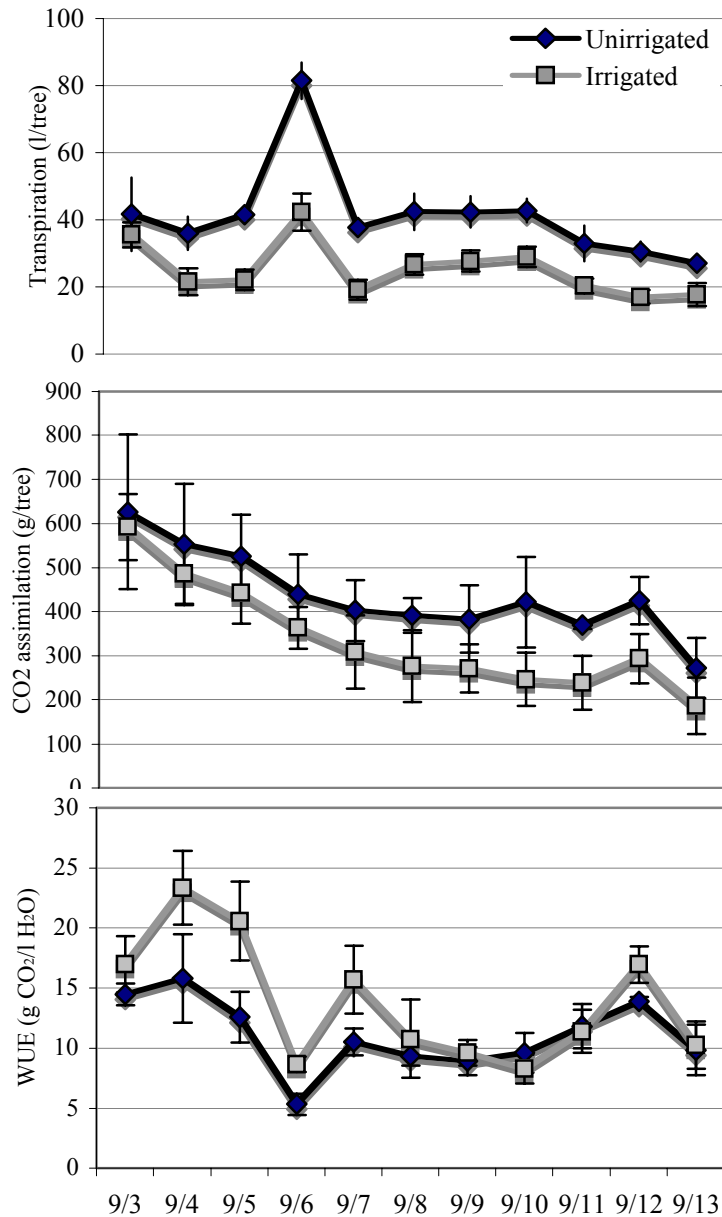


Figure 2. Whole-canopy transpiration (l/tree), CO₂ assimilation (g/tree), and water use efficiency (g CO₂/l H₂O) of irrigated and unirrigated ‘Fuji’/’M.26’ trees. Each data point is the mean ± standard error of 3 trees.

both treatments followed a similar trend over the course of the trial (Fig. 2). In general, unirrigated trees exhibited higher rates of transpiration – overall mean canopy transpiration was 68% higher from unirrigated trees. This may not reflect a treatment effect but have been related more to differences in canopy leaf area. Data are presented as liters of water per tree per day and do not account for potential differences in leaf area. With the exception of the 6th of September, whole-tree water use was fairly consistent over the 10 day sample period. Unirrigated trees transpired between 27 l and 43 l, irrigated trees transpired between 17 l and 36 l. In addition, there was little variability among replicates in transpiration.

Canopy CO₂ assimilation for all trees declined throughout the trial period (Fig. 2). Assimilation was roughly 50% - 60% lower at the end of our trial compared to the outset. The cause of this decline is not clear but not likely related to any change in canopy/stomatal conductance because transpiration was not affected. Our treatments may have elicited a metabolic response not related to canopy/soil water status. Interestingly, we did not document a similar decline in single leaf NCER over the same period. Overall, canopy CO₂ assimilation was about 29% higher from unirrigated trees (437 g/tree vs. 337 g/tree). This again may be related more to differences in canopy area rather than a direct treatment effect.

Moreover, trees exhibited significant

variability in daily CO₂ assimilation – we documented greater than 3-fold differences among individual trees (data not shown). Differences may be related to canopy architecture, light distribution/interception, and leaf area. Crop load (fruit number and yield) was similar among all trees (data not shown) and therefore, presumed to be irrelevant with respect to CO₂ assimilation. In addition, key environmental conditions were similar throughout the trial (Fig. 3, Table 1).

Tree water use efficiency (WUE, g CO₂/l H₂O) was higher for irrigated trees overall (Fig. 2). Mean WUE was ca. 27% higher in irrigated trees. This was due in large part to lower rates of transpiration, rather than differences in CO₂ assimilation. However, in the latter half of the trial, WUE was similar for both treatments.

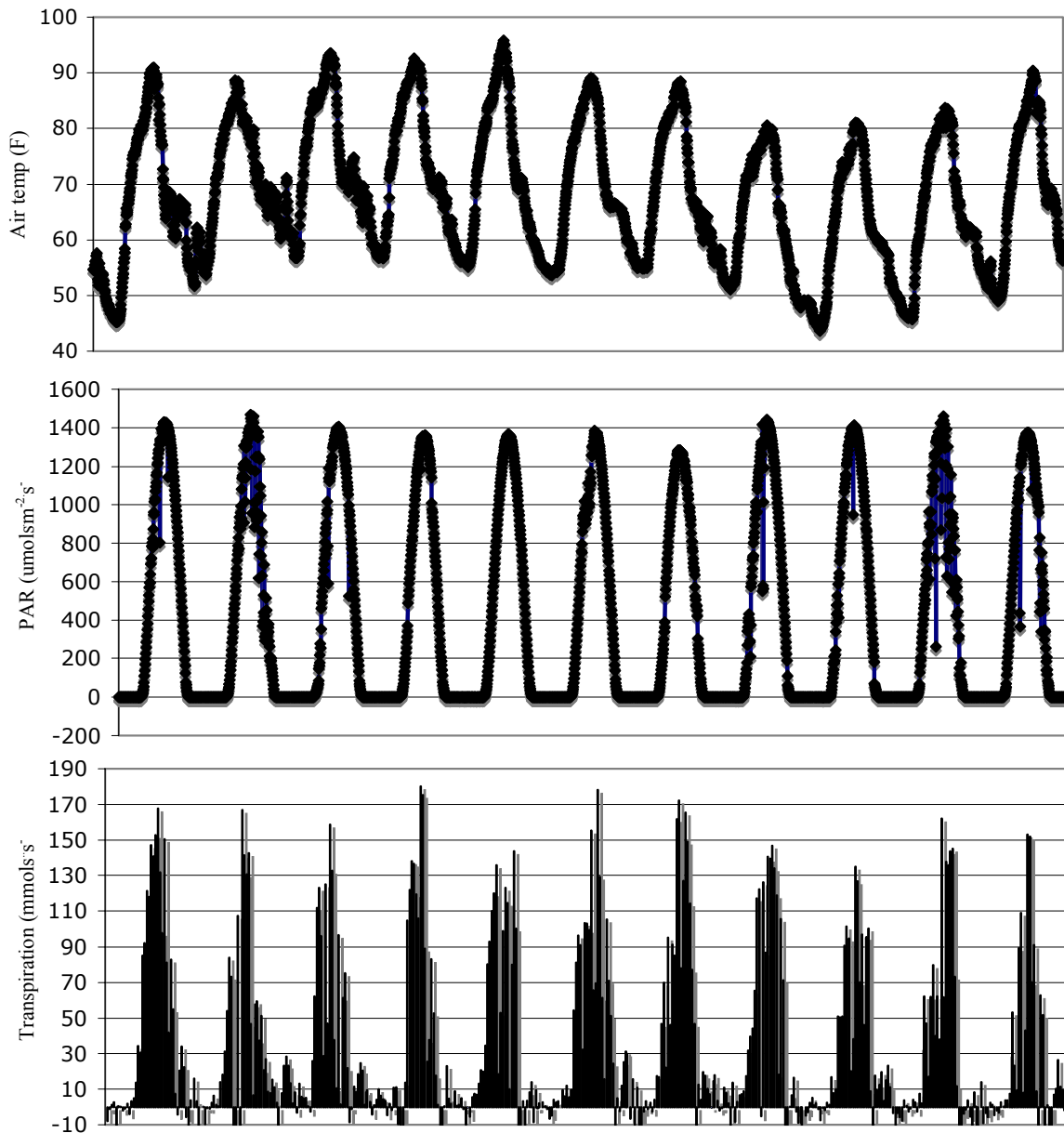


Figure 3. Trend in air temperature (F), photosynthetically active radiation ($\mu\text{mol m}^{-2} \text{s}^{-1}$), and whole-canopy transpiration (mmol s^{-1}) of a representative tree throughout the 10-day period of measuring canopy gas exchange. Data are from the 3rd of September to the 13th of September.

Table 1. Environmental conditions between 3rd and 13th of September, 2006 during measurements of canopy gas exchange.

	3-Sept	4-Sept	5-Sept	6-Sept	7-Sept	8-Sept	9-Sept	10-Sept	11-Sept	12-Sept	13-Sept
Mean air temp (F)	68.9	72.9	73.5	73.7	70.5	69.9	62.7	59.8	64.2	69.0	61.0
Max air temp (F)	80.8	87.3	88.8	90.6	85.4	85.7	73.5	75.1	78.6	86.2	74.3
Min air temp (F)	56.2	58.3	59.1	60.4	57.0	56.2	49.7	44.3	47.9	54.4	47.0
Solar radiation (MJ/M2)	10.73	12.1	11.96	12.26	12.16	11.31	12.4	12.13	11.5	11.82	8.33