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ABSTRACT

Processed calcined kaolin particle films (PKPF) have been shown to repel some insects and reduce various environmental stresses often resulting in improved yield and quality of horticultural crops. PKPF studies in the literature have various water use efficiency (WUE) responses. The purpose of this study was to evaluate 2 application rates of PKPF (3% and 12%) on the whole tree water use efficiency and compare gas exchange values with carbon isotopic discrimination and seasonal water use efficiency in order to determine if there are consistent effects on WUE in apple and infer the mechanisms of action. Fruit yield and total biomass influence WUE whether WUE is measured directly in whole plant chambers, estimated by biomass/unit evapotranspiration, or Δ^{13} C. Δ^{13} C analysis of WUE generally found that the untreated control had lower values and therefore higher WUE than PKPF treatments. Whole plant gas exchange analysis of WUE generally lacked the sensitivity to identify the treatment differences observed using Δ^{13} C analysis and those treatment differences identified were contrary to Δ^{13} C analysis. A lysimeter study supported the reduced WUE of PKPF treated plants by demonstrating increased E with a range of PKPF residue amounts. The lysimeter results suggest that for well-watered and presumably non-stressed conditions, gas exchange is limited by leaf temperature and the concomitant leaf-air vapor pressure deficit that influence stomatal conductance. When leaf temperature is reduced by PKPF, the response results in increased stomatal conductance and gas exchange, as measured by increased E. The reduction in WUE by PKPF treatment is balanced by an increase in overall gas exchange and increased yield and quality.

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1. Introduction

Processed calcined kaolin particle films (PKPF) reduced insect, heat, photosynthetically active radiation (PAR) and ultraviolet radiation (UV) stress in plants due to the reflective nature of the particles (Glenn and Puterka, 2005). In addition PKPF increased the microbial biomass on the leaf surface (Glenn et al., 2015) and together the particles and microbial biomass may aid in the mitigation of ozone damage (Glenn, 2016). The particle film alters reflected infrared (IR), PAR, and UV radiation compared to an untreated plant (Glenn et al., 2002; Glenn, 2016) resulting in reduced canopy temperatures and increased intra-canopy PAR (Glenn and Puterka, 2007; Rosati et al., 2006, 2007; Wünsche et al.,

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2004) which has improved fruit color and mass (Glenn and Puterka, 2007; Glenn 2016). Reduced canopy temperature can potentially reduce transpiration (Boari et al., 2015; Glenn, 2010; Jifon and Syvertsen, 2003; Moftah and Al-Humaid, 2005; Steiman et al., 2011) which may alter water use efficiency (WUE). Studies of leaf-level water use efficiency (WUE_{leaf}) in different crops demonstrate that PKPF can increase (Tworkoski et al., 2002; Jifon and Syvertsen, 2003; Moftah and Al-Humaid, 2005; Boari et al., 2014; Cantore et al., 2009; Prive et al., 2006; Boari et al., 2015), have no effect (Denaxa et al., 2012; Roussos et al., 2010; Gindaba and Wand, 2007a,b; Glenn et al., 2010; Prive et al., 2007; Steiman and Bittenbender, 2007) or decrease (Le Grange et al., 2002) WUE. There are fewer whole canopy and carbon isotopic discrimination studies evaluating PKPF effects on WUE and those demonstrate both increased (Cantore et al., 2009; Glenn et al., 2010), no effect (Lombardini et al., 2005; Steiman and Bittenbender, 2007) and decreased (Glenn 2010; Glenn et al., 2003) WUE.

The reflection of PAR by the particle film at the leaf level is compensated in varying degrees by diffusion of PAR into the interior of the canopy. The combined particle film effects of reduced canopy temperature and increased diffusion of PAR into the interior of the canopy can increase whole canopy photosynthesis (Glenn et al., 2003; Glenn, 2009, 2010) or have no effect (Wünsche et al., 2004). The effect of PKPF on yield studies is similarly inconsistent. Excluding sunburn effects, many studies show increased yield and fruit quality of horticultural crops (Aly et al., 2010; Glenn, 2009, 2010; Glenn and Puterka, 2007; Glenn et al., 2001, 2003, 2005; Lapointe et al., 2006; Cantore et al., 2009; Pace et al., 2007; Wand et al., 2006; Saavedra et al., 2006; Steiman and Bittenbender, 2007; Boari et al., 2014; Ergun, 2012; Khaleghi et al., 2015; Lalancette et al., 2005; Shellie and Glenn, 2008; Shellie and King, 2013; Sugar et al., 2005). There are also studies demonstrating no or negative effects of PKPF on fruit yield and quality (Schupp et al., 2002; Gindaba and Wand, 2005; Kahn and Damicone 2008; Lombardini et al., 2005; Russo and Diaz-Perez, 2005). Based on the methods of these studies it is most likely that the particle films had a residue density of $1-4 \text{ g m}^{-2}$ based on application rates ranging from 12 to 50 kg/ha. The purpose of this study was to evaluate 2 application rates of PKPF (3% and 12%) on the whole tree water use efficiency and compare gas exchange values with carbon isotopic discrimination and seasonal water use efficiency in order to determine if there are consistent effects on WUE in apple and infer the mechanisms of action.

2. Methods and materials

2.1. Field plots and treatment application

The apple orchard was a moderate density planting (500 ha⁻¹) of 'Empire'/'Malling7A' planted in 1992 at the USDA/ARS Appalachian Fruit Research Station, Kearneysville, WV. Tree water requirements were based on 70% of pan evaporation (Glenn, 1995, 1999). Irrigation treatments consisted of two drip emitters per tree providing the daily water needs. Treatments were randomly assigned in 2005 in a split-plot block design with irrigation as the main plot and PKPF treatment as the subplot with six, two tree replicates. In all years, the trees were hand-thinned post-bloom. Trees received a PKPF treatment (3% or 12%) or were not treated. All treatments were over-sprayed with conventional pesticides to protect from disease or insect damage. Conventional orchard practices were used in tree training, mowing, nutrition, and weed control.

Apple trees received applications of a highly reflective, white, calcined hydrophilic particle based on the kaolin mineral (PKPF treatment was Surround WPTM, NovaSource, a division of Tessenderlo Kerley Inc. Phoenix, AZ) in addition to a conventional pesticide spray program. Surround WP was applied at the rate of 25 kg/ha and 100 kg/ha for 3% and 12% (w/v) during the 2005–2010 growing seasons. The Surround WP treatments were applied in a spray volume of $833 L ha^{-1}$ (approximately 50% of tree row volume) using an air blast sprayer at a ground speed of 3.2 km/h. There was an untreated control treatment. PKPF treatments were applied every 2 weeks following petal fall until 2–3 weeks before harvest.

2.2. Field measurements

At harvest, all fruit were weighed and counted for each tree in a plot. Fruit from each tree were processed with an electronic grader that counted and weighed each fruit.

Following harvest, the trees were covered with a netting to capture all the leaves when they abscised. The leaves were collected and air-dried at 60 °C for approximately 1 week. At field sampling, a subsample of approximately 1 kg fresh weight was separated, leaf area was measured, and the ratio of air-dried weight:leaf area calculated. This ratio was used to convert the total air-dried weight of each tree to total leaf area. Pruning weights from each tree were collected in the dormant season and weighed. Mean fruit dry weight (at 60 °C) was 17% fresh weight and mean wood dry weight (at 60 °C) was 40% pruning weight in the field. Biomass was the dry weight sum of leaf, fruit and dormant season pruning.

Photosynthetically active radiation (PAR), relative humidity, wind speed, pan evaporation, and air temperature were measured at a weather station approximately 500 m from the measurement site. Pan evaporation was measured 3 times/week and the seasonal transpiration potential (E_f) was calculated as 0.8 X seasonal cumulative pan evaporation. Radiation use efficiency for each year was calculated as biomass (kg/tree)/canopy area (m²)/seasonal PAR interception (G]).The growing season was May to September.

Whole tree gas exchange was measured in open-top chambers similar to Glenn (2010). The whole canopy chamber was constructed of 0.762-mm-thick polycarbonate (Makrolon GP; Sheffield Plastics Inc.—Bayer Material Sciences Pittsburgh, PA) in a rigid cube $2.4 \times 2.4 \times 2.4$ m. A framed polycarbonate pitched roof covered the cube and the floor was a polyethylene tarp material split to the center to slide onto the tree and was sealed with a Velcro strip. Details of the gas exchange chambers are described in Glenn (2010).

Diurnal data were collected: 2005 DOY's 202–226; 2006 DOY's 217–225; 2007 DOY's 255–263; 2008 DOY's 238–244; 2009 DOY's 225–239; 2010 DOY's 241–242 and 264–266. During each sampling, eight trees, including two or three trees of each treatment, were simultaneously measured for multiple days. After sampling, the chambers were moved to eight other trees multiple times providing four single tree replications of each treatment for whole tree gas exchange. Data were collected for 24 h each day but only data from 1000 to 1600HR, were analyzed. PKPF treatment effects were analyzed over days and hours using analysis of covariance in which yield and biomass were the independent variables. Data were compared using PDIFF which compares least squares means from the analysis of covariance. Treatment means were compared using Fisher's protected least significant difference (LSD), $P \le 0.05$.

Twenty mid-shoot leaves per tree were collected in late July to early August, washed with deionized water and air dried at 60° C for approximately one week. The tissue was re-dried at 60°C for 72 h, ground, and analyzed for ¹³C content (University of California, Davis Stable Isotope Facility, Department of Plant Sciences, One Shields Ave, Mail Stop 1, Davis, CA 95616, USA). Carbon isotope discrimination (Δ^{13} C) was calculated according to Farquhar et al. (1989). The carbon dioxide isotope composition in air (δ^{13} C_{air}) was assumed to be -7.8 parts per thousand (Francey et al., 1995).

2.3. Lysimeter study 2015

Two weighing lysimeters (Cardinal Scale Co., Webb City, MO; model FS-8; see Glenn et al., 1996) were used to measure whole tree transpiration by measuring weight loss from 9 AM to 5 PM. Tenyear-old 'Empire' apple trees were transplanted to plastic apple bins $(1.1 \text{ m} \times 1.1 \text{ m} \times 0.7 \text{ m}; \text{ L} \times \text{W} \times \text{D})$ in 2008 and grown with supplemental irrigation and fertilization under field conditions. The apple containers were placed directly on the surface of the lysimeter; the surface of the lysimeter was covered with a waterproof covering. During the study, irrigation was applied at midnight and drainage was completed by sunrise. Baseline data from both untreated trees was measured DOY's 186-193 and the tree to be treated had 70.12% of the mean E compared to the control tree. The treated tree E was normalized by dividing its E by 0.7012. Applications of Surround were applied with an orchard sprayer similar to the field studies although multiple applications were made to achieve greater and greater residue levels. The following residue amounts were measured: DOY's 199–202, 0.5 g/m²; DOY's 203-207, 1.3 g/m²; DOY's 208-211, 2.8 g/m²; DOY's 212-215, 4.9 g/m²; DOY's 216–217, 3.95 g/m² (reduced by rain); DOY's 220–221, 15.8 g/m²; DOY's 222–223, 7.0 g/m² (reduced by rain);

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