Foliar Boron and Nickel Applications Reduce Water-stage Fruit-split of Pecan

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Abstract. Water-stage fruit-split (WSFS) is a relatively common and often major problem of certain pecan (Carya illinoiensis (Wangenhn.) K. Koch) cultivars (e.g., ‘Schley’, ‘Oconee’, ‘Sumner’, ‘Wichita’, ‘Frotcher’, and ‘Farley’) and, to a lesser degree, by certain relatively thick-shelled cultivars (e.g., ‘Cape Fear’ and ‘Elliott’). The malady occurs when the ovary wall (shell), underlying testa (seedcoat), and sometimes the involution (shuck) splits about the time of the initiation of kernel (cotyledon) filling and shell hardening, resulting in abortion and drop of damaged fruit within 7 d after splitting (Wood and Reilly, 1999).

The WSFS malady is highly erratic with incidence and severity varying depending on cultivar, location, and year (Prussia et al., 1985). Crop loss can be severe in certain years (Storey, 1969; Worley and Taylor, 1972) and nearly absent in others. Similar fruit-splitting disorders occur on other crops such as apple (Malus domestica Borkh.; Goode et al., 1975), cherry (Prunus avium L.; Verner and Blodgett, 1931), and plum (Prunus domestica L.; Mrozek and Burkhardt, 1973), but they appear to be largely associated with fruit flesh rather than with seed. In the case of pecan, WSFS occurs during the “late water stage,” a time when turgor pressure of the liquid endosperm is high (Prussia et al., 1985) and the shell is beginning to become rigid (Allison et al., 1987) with lignin deposition. This typically occurs during mid-August for susceptible cultivars growing in the southeastern United States.

The malady is typically associated with rainfall occurring at the initiation of shell hardening (Storey, 1969). Wood and Reilly (1999) identified two episodes of WSFS for ‘Wichita’ pecan, the major episode being triggered by increased water availability resulting from rainfall (or potentially irrigation) and potentially a relatively minor event triggered by “high humidity/low light.” They also noted that a variety of cultural (e.g., irrigation scheduling and amounts), environmental factors (e.g., rainfall, relative humidity, sunlight/shading, soil characteristics), and tree characteristics (crop load and distribution, nutrient element status) potentially influence WSFS (Wood and Reilly, 1999).

The endosperm of pecan fruit develops from the central cell of the megagametophyte after fertilization by the second nuclear sperm cell migrating from the pollen grain. This tripliod nuclear endosperm is located in the proximal zone of a cytoplasm that encompasses a large central vacuole (i.e., endosperm coenocyte) that is filled with a pressurized aqueous solution comprised primarily of elemental ions and sugars ( Olsen, 2001).

Turgor pressure generated by solutes forces the testa into the void generated as a result of ovary wall expansion as the fruit grows. This solution-filled central vacuole is present from soon after fertilization until about the time of shell hardening, when acceleration of centripetal growth of alveolus cell layers begins to completely fill the central vacuole to form the cotyledons, but is not particularly noticeable until within a couple weeks before the initiation of shell hardening. It is usually about the time of the formation of radial microtubular systems to deposition of a couple layers of alveolus that WSFS occurs (B.W. Wood, personal observation). Control of turgor pressure exerted by the central vacuole of the nuclear endosperm against the various fruit tissues is at least partially through movement of potassium (K) in and out of the endosperm solution. The movement of K is potentially influenced by several factors, one apparently being the availability of boron (B) within cellular membranes (Parr and Loughman, 1983; Schon et al., 1991). Thus, inadequate B within certain cells and tissues of the developing fruit potentially affects WSFS through its effect on rapid influx or efflux of K.

Foliar-applied B has been previously shown to increase fruit retention and fruit quality in a number of perennial tree fruit crops, including pecan ( Wells et al., 2008). A common aspect of B-deficient plants is brittle cell walls with supraperom B concentrations enabling greatly enhanced elasticity (Blevins and Lukaszewski, 1998). Foliar-applied B appears to be highly mobile in some tree species (Picchioni et al., 1995). Thus, timely application of foliar B may potentially influence cell wall elasticity of fruit tissues.

Other potential nutritional factors affecting WSFS include the essential trace micronutrients that link either indirectly or directly to lignification [manganese (Mn), iron (Fe), zinc (Zn), copper (Cu), and nickel (Ni)]. Because xylem connections to developing fruit are absent or poorly formed, their availability to the developing seed is primarily dependent on phloem transport. Certain micronutrients are only variably mobilized in the phloem [Fe, Zn, Cu, molybdenum (Mo), Ni, and cobalt (Co)], whereas others are only conditionally mobile [calcium (Ca), B, and Mn; Welch, 1999]. Poor plant Ni nutrition potentially affects primary metabolism in such a way that endogenous availability of certain amino acids, organic acids, and acetate-CoA can potentially limit lignification (Bai et al., 2006), and pecan trees in the southeastern United States can exhibit an early-season Ni deficiency (Wood et al., 2004a, 2004b, 2004c). The nature of the interaction between micronutrients and WSFS is unknown, especially for B and Ni.

Crop loss to WSFS is minimized, but not totally prevented, by managing tree water relations to minimize the severity and duration of water stress during the last 2 weeks of fruit sizing and by crop-load thinning. The notion of incorporating a micronutrient component to WSFS management has not been studied. We report here the influence of foliar...
micronutrient sprays of B and Ni on WSFS of pecan.

Materials and Methods

Elemental composition of liquid endosperm. Liquid endosperm was collected in mid-Aug. 2004 from 'Wichita' fruit within the same fruiting cluster as WSFS fruit on the day the WSFS occurred. Developing fruit were surgically excised to expose the non-ruptured seedcoat, at which time a hypodermic syringe was used to penetrate the liquid endosperm and the aqueous solution withdrawn and stored until analysis. Samples from six fruit were bulked for analysis. Elemental analysis was by the University of Georgia Plant and Tissue Analysis Laboratory using standard techniques for water analysis.

Influence of boron, nickel, and boron + nickel on water-stage fruit-split—Cape Fear. The hypothesis that timely fertilization with either B or Ni can reduce WSFS in relatively thick-shelled cultivars was tested on 'Cape Fear'. Studies were conducted in a 20-year-old commercial 'Cape Fear' pecan orchard in Berrien County, GA, growing on a Tifton loamy sand (fine-loamy, siliceous, thermic Plinthic Paleudult) soil. The study spanned over 3 years. Orchard trees were spaced 18.3 x 18.3 m and were drip-irrigated as needed throughout the growing season. Treated trees did not exhibit any visible symptoms of B or Ni deficiency, and midsummer leaf concentrations were within the visible symptom-based sufficiency range for pecan [i.e., 15 to 50 mg kg\(^{-1}\) for B (Plank, 1988) and greater than \(\approx 1\) mg kg\(^{-1}\) for Ni] (Nyczepir et al., 2006). The experiment consisted of three treatments structured as a randomized complete block. Each block was comprised of five trees per experimental unit. There were three blocks in the 2005 study and four blocks in the 2006 to 2007 study. The five sample trees for each treatment were randomly selected from within an orchard row with treated rows being separated by unsprayed border rows. All data are presented as the mean response of the five sampled trees per plot. Treated trees received annual foliar B and Ni treatments. Data were analyzed by analysis of variance for each individual year with mean separation by Fisher's protected least significant difference. Treatment consisted of Top Side Liquid Boron (Triangle Chemical, Macon, GA), a commercial product containing 6% B from tetraboric acid (\(\text{H}_2\text{B}_4\text{O}_7\)) applied at a concentration of 84 mg L\(^{-1}\) actual B as an aqueous spray. The Ni treatment was Nickel Plus (NIPAN, LLC., Valdosta, GA), a commercial product containing 6% Ni as Ni-lignosulfonate. Ni was applied at a concentration of 84 mg L\(^{-1}\) (NiPlus at 1169 mL ha\(^{-1}\)) in water and an nonionic surfactant (0.05% BioSurf). Ni was applied with an airblast sprayer with the first application going on 15 Apr. with a second being applied 60 d later, and a third 4 d after the second application for a total of three applications during the canopy expansion phase of growth. All treatments were compared with a nontreated control. Applications were made with a commercial air-blast sprayer delivering 935 L spray solution per hectare. Boron treatments were applied before pistil receptivity (=3 weeks before anthesis) [stem elongation stage 31, 39/60; flowering stage 69; fruit stages 72, 75 (Finn et al., 2007)] and continued every 14 d for a total of five applications. Nickel applications were applied at pasture stage in mid-April and again 60 d later. The combined B and Ni applications were as described for B and Ni.

The impact of B and Ni treatments on WSFS was assessed by estimating crop load and the "incidence" and "severity" of WSFS. Crop load was estimated by counting 25 random terminals per tree on five trees within each plot and dividing the number of fruiting terminals by the total number of terminals counted per tree. Incidence is defined as the percentage of clusters with one or more WSFS nuts. Severity is defined as the percentage of WSFS nuts per cluster. For 2005 incidence and severity estimates, fruiting terminals on 20 random terminal shoots per tree, with 10 on each of the east and west sides (20 total branches) of the canopy, were flagged for each of the five data trees in each plot. All counts were made in early August (fruit stage \(\approx 79\)). Incidence of WSFS was estimated by counting the number of flagged clusters with WSFS nuts and dividing by the total number of clusters counted. Severity of WSFS was estimated by counting the number of WSFS nuts per cluster and dividing by the total number of nuts within the cluster. In 2006 and 2007, all WSFS nuts on the ground beneath the tree canopy in a 12.4-m\(^2\) area were counted. A WSFS severity index rating was estimated by dividing the number of split nuts per tree by the percentage of fruiting terminals. All data are presented as the mean of the five sampled trees per plot.

Results

Elemental composition of liquid endosperm. The liquid endosperm of developing 'Wichita' fruit at time of WSFS contained several elements in solution (Table 1). The concentrations of elements was such that K > Ca > Mg > P > Zn > Cu > Mn > Fe > B > Na > aluminum > Ni. The hypothesis that timely fertilization with Ni can reduce WSFS in a relatively thin-shelled cultivar was tested on 'Sumner'. Studies were conducted in a 7-year-old commercial-like 'Sumner' orchard in Peach County, GA (latitude 32°39'54" N, longitude 83°44'31" W) growing on a Faceville fine sandy loam (fine, kaolinitic, thermic Typic Kandiudult) soil. The study was performed in 2006. Orchard trees were spaced 4.6 x 9.2 m and were drip-irrigated as needed throughout the growing season. Treated trees did not exhibit visible symptoms of Ni deficiency, and midsummer leaf concentrations were within the visible symptom-based sufficiency range for pecan (i.e., greater than \(\approx 1\) mg kg\(^{-1}\) for Ni) (Nyczepir et al., 2006). The experiment consisted of two treatments structured as a randomized complete block. Each block was comprised of two treatments with one tree per experimental unit. There were 16 blocks with trees blocked by both tree diameter and estimated crop load. Data were analyzed by ANOVA. Blocks were within one of 16 tree rows comprised of 48 trees with a border row between treatment rows. Because it was necessary to begin treatment of trees during the canopy expansion phase [stem elongation stage 31, 39/60; flowering stage 69 (Finn et al., 2007)], before crop load could be accurately estimated, all 48 trees per block were treated for all three Ni sprays. During early August, before onset of WSFS, treated and nontreated trees were identified and assigned to block based on trunk caliper and crop load. Treatments consisted of Ni as Nickel Plus (NIPAN, LLC.), a commercial product containing 6% Ni as Ni-lignosulfonate. Ni was applied at a concentration of 84 mg L\(^{-1}\) (NiPlus at 1169 mL ha\(^{-1}\)) in water and a nonionic surfactant (0.05% BioSurf). Ni was applied with an airblast sprayer with the first application going on 15 Apr. with a second being applied 60 d later, and a third 4 d after the second application for a total of three applications during the canopy expansion phase of growth. All treatments were compared with a nontreated control. Applications were made with a commercial air-blast sprayer delivering 831 L spray solution per hectare. WSFS occurred \(\approx 16\) Aug. at fruit development stage 79 (Finn et al., 2007). The proportion of fruit exhibiting WSFS was determined in early September by counting all WSFS fruit on the ground beneath the tree canopy and dividing by the total number of fruit retained by the tree. Thus, counting all fruit and counting by both tree trunk diameter and crop load provided relatively high resolution of treatment effects.

Influence of nickel on water-stage fruit-split—'Sumner'. The hypothesis that timely fertilization with Ni can reduce WSFS in a relatively thin-shelled cultivar was tested on 'Sumner'. Studies were conducted in a 7-year-old commercial-like 'Sumner' orchard in Peach County, GA (latitude 32°39'54" N, longitude 83°44'31" W) growing on a Faceville fine sandy loam (fine, kaolinitic, thermic Typic Kandiudult) soil. The study was performed in 2006. Orchard trees were spaced 4.6 x 9.2 m and were drip-irrigated as needed throughout the growing season. Treated trees did not exhibit visible symptoms of Ni deficiency, and midsummer leaf concentrations were within the visible symptom-based sufficiency range for pecan (i.e., greater than \(\approx 1\) mg kg\(^{-1}\) for Ni) (Nyczepir et al., 2006). The experiment consisted of two treatments structured as a randomized complete block. Each block was comprised of two treatments with one tree per experimental unit. There were 16 blocks with trees blocked by both tree diameter and estimated crop load. Data were analyzed by ANOVA. Blocks were within one of 16 tree rows comprised of 48 trees with a border row between treatment rows. Because it was necessary to begin treatment of trees during the canopy expansion phase [stem elongation stage 31, 39/60; flowering stage 69 (Finn et al., 2007)], before crop load could be accurately estimated, all 48 trees per block were treated for all three Ni sprays. During early August, before onset of WSFS, treated and nontreated trees were identified and assigned to block based on trunk caliper and crop load. Treatments consisted of Ni as Nickel Plus (NIPAN, LLC.), a commercial product containing 6% Ni as Ni-lignosulfonate. Ni was applied at a concentration of 84 mg L\(^{-1}\) (NiPlus at 1169 mL ha\(^{-1}\)) in water and a nonionic surfactant (0.05% BioSurf). Ni was applied with an airblast sprayer with the first application going on 15 Apr. with a second being applied 60 d later, and a third 4 d after the second application for a total of three applications during the canopy expansion phase of growth. All treatments were compared with a nontreated control. Applications were made with a commercial air-blast sprayer delivering 831 L spray solution per hectare. WSFS occurred \(\approx 16\) Aug. at fruit development stage 79 (Finn et al., 2007). The proportion of fruit exhibiting WSFS was determined in early September by counting all WSFS fruit on the ground beneath the tree canopy and dividing by the total number of fruit retained by the tree. Thus, counting all fruit and counting by both tree trunk diameter and crop load provided relatively high resolution of treatment effects.
Table 1. Elemental composition of the liquid endosperm solution from developing ‘Wichita’ pecan fruit at time of water-stage fruit-split (WSFS) during mid-Aug. 2004.

<table>
<thead>
<tr>
<th>Element</th>
<th>Conc (mg-L⁻¹)</th>
<th>Relative composition (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aluminum</td>
<td>0.07</td>
<td>0.01</td>
</tr>
<tr>
<td>Boron</td>
<td>0.33</td>
<td>0.01</td>
</tr>
<tr>
<td>Cadmium</td>
<td>&lt;0.005</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Calcium</td>
<td>578.6</td>
<td>18.7</td>
</tr>
<tr>
<td>Chromium</td>
<td>&lt;0.005</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Copper</td>
<td>0.89</td>
<td>0.29</td>
</tr>
<tr>
<td>Iron</td>
<td>0.40</td>
<td>0.01</td>
</tr>
<tr>
<td>Magnesium</td>
<td>252.6</td>
<td>8.2</td>
</tr>
<tr>
<td>Manganese</td>
<td>0.41</td>
<td>0.01</td>
</tr>
<tr>
<td>Molybdenum</td>
<td>&lt;0.005</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Nickel</td>
<td>&lt;0.005</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Phosphorus</td>
<td>111.5</td>
<td>3.6</td>
</tr>
<tr>
<td>Potassium</td>
<td>2142.0</td>
<td>69.4</td>
</tr>
<tr>
<td>Sodium</td>
<td>0.10</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Zinc</td>
<td>1.35</td>
<td>0.04</td>
</tr>
</tbody>
</table>

Table 2. Crop load (percentage of fruiting terminals) and effect of micronutrient sprays on water-stage fruit-split (WSFS) incidence (percentage of fruiting terminals exhibiting WSFS) and severity (percentage of WSFS nuts per cluster) on ‘Cape Fear’ pecan in 2005.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Crop load (% fruiting terminals)</th>
<th>WSFS incidence (%)</th>
<th>WSFS severity (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Untreated</td>
<td>60 a</td>
<td>33.6 a</td>
<td>42.1 a</td>
</tr>
<tr>
<td>Boron</td>
<td>60 a</td>
<td>22.0 a</td>
<td>24.0 b</td>
</tr>
<tr>
<td>Nickel</td>
<td>59 a</td>
<td>26.2 a</td>
<td>26.9 b</td>
</tr>
<tr>
<td>Boron + nickel</td>
<td>58 a</td>
<td>25.3 a</td>
<td>31.0 ab</td>
</tr>
</tbody>
</table>

These data indicate that the severity of WSFS exhibited by pecan trees partially links to nutrient deficiencies that affect tissues of the developing fruit. Movement of nutrients from the maternal plant to fruit sink tissues is partially controlled by transport through phloem vessels, because no mature xylem vessels extend completely into the ovary’s tissues (Welch, 1999). The relatively low transpiration rate of developing fruit and humid conditions of tissues contained within the ovary wall also contribute to little movement of mineral nutrients into ovary tissues. Thus, phloem connections to apoplastic spaces adjoining the testa, subsequent absorption by testa cells, and movement within the symplasm of testa and nuclear endoplasmic reticulum are critical to meeting the needs of the developing nuclear endoplasm. A similar situation exists for the developing ovary wall.

The relative abundance of K in the liquid-filled vacuole of the nuclear endoplasmic reticulum indicates that K⁺ is likely the key factor regulating the turgor pressure exerted by the vacuolar liquid endosperm within the developing pecan fruit; consequently, there must also exist a K-associated pressure-regulating mechanism to prevent excessive turgor pressure from rupturing the vacuole. We therefore postulate that factors affecting the speed of transmembrane-associated influx or efflux of K⁺ are key to predisposing fruit to WSFS. Although K⁺ readily translocates in phloem vessels to developing ovary tissues, B is only conditionally mobile and Ni is variably mobile, hence leading to potential deficiency within developing tissues (Welch, 1999).

The observed influence of either B or Ni fertilization on severity of WSFS indicates that there is indeed a potential deficiency in either B or Ni sufficient to lead to suboptimal processes involved in the development of certain ovary tissues that predisposes developing fruit to WSFS. This limitation could be the result of low bioavailability in the soil solution, poor root uptake, limitations on phloem loading or phloem sap transport from source to ovary tissues, uploads of sap nutrients within ovary tissues, or phloem-xylem nutrient element exchange processes (Welch, 1999).

Although the incidence of WSFS was unaffected by micronutrient sprays, WSFS severity was reduced for ‘Cape Fear’, a relatively thick-shelled cultivar, in each of the three study years (2005 to 2007) by foliar B application. Severity of WSFS was also reduced in 2005 on ‘Cape Fear’, and in 2006 on ‘Sumner’, by foliar Ni application. The mechanism by which B and Ni influence the severity of WSFS is speculative; however, circumstantial evidence pertaining to the observed abundance of K in liquid endosperm solution, the apparent role of B in K transport across membranes (Brown et al., 2002; Parr and Loughman, 1983; Schon et al., 1991), the influence of B on cell wall elasticity (Blevins and Lukaszewski, 1998), and the potential influence of Ni on lignification (Bai et al., 2006; Wood et al., 2004a) collectively support multiple mechanisms in which these two trace elements can individually influence WSFS. Observations by Piccioni et al. (1995) that foliar-applied B is highly mobile in certain tree fruit species appear to be also true for pecan in that foliar B sprays reduced structural damage to developing fruit long after B was applied to foliage.

Water-stage fruit-split is associated with an interaction between turgor pressure and shell strength (Allison et al., 1987; Prussia et al., 1985; Wood and Reilly, 1999). There is evidence that B enhances the movement of K⁺ across cell membranes (Brown et al., 2002). Potassium is involved in the water...
relations, charge balance, and osmotic pressure within cells and across membranes (Havlin et al., 2005). Therefore, B might potentially exert an indirect influence on the osmotic pressure within plant cells by facilitating the movement of K⁺ across the membrane of the vacuole of the nuclear endosperm. Such a relationship might well influence turgor pressure within the pecan fruit in the late water-stage, thus affecting WSFS. Dell and Huang (1997) suggested that when B was limited later in fruit development, symptoms of abnormal fruit growth may be apparent, thus supporting the observation of a B association to WSFS in pecan. Further evidence is provided in that B deficiencies have been previously linked to a premature testa staining disorder in avocado (Harkness, 1959) and misshapen, cracked, or bleeding fruit of mango (Ram et al., 1989).

Although crop load did not differ between treatments within years, this factor was incorporated into the severity rating used in 2006 and 2007 to account for its potential effect. The severity of WSFS has previously been shown to increase with increasing crop load (Wood and Reilly, 1999). The increased severity of WSFS in ‘Cape Fear’ in all treatments during 2007 may have been influenced to a greater extent by environmental conditions at the site than by crop load. Although environmental conditions were not measured at the study site, the 2007 pecan-growing season in Georgia was characterized by early-season drought and plentiful rainfall during late fruit development. Such conditions would enhance the likelihood of WSFS, especially under conditions of a heavy crop load. Wood and Reilly (1999) suggest that dry soil 2 weeks before entering the water-split stage can enhance WSFS.

Previous studies have suggested that cultural practices such as crop thinning and management of soil moisture can reduce, but not eliminate, the WSFS problem (Allison et al., 1987; Prussia et al., 1985; Storey, 1969; Wood and Reilly, 1999; Worley and Taylor, 1972). Our results indicate that timely foliar application of either B or Ni can potentially reduce the severity of crop loss to WSFS in certain orchard situations and may be useful components of strategies to reduce WSFS losses in commercial orchard enterprises. These observations lead us to postulate that WSFS in pecan is partially controlled by a timely interplay between B and K flux within the nuclear endosperm and the roles of B and Ni in lignification of the ovary wall, because these processes interact with sudden changes in the balance of available water from the maternal plant to the vacuole of the nuclear endosperm of the developing fruit. Efforts to reduce crop losses to WSFS might well benefit from strategies to ensure that ovary tissues are well supplied with bioavailable forms of B, Ni, K, and perhaps other trace nutrient elements in certain orchard situations.

**Literature Cited**


