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Seasonal Variation in Mineral Nutrient Concentration of Primocane and Floricane Leaves in Trailing, Erect, and Semierect Blackberry Cultivars

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Abstract*. Floricane-fruiting blackberry (*Rubus* L. subgenus *Rubus,* Watson) cultivars, 'Marion', 'Black Diamond', 'Onyx', 'Columbia Star' (early-season trailing types), 'Ouachita' (erect, midseason), and 'Triple Crown' and 'Chester Thornless' (semierect, late season) were studied for 2 years to determine whether these cultivars and types of blackberry should be sampled at a certain stage of development or time of season to best evaluate plant nutrient status. Leaf nutrient standards are based on primocane leaves in most countries, but there is interest in using floricane leaves. Primocane leaves were sampled every 2 weeks from late May through early October, whereas leaves on fruiting laterals (floricane) were sampled every 2 weeks from early May through fruit harvest. Leaves were analyzed to determine the concentration of macronutrients and micronutrients. The pattern of change in primocane leaf nutrient concentration varied between the trailing and the later-fruiting erect and semierect types, particularly for P, K, Ca, Mg, B, and Mn, where leaf levels were higher in the late season for the erect and semierect cultivars (except for P and K which were lower). Nutrient concentrations in floricane leaves decreased during growth and development of the lateral and fruiting season for N, P, K, and S, but increased for most other nutrients in all blackberry types. Floricane leaf N and K declined most rapidly during the fruit development period in all cultivars. Sampling of floricane leaves is not recommended, particularly for trailing types, as there are no sufficiency standards. In primocane leaves, the nutrients that did not show significant changes in concentration during the currently recommended sampling period of late July to early August were N, Mg, K, Ca, S, B, Mn, and Zn, but only in 2014. Leaf P, Fe, and Al were stable during this period in both years. In contrast, when sampling in mid to late August, leaf N, Mg, Fe, Mn, and Al were stable in both years and leaf K, Ca, S, B, Cu, and Zn were stable in one of the 2 years. We thus propose changing the recommended sampling time to mid to late August for these diverse blackberry cultivars. The current sufficiency standards for primocanes did not encompass the blackberry types and cultivars studied here, suggesting the standards may need to be revised for this region.

About 6000 ha of blackberry (*Rubus* L. subgenus *Rubus*, Watson) were harvested in the United States in 2012, with 42% of this production located in Oregon (U.S. Department of Agriculture, 2014). Oregon is the leading producer of trailing blackberry in the United States but also produces erect and semierect types for the fresh market. The growth habit and fruiting season of trailing, erect, and semierect blackberry differ considerably (Strik and Finn, 2012). In Oregon, the fruiting season of trailing cultivars ranges from late June through July with erect and semierect types fruiting from late July through August and early August through early October, respectively. All of these blackberry types produce biennial canes. The primocanes of these floricane-fruiting cultivars are vegetative in their first year of growth. In their second year, when they are called floricanes, they flower, fruit, and then senesce. In trailing types, primocanes are not self-supporting and are trained along the ground under the floricane canopy until trained to the trellis after fruit harvest and floricane pruning. By contrast, the primocanes of erect and semierect types are self-supporting and are summer pruned (tipped) to encourage branching. Summer pruning of primocanes affects leaf nutrient levels in primocane-fruiting blackberry (Strik, 2015).

Commercial blackberry growers are encouraged to develop fertilization programs based on general guidelines for nitrogen (N) fertilizer, in which rates increase from the planting year to maturity (Hart et al., 2006). Adjustments of fertilizer N and other macronutrients and micronutrients are based on the periodic soil nutrient analysis, observations of plant growth, and annual leaf tissue analysis (Bolda et al., 2012; Bushway et al., 2008; Fernandez and Ballington, 1999; Hart et al., 2006; Krewer et al., 1999). In floricane-fruiting blackberry and raspberry, leaf sampling of primocanes in mid to late-season informs growers of plant nutrient requirements for fruit production the following season, when the primocane becomes a floricane.

Leaf sampling for tissue nutrient analysis is recommended for primocanes from May to August (Bolda et al., 2012), "following harvest" (Fernandez and Ballington, 1999), the first week of August (Bushway et al., 2008), or late July to early August (Hart et al., 2006). The recommended nutrient sufficiency levels are similar among these currently available nutrient management guides and all have the same standards and sampling time recommendations regardless of the blackberry type. Strik (2015) recommended that primocane-fruiting blackberry be sampled at the early green-fruit stage (about 8 weeks after summer pruning) than a particular calendar date and suggested the leaf sufficiency range for phosphorus (P) and potassium (K) may need to be lowered for this crop.

Primocane leaf nutrient levels have been shown to vary over the growing season in erect (Clark et al., 1988) and trailing (Mohadjer et al., 2001) floricane-fruiting blackberry, primocane-fruiting blackberry (Strik, 2015), and floricane-fruiting raspberry (Hughes et al., 1979; John and Daubeny, 1972; John et al., 1976; Kowalenko, 1994; Wright and Waister, 1980). Although floricane leaves in blackberry have been shown to change over the fruiting season, standards for this leaf tissue type have only been developed in Brazil (Pereira et al., 2015).

Cultivars of blackberry (Fernandez-Salvador et al., 2015a, 2015b, 2015c; Dixon et al., 2016; Harkins et al., 2014; Strik, 2015) and raspberry (John and Daubeny, 1972; John et al., 1976) differed in primocane leaf nutrient levels when sampled in midseason. By contrast, Clark et al. (1988) found no difference among three erect blackberry cultivars in leaf nutrient levels and speculated that this was because of their similar parentage.

The objective of this study was to evaluate the impact of sample date on primocane leaf nutrient concentration in trailing, erect, and semierect blackberry cultivars with a goal of establishing the ideal sampling time for these crops. In addition, we studied the impact of sampling time during fruiting lateral development and fruiting on the nutrient concentration in the floricane to assess whether this might offer an alternative sampling method in these types of blackberry.

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Materials and Methods

Study site. The study was conducted in 2013 and 2014, in a mature planting at Oregon State University's North Willamette Research and Extension Center, Aurora, OR [lat. 45°16′47′′N, long. 122°45′23″W; USDA hardiness zone 8b (U.S. Department of Agricultural Research Service, 2014); the weather records for this site can be viewed at U.S. Department of Interior (2014)]. The soil is mapped as a Willamette silt loam, classified as a fine-silty, mixed, superactive, mesic, Pachic, Ultic, and Argixeroll.

Plants were established in Spring 2010 except for 'Triple Crown' (planted Oct. 2010) and 'Columbia Star' (planted Sept. 2011) at 1.5 m in the row with 3 m between rows (2222 plants/ha). A permanent grass cover crop grew between the rows. The in-row area was maintained (bare soil) with preemergent herbicides and hoeing, as needed. Plants were irrigated with a single line of drip tubing (UniRam; Netafim USA, Fresno, CA) containing pressure-compensating emitters (1.9 $L \cdot h^{-1}$ in-line) spaced every 0.6 m.

Soil testing. Soil samples were collected on 12 Nov. 2013 and 21 Oct. 2014 using a 2.4-cm diameter, 0.5-m long, slotted, open-side, chrome-plated steel soil probe (Soil Sampler Model Hoffer; JBK Manufacturing, Dayton, OH). Soil was sampled to a depth of 0.3 m at the center of the row, ≈0.3 m from the crown between plants and within the water emitter drip zone or fertilization area. A pooled sample was collected for the site (not replicated) and was sent for analysis of macronutrient and micronutrient concentration and pH to Brookside Laboratories Inc. (New Bremen, OH) (Table 1).

Cultivars. Four trailing blackberry cultivars ('Black Diamond', 'Columbia Star', 'Marion', and 'Onyx'), one erect type ('Ouachita'), and two semierect types ('Chester Thornless' and 'Triple Crown') were studied.

Production system. The trailing cultivars were grown in an alternate year production system (Strik and Finn, 2012), but only plants in the fruiting or "on year" were sampled in 2013 and 2014. Plants were trained on a two-wire vertical trellis system in each row with the wires attached to steel posts at 1.0 and 1.6 m above the ground. New primocanes were trained along the ground in the row, under the floricane canopy during the growing season, per standard practice. In the erect and semierect blackberry cultivars, primocanes were tipped at a height of ≈1.0–1.5 m per standard practice to encourage branching. Floricanes were removed in autumn (after they had senesced). Primocanes were then trained to the same two-wire vertical trellis system as the trailing cultivars by wrapping and tying as necessary. Primocane branches were pruned to 1 m, in winter, as needed.

Fertilizer nutrients were applied per standard recommendations (Hart et al., 2006) and using results from soil analysis (Table 1). In 2013, 79 $kg \cdot ha^{-1}$ N was applied in a split application with 34 $kg \cdot ha^{-1}$ on 3 Apr. (8N–7.5P–14K) and 45 $kg \cdot ha^{-1}$ on 28 May (40N–0P–0K–5Mg). In 2014, 90 $kg \cdot ha^{-1}$ N was applied in two equal portions on 27 Mar. and 5 June (16N–7P–13K). An additional 28 $kg \cdot ha^{-1}$ N was applied to erect and semierect cultivars on 7 July 2014 (16N–7P–13K). These granular fertilizers were broadcast within the in-row area and were washed into the soil through rainfall (data not shown). Copper fungicide (Nu-Cop; Albaugh Inc., Ankeny, IA) was applied to all plants (0.4 $kg \cdot ha^{-1}$ of Cu) on 24 Mar. 2014 to control the cane diseases purple blotch [*Septocyta ruborum* (Lib.) Petr.] and cane rust [*Kuehneola uredines* (Link) Arthur].

Leaf sampling. Tissue samples for nutrient testing were collected approximately every 2 weeks by choosing the most recent fully expanded leaves on primocanes (20–21 May to 6–7 Oct. 2013 and 2014) and floricane laterals (trailing: 6–7 May to 28–29 July; erect and semierect: 6–7 May to 6–7 Oct. 2013 and 2014) for a total of 11 samples in each year for primocanes (Table 2). 'Black Diamond' and 'Ouachita' primocanes grew earlier in the spring and were thus sampled 2 weeks before those of the other cultivars. Stages of plant development and fruiting season were recorded and are presented in Table 2. The rate of plant development and fruiting season did not appear to differ between years (data not shown). Yield data were not recorded, but the field was observed to have a good, typical commercial yield for these cultivars in Oregon.

About 6 or 12 of the most recent, fully expanded primocane and fruiting lateral leaves, respectively, including petioles, were sampled per plot on each date and were left unwashed per standard recommendation (Hart et al., 2006). Sampled leaves were priority shipped to Brookside Laboratories for analysis. Leaf N was determined using a combustion analyzer with an induction furnace and a thermal conductivity detector (Gavlak et al., 1994). Other nutrients, including P, K, calcium (Ca), magnesium (Mg), aluminum (Al), B, copper (Cu), manganese (Mn), iron (Fe), and zinc (Zn) were determined using an inductively coupled plasma spectrophotometer after wet ashing the samples in nitric/perchloric acid (Gavlak et al., 1994).

Data analysis. The treatments were arranged in a completely random design with three replicates of three-plant plots. Data were analyzed by tissue type (primocane or floricane) separately, as our goal was not to compare canes, but to determine changes in leaf nutrient concentration over the sampling period and assess optimal sampling times. In addition, there is evidence that floricanes and primocanes are independent in blackberry (Bryla and Strik, 2008). On each sample date, leaf nutrient data were first analyzed for the effect of year and cultivar using PROC MIXED (SAS version 9.3) with year as the main effect ($n = 2$) and cultivar as the subplot effect ($n = 4$) with a Satterthwaite approximation used, as needed, for main effect comparisons.

Three sample dates (from late July to late August) that bracketed the recommended tissue nutrient sampling time for blackberry in our region (Hart et al., 2006), were analyzed to determine the effect of year, cultivar, and sample date. Mean comparisons were performed using least-square means. Contrasts were used to determine differences between early- and late-fruiting types (trailing vs. erect and semierect) as well as differences between erect and semierect types during this period.

PROC UNIVARIATE and the Shapiro–Wilk procedure were used to assess normality of the data for all the aforementioned analyses. As the tissue concentration of many nutrients was not normally distributed, a log transformation was used to improve homogeneity of variance and to assess proportional effects. Data were back transformed for presentation.

Results and Discussion

Rate of development and fruiting season. Key phenological stages and fruiting season are presented for the cultivars studied for each tissue sample date (Table 2). There were no observed differences among key phenological stages between years (data not shown). The trailing cultivars Black Diamond, Columbia Star, Marion, and Onyx had the earliest fruiting seasons, followed by the erect cultivar Ouachita and then the semierect cultivars Triple Crown and Chester Thornless (Table 2).

Year effect. There was a significant main effect of year on primocane leaf nutrient concentration of the cultivars studied on many sample dates through the season for all nutrients (data not shown). In general, the concentration of N, S, B, Fe, and Al was higher and that of P, Mg, K, Ca, Mn, and Cu was lower on one or more sample dates in 2013 than in 2014.

Table 1. Soil nutrient pH, organic matter content, and nutrient levels when sampled in Nov. 2013 and Oct. 2014 at Oregon State University's North Willamette Research and Extension Center, Aurora, OR (unreplicated).

			Ppm													
Yr	pH	Organic matter (%)	NH_4^-N	NO_3^-N	P[z]	K	Ca	Mg	S	B	Fe	Mn	Cu	Zn	Al
2013	5.5	2.9	8.5	1.5	395	333	934	240	19	0.48	354	34	9.4	3.8	1375
2014	5.8	3.1	5.5	4.2	278	312	1159	284	22	0.43	365	35	8.8	3.9	1296

[z]P tested using Bray I method (Brookside Laboratories Inc., New Bremen, OH).

Table 2. Developmental stages for primocanes and floricanes through the season (2013 and 2014 averaged) for floricane-fruiting blackberry types and cultivars studied at Oregon State University's North Willamette Research and Extension Center, Aurora, OR.

Blackberry type and cultivar	Approximate stage of development on each date[z] 15 May	1 June	15 June	1 July	15 July	1 Aug.	15 Aug.	1 Sept.	15 Sept.	1 Oct.	15 Oct.
Floricane-fruiting trailing											
Black Diamond	Late fruit set	Mid green fruit	Late red fruit	Harvest	Harvest	End of harvest	P[y] growing	P growing	P growing	P growing	P growing
Columbia Star	Late fruit set	Mid green fruit	Late red fruit	Harvest	Harvest	End of harvest	P growing	P growing	P growing	P growing	P growing
Marion	Late bloom	Early green fruit	Early red fruit	Begin harvest	Late harvest	P growing	P growing	P growing	P growing	P growing	P growing
Onyx	Full bloom	Early green fruit	Early red fruit	Late red fruit	Harvest	Late harvest	P growing	P growing	P growing	P growing	P growing
Floricane-fruiting erect											
Ouachita	Very early bloom	Late bloom to early green	Late green fruit	Late green to first red	Late red to shiny black	Harvest	Harvest	Late harvest	P growing	P growing	P growing
Floricane-fruiting semierect											
Triple Crown	P growing	Early bloom	Late bloom to early green	Mid green fruit	Late red to shiny black	Late red to shiny black	First harvest	Harvest	Harvest	End of harvest	P growing
Chester Thornless	P growing	Early bloom	Early green	Late green to first red	Late red fruit	Late red to shiny black	Late red to shiny black	Harvest	Harvest	Late harvest	P growing

[z]Approximate stage of development is provided. The beginning and end of fruit harvest for a particular cultivar may have occurred between the dates provided. Primocane leaves were sampled for tissue analysis on 6 May, 19–20 May, 2–3 June, 16–17 June, 30 June–1 July, 14–15 July, 28–29 July, 12 Aug., 25–27 Aug., 8–10 Sept., 22–23 Sept., and 6–7 Oct. (depending on year). Leaves on fruiting laterals were sampled for tissue analysis on 6 May, 19–20 May, 2–3 June, 16–17 June, 30 June–1 July, 14–15 July, and 28–29 July for all blackberry types and additionally on 12 Aug., 25–27 Aug., 8–10 Sept., 22–23 Sept., and 6–7 Oct. for the erect and semierect cultivars.

[y]P = primocane; primocane growth would have slowed toward the end of the season (e.g., 1 Oct.) as temperatures declined in autumn.

There was an effect of year on floricane leaf nutrient concentration of the cultivars studied on many sample dates for all the nutrients (data not shown). The concentrations of K, B, Fe, Zn, and Al were higher, whereas N, P, Mg, Ca, S, and Mn were lower on at least one sample date in 2013 than in 2014.

The pattern of the change in primocane and floricane leaf nutrient concentration within each cultivar was relatively similar through the season between years (data not shown), but there was an effect of year and cultivar, and sometimes an interaction between year and cultivar found for several nutrients and sample dates. We have chosen not to show both years of data for seven cultivars and 11 primocane and 7–11 floricane sample dates. Instead, we are presenting data for the 2014 growing season to show the pattern of change over time. In Table 3, we present year by cultivar interactions for primocanes on select dates that bracket the range in current tissue nutrient sampling recommendations (Hart et al., 2006).

Primocane leaf nutrient concentration. There was a significant effect of cultivar on primocane leaf N concentration in early spring and toward the later part of the growing season (Fig. 1). 'Black Diamond', 'Columbia Star', 'Marion', and 'Onyx' had lower leaf N in mid-June than the erect and semierect cultivars, whereas 'Chester Thornless' and 'Triple Crown' had a higher and lower leaf N, respectively, in the late summer and autumn. Leaf N declined from spring to summer in all the cultivars and was then variable ('Chester Thornless') or remained relatively stable (all others).

Leaf P declined through the season in 'Ouachita', 'Triple Crown', and 'Chester Thornless', whereas concentrations increased in late summer in most of the trailing cultivars to levels significantly higher than in the erect and semierect cultivars (Fig. 1). 'Onyx' was similar to the other trailing cultivars until mid-August when the P concentration in primocane leaves declined to levels similar to those measured in 'Chester Thornless'.

Leaf K concentration declined from spring to autumn in 'Ouachita' (except for one increase in late July) and 'Triple Crown', as was reported for primocane-fruiting blackberry by Strik (2015), but in 'Chester Thornless' levels were the lowest in late summer. 'Columbia Star', 'Marion', and 'Black Diamond' had the highest leaf K concentration in late summer to autumn (Fig. 1), perhaps because the fruiting season was over in these cultivars (Table 2), although leaf K in 'Onyx' was highest in the spring.

Leaf Mg and Ca concentrations increased overall from spring to autumn in the erect and semierect cultivars, although levels did fluctuate during the summer, whereas in trailing cultivars levels were

Table 3. The effect of year and cultivar on primocane leaf nutrient concentration of floricane-fruiting trailing, erect, and semierect cultivars at the North Willamette Research and Extension Center, Aurora, OR and sampled on three dates in late July–early Aug. 2013 and 2014.

	N[z]		P		Mg		K		Ca		S		B		Fe		Mn		Cu		Zn		Al	
	%																		ppm					
Tissue standards[y]	2.3–3.0		0.19–0.45		0.3–0.6		1.3–2.0		0.6–2.0		0.1		30–70		60–250		50–300		6–20		15–50		n/a	
Year																								
2013	2.24		0.18 b[x]		0.42 a		1.06		0.77		0.14		45		223 b		200		6.9 b		29		193 b	
2014	2.20		0.21 a		0.41 b		1.05		0.85		0.14		46		334 a		239		7.9 a		30		305 a	
Date[w]	2013	2014	2013	2014	2013	2014	2013	2014	2013	2014	2013	2014	2013	2014	2013	2014	2013	2014	2013	2014	2013	2014	2013	2014
7	2.53 a	2.36 ab	0.18 bc	0.22 a	0.49 a	0.40 b	1.10 ab	1.14 a	1.01 a	0.70 b	0.17 a	0.15 b	61 a	37 c	173 b	236 a	271a	180 cd	6.9 c	7.1 c	33 a	29 b	139 b	203 a
8	2.13 cd	2.29 bc	0.17 c	0.23 a	0.40 b	0.41 b	0.97 c	1.08 ab	0.69 b	0.70 b	0.14 bc	0.15 b	40 c	41 c	168 b	408 a	171 d	206 bc	6.3 d	7.9 ab	26 c	31 ab	143 b	379 a
9	2.16 cd	2.00 d	0.19 b	0.19 b	0.41 b	0.41 b	1.09 ab	0.99 bc	0.65 b	1.10 a	0.13 cd	0.12 d	36 c	57 b	318 b	312 a	170 d	311 ab	7.4 bc	8.3 a	29 b	30 ab	289 b	288 a
Cultivar			2013	2014	2013	2014	2013	2014	2013	2014	2013	2014			2013	2014			2013	2014	2013	2014	2013	2014
Black Diamond	1.97 d		0.171 fgh	0.214 bc	0.36 ef	0.36 ef	0.89 ef	0.97 cde	0.66 efg	0.68 ef	0.12 gh	0.11 gh	43 bc		334 b	505 a	192 bc		6.5 g	8.2 ab	22 j	27 ghi	302 b	456 a
Chester Thornless	2.42 a		0.187 defg	0.216 bc	0.51 ab	0.46 bc	1.08 bc	1.00 cde	0.91 abc	1.08 ab	0.18 ab	0.17 abc	50 ab		213 bc	233 bc	270 a		8.4 a	8.8 a	31 cdef	29 efgh	182 bcd	204 cd
Columbia Star	2.11 cd		0.190 def	0.237 b	0.33 fgh	0.29 h	1.29 a	1.45 a	0.57 fg	0.51 g	0.12 gh	0.12 fgh	39 c		236 c	257 bc	170 c		7.1 de	8.0 ab	26 hi	25 ij	209 cd	242 bc
Marion	2.29 ab		0.200 cde	0.294 a	0.40 de	0.30 gh	1.08 bc	1.30 ab	0.92 abc	0.58 efg	0.11h	0.13efg	39 c		229 bc	238 bc	181 c		6.1 g	8.3 abc	30 defg	39 a	199 bcd	218 bc
Onyx	2.36 a		0.184 efgh	0.209 cd	0.36 efg	0.40 de	0.97 def	0.86 f	0.75 de	0.83 bcd	0.13e	0.13ef	46 c		222 bc	576 a	215 bc		6.7 efg	7.6 bcd	28 fghi	26 hi	194 bcd	544 a
Ouachita	2.14 ab		0.165 h	0.172 gh	0.49 bc	0.43 cd	1.09 bc	1.09 cd	0.75 cde	0.81 cde	0.17 bcd	0.16 d	42 c		171 cd	192 cd	217 bc		7.0 ef	7.1 cde	36 ab	33 abcd	146 d	166 de
Triple Crown	2.21 bc		0.170 gh	0.195 def	0.58 a	0.50 b	0.98 cdef	1.05 cd	0.91 abc	1.13 a	0.19 a	0.17 cd	57 a		132 d	182 cd	264 ab		6.4 fg	6.7 efg	32 bcde	34 abc	100 e	161 de
Significance[v]																								
Year	NS		0.0017		0.0218		NS		NS		NS		NS		0.0186		NS		0.0023		NS		0.0151	
Date	0.0006		0.0367		0.0444		NS		0.0167		<0.0001		0.0325		0.0023		NS		0.0014		0.0438		0.0004	
Year × date	0.0297		0.0005		0.0178		0.0227		0.0003		0.014		0.0001		0.0129		0.001		0.0048		0.0031		0.0087	
Cultivar	<0.0001		<0.0001		<0.0001		<0.0001		<0.0001		<0.0001		<0.0001		<0.0001		0.0044		<0.0001		<0.0001		<0.0001	
Year × cultivar	NS		0.0119		0.0075		0.0348		0.022		0.0099		NS		0.0103		NS		0.001		0.0038		0.0035	
Date × cultivar	0.0271		0.0001		NS		0.0003		<0.0001		0.0001		<0.0001		NS		0.0001		<0.0001		NS		0.0267	
Year × date × cultivar	NS		NS		NS		NS		NS		NS		0.0367		0.0005		0.0369		NS		NS		<0.0001	
Trailing vs. other	0.0013		<0.0001		<0.0001		NS		<0.0001		<0.0001		0.0001		<0.0001		0.0005		NS		<0.0001		<0.0001	
Erect vs. semierect	NS		0.0003		0.0087		NS		0.0006		0.0009		0.0002		NS		0.0415		0.0313		0.0144		NS	

[z]N = nitrogen; P = phosphorus; Mg = magnesium; K = potassium; Ca = calcium; S = sulfur; B = boron; Fe = iron; Mn = manganese; Cu = copper; Zn = zinc; Al = aluminum.
[y]Recommended sufficiency range for blackberry when sampled in late July to early August (Hart et al., 2006); no sufficiency levels are available for aluminum (n/a).
[x]Means followed by the same letter within treatment or the interaction are not significantly different (LSmeans) ($P > 0.05$).
[w]Sampling dates for 7, 8, and 9 were 29 July, and 12 and 27 Aug. 2013, respectively, and 28 July, and 12 and 25 Aug. 2014, respectively.
[v]Nonsignficant (NS) or actual *P* value provided when significant by analysis of variance.

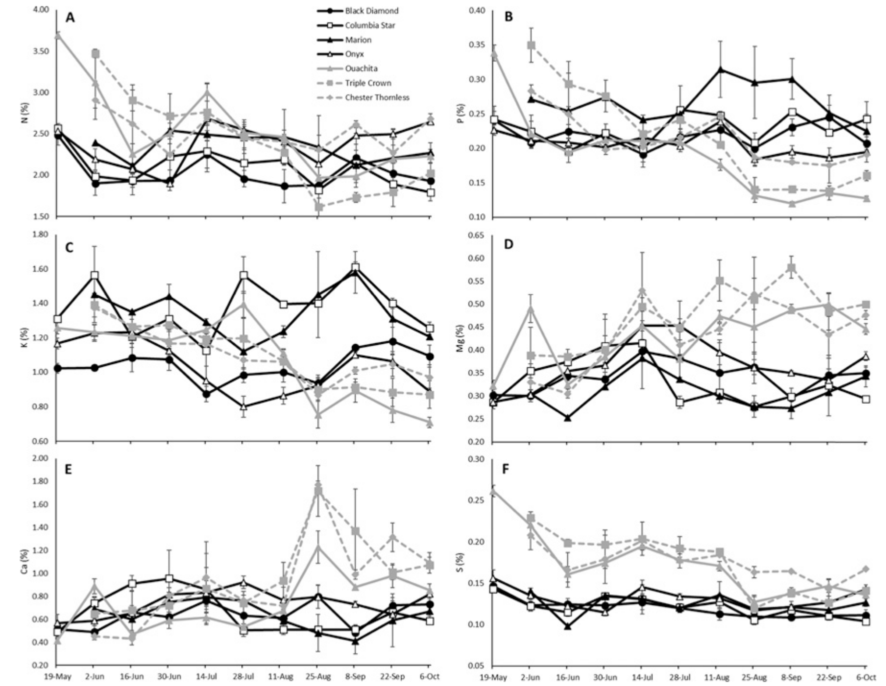

Fig. 1. The effect of sample date over the growing season on the concentration of macronutrients in primocane leaves of floricane-fruiting, trailing ('Black Diamond', 'Columbia Star', 'Marion', and 'Onyx'), erect ('Ouachita'), and semierect ('Chester Thornless' and 'Triple Crown') blackberry at the North Willamette Research and Extension Center, Aurora, OR, 2014. **A** = nitrogen; **B** = phosphorus; **C** = potassium; **D** = magnesium; **E** = calcium; **F** = sulfur. Bars indicate standard error for cultivar.

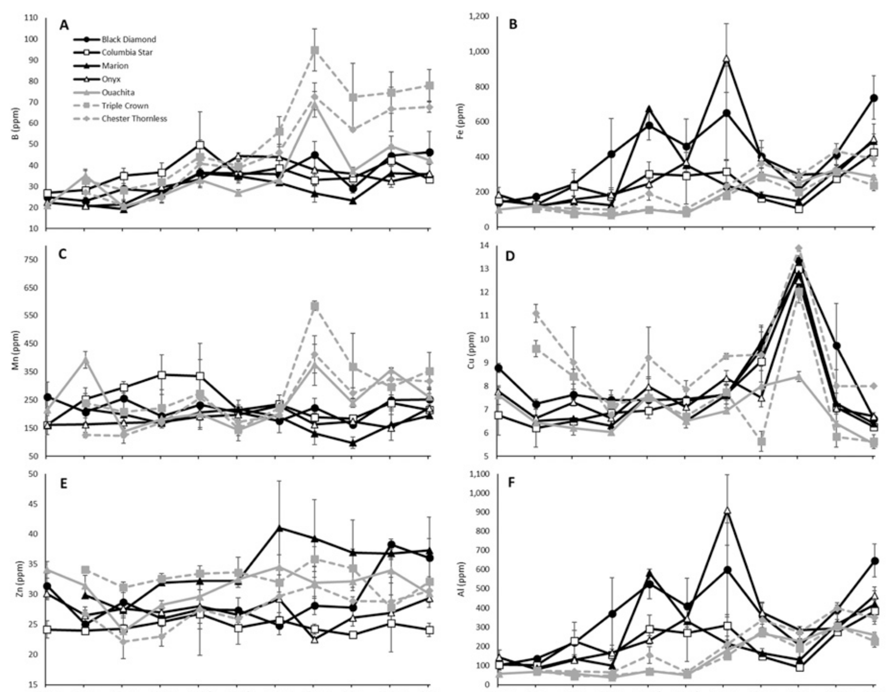

Fig. 2. The effect of sample date over the growing season on the concentration of micronutrients in primocane leaves of floricane-fruiting, trailing ('Black Diamond', 'Columbia Star', 'Marion', and 'Onyx'), erect ('Ouachita'), and semierect ('Chester Thornless' and 'Triple Crown') blackberry at the North Willamette Research and Extension Center, Aurora, OR, 2014. **A** = boron; **B** = manganese; **C** = iron; **D** = copper; **E** = zinc; **F** = aluminum. Bars indicate standard error for cultivar.

relatively flat through the season and were lower in late summer. Peaks in leaf Ca concentration during the season may have been related to variability in leaf sampling caused by summer pruning (tipping) as reported for primocane-fruiting blackberry (Strik, 2015). The trailing blackberry cultivars had a considerably lower leaf S concentration than the erect and semierect cultivars, especially from spring to midsummer (Fig. 1).

The pattern of change in leaf B was similar amongst the blackberry cultivars in the spring to midsummer (Fig. 2). However, leaf B increased rapidly from late July to the end of August in the erect and semierect cultivars to levels significantly higher than in the trailing cultivars, particularly in 'Chester Thornless' and 'Triple Crown'. A similar pattern was observed for leaf Mn. Leaf Cu was relatively stable in the early season except for the semierect cultivars, but increased for all cultivars except 'Ouachita' in the late season. Leaf Zn concentration generally increased through the season and was highest ('Marion') and lowest ('Columbia Star') in the trailing cultivars on many sample dates.

Primocane leaf Fe and Al concentrations were much higher in the trailing cultivars than the other blackberry types likely because there was dust (soil) on these leaves and leaves are not washed before tissue analysis (Hart et al., 2006); trailing blackberry primocanes are not self-supporting and were laying on the soil during the sampling period as opposed to the erect growth habit of 'Ouachita', 'Triple Crown', and 'Chester Thornless'.

Sampling time recommendations. The current tissue sampling recommendations for blackberry vary by region (Bolda et al., 2012; Bushway et al., 2008; Fernandez and Ballington, 1999; Hart et al., 2006). Sampling is recommended for a time when tissue nutrient concentrations are relatively stable to minimize variation. Rather than a specific date, Strik (2105) recommended that primocane-fruiting, erect blackberry cultivars be sampled at the bloom to green-fruit stage of growth. In the erect and semierect cultivars we studied, the only nutrients that appeared to have relatively stable concentrations in the late July to early August sampling period were N, K, S, Mn, Zn, and Cu (Figs. 1 and 2). However, by late July to early August, some nutrients started to increase (Ca, Mg, B, and Al) or decrease (P) in concentration.

Tissue nutrient concentrations for the three primocane leaf sampling dates ranging from the end of July to late August of each year were compared for all cultivars (Table 3). We chose these dates to correspond to a range similar to what is currently recommended for sampling in our region (Hart et al., 2006). There was a significant year by sampling date interaction for all nutrients. When comparing sampling in late July to early August (dates 7 and 8 in Table 3), there was no sampling date effect for N, Mg, K, Ca, S, B, Mn, and Zn (only in 2014); P, Fe, and Al in both years; primocane leaf Cu was not stable during this period in either year. However, when the sampling period was mid to late August (dates 8 and 9, Table 3), primocane leaf N, Mg, Fe, Mn, and Al were stable in both years, leaf K, Ca, S, B, Cu, and Zn were stable in one of the years of study, whereas leaf P was not stable in either year.

Leaf N concentration declined from late July to mid-August, particularly in 2013 (Table 3), and leaf N, on average, was only within the current recommended sufficiency levels on the first sample date (both years). Leaf P was below current sufficiency levels on the first two sample dates in 2013, but was at the low end of the range on all dates. Average leaf K concentration was below current sufficiency levels on all sampling dates despite sufficient soil K (Table 1) and fertilization with K at recommended levels. Leaf K declined during the late July to late August sampling period in 2014 but only declined from late July to early August in 2013 before increasing again in late August (Fig. 1). Primocane leaf Fe and Mn were above recommended sufficiency levels on one or more later sampling dates (Table 3), perhaps because of the presence of dust on the leaves (Fe) and a soil pH that was at the low end of the recommended range for blackberry (Hart et al., 2006). All of the other nutrients were within current sufficiency levels during the late July to late August sample period.

Cultivar had a significant effect on the nutrient concentration of primocane leaves when sampled from late July to late August (Table 3). The trailing cultivars had a lower average leaf N than the erect and semierect cultivars (which did not differ). There was a cultivar × date interaction, because leaf N in 'Onyx', 'Columbia Star', 'Triple Crown', and 'Ouachita' declined rapidly from mid to late August, whereas leaf N changed relatively little during this period in the other cultivars (Fig. 1). All cultivars except 'Chester Thornless' and 'Onyx' had an average primocane leaf N concentration below the current recommended standards (when sampling from late July to late August; Table 3).

Average leaf P was below current sufficiency levels in all cultivars in 2013 and in 'Ouachita' in 2014 (Table 3). Trailing cultivars had higher leaf P than the erect and semierect cultivars, whereas the erect cultivar, Ouachita, had a lower leaf P than the semierect cultivars. Leaf P increased during the early sampling period for 'Marion', 'Chester Thornless', and 'Onyx', whereas levels remained relatively stable or declined in the other cultivars (Fig. 1).

Leaf Mg concentration was lower in the trailing cultivars than the other types and was higher in the semierect cultivars than in the erect cultivar, Ouachita (Table 3). Although blackberry type did not affect leaf K, there was a strong effect of cultivar. 'Columbia Star' had the highest leaf K and 'Black Diamond' and 'Onyx' the lowest, depending on year. All cultivars had a leaf K below the current sufficiency levels except for 'Columbia Star' and 'Marion' in 2014 (Table 3). Although there was no date × cultivar interaction during the late July to late August sampling period (Table 3), leaf K was higher in the late season for all trailing cultivars except 'Onyx' as compared with the erect and semierect cultivars that were still fruiting at this time (Fig. 1; Table 2). It is possible that uptake of K from the soil preferentially goes to the floricane during the fruiting period. On average, trailing cultivars had lower leaf Ca than erect and semierect cultivars and 'Ouachita' had a higher leaf Ca than the semierect cultivars (Table 3). There was a sampling date × cultivar interaction during the late July to late August period because leaf Ca increased from mid to late August in the erect and semierect cultivars considerably compared with the trailing cultivars (Fig. 1). Leaf Ca levels are affected by tipping date and resulting leaf age at sampling in erect blackberry (Strik, 2015).

The trailing blackberry cultivars had lower leaf B, Mn, and Zn and higher leaf Fe and Al than the erect and semierect cultivars from late July to late August (Table 3). The semierect cultivars had higher leaf B, Mn, and Cu, but lower Zn than the erect cultivar Ouachita. Leaf B and Mn increased in the erect and semierect cultivars on the late August sample date relative to the trailing cultivars (Fig. 2). Leaf Cu was significantly lower in 'Triple Crown' than the other cultivars on the late August sample date (Fig. 2). 'Onyx' and 'Columbia Star' had a lower leaf Al than the other cultivars on the late August sample date (Fig. 2).

Based on current sufficiency levels and the pattern of leaf nutrient change over the season, we do not recommend sampling the late-fruiting floricane cultivars 'Ouachita', 'Triple Crown', or 'Chester Thornless' at a particular developmental stage as was suggested by Fernandez and Ballington (1999; "following harvest") or for primocane-fruiting blackberries (Strik, 2015). The primocane leaf nutrient concentrations measured in this study were below the published recommended standards for blackberries (Bolda et al., 2012; Bushway et al., 2008; Fernandez and Ballington, 1999; Hart et al., 2006) for P and K (all regions), N (only in Oregon; Hart et al., 2006), and Mg and S (only in the northeastern United States; Bushway et al., 2008). Leaf Fe (all regions) and Mn (California and northeastern United States; Bolda et al., 2012; Bushway et al., 2008) were higher than current standards.

Floricane leaf nutrient concentration. The effect of cultivar on floricane (fruiting lateral) leaf tissue nutrient concentration over the 2014 growing season is shown in Figs. 3 and 4. Floricane leaf N concentration declined during the floricane season from early-fruiting lateral development (6 May) through fruiting and early floricane senescence in all of the blackberry types and cultivars (Fig. 3). Nitrogen may have declined as N was mobilized to the developing fruit. Leaf N levels were similar to those reported for floricane leaves of 'Tupy' and 'Xavante' erect blackberry in Brazil (Pereira et al., 2015) and 'Black Diamond' and 'Marion' in Oregon (Dixon et al., 2016). The pattern of change in leaf S in the floricanes was similar to leaf N (Fig. 3).

Floricane leaf P declined rapidly in the early spring for all cultivars, but was then relatively stable from early June through floricane senescence, except in 'Marion' where leaf P dropped from early to late July. 'Black Diamond' had the greatest leaf P in June and early July, whereas 'Chester Thornless' had a significantly higher leaf P in the late summer than the other erect and semierect cultivars (Fig. 3).

Leaf K in the fruiting lateral declined from spring to summer in the early-fruiting trailing cultivars and the midseason erect cultivar Ouachita, whereas leaf K did not start declining much in the semierect cultivars until late July (Fig. 3) when fruit coloring began (Table 2). 'Black Diamond', 'Columbia Star', and 'Onyx' leaf K increased during the last 2 weeks of July just before the onset of senescence. Floricane leaf K concentrations were similar to those reported in other erect cultivars (Pereira et al., 2015) and in 'Black Diamond' (Dixon et al., 2016); however, leaf K for 'Marion' was higher in our study than what was previously reported by Dixon et al. (2016). Leaf K declined during fruiting likely because of the relatively high K content of blackberry fruit (Dixon et al., 2016; Harkins et al., 2014).

Floricane leaf Mg increased from spring to the beginning of the fruiting season in the erect and semierect cultivars, whereas leaf Mg stayed relatively stable in the early-fruiting trailing cultivars (Fig. 3). There was a very similar pattern in leaf Ca for all types and cultivars studied with increasing levels through the season. The reason for the decline in leaf Ca in the floricanes of erect and semierect cultivars in late August is not clear, but this decline was not observed in 2013 for these cultivars (data not shown). Floricane leaf Ca and Mg concentrations likely increased through the season because these leaves were aging, as has been reported in older leaves of raspberry (Hughes et al., 1979) and blackberry (Strik, 2015), and there are relatively low concentrations of these nutrients in trailing blackberry fruit (Dixon et al., 2016; Harkins et al., 2014).

In general, leaf B, Mn, Fe, and Al concentration increased during the floricane season, with the exception of lower values of leaf B and Mn for the erect and semierect cultivars either just after fruit harvest or during fruit harvest, respectively (Fig. 4). Leaf Cu concentrations decreased slightly in the early season, but increased and were more variable for the later-fruiting cultivars from August to October. Leaf Zn levels declined in May and then increased slightly or remained relatively stable during fruiting, depending on cultivar.

Nutrient sufficiency standards have not been developed for floricane leaves of any type of blackberry in North America. Because the nutrient concentration in floricane leaves of trailing cultivars just before fruit harvest (approximately mid-June to 1 July) was greater than published primocane leaf sufficiency levels (Hart et al., 2006) for most nutrients, floricane-specific standards would

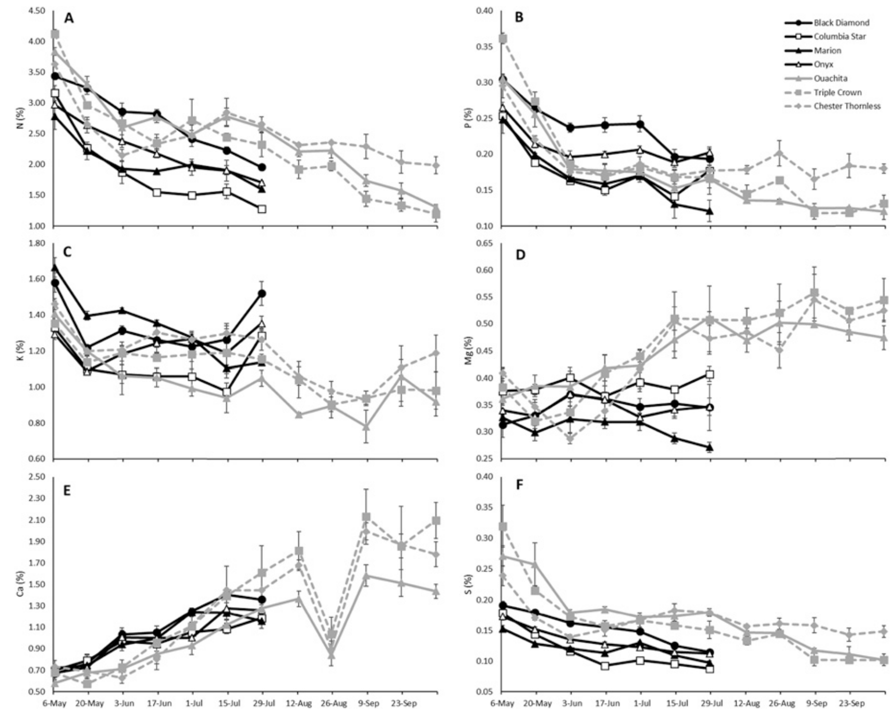

Fig. 3. The effect of sample date over the growing season on the concentration of macronutrients in floricane leaves of trailing ('Black Diamond', 'Columbia Star', 'Marion', and 'Onyx'), erect ('Ouachita'), and semierect ('Chester Thornless' and 'Triple Crown') blackberry at the North Willamette Research and Extension Center, Aurora, OR, 2014. **A** = nitrogen; **B** = phosphorus; **C** = potassium; **D** = magnesium; **E** = calcium; **F** = sulfur. Bars indicate standard error for cultivar.

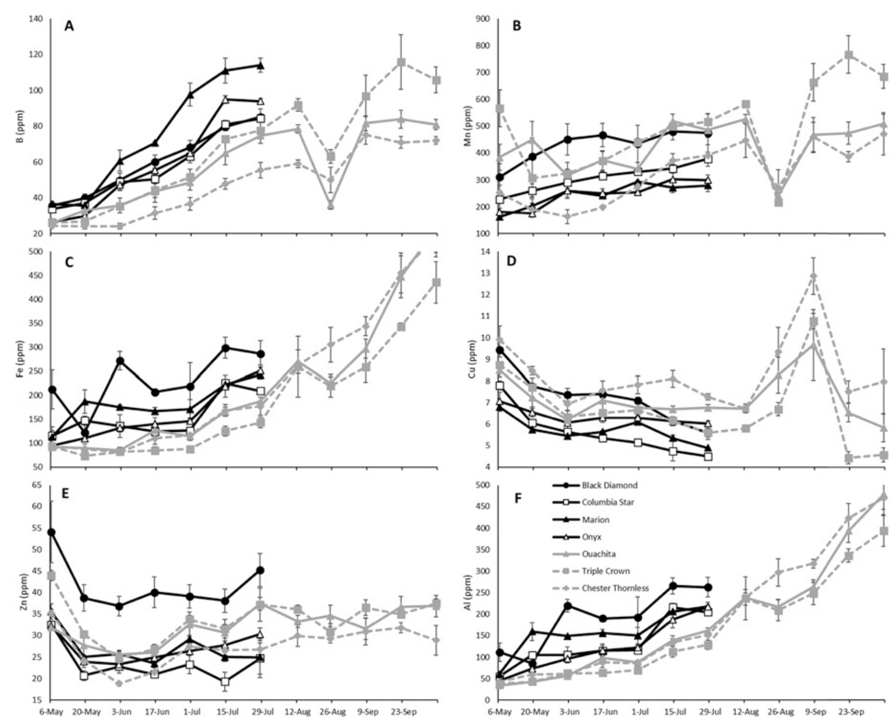

Fig. 4. The effect of sample date over the growing season on the concentration of micronutrients in floricane leaves of floricane-fruiting, trailing ('Black Diamond', 'Columbia Star', 'Marion', and 'Onyx'), erect ('Ouachita'), and semierect ('Chester Thornless' and 'Triple Crown') blackberry at the North Willamette Research and Extension Center, Aurora, OR, 2014. **A** = boron; **B** = manganese; **C** = iron; **D** = copper; **E** = zinc; **F** = aluminum. Bars indicate standard error for cultivar.

need to be developed. However, we do not recommend floricane leaves be used to manage fertilizer programs as the concentrations of most nutrients were rapidly changing during the lateral and fruit developmental period. By contrast, the floricane leaf concentrations of most nutrients in the erect and semierect cultivars were within the current primocane sufficiency levels (Table 3) when sampled in late July to early August (just before fruiting, Table 2), except for floricane leaf K (lower) and Fe and Mn (higher) (Fig. 4).

Conclusions

Our findings indicate that the growth habit or fruiting season (or both) of the three floricane-fruiting blackberry types as well as the cultivar grown affect primocane tissue nutrient concentrations. We thus recommend blackberry types and cultivars be sampled separately for tissue analysis. We do not recommend sampling floricane leaves to manage fertilizer programs in any blackberry type in North America, as standards for these diverse types have not been developed.

The currently recommended primocane leaf tissue sampling period of late July to early August (Hart et al., 2006) was not consistently ideal for all nutrients in these diverse blackberry cultivars. In our study, all nutrients except Cu were stable when sampling in the late July to early August period in 2014, but only P, Fe, and Al were stable during this period for both years. However, if sampling was delayed slightly, to mid to late August, more nutrients were stable during this period for both years of the study (N, Mg, Fe, Mn, and Al). Considering leaf N is a key nutrient sampled to adjust fertilizer programs, we recommend shifting the recommended primocane sampling time to this slightly later period.

The primocane leaf nutrient concentrations measured in this study were below (P, K, and N) or above (Fe) current standards for our region (Hart et al., 2006). Our findings thus suggest that changing the sufficiency levels for these diverse blackberry types would aid growers in managing their fertilizer nutrient programs, as proposed by Strik and Vance (2016).

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