Are Boron levels in sandy soils adequate to ensure reliable fruit set and would the application of Boron have a beneficial effect?



FINAL REPORT FOR INCUBATOR PROJECT

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1. Abstract

Optimum nutrition at flowering and fruit set is crucial for grapevines to attain desired reproductive capacity. Boron (B) is reported to be involved in many aspects of plant reproductive physiology. This project aimed to investigate the effect of B as a foliar application at three concentrations (0.025%, 0.05%, 0.1%) and two phenological stages (EL-16, EL-23) on fruit-set of grapevines in the Riverland region in South Australia. A Verdelho block with a history of poor fruit set and documented foliar and soil B deficiency was used for this study. The foliar B application did not increase B concentrations either in petioles or in inflorescences in alignment with the treatments. Fruit set percentage, coulure index and millerandage index were not significantly improved following a single season of foliar treatments. Soil B concentration was highly variable across the trial block, and likely masked the effect of the foliar B treatments. Moreover, vine Fe, Mg^{2+} and Ca^{2+} were deficient. Additionally, hail and storm events around flowering and fruit set period likely impacted the development of the reproductive tissues. Drawbacks in conducting nutrition trials for a single season have been identified and recommendations have been made.

2. Executive summary

Foliar boron (B) application during floral development and anthesis has been proven effective at promoting flowering and increasing fruit set and yield in a variety of perennial tree crops. Observation by industry in the Riverland region found a positive response to the application of foliar boron at flowering in some crops such as almonds, where the tissue B concentration was marginal. Soil B availability is influenced by many factors such as soil texture, pH, soil moisture, and interactions with other nutrients. Many of the Riverland soils are low in B, which can be attributed to its sandy texture and the consequent leaching of B into the lower layers of soil. This project investigated the effect of foliar B applications in the form of boric acid at two critical phenological stages on (a) fruit set and yield parameters and (b) concentrations of B and other mineral elements in inflorescences and petioles at three sampling times of grapevine cultivar Verdelho grown in the Riverland region. The aim was to determine when and at what concentration B application will be most effective at improving fruit set and yield.

Soil sampling pointed towards variable B concentration across the block, and minimal impact of foliar B spray on fruit set and yield parameters. Despite foliar application, B concentration in the flowers and petioles was not significantly increased compared to the control, indicating that the applied B was not efficiently and/or uniformly absorbed by the plants, or if it was absorbed, potentially translocated to organs other than inflorescences and petioles. This suggests that higher boric acid concentration in the spray may be required or, alternatively,

the addition of an adjuvant may be necessary to improve the efficiency of uptake. The 2021-22 season was a particularly harsh season with storms and hail incidents during the flowering and fruit set period in the Riverland Region. Canopy damage likely had an adverse effect on fruit set, complicating interpretation. This suggests the requirement for trials over multiple seasons to not only deal with the complex dynamics of internal nutrient mobilisation and partitioning within the vine, but also the additional complexity of environmental conditions.

3. Background and Aims

Fruit set and yield of grapevines are underpinned by successful floral development, pollination, pollen tube growth and fertilisation (Staudt and Kassemeyer 1984; May 2004). Nutrient deficiencies at the reproductive development stages can affect the success of fertilisation and the subsequent conversion of an ovary to a berry (May 2004). It has been reported by many authors that B is essential for flower fertility, pollen germination and pollen tube growth (Hanson, et al. 1985; Alva, et al. 2015; Wells, et al. 2008). The requirement of B seems to be higher in reproductive organs than vegetative organs (Reid 2014) and, in the field, reproductive growth can be compromised due to low soil boron without symptoms on the vegetative components (Wells, et al. 2008). The benefits of foliar B application on fruit set and yield have been well documented in perennial tree crops (Hanson, et al. 1985; Nyomora, et al. 1999; Perica, et al. 200; Usenik and Stampar 2007). However, there is a great diversity in the consequences of low B on reproductive growth among species (Erdogan and Aygun 2008), and the effects of foliar-applied B on grapevines are poorly understood.

Impaired fruit set in grapevines is a major concern in the Riverland region, where most of the vine growing soils are sandy and potentially low in B. Despite a well-documented positive effect of foliar B application on fruit set and yield of perennial fruit trees, effect of B on grapevines is poorly understood. Better understanding on the optimum time and concentration of B foliar application is also critical in developing corrective nutritional spray programmes to achieve consistent and sustainable fruit set and yield.

The objective of this project was to provide new insights into the potential of foliar B application to improve fruit set and yield of grapevines grown in soils with limited B availability. This project tested the hypothesis that the effective fertilisation and fruit set in grapevines, especially in sandy soils, can be improved by foliar B application during the reproductive development of grapevines. Following are the specific aims of this project:

1. Assess the effect of foliar B application at EL-16 and EL-23 on fruit set, coulure index and millerandage index.

2. Assess the effect of three different concentrations of B in the form of boric acid on the above parameters.
3. Examine B concentrations in the soil, and in the vegetative and reproductive organs before and after sprays and correlate with fruit set.

4. Materials and Methods

The site was located at Kingston Conundrum Vineyard, in the Riverland region in South Australia (34°17'36.2"S 140°43'57.9"E). Monthly rainfall and evapotranspiration data obtained from the Australian Bureau of Meteorology Moorook weather station are given in Supplementary Fig. 1. The trial consisted of Verdelho vines on own roots planted in 2006-2007. The spacing between vines was 2 m and between rows was 3 m with north-south row orientation. Vines were spur-pruned and trained along a single horizontal wire above a bilateral cordon. All vines were managed in the same way in terms of irrigation and nutrition. The trial block was isolated, and in the year of the trial, not included in the annual pre-flowering trace element sprays.

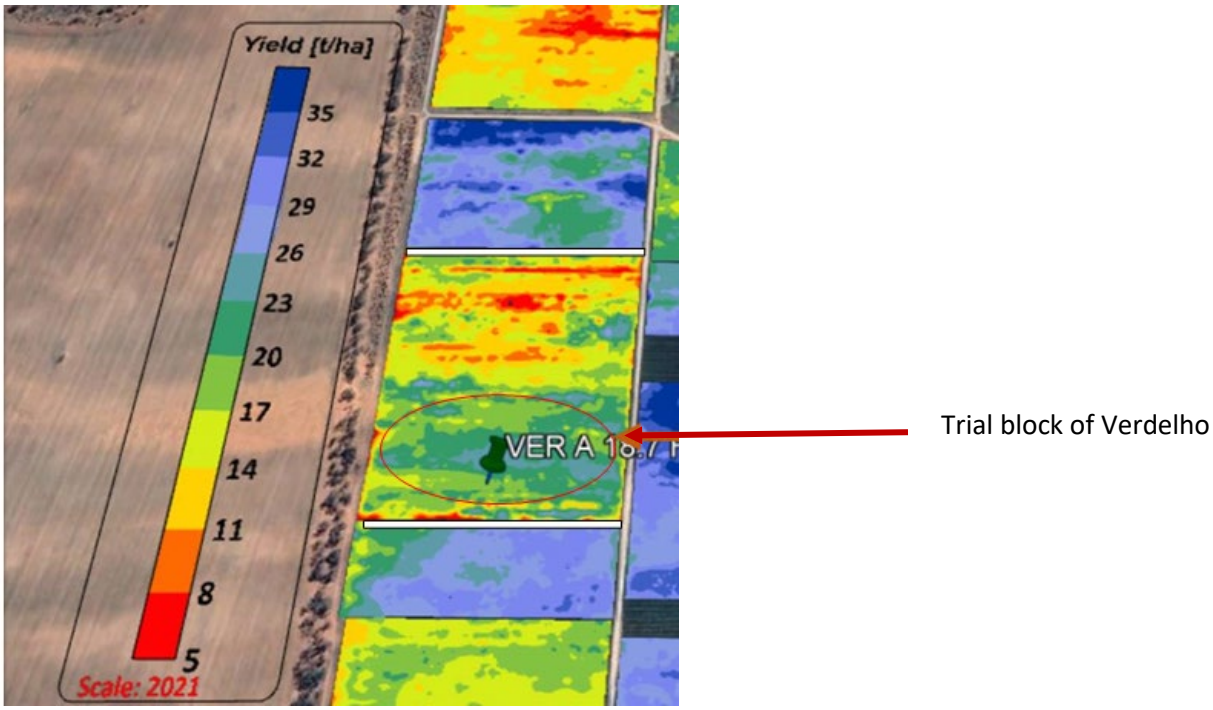


Figure 1. Yield map of the trial block, showing the colour range from red to blue corresponding to low yield (5t/ha) to high yield (35 t/ha).

The trial design included treatments of boric acid 0.025% at EL-16, boric acid 0.05% at EL-16, boric acid 0.1% at EL-16, boric acid 0.025% at EL-23, boric acid 0.05% at EL-23, boric acid 0.1% at EL-23 and a no spray control in a replicated and randomized vineyard design.

The trial site consisted of four rows of three vines in a panel. Each of the seven treatments was replicated four times within the trial site with one replicate in each row.

4.1 Plant material and experimental design



Figure 2. Verdelho vines at EL:13



Figure 3. Verdelho vines at EL:26-27

4.2. Soil

The principal soil types in the block consisted of river valley soils, consisting of sandy loams over clay subsoils. Soil samples were taken prior to treatments. Four replicate soil core samples at 0-30cm and 30-60 cm depth were collected from each of four locations (northeast, northwest, southeast, southwest) across the block at the beginning of the trial. Soil samples were air dried at 60°C for 5 days, ground and sieved (2mm mesh size). Exchangeable cations (Na, K, Ca and Mg) were extracted in 1 M ammonium chloride at pH 7.0, and total boron was extracted in concentrated nitric acid. These minerals were analysed by using inductively

coupled plasma atomic emission spectroscopy (ICP-AES) at the Environmental and Analytical Laboratories at Charles Sturt University, Wagga Wagga. Soil electrical conductivity (EC1:5) (SevenCompact Cond meter, Mettler Toledo, Port Melbourne, Australia) and pH (Hanna precision pH meter Model pH 211, Hanna instruments, Melbourne, Australia) were also determined.

4.3. Assessment of reproductive performance

Reproductive performance of each vine was assessed with the method described in Dry et al. (2010). Briefly, two inflorescences per vine from the three vines of each treatment in each replicate row (24 bunches/treatment) were enclosed with fine mesh bags and secured with plastic ties before flowering [stage EL–16, Coombe (1995)]. After the completion of flowering (EL–26), the bags were removed, and flower caps were counted to estimate the number of flowers per inflorescence. Berry number was determined by counting the berries at veraison; seeded berries, seedless berries and live green ovaries (LGO) were counted separately for each bunch (Figure 1). Percent fruit set (the number of flowers that turned into berries, including seeded berries and seedless berries), Coulure index (the proportion of flowers that dropped off from a bunch without turning into a berry or a LGO), and Millerandage index (the proportion of LGOs and seedless berries relative to seeded berries) were calculated as described by Dry et al. (2010) by using the following formulas (Collins and Dry 2009).

$$\text{Fruitset (\%)} = \left(\frac{\text{total berry number per bunch}}{\text{number of flowers per inflorescence}}\right) \times 100$$

$$\text{'Coulure Index'} = 10 - \left\{\frac{(\text{no. of seeded berries per bunch} + \text{no. of seedless berries per bunch} + \text{no. of LGOs per bunch}) \times 10}{\text{no. of flowers per inflorescence}}\right\}$$

$$\text{'Millerandage Index'} = 10 - \left\{\frac{(\text{no. of seeded berries per bunch} \times 10)}{(\text{no. of seeded berries per bunch} + \text{no. of seedless berries per bunch} + \text{no. of LGOs per bunch})}\right\}$$

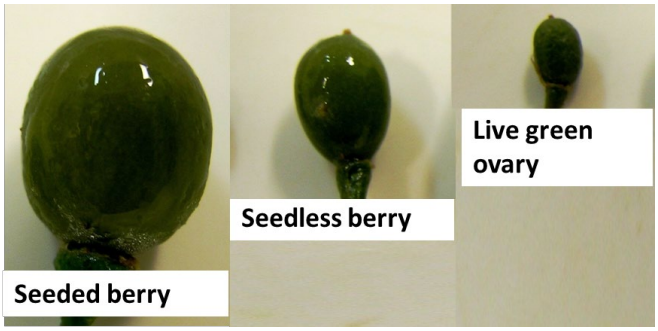


Figure 4. Seeded berry, which contained one to four seeds, seedless berry with no seeds, and live green ovary.

4.4. Tissue nutrient analysis

Petioles and inflorescences were sampled at three time points including: EL-15 (before the first spray treatments), EL-23 (two weeks after the first spray treatments and EL-28 (two weeks after the second spray treatments). For each replicate, two petioles and two inflorescences per vine from the basal bunch node position were collected from the three-vine panels. Petioles and inflorescences were washed in distilled water and blot dried using paper towels. From the inflorescences, whole flowers and whole berries were separated from the rachis. Petioles, flowers and berries were dried at 60°C for one week and ground using a tissue lyser (TissueLyser II, Qiagen, Chadstone, Victoria). 100 mg of ground tissue was analysed for calcium (Ca), magnesium (Mg), potassium (K), phosphorous (P), iron (Fe), manganese (Mn), boron (B), copper (Cu), zinc (Zn), sulfur (S), and sodium (Na) by inductively coupled plasma atomic emission spectroscopy (ICP-AES) at the Environmental and Analytical Laboratories at Charles Sturt University, Wagga Wagga.

4.5. Statistical methods

A two-way ANOVA, followed by Tukey's multiple comparison test was applied to the petiole and flower/berry nutrient concentration data to compare the main effects of treatments and sampling time points. Two-way ANOVA, followed by Tukey's multiple comparison test was applied to the soil characteristics to compare the main effects of soil depth and different locations of the trial block. One-way ANOVA followed by Tukey's multiple comparison test was applied to the reproductive performance parameters for comparing the treatment effects. *P* values <0.05 were considered significant. All analyses were performed using GraphPad Prism 8 (GraphPad Software, San Diego, California, USA).

5. Results and Discussion

5.1 Soil properties

Significant differences in soil characteristics were observed across the block (Table 1). Total B was significantly different across the four different locations of the trial block. The northeast location had the highest soil B concentration compared to the other locations. Similarly, there was a significant difference in total B between two layers of the soil. The top layer (0-30cm) of the soil had higher B concentration than the bottom layer (30-60 cm). pH, Ca and Mg concentrations were uniform across the block. Similarly soil EC was uniform across the block but was higher in the top layer of the soil. Soil K and Na concentrations were uniform across the block but both minerals were higher in the top layer of the soil.

Table 1. Soil properties at two different depths at four locations of the trial block.

Soil properties	Different locations in the trial block				***Depth means***	***P-values***
	Northeast	Northwest	Southwest	Southeast		
pH						
pH 0-30 cm	7.8	7.8	7.8	7.9	***7.8***	ns$^{(L*D)}$
pH -30-60 cm	7.9	7.6	7.9	8.0	***7.8***	ns$^{(D)}$
Location means	***7.9***	***7.7***	***7.9***	***8.0***		ns$^{(L)}$
EC (µS/cm)						
EC 0-30 cm	494	495	485	494	***491^{a}***	ns$^{(L*D)}$
EC 30-60 cm	455	465	460	452	***457^{b}***	0.0018$^{(D)}$
Location means	***475***	***480***	***472***	***473***		ns$^{(L)}$
Total B (mg/kg)						
Total B 0-30 cm	15.0	8.5	5.0	3.0	***7.9^{a}***	ns$^{(L*D)}$
Total B 30-60 cm	8.0	6.5	2.8	3.0	***5.1^{b}***	0.0203$^{(D)}$
Location means	***11.5^{a}***	***7.5^{b}***	***3.8^{c}***	***3.0^{c}***		0.0002$^{(L)}$
Ca (mg/kg)						
Ca 10-30 cm	1657	3280	2493	1503	***2233***	ns$^{(L*D)}$
Ca 30-60 cm	2470	3738	2980	4103	***3323***	ns$^{(D)}$
Location means	***2063***	***3509***	***2736***	***2803***		ns$^{(L)}$
K mg/kg						
K 0-30 cm	478	577	333	514	***475^{a}***	ns$^{(L*D)}$
K 30-60 cm	261	294	233	295	***270^{b}***	0.0003$^{(D)}$
Location means	***370***	***436***	***283***	***405***		ns$^{(L)}$
Mg mg/kg						
Mg 0-30 cm	161	201	163	126	***163***	ns$^{(L*D)}$
Mg 30-60 cm	166	195	187	180	***182***	ns$^{(D)}$
Location means	***164***	***198***	***175***	***153***		ns$^{(L)}$
Na mg/kg						
Na 0-30 cm	40	64	38	46	***47^{a}***	ns$^{(L*D)}$
Na 30-60	31	33	30	35	***32^{b}***	0.0011$^{(D)}$
Location means	***36***	***48***	***34***	***40***		ns$^{(L)}$

Two way - ANOVA of depth (D) and location (L) effects on soil properties. *P* values <0.05 were considered significant. Different superscript letters show significant difference.

5.2 Effect of Boron spray on tissue nutrient concentrations

There was no significant difference in the inflorescence nutrient concentration between B spray treatments, however there was a significant effect of sampling time on inflorescence nutrient concentration (Tables 2 & 3). B and Fe were highest at EL-23 and lowest at EL-28. Cu, Mn and Zn were highest at EL-15 and lowest at EL-28. Macro nutrient concentrations were also significantly different between the sampling time points. K, Mg, Ca, P, S and Na were the lowest at EL-28. Mg, P and S were the highest at EL-15 while K, Ca and Na were higher at both EL-15 and EL-23 compared to EL-28. In the inflorescences, the interactions of

sampling time point X treatment were significant for the concentrations of mineral elements B, K, Ca and S, indicating that there was a combined effect of sampling time point and treatment on the concentration of these minerals.

Table 2. Micronutrient concentration in the inflorescences

Micro nutrients/three time points	Treatments							*Sampling time means*	*P* value
	control	0.025% B at EL-16	0. 05% B at EL-16	0.1% B at EL-16	0.025% B at EL-23	0.05% B at EL-23	0.1% B at EL-23		
B mg/kg									
EL-15	37	38	42	40	44	44	44	***41^{b}***	0.0322$^{(S*T)}$
EL-23	54	54	52	56	44	44	44	***50^{a}***	<0.0001$^{(S)}$
EL-28	34	33	32	29	27	32	32	***31^{c}***	ns$^{(T)}$
Treatment means	***42***	***42***	***42***	***42***	***38***	***40***	***40***		
Cu mg/kg									
EL-15	40	44	42	43	42	41	37	***41^{a}***	ns$^{(S*T)}$
EL-23	33	34	34	36	34	36	39	***35^{b}***	<0.0001$^{(S)}$
EL-28	15	15	15	16	15	15	16	***15^{c}***	ns$^{(T)}$
Treatment means	***29***	***31***	***31***	***31***	***30***	***31***	***30***		
Fe mg/kg									
EL-15	75	80	78	78	79	79	59	***75^{b}***	ns$^{(S*T)}$
EL-23	137	135	144	137	147	144	149	***141^{a}***	<0.0001$^{(S)}$
EL-28	35	35	35	35	36	38	38	***35^{c}***	ns$^{(T)}$
Treatment means	***82***	***83***	***86***	***83***	***87***	***87***	***82***		
Mn mg/kg									
EL-15	176	214	190	223	233	211	169	***202^{a}***	ns$^{(S*T)}$
EL-23	145	167	165	170	169	158	148	***160^{b}***	<0.0001$^{(S)}$
EL-28	62	66	65	72	73	73	73	***69^{c}***	ns$^{(T)}$
Treatment means	***128***	***149***	***140***	***155***	***158***	***147***	***130***		
Zn mg/kg									
EL-15	58	62	59	64	63	63	55	***60^{a}***	ns$^{(S*T)}$
EL-23	37	42	39	42	40	38	40	***39^{b}***	<0.0001$^{(S)}$
EL-28	17	18	17	17	18	18	19	***17^{c}***	ns$^{(T)}$
Treatment means	***38***	***41***	***38***	***41***	***40***	***39***	***38***		

Two way - ANOVA of the influence of three sampling time points EL-15, EL-23 and EL-28 (S) and seven treatments (T) on the inflorescence nutrients. *P* values <0.05 were considered significant. Different superscript letters show significant difference.

Table 3. Macronutrient concentration in the inflorescences

Macro nutrients/three time points	Treatments: control	0.025% B at EL-16	0. 05% B at EL_16	0.1% B at EL-16	0.025% B at EL- 23	0.05% B at EL- 23	0.1% B at EL-23	*Sampling time means*	*P* value
K mg/kg									
EL-15	17669	18377	18186	18547	17996	18417	13273	***17495***^{a}	0.0467$^{(S*T)}$
EL-23	16627	17384	18031	19213	18276	18745	18867	***18163***^{a}	<0.0001$^{(S)}$
EL-28	9366	9223	9604	9538	9642	9576	9508	***9494***^{b}	ns$^{(T)}$
Treatment means	***14554***	***14995***	***15274***	***15766***	***15305***	***15579***	***13883***		
Mg mg/kg									
EL-15	3170	3312	3214	3240	3399	3347	2469	***3164***^{a}	ns$^{(S*T)}$
EL-23	2733	2761	2658	2934	2839	3021	2932	***2840***^{b}	<0.0001$^{(S)}$
EL-28	1417	1403	1370	1399	1415	1484	1481	***1424***^{c}	ns$^{(T)}$
Treatment means	***2440***	***2492***	***2414***	***2524***	***2551***	***2617***	***2294***		
Ca mg/kg									
EL-15	18671a	19626a	18504a	19361a	20027a	19906a	12835b	***18419***^{a}	0.0007$^{(S*T)}$
EL-23	15363	16197	16539	18349	18194	18598	18727	***17424***^{a}	<0.0001$^{(S)}$
EL-28	8828	8980	8691	9130	9039	9235	9427	***9047***^{b}	ns$^{(T)}$
Treatment means	***14287***	***14934***	***14578***	***15613***	***15753***	***15913***	***13663***		
Na mg/kg									
EL-15	106.4	108.5	103.5	104.2	95.98	103.9	66.05	***98***^{a}	ns$^{(S*T)}$
EL-23	79.43	81.13	89.2	85.45	90.55	87.38	90.58	***86***^{a}	<0.0001$^{(S)}$
EL-28	15.3	10.43	12.2	12.25	14.55	15.35	17	***13***^{b}	ns$^{(T)}$
Treatment means	***67.03***	***66.67***	***68.29***	***67.3***	***67.03***	***68.86***	***57.88***		
P mg/kg									
EL-15	4867	5155	5287	5459	5281	5207	4331	***5084***^{a}	ns$^{(S*T)}$
EL-23	3682	3759	3720	3807	3594	3660	3856	***3725***^{b}	<0.0001$^{(S)}$
EL-28	2151	2190	2193	2231	2202	2214	2179	***2194***^{c}	ns$^{(T)}$
Treatment means	***3567***	***3701***	***3733***	***3832***	***3692***	***3693***	***3455***		
S mg/kg									
EL-15	4477	4604	4604	4619	4543	4466	3212	***4361***^{a}	0.0005$^{(S*T)}$
EL-23	3051	3251	3359	3578	3514	3597	3752	***3443***^{b}	<0.0001$^{(S)}$
EL-28	1818	1859	1867	1856	1897	1906	1917	***1874***^{c}	ns$^{(T)}$
Treatment means	***3115***	***3238***	***3277***	***3351***	***3318***	***3323***	***2961***		

Two way - ANOVA of the influence of three sampling time points EL-15, EL-23 and EL-28 (S) and seven treatments (T) on the inflorescence nutrients. *P* values <0.05 were considered significant. Different superscript letters show significant difference.

In petioles, the concentration of all the nutrients including micro and macro nutrients were significantly different between the three sampling time points (Tables 4&5). This is consistent with the previous reports on nutrient accumulation in various tissues during various growth stages in grapevines (Baby et al. 2021). B concentration was the highest at EL-15. Cu, Mn and Zn were highest at EL-28 and Cu was lowest at EL-23 and Zn was lowest at EL-15. Fe

was higher at EL-15 and EL-23 compared to EL-28. Ca and Mg were highest at the EL-28 and Mg was lowest at EL-15. Ca concentration was similar at EL-15 and EL-23. P and Na concentrations were highest at EL-15 and lowest at EL-28. The sampling time point x treatments interaction was significant for the following mineral element concentrations in petioles: B, Mn, Zn, Ca, K, Mg, Na, P and S, indicating that there was a combined effect of sampling time point and treatment on the concentration of these minerals. There were some differences in the concentration of most of the minerals in the petioles between each B spray treatment, including in petiole B concentration, but that cannot be attributed to the treatment effect as there was no correlation between the concentrations of these nutrients in petioles with petiole B concentration (data not shown). It is reasonable to suggest that the difference in the nutrient concentrations between the treatment group might be due to the soil nutrient effect or from the nutrient reserves of the sampled vine itself.

In our study, the concentrations of Cu, Mn, Zn, K and P at EL-23 were at adequate levels in the petioles according to the grapevine petiole analysis standards at flowering, as per Reuter and Robinson (1997) (Table 6). However, Fe, Mg and Ca were lower than the adequate level across the trial block. Petiole B concentration at sampling time EL-23 was lower than the adequate level for B spray treatments 0.1% B at EL-16, 0.025 % B at EL-23, 0.05% B at EL-23 and 0.1% B at EL-23, while it was adequate, however, for the control and other B spray treatments 0.025% B at EL-16 and 0.05% B at EL-16).

Table 4. Micronutrient concentration in the petioles

	Treatments								
Macro nutrients/three time points	control	0.025% B at EL-16	0. 05% B at EL-16	0.1% B at EL-16	0.025% B at EL-23	0.05% B at EL-23	0.1% B at EL-23	***Sampling time means***	***P* value**
B mg/kg									
EL-15	78	58	60	66	60	48	48	***60^{a}***	<0.0001$^{(S*T)}$
EL-23	37	37	35	25	20	17	24	***27^{c}***	<0.0001$^{(S)}$
EL-28	42	33	41	38	30	42	34	***37^{b}***	<0.0001$^{(T)}$
Treatment means	***52^{a}***	***42^{b}***	***45^{b}***	***42^{b}***	***36^{c}***	***35^{c}***	***35^{c}***		
Cu mg/kg									
EL-15	9	9	8	8	8	9	10	**9^{b}**	ns$^{(S*T)}$
EL-23	6	7	8	7	6	7	9	**7^{c}**	<0.0001$^{(S)}$
EL-28	11	11	12	12	11	11	11	**11^{a}**	0.0344$^{(T)}$
Treatment means	**9^{ab}**	**9^{ab}**	**9^{ab}**	**9^{ab}**	**8^{b}**	**9^{ab}**	**10^{a}**		
Fe mg/kg									
EL-15	18	16	12	12	14	11	17	**14^{a}**	ns$^{(S*T)}$
EL-23	14	12	13	10	14	13	13	**13^{a}**	<0.0001$^{(S)}$

EL-28	11	7	5	6	5	6	5	**6^{b}**	0.0108$^{(T)}$
Treatment means	**14^{a}**	**11^{ab}**	**10^{bc}**	**8^{cd}**	**7^{cd}**	**6^{d}**	**7^{cd}**		
Mn mg/kg									
EL-15	79	93	89	100	102	100	94	**94^{b}**	0.0006$^{(S*T)}$
EL-23	88	112	119	88	83	65	60	**88^{b}**	<0.0001$^{(S)}$
EL-28	104	128	108	124	128	114	119	**118^{a}**	0.0076$^{(T)}$
Treatment means	**91^{b}**	**111^{a}**	**106^{ab}**	**104^{ab}**	**105^{ab}**	**93^{ab}**	**91^{b}**		
Zn mg/kg									
EL-15	28	31	29	32	31	35	31	**31^{c}**	<0.0001$^{(S*T)}$
EL-23	43	44	49	35	30	26	34	**37^{b}**	<0.0001$^{(S)}$
EL-28	41	44	40	46	44	40	41	**42^{a}**	ns$^{(T)}$
Treatment means	***37***	***40***	***40***	***38***	***35***	***34***	***35***		

Two way - ANOVA of the influence of three sampling time points EL-15, EL-23 and EL-28 (S) and seven treatments (T) on the petiole nutrients. *P* values <0.05 were considered significant. Different superscript letters show significant difference.

Table 5. Macronutrient concentration in the petioles

Macro nutrients/three time points	Treatments							***Sampling time means***	***P* value**
	control	0.025% B at EL-16	0. 05% B at EL-16	0.1% B at EL-16	0.025% B at EL-23	0.05% B at EL-23	0.1% B at EL-23		
Ca mg/kg									
EL-15	10529	11624	10953	10945	10939	10959	10977	***10990^{b}***	<0.0001$^{(S*T)}$
EL-23	14115	15588	15785	12229	10344	8509	7385	***11993^{b}***	<0.0001$^{(S)}$
EL-28	18300	18971	18088	18978	19602	18636	18661	***18748^{a}***	0.0006$^{(T)}$
Treatment means	***14315^{ab}***	***15394^{a}***	***14942^{a}***	***14051^{ab}***	***13629^{ab}***	***12701^{b}***	***12341^{b}***		
K mg/kg									
EL-15	30980	29911	35396	30250	31753	30814	24278	***30483^{b}***	<0.0001$^{(S*T)}$
EL-23	23768	35003	42509	38793	35223	39590	47885	***37539^{a}***	<0.0001$^{(S)}$
EL-28	14144	13441	12603	14094	13767	13243	13206	***13500^{c}***	0.0138$^{(T)}$
Treatment means	***22964^{b}***	***26119^{ab}***	***30169^{a}***	***27712^{ab}***	***26914^{ab}***	***27882^{ab}***	***28456^{ab}***		
Mg mg/kg									
EL-15	2442	2335	2173	2290	2488	2302	2148	***2311^{c}***	<0.0001$^{(S*T)}$
EL-23	4080	3878	3724	2819	2265	1942	1854	***2937^{b}***	<0.0001$^{(S)}$
EL-28	3574	3638	3324	3434	3559	3616	3411	***3508^{a}***	<0.0001$^{(T)}$
Treatment means	***3365^{a}***	***3284^{ab}***	***3074^{ab}***	***2848^{bc}***	***2771^{bcd}***	***2620^{d}***	***2471^{d}***		
Na mg/kg									
EL-15	770	302	313	342	224	296	216	***352^{a}***	<0.0001$^{(S*T)}$
EL-23	156	243	236	280	207	159	171	***207^{b}***	<0.0001$^{(S)}$
EL-28	119	114	98	133	109	111	115	***114^{c}***	<0.0001$^{(T)}$
Treatment means	***348^{a}***	***220^{bc}***	***215^{bc}***	***251^{b}***	***180^{c}***	***188^{bc}***	***167^{c}***		
P mg/kg									
EL-15	4285	4617	4497	4862	5004	4768	4800	***4690^{a}***	0.0148$^{(S*T)}$

EL-23	3376	3763	3992	3118	2329	2037	4221	***3262^{b}***	<0.0001$^{(S)}$
EL-28	2293	2388	2178	2855	2389	2438	2809	***2478^{c}***	ns$^{(T)}$
Treatment means	***3318***	***3589***	***3555***	***3612***	***3241***	***3081***	***3943***		
S mg/kg									
EL-15	1406	1484	1395	1297	1344	1470	1421	***1402***	0.0027$^{(S*T)}$
EL-23	1352	1490	1498	1172	863	763	1746	***1269***	ns$^{(S)}$
EL-28	1217	1275	1185	1369	1222	1278	1294	***1263***	0.0105$^{(T)}$
Treatment means	***1325^{ab}***	***1416^{ab}***	***1359^{ab}***	***1280^{ab}***	***1143^{b}***	***1171^{b}***	***1487^{a}***		

Two way - ANOVA of the influence of three sampling time points EL-15, EL-23 and EL-28 (S) and seven treatments (T) on the petiole nutrients. *P* values <0.05 were considered significant. Different superscript letters show significant difference.

Table 6. Grapevine petiole analysis standards at flowering adapted from Reuter and Robinson (1997)

Element	Adequate
Nitrogen (mg/kg)	8000-11000
Phosphorous (mg/kg)	2500-5000
Potassium (mg/kg)	18000-30000
Calcium (mg/kg)	12000-25000
Magnesium (mg/kg)	>4000
Sodium (mg/kg)	1000-3000
Manganese (mg/kg)	30-60
Zinc (mg/kg)	>26
Copper (mg/kg)	6-11
Boron (mg/kg)	35-70
Iron (mg/kg)	>30

5.3 Effect of Boron spray on fruit set and hen and chicken berries

Effect of B spray treatments was not obvious on fruit set %, coulure index or millerandage index (Figure 3). This can be due to several factors: the variability in B concentration in the soil, no increase in B concentration in the tissues consistent with the treatments, deficiencies of other nutrients such as Fe, Ca and Mg in the vines during floral development and flowering, and hail damage as evidenced by torn leaves.

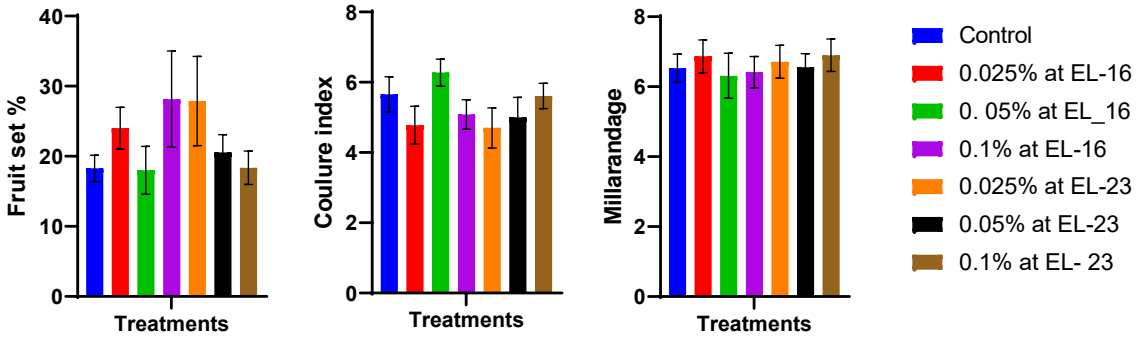


Figure 3. Influence of B spray treatments on fruit set percentage, coulure index and millerandage index. Vertical bars represent mean ± s.d (n = 24 replicates). One-way ANOVA was performed for each of fruit set, coulure and millerandage indices. There were no significant differences between treatments at *P* values <0.05.

6. Conclusion and recommendations

This project investigated the effect of Boron foliar application on fruit-set of grapevines in the Riverland region. No significant treatment effect on the reproductive performance of grapevines was observed. Soil composition analysis showed variability across the trial site especially in total B concentration, indicating that the soil B had more influence on the observed fruit set parameters than the foliar B applications. Additionally, the foliar B application didn't increase B concentrations either in petioles or in inflorescences in alignment with the treatments, suggesting that the B sprays may not have been absorbed efficiently into the tissues. Adding an adjuvant in the sprays might be advantageous to ensure B is absorbed efficiently, however further studies are required to test this. An alternative interpretation is that where B was absorbed, it translocated from the leaves to other growing tissues or to the nutrient reserve storage within the perennial components of the vine, as opposed to the reproductive tissues. Unexpected environmental events such as storms, along with the complexities of understanding nutrient dynamics in a large perennial system, points towards the requirement for multi-season trials. It is not unusual that several seasons of repetitive nutrition applications before a response in physiological performance on growth is observed (Schreiner & Skinkis, 2013). The apparent Fe, Mg and Ca deficiencies may have further interfered with fruit set (Ashley 2011). This may have been avoided if a pre-season of adequate nutrition was applied to the vines. The lag-time between sample collection, processing, analyses, and data interpretation also favours a minimum of two seasons, permitting a more comprehensive understanding of the block in the first season and the application of tailored treatments in the second season.

7. Extension

Despite the absence of clear treatment effects on the fruit set parameters of grapevines in this study, nutrition is undoubtedly an important cornerstone of grapevine management. The role of nutrition on reproductive development, along with its complexities, will be communicated with the industry partners in the Riverland Region, SA. Recommendations for better nutrition management strategies especially for improving fruit set and yield of grapevines in sandy soils will be communicated through an industry and scientific article.

8. Researchers benefit and feedback

This project provided me with a great opportunity to collaborate with the industry partners in Riverland Region, in South Australia. There were challenges of not being able to visit the trial site and set-up the field trial by myself due to COVID-19 travel restrictions. However, regular online meetings and communication with the industry partners helped me to understand the region-specific challenges for vineyard nutrition management, and I was able to design and coordinate the trial. Working in this project improved my project management skills, communication skills and my confidence in managing research projects.

This trial showed a limitation to draw conclusions based on a single season study, especially for nutrition trials because of the complexity of the topic. As the reproductive development of grapevines spans over two seasons and interact with many factors such as soil and weather conditions, a longer study including multiple seasons is recommended for nutrition trials.

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Supplementary Figure 1. Monthly rainfall and evapotranspiration data during the growing season.

