SUPPLEMENTARY MATERIALS FOR

Distribution of 3-Isobutyl-2-methoxypyrazine across rachis components of *Vitis vinifera* Shiraz and Cabernet Sauvignon

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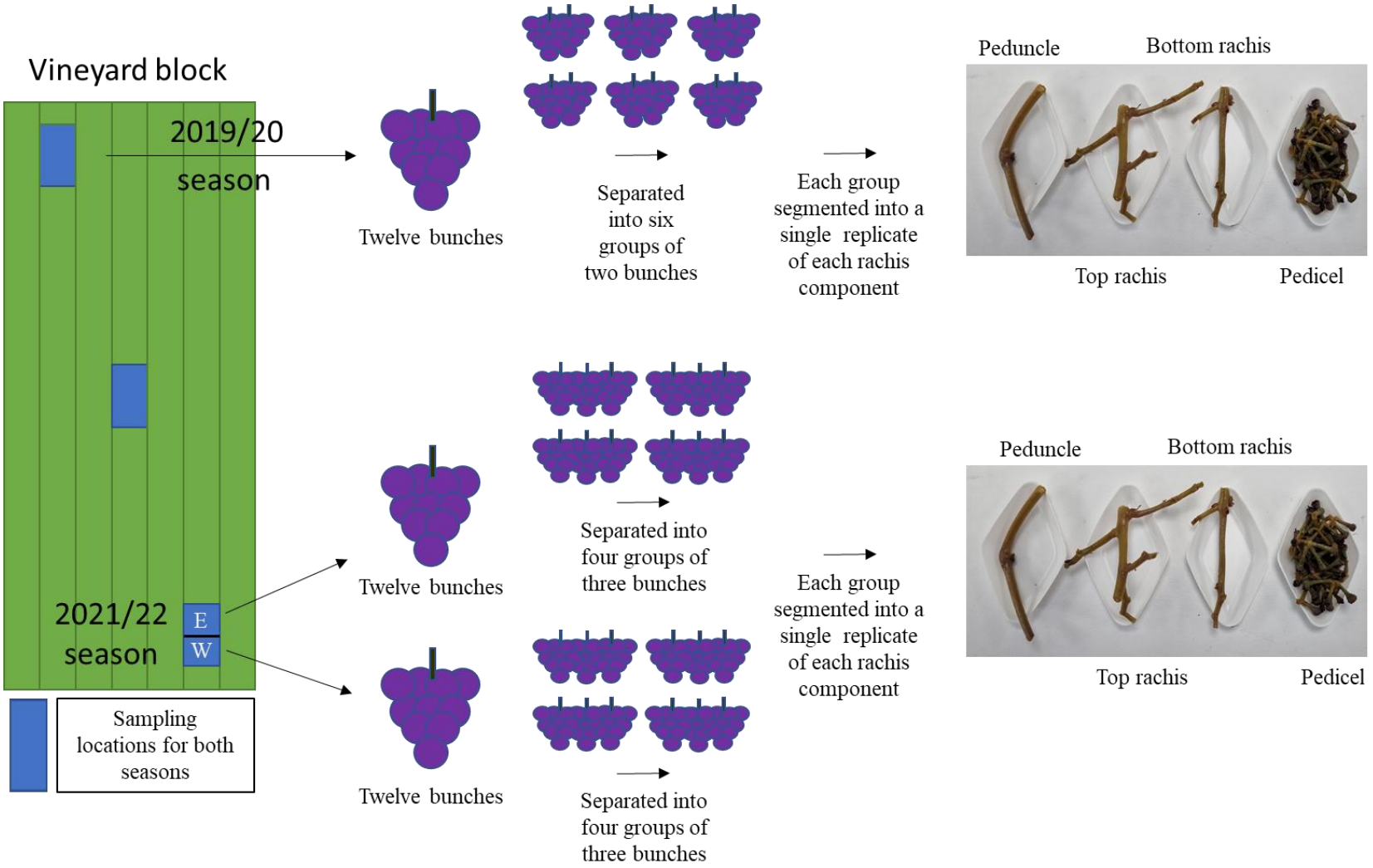

Figure S1. Overview of sampling methodology for Shiraz grapes from the Barossa Valley in the 2019/20 and 2020/21 seasons. At veraison and maturity in the 2019/20 growing season, twelve bunches were sampled from each location and separated into six groups of two bunches before being segmented into rachis components (peduncle, top rachis, bottom rachis, pedicel). At veraison and maturity in the 2020/21 growing season, twelve bunches were taken from each east (E) and west (W) sub-region in every sampling location before being separated into four groups of three bunches and segmented into rachis components. Note for both growing seasons, sample numbers were doubled at flowering due the small size of rachis at this phenological stage.

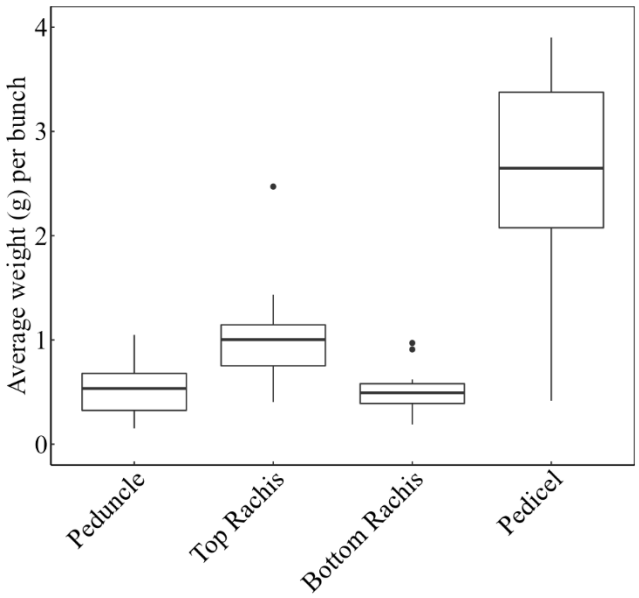

Figure S2. Average weight (g) of rachis components from Shiraz bunches (n = 26) sampled at harvest in 2022 from Wrattonbully, South Australia.

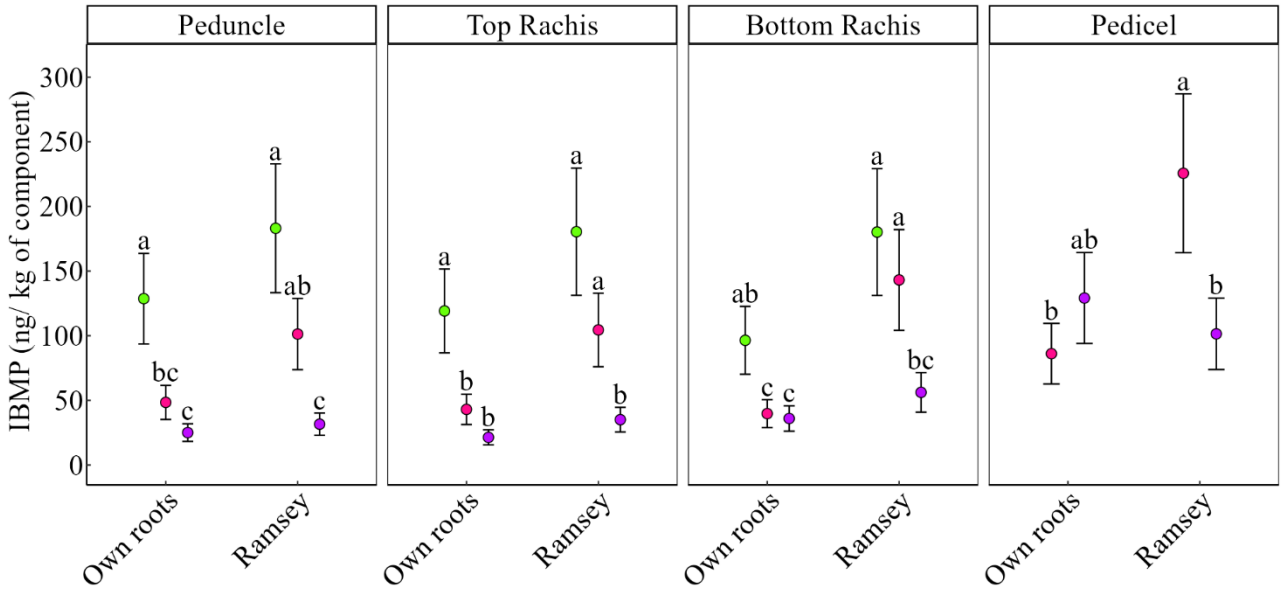

Figure S3. Estimated marginal means of IBMP (ng/kg of component ± SE) in the peduncle, top rachis, bottom rachis, and pedicel from Shiraz rachis sampled during the 2019/20 and 2021/22 vintages, considering the interaction effect between rootstock (own roots and Ramsey) and berry maturity (flowering (• green), veraison (• red), and harvest (• purple)). Bars sharing the same letter within a component are not significantly different (linear mixed model, α = 0.05, Bonferroni-adjusted). Note pedicel material was not sampled at flowering.

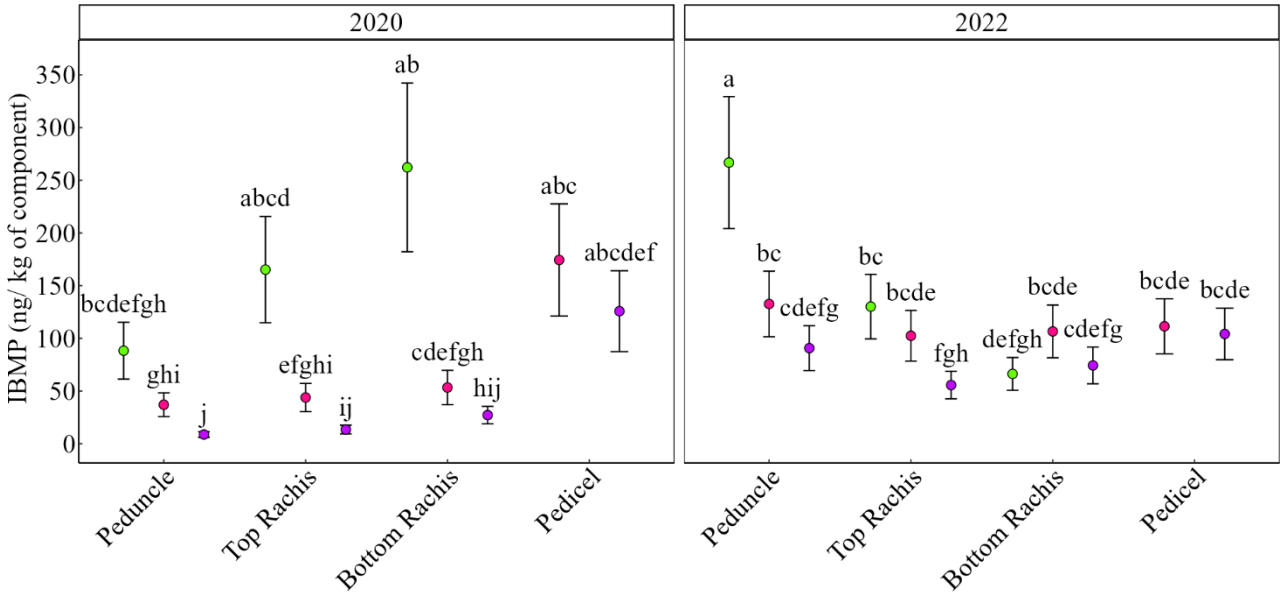

Figure S4. Estimated marginal means of IBMP (ng/kg of component ± SE) in the peduncle, top rachis, bottom rachis, and pedicel of Shiraz rachis sampled during the 2019/20 and 2021/22 vintages at (flowering (• green), veraison (• red), and harvest (• purple)) considering a three-way interaction effect between component, vintage, and berry maturity. Bars sharing the same letter between the plots are not significantly different (linear mixed model, $\alpha = 0.05$, Bonferroni-adjusted). Note pedicel material was not sampled at flowering.

Table S1. Overview of rootstock, average degrees Brix at harvest, average winter rainfall, and climate class as determined according to the Huglin index for each viticultural region and season.

Region	Year	Rootstock	Average °Brix at harvest	Average winter rainfall (mm)[†]	Climate class (Huglin index)
Barossa Valley	2019/20	Own roots	28.2	149	Temperate warm (2379)
		Ramsey	26.0		
	2021/22	Own roots	29.8	151	Temperate warm (2252)
		Ramsey	27.3		
Coonawarra	2019/20	110 Richter	24.5	234	Temperate (1993)

[†] Average winter rainfall (June – August) was obtained using data from Australian BOM weather stations.

Table S2. Average leaf area index (LAI) values for Shiraz vines grown in the Barossa Valley over the 2020/21 season.

Rootstock	Row	Sub-block[†]	LAI[‡]
Own roots	42	W	2.40
		E	3.20
	45	W	3.19
		E	3.97
	48	W	3.07
		E	3.92
Ramsey	53	W	2.85
		E	2.31
	56	W	2.07
		E	2.58
	59	W	1.50
		E	2.01

[†] Each sub-block consisting of six vines was divided into east (E) and west (W) regions. [‡] LAI was recorded on the 17th of January 2022 with a LICOR LAI-2200C Plant Canopy Analyser.

Table S3. Average concentration of 3-isobutyl-2-methoxypyrazine (IBMP) (ng/kg ± SD) in different rachis components of Shiraz sampled from the Barossa Valley at flowering (80% cap fall), 50% veraison, and harvest in the 2019/20 and 2021/22 growing seasons.

Component	Rootstock	Flowering		Veraison		Harvest	
		2019/20	*2021/22*	*2019/20*	*2021/22*	*2019/20*	*2021/22*
Peduncle	Own roots	70.3 ± 17.1	263 ± 105	47.8 ± 37.7	127 ± 85.6	20.0 ± 19.8	90.9 ± 56.0
	Ramsey	116 ± 17.0	305 ± 87	57.2 ± 7.47	207 ± 103	31.5 ± 32.7	141 ± 75.0
Top Rachis	Own roots	150 ± 45.1	107 ± 42.2	56.7 ± 47.4	96.4 ± 65.3	34.5 ± 42.2	56.7 ± 59.2
	Ramsey	193 ± 34.9	180 ± 68.1	90.0 ± 52.1	179 ± 102	50.4 ± 53.3	126 ± 78.3
Bottom Rachis	Own roots	207 ± 72	50.3 ± 17.6	49.1 ± 35.2	93.3 ± 60.9	60.8 ± 60.9	57.4 ± 57.3
	Ramsey	359 ± 64.4	95.7 ± 28.9	154 ± 85.4	200 ± 118	121 ± 122	181 ± 114
Pedicel	Own roots	N/A[†]	N/A	169 ± 121	82.9 ± 39.8	231 ± 73.8	91.0 ± 47.8
	Ramsey	N/A	N/A	302 ± 34.7	187 ± 118	262 ± 253	159 ± 84.1

[†] N/A, samples were not collected at this time point

Table S4. Average concentration (ng/kg ± SD) of 3-isobutyl-2-methoxypyrazine (IBMP), 3-isopropyl-2-methoxypyrazine (IPMP), and 3-*sec*-butyl-2-methoxypyrazine (SBMP) in different rachis components of Shiraz sampled from the Barossa Valley at harvest in 2022 from control (ambient light) and box (light excluded) experiments.

Analyte	Rootstock	Peduncle		Top Rachis		Bottom Rachis		Pedicel	
		Box	Control	Box	Control	Box	Control	Box	Control
IBMP	Own roots	416 ± 219	91.0 ± 56.0	666 ± 522	56.7 ± 59.2	878 ± 625	57.4 ± 57.3	811 ± 410	91.0 ± 47.8
	Ramsey	614 ± 252	141 ± 75.0	1310 ± 493	126 ± 78.3	2320 ± 1160	181 ± 84.1	1260 ± 578	159 ± 84.1
IPMP	Own roots	nq[†]	nq	nq	nq	nq	nq	nq	nq
	Ramsey	61.3 ± 53.4	nq	64.6 ± 64.5	nq	90.5 ± 95.7	nq	72.8 ± 33.6	nq
SBMP	Own roots	nq	nq	nq	nq	nq	nq	nq	nq
	Ramsey	40.7 ± 31.8	nq	41.9 ± 34.2	nq	44.2 ± 37.7	nq	46.7 ± 16.8	nq

[†] nq, samples were below level of quantitation (LOQ). The LOQ values (ng/kg) were 0.44 for IBMP, 0.37 for IPMP, and 0.48 for SBMP.

Table S5. Average concentration of 3-isobutyl-2-methoxypyrazine (IBMP) (mean ± SD) in rachis components at 80% veraison and harvest from Reynella and SA125 Cabernet Sauvignon clones grown in the Coonawarra over the 2019/20 growing season.

Component	Clone	Veraison	Harvest
Peduncle	SA125	54.9 ± 12.7	35.8 ± 6.99
	Reynella	70.9 ± 22.2	63.4 ± 18.3
Top rachis	SA125	175 ± 32.3	91.8 ± 22.4
	Reynella	148 ± 33.5	111 ± 26.9
Bottom rachis	SA125	211 ± 16.6	89.7 ± 37.1
	Reynella	182 ± 72.4	133 ± 25.0
Pedicel	SA125	307 ± 66.7	174 ± 34
	Reynella	259 ± 84.9	244 ± 119