Botryosphaeria canker of grapevines in California

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Perennial cankers and consequent grapevine dieback are a major problem in California vineyards. Grapevine dieback symptoms include dead spurs and cordon due to canker formation in the vascular tissue. For many years, *Eutypa lata* (Pers.:Fr.) Tul. & C. Tul., the causal agent of Eutypa dieback, has been thought to be the most important canker and dieback causing agent of grapevines not only in California but worldwide. However, recent studies have also identified *Botryosphaeria* species as important grapevine pathogens capable to cause cankers and other dieback symptoms such as wood streaking, shoot dieback, cane bleaching, bud necrosis and graft failure in all major viticulture regions throughout the world (Larignon et al. 2001; Phillips, 2002; van Niekerk et al. 2004; Taylor et al. 2005). Botryosphaeria canker was first reported in California in 1987, and was shown to be prevalent in the warm growing areas of southern California (Leavitt, 1987). However, Botryosphaeria canker has remained overlooked in California due to the similarity of the symptoms to those caused by *E. lata*. Canker diseases decrease vineyard longevity, reduce yields and increase production costs as a result of cultural and chemical preventive measures as well as removing diseased/dead wood from the vine after infection. In California, the overall loss in net income for wine grape caused by Eutypa dieback and Botryosphaeria canker disease was estimated to be over $260 million per annum (Siebert, 2001).

In order to know the current status of *Botryosphaeria* species involved in grapevine dieback in California, as well as evaluate their incidence and importance in the state, a three year field survey study was conducted throughout the main grape-growing areas of California. Over 1,700 perennial cankers from spurs, cordons and/or trunks were collected from the predominant wine, table and raisin grape cultivars. Fungal isolation showed *Botryosphaeria* species as the most prevalent fungi associated with cankers in California (Table 1). Only in Napa, Solano, Stanislaus and San Joaquin, was the incidence of *Eutypa lata* significantly higher than *Botryosphaeria* (Table 1). Based on morphological characters and partial sequence analysis of ITS and beta-tubulin genes, 9 *Botryosphaeria* species were identified from cankers of grapevines in California, including *B. australis*, *B. dothidea*, *B. lutea*, *B. obtusa*, *B. parva*, *B. rhodina*, *B. sarmentorum*, *B. stevensii* and *B. viticola* (Úrbez-Torres et al 2006; Úrbez-Torres et al, 2007).

Pathogenicity and epidemiology of *Botryosphaeria* spp. in vines have been the subject of confusion for many years. *Botryosphaeria* spp. have been largely considered saprophytes or secondary colonizers in grapevines (Phillips, 2002). In order to determine the importance of Botryosphaeria species in grapevine health in California, pathogenicity tests were conducted in mature vines. Dormant grapevines cv. Chardonnay were pruned to 4-5 buds. Fresh pruning wounds were inoculated using mycelium plugs of the nine *Botryosphaeria* species found in California. Internal lesions were measured 12 months after inoculation. Preliminary results verified the pathogenicity of the nine *Botryosphaeria* species, however, virulence varied among species. Isolates of *B. rhodina* were the most virulent based on extent of spread in the wood while isolates of *B. obtusa* were the least virulent. More studies designed to characterize the virulence of *Botryosphaeria* species on grapevines in California are currently underway.

Up to this date, no information was available about the epidemiology and mode of inoculum dispersal for *Botryosphaeria* species in California. Consequently, spore traps are being using to determine what environmental conditions favor the release of *Botryosphaeria* inoculum. Spore traps were placed in selected vineyards in five different locations, including Napa, Colusa, San Joaquin, San Luis Obispo and Riverside Counties. Spores are being trapped using microscope slides covered with a thin layer of Vaseline and placed on grapevine cordons. Preliminary results have shown that *Botryosphaeria* spores were mainly trapped following rainfall events or overhead sprinkler irrigation. Interestingly, *Botryosphaeria* spores were also trapped in some locations in the absence of rainfall or irrigation suggesting that other environmental factors may contribute to spore release.

These findings confirm Botryosphaeria species as the most prevalent fungi associated with grapevine cankers in California. Nine *Botryosphaeria* species have been identified in California, and *in vivo* and *in vitro* pathogenicity tests have revealed all 9 *Botryosphaeria* species to be important pathogens of wine

and table-grape cultivars in California. These findings indicate the importance of *Botryosphaeria* species as grapevine pathogens and that the role of these fungi in grapevine health needs to be more carefully considered.

References

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Table 1. Incidence of *Botryosphaeria* spp. in the main grapevine-production areas of California

Grapevine region	Vineyards[b]	No. (%) *Bot.*[c]	Total[d]	Number (%) of cankers yielding[a]				
				A[e]	B[f]	A + B[g]	*E. lata*	*P. viticola*
North Coast								
Mendocino Co.	8	8 (100)	91	37 (41)	11 (12)	48 (53)	11 (12)	0
Napa Co.	11	10 (91)	113	30 (26)	13 (11)	43 (38)	54 (48)	0
Sonoma Co.	17	17 (100)	192	85 (44)	13 (7)	98 (51)	41 (21)	0
Total	36	35 (97)	396	152 (38)	37 (9)	189 (48)	106 (27)	0
Central Coast								
Santa Clara Co.	2	2 (100)	23	10 (43%)	7 (30)	17 (74)	4 (17)	0
San Benito Co.	6	6 (100)	65	49 (75%)	9 (14)	58 (89)	6 (9)	1 (1)
Monterey Co.	6	6 (100)	65	44 (68)	4 (6)	48 (74)	13 (20)	0
San Luis Obispo Co.	8	8 (100)	80	37 (46)	7 (9)	44 (55)	15 (19)	0
Total	22	22 (100)	233	140 (60)	27 (12)	167 (72)	38 (16)	1 (0.5)
South Coast								
Santa Barbara Co.	7	7 (100)	81	58 (72)	16 (20)	74 (91)	1 (1)	3 (4)
Mountain Counties								
El Dorado Co.	2	2 (100)	20	9 (45)	2 (10)	11 (55)	2 (10)	0
Amador Co.	3	3 (100)	32	13 (41)	4 (12)	17 (53)	1 (3)	1 (3)
Total	5	5 (100)	52	22 (42)	6 (11)	28 (54)	3 (6)	1 (2)
Sacramento Valley								
Yolo Co.	4	4 (100)	42	10 (24)	11 (26)	21 (50)	9 (21)	0
Solano Co.	5	5 (100)	53	2 (4)	11 (21)	13 (24)	39 (74)	0
Sacramento Co.	7	7 (100)	78	35 (45)	28 (36)	63 (81)	11 (14)	3 (4)
Total	16	16 (100)	173	47 (27)	50 (29)	97 (56)	59 (34)	3 (2)
Northern San Joaquin Valley								
San Joaquin Co.	7	5 (71)	65	4 (6)	6 (9)	10 (15)	34 (52)	0
Stanislaus Co.	5	5 (100)	62	20 (32)	6 (10)	26 (42)	29 (47)	0
Merced	10	10 (100)	116	63 (54)	21 (18)	84 (72)	4 (3)	1 (1)
Total	22	20 (91)	243	87 (36)	33 (13)	120 (49)	67 (27)	1 (0.5)
Southern San Joaquin Valley								
Madera Co.	18	10 (55)	198	27 (14)	5 (2)	32 (16)	17 (9)	16 (8)
Fresno Co.	13	8 (61)	120	10 (8)	3 (2)	13 (11)	1 (1)	63 (52)
Kern Co.	6	4 (67)	42	5 (12)	1 (2)	6 (14)	5 (12)	5 (12)
Tulare Co.	4	4 (100)	41	8 (19)	0	8 (19)	0	19 (46)
Total	41	26 (63)	401	50 (12)	9 (2)	59 (15)	23 (6)	103 (26)
South California								
Riverside Co. (desert area)	17	17 (100)	156	82 (52)	2 (1)	84 (54)	0	3 (2)
Total California	166	148 (89)	1,735	638 (38)	180 (10)	818 (47)	297 (17)	115 (7)

[a] Percentage of the total number of cankers sampled per county.
[b] Number of vineyards sampled.
[c] Number of vineyards (and percentage of the total number of vineyards sampled per county) yielding *Botryosphaeria* spp.
[d] Total number of cankers collected.
[e] A = number of cankers from which a *Botryosphaeria* sp. only was isolated.
[f] B = number of cankers from which a *Botryosphaeria* sp. was isolated together with other grapevine wood fungal pathogens, such as *Eutypa lata*, *Phomopsis viticola*, *Phaeoacremonium* spp., *Phaeomoniella chlamydospora*, and other species of ascomycetes.