

CONTROLLING FUSARIUM WILT OF STRAWBERRIES BY ANAEROBIC SOIL DISINFESTATION

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Anaerobic soil disinfestation (ASD) has been shown to be effective in controlling Verticillium wilt and to be able to provide marketable fruit yields comparable to fumigation in California (CA) strawberry systems (Shennan et al., 2014a). ASD also reduced charcoal rot mortality in strawberries caused by *Macrophomina phaseolina* by 50% two years in a row in an organic site in southern CA (Shennan et al., 2015). In the 2014-15 season, the ASD-treated berry acreage exceeded 1,000 acres in California; more than doubled from the previous season.

Fusarium wilt caused by *Fusarium oxysporum* f. sp. *fragariae* (F.o.f), an emerging lethal disease of strawberries in CA, can also be controlled by ASD. However, a study has shown that higher soil temperatures are necessary to control this disease by ASD (> 300 cumulative hours above 86 °F at 8" soil depth during the treatment (Yonemoto et al., 2006). In the 2013-14 season, we demonstrated that ASD summer flat treatment using 9 t/ac of rice bran and clear TIF can exceed the temperature threshold resulting in reduced F.o.f. soil densities (Table 1 and Fig. 1 left) and disease symptoms at Watsonville, CA. However, fruit yield was reduced by the flat ASD treatment, likely resulting from an unexpectedly low-N supply from the added rice bran (Shennan et al., 2014b).

Here we report on the 2014-15 trial at the same site that aimed to: 1) demonstrate whether ASD summer flat (ASD-flat) treatment can consistently control Fusarium wilt of strawberries, 2) monitor the effect of ASD-flat on soil N dynamics and 3) examine whether changes in soil microbial community composition are responsible for soilborne disease suppression by ASD in the field trial.

Field trial: A replicated trial included ASD-flat with rice bran 9 t/ac (ASD-flat RB9), ASD-flat with molasses 6 t/ac (ASD-flat-ML6), chloropicrin 300 lbs/acre (Pic 300) and untreated check (UTC) as main plots, and with and without pre-plant fertilizer (PPF) as sub plots was established on a sandy-loam Monterey Bay Academy (MBA) site infested with F.o.f. For ASD-flat-RB9 plots, rice bran was broadcasted, rotor-tilled, and covered with TIF clear tarp and had 7 lines of drip tapes applied on September 3. For ASD-flat-ML6 plots, molasses was injected via drip tapes under TIF on Sep 3 and 4. On October 23, beds were listed and pre-plant fertilizer (1,000 lbs/ac of 18-8-13, 7-9 months slow release) was shanked into beds of PPF sub plots. Strawberry cv. Albion was planted on November 18. Unlike the 2013-14 season, due to the late start of the treatment, soil temperatures in ASD-flat-RB9 barely reached the cumulative temperature threshold and did not

reach the threshold in ASD-flat-ML6 where additional water was used to inject molasses (Table 1). As a result, ASD treatments did not reduce F.o.f. in the soil compared to UTC regardless of type of C-source whereas Pic 300 reduced populations significantly (Figure 1 right). Reflecting the soil F.o.f. level, wilt score (1: healthy – 5: 76-100% leaves dead) was lowest for Pic 300 and no difference was observed between ASD and UTC though use of PPF slowed disease progression until May (Fig. 2 left). Pic 300 possessed the highest yield, while yields for ASD-flat-RB9 was ~50%, and ASD-ML6 and UTC was ~30% of Pic 300 (Fig. 2 right). Soil inorganic N dynamics in 0"-6" soil in ASD-RB9 plots during the growth season showed a similar low level with the previous season (Fig. 3). Changes in soil inorganic N distribution across the soil profile (0"-24") before and after the flat ASD treatment indicated mineralized N accumulation in the subsoil (6"-12" depth) rather than top soil (0"-6" depth) (data not shown). T-RFLP analysis indicated that a distinctive soil bacterial community was formed by each main treatment immediately post-treatment though it did not affect the soil F.o.f. population (Fig. 4).

In summary, to reduce Fusarium wilt by ASD, summer flat ASD must start by mid-August at latest in the central coast of CA. In controlled environment experiments (32/24 °C, 2 weeks), ASD with grass effectively suppressed populations of an introduced F.o.f. isolate and suppressed wilt (Fig. 5). Effectiveness of summer flat ASD with varying C-source on F.o.f. will be examined in the next field trial. N in C-source for summer flat ASD may not be efficiently used by strawberries. Thus, a low-N C-source would be more appropriate for the summer flat ASD. A shift in bacterial community induced by ASD was not related to immediate disease suppression in this case though other studies indicate that it may be related to the development of disease suppressiveness. At this point, use of a resistant cultivar, crop rotation, good sanitation and a combination of all three are the most effective non-fumigant Fusarium wilt management strategies for strawberries in the central coastal CA.

Selected References:

- Shennan, C., J. Muramoto, M. Mazzola, D. Butler, E. Roskopf, N. Kokalis Burelle, K. Momma, Y. Kobara, and J. Lamers. 2014a. Anaerobic Soil Disinfestation for Soil Borne Disease Control in Strawberry and Vegetable Systems: Current Knowledge and Future Directions. *Acta Horticulturae (ISHS)* 1044:165-175.
- Shennan, C., J. Muramoto, M. Zavatta, and M. Mazzola. 2014b. Non-fumigant approaches for controlling fusarium wilt and charcoal rot of strawberry. 2014 Annual International Research Conference on Methyl Bromide Alternatives and Emissions Reductions:11-1- 11-4.
- Yonemoto, K., K. Hirota, S. Mizuguchi, and K. Sakaguchi. 2006. Utilization of the sterilization by soil reduction in an open air field and its efficacy against Fusarium wilt of strawberry. *Proc. Assoc. Pl. Protec. Shikoku*:15-24 (in Japanese with English summary).

Table 1. ASD conditions of the summer flat ASD trials (the 2013-14 and 2014-15 trials) at MBA, Watsonville, CA.

Year	Treatment	Period (days)	Cum Eh < 200 mV hrs	Cum soil temp > 86 ° F hrs	Water added ac-inches
2013-14	ASD rice bran 9 t/ac	8/14 - 9/23 (40)	224,578	980	3.3
2014-15	ASD rice bran 9 t/ac	9/3 - 9/26 (23)	125,000	348	2.3
	ASD molasses 6 t/ac		116,000	211	5.9
	UTC		Not measured	81	0.0
Threshold			> 50,000*	> 300**	

* For *Verticillium dahliae* at 77 ° F (Shennan et al., 2007).
 ** For *Fusarium oxysporum* f. sp. *fragariae* at 8" soil depth (Yonemoto et al., 2006).

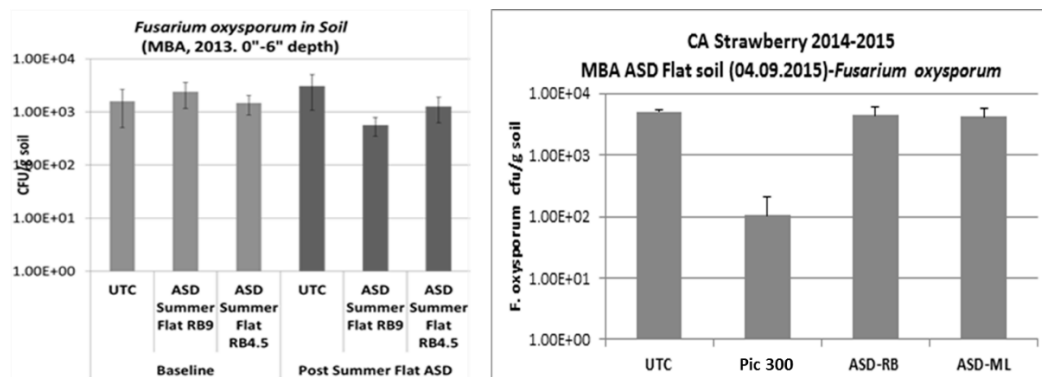


Figure 1. *Fusarium oxysporum* population in soil at post-treatment in the 2013-14 (left, black) and 2014-15 (right) trials.

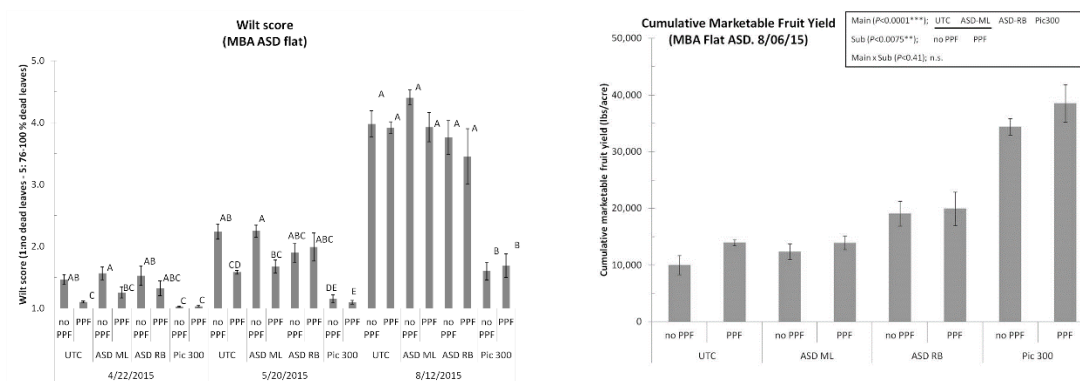


Figure 2. Wilt score development in ASD flat trial (left). Wilt score 1 = healthy plant, 2 = 1-25% dead leaves, 3 = 26-50% dead leave, 4 = 51-75% dead leaves, 5 = 76-100% dead leaves. Cumulative marketable fruit yield in ASD flat trial (right). No significant difference between variables on the same line by Tukey's-HSD test ($P < 0.05$).

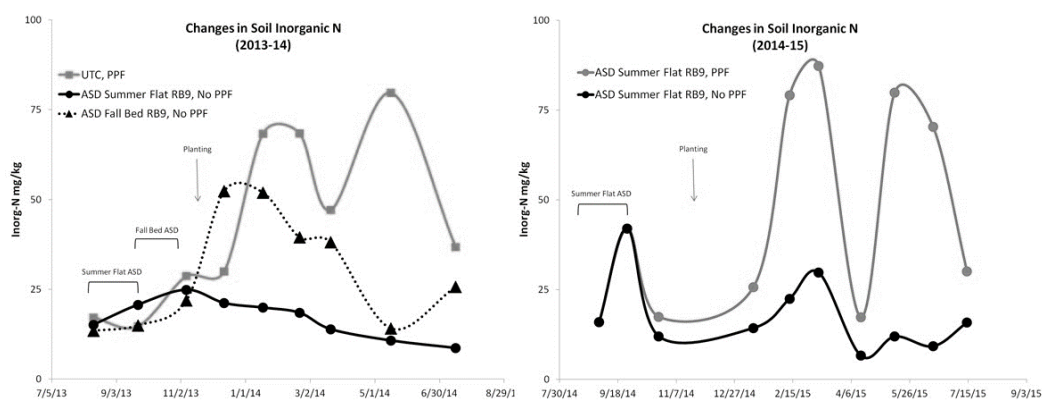


Figure 3. Soil inorganic N dynamics (0"-6" depth) in selected plots of the 2013-14 trial (left) and the 2014-15 trial (right). In the 2013-14 trial, PPF (640 lbs/ac of 18-6-12, 12-14 months slow release) was applied only to UTC. In the 2014-15 trial, PPF (1,000 lbs/ac 18-8-13, 7-9 months slow release) was applied to PPF sub plots.

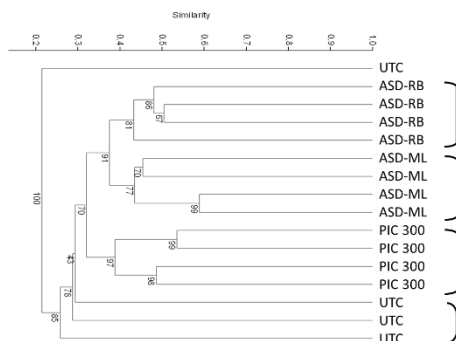


Figure 4. Effect of soil treatments on relative similarity of bacterial communities as determined by analysis of T-RFLP fragment derived data.

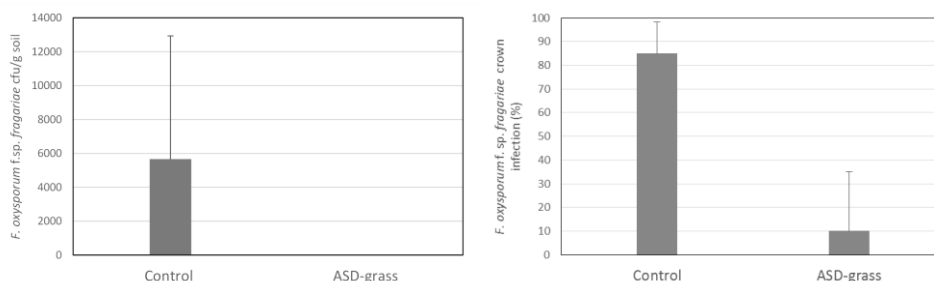


Figure 5. Populations of *F. oxysporum* f. sp. *fragariae* and resulting crown infection of strawberry as affected by ASD conducted using 9 tons/acre of dry grass as the carbon input. Error bars indicated one standard deviation of the mean.