Compost Practices for Control of Grape Powdery Mildew (Uncinula necatòr Andreas Tränkner and William Brinton

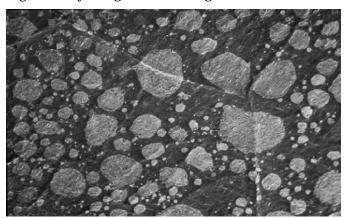
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INTRODUCTION

Powdery mildew causative agent *Uncinula necator* is a widespread obligate parasite which infects practically all assimilative grape organs. Under favorable spreading conditions typical spreading white conidia tufts are easy to recognize. Infected inflorescences and young leaves are retarded in growth and the older vine leaves are especially impacted with reduced photosynthesis (Lakso et al., 1982). Economic loss from downy mildew results from a whole set of factors. Early infection of the grapes causes berries to dry out, while late infection causes them to split open (Nieder, 1988, 1989). The latter particularly paves the way for post-infection attack by *Botrytis cinerea*. In addition to these effects, the canes of infected plants show slowed growth and measurably decreased frost resistance. Finally, metabolites produced by the fungus impart an off-flavor to wine (Pool et al., 1984).

The optimal temperatures for spread of *U. necator* is between about 20° and 27°C (68 and 80F). At higher temperatures fungal-mycelial growth is inhibited, and dying off is evident from prolonged heat stress at 35°C (95F) or above. High humidity is of advantage for fungal conidia formation, but unfortunately conidia germination is almost independent of humidity (Stenzel, 1955; Yarwood, 1978). Thus, the fungus is well adapted to fluctuation in weather conditions. For these and other reasons, predicting *U. necato*r infection is difficult and there are hardly any reliable forecasting models.

Control of *Uncinula necator* infection is possible using a variety of agents including sulfur formulations,



Magnification of bacterial colonies on grape leaf sprayed with compost tea (Source: Woods End Research Lab)

synthetic fungicides or certain combined preparations. Sulfur has been successfully used since the middle of the eighteenth century and is still considered to be an effective agent in the form of dusts or liquid formulations (Holst, 1986; Riffiod, 1987). However, such natural or organic-type treatments must be repeated frequently for adequate control. For this reason, synthetic fungicides with much longer effectiveness dominate in conventional viticulture. However, due to the emergence and rapid increase of fungal resistance to synthetic fungicides within the past decade (Buchenauer & Hellwand, 1985; Boubals, 1987; Payan et al., 1989), increased endeavors are being undertaken to develop biological control measures.

BACTERIA AND COMPOST FOR CONTROL

Schönbeck (1982) was one of the earliest to show the potential usefulness of applications of bacterial culture filtrates for control of obligate parasites, attributing these effects to "induced-resistance" also called systemic acquired resistance (SAR). However, attempts to obtain satisfactory control using bacterial filtrates in the Weinberg region has proven largely unsuccessful and the researchers conclude that improvements in product formulation and in the method of application are necessary. Similarly, Kast (1985) reported that he could not obtain any significant reduction of *Uncinula* infection with 16 applications of such bacterial filtrates.

Up until recently, possible microbiological control approaches have appeared limited to the use of hyperparasites. Sztejnberg et al. (1989) and Gadoury & Pearson (1988) reported a naturally occurring parasitization of *Uncinula* by *Ampelomyces quisqualis*. Effects of the mycoparasite *Tilletiosis* sp. against powdery mildew were also observed (Hoch & Provvidenti, 1979). However, to our knowledge, very few experiments have been conducted under actual field conditions using hyperparasites for control of *Uncinula*.

In this paper, we discuss a new angle on a specific use of composts for control of disease. Here, we present the use of aqueous, microbiologically active extracts of composts against the pathogen *U. necator*. Compost extracts, also called compost teas, have been discussed by the authors in a previous issue of Biodynamics and elsewhere (Brinton, et al., 1995-1996). Generally speaking, compost teas are made by steeping mature but not overly-mature compost in water for a period of 3-12 days after which the filtered material is sprayed undiluted. The optimal period partly depends on the com-

post and the pathogen-system being dealt with. We normally expect to obtain levels of microorganisms in compost extracts of the order of 108 - 1010 range, and the important microbial groups are both aerobic and facultative anaerobic genera. When sprayed, the extracts coat the leaf with live bacterial colonies (see photo on preceding page.)

Table 1— Effect of Watery-Extracts of Grape-Pomace and Cattle Manure Composts (two extraction times) on Uncinula infection of Greenhouse-Grown Grapes.

COMPOST Treatment	Assessment Aug. 8	Assessment Aug 22		Assessment Sept. 5	
	Incidence (%)	Incidence (%)	Severity (%)	Incidence (%)	Severity (%)
Control (none)	33.4 a*	67.7 a	18.6 a	82.0 a	46.6 a
Pomace (3 day extr)	10.0 b	31.2 b	8.2 b	39.7 b	17.7 b
Pomace (7 day extr)	6.4 b	32.4 b	8.2 b	46.0 b	24.4 b
Cattle (3 day extr)	1.1 c	6.1 c	1.6 c	10.5 c	8.7 c
Cattle (7 day extr)	0.0 c	4.0 c	1.2 c	8.6 c	2.3 d

Values within a column with differing letters are significantly different at p=0.05% according to Duncan's test. Source: Fischer and Weltzien, unpublished.

RESEARCH STUDIES

In a germinal study in Bonn, compost extracts were prepared from grape pomace compost and cattle manure compost and tested on greenhouse grown grape vines for control of *U. necator*. Both composts were extracted for 3—7 days to obtain the spray concentrate. The incidence of infection by mildew was rated following repeated applications of the extracts and compost extracts gave significant reductions in comparison with control at all evaluation times (see Table 1).

The extracts from cattle manure compost show significantly better effects against mildew than such from a purely vegetal compost, the grape pomace compost. These clear differences appeared at all assessment intervals and related to infection frequency as well as to severity of infection. With regard to the optimization of the extraction times, there was no unequivocal evidence supporting longer extraction times.

In Table 2 we show results of another greenhouse experiment in which the action of horse-compost extracts on powdery mildew is compared with treatment of a standard wettable sulfur preparation.

Table 2— Effect of Compost Extract and Sulfur (4x) Treatments on Incidence of <u>Uncinula necator</u> on Greenhouse-Grown Grapes

Treatment	Leaf infection (%) (late season)	
Control (no treatment)	82 a*	
Horse-compost extract (3 day extr)	8 b	
Wettable sulfur preparation	.6 b	
Horse-compost extract + wettable sulfur	5 b	

T* Values with different letters differ significantly by the Duncan test (p=0.05%) Source: Ketterer, 1990

Clearly, a strongly suppressive effect against *U. necator* of the horse manure-compost tea comparable to sulfur is observed here. The level of mildew infection seen on control vines has been restricted to a less than one tenth residual level with the treatment.

It is often presumed that extracts containing microorganisms exert an action on the fungal conidia germination of U. necator. To test this, conidia germination rate was studied following a 48 hour induction period of the extracts (See Table 3).

Table 3— Effect of compost extracts on Powdery Mildew conidia germination (Uncinula necator)

Treatment	Conidia germination (%)	
Control (no treatment)	62 a*	
Pomace Compost Tea (3 day extract)	23 b	
Pomace Compost Tea (7 day extract)	22 b	
Cattle-Manure Compost tea (3 day extract)	14 c	
Cattle Manure Compost Tea (7 day extract)	13 c	

Values within columns with differing letters are significantly different according to the Duncan test (p=0.05%)

This and other data show that a significantly reduced germination rate in comparison with control could be observed in all extract variants. A striking agreement of the results of conidia germination with that of the level of *Uncinula* infection in greenhouse vines was determined. The extracts from cattle manure compost exerted a clearly more suppressive action on the conidia germination in comparison with those of the grape pomace compost. With regard to the effect of varying extraction times, neither of the two times studied gave a clear preference with either fungus germination or leaf infection incidence.

Histological-biochemical studies were performed and yielded no evidence that would indicate induced resistance as an action mechanism in *Uncinula* control in these trials. Neither differences in papilla formation nor in lignifications in the cell wall area could be detected as indications of post-infection defensive reactions between the individual varieties studied. Thus, according to these findings, we conclude that a direct action of compost extracts on conidia germination predominates in powdery mildew systems, and consequently a completely different mode of action is evident in contrast to other mildews we have studied such as *Sphaerotheca fuliginea*, cucurbit mildew. Case by case,

we are finding compost exerts various and different modes of bio-control where disease is concerned.

The compost extracts studied here with their antagonist flora were primarily optimized previously for control of *Plasmopara viticola* (downy mildew), the major grape disease in Europe. Hence the treatment variations and optimization practices we have developed for P. viticola would apply here. A later report will present the findings from these optimization trials. Basically short term extracts (3 days) were found to be ideal, and supplementation of the teas with yeast-extract and saccharose significantly improved the results.

Over a two-year period, heavy infection pressure was observed for *Uncinula necator* in two grape growing regions, and field experimental parcels were therefore established. Satisfactory effects from compost extracts were attained (see Table 4).

Only a moderate *Uncinula* infection pressure appeared in 1989 in an experimental parcel in Württemberg. Here extract applications of cattle manure compost reduced leaf infection from 20 to 6%. The addition of *Trichoderma harzianum* and *Mortierella alpinia* cultured separately in the laboratory and mixed with the extracts prior to application, gave no statistically significant increase in control beyond that of the compost extract.

Applications of cattle-manure compost extract proved to be very successful in the control of *U. necator* in field studies in the Ahr wine growing area (Table 4). The treatments reduced fungal leaf infection to one control vines. The compost extracts were more effective in these trials than wettable sulfur, which gave a 50% reduction.

Table 4— Effect of cattle-manure-compost extracts on the incidence in grape leaves of <u>Uncinula necator</u> as determined in field-plots in the Württemberg and Ahr wine growing regions (after Sackenheim, 1993).

	% Incidence of Infection		
TREATMENT	Württemberg Region	Ahr Region	
Control	20.4 a*	65.3 a	
Cattle-Manure compost extract	6.3 b	6.9 b	
Compost extract + microorganisms ¹	4.1 b	*	
Wettable sulfur		32,2 h	

Values within a column with different letters are significantly different according to the Duncan test (p=0.05%)

These trials were followed with a field study employing a 2-6 month old horse-manure-compost extract instead of the cattle manure compost previously used. The extracts were additionally supplemented in order to promote antagonistic microorganisms with the aid of nutrient additives. The results indicate that the horse-compost extract was fairly unsatisfactory for reducing powdery mildew infection (Table 5).

Table 5— Effect of horse-manure-compost extracts with and w/o added nutrients on infection level of <u>Uncinula necator</u> in the Württemberg wine growing area.

Treatment	% Infection
Control (no treatment)	57.6 a*
Horse-compost extract	40.2 b
Horse-compost extract + saccharose (1%)	31.6 b
Horse-compost extract + saccharose + yeast	34.7 b
Fungicides	2.4 c

Values within a column with different numbers are significantly different according to the Duncan test (p=0.05%)

Source: Sackenheim, 1993

Due to large variances in the experimental results, even variants which tended to be successful did not appear significantly better with the nutrient additives. In this case, one of the reasons for the poor suppressive action against *U. necator* is the relatively advanced age of the horse manure compost employed here. As experiments with other host-pathogen systems have shown, younger (2 months) rather than older composts

are basically better suited for extract preparation. More effort is needed to identify the appropriate maturity stage for such composts. The authors are working with the Solvita test, bioassays and other microbial tests to identify maturity factors that can be reliably used to predict these potential effects.

Growers who supplement their compost extracts with additives frequently complain of high odor levels. In

Trichoderma harzianum and Mortierella alpinia (batch cultured)

order to avoid this disturbing result, a new strategy for improving the action of compost extracts was recently studied. Here, we select nutrients which would normally be used to promote the growth of the desired antagonistic microorganisms. These are not, however, added at the beginning of the extraction period, but rather added to the compost extract just prior to being sprayed. The strategy behind this is to promote the desired microbes in the phyllosphere of the grape leaf. It also supports the indigenous microorganisms from the compost and their ability to compete on the grape leaf is raised. For this purpose glucose and bouillon are used as nutrients, while the addition of emulsified rape oil has been found to improve weather resistance.

The results of these trials are seen in the following table (Table 6). Whereas the extract from horse manure compost attained approximately 50% reduction in infection, the combined use of the extract with casein and a rape oil was extremely successful. This combination which lowered leaf infection by *U. necator* to 8% was as successful as a spraying sequence with conventional fungicides. Oil and CASO bouillon alone had no influence on the occurrence of infection. This suggests that the compost extracts are necessary for enriching antagonistic phyllosphere microflora, and the supplementation reinforces the biological effect by improving longevity.

Table 6— Effect of compost extracts and various additives for control of <u>Uncinula necator</u> infection of grape leaves in the Ahr valley.

Treatment	Leaf infection (%)	
Control	62 a**	
Horse-compost extract	32 b	
Horse-compost extract + glucose	33 b	
Horse-compost extract + CASO + rape oil	8 c	
CASO bouillon	50 ab	
Rape oil	57 a	
Fungicide	4 c	

Values with different letters are significantly different according to the Duncan test (p=0.05%)

In order to study the effect of antagonistic microorganisms in detail, the phyllosphere population of grape leaves was determined 3-4 days following sprayings of compost extracts. Leaf samples were detached from the middle region of the vine, washed and quantitatively evaluated for microorganisms. Total germ count was established, and pseudomonads— an important group of antagonists— were evaluated via a selective culture medium. Aerobic spore formers were determined after short heating to 80° C with subsequent cultural plating. The total germ count gives information on the total microorganism community. The aerobic spore -former count was interesting, since these organisms exhibit a special adaptability to leaf surface conditions.

The analysis of the horse-compost-extract showed total bacteria count lay between 3.4×10^6 to 1.4×10^7 /ml. The number of pseudomonads was found to be between 1.2 and 6.5 x 106/ml. Of the aerobic spore formers, we found 3.9 to 4.4 x 105 in ready-to-spray extract. Following this, leaf populations were evaluated. In six out of 8 spray terms, and despite considerable variations in the weather, we found a a significant increase of the total natural leaf-surface bacteria count

(2.9 x 104/g leaf mass) to a power of ten higher that could be attributed to the extract treatments. No significant changes over against the natural population of the phyllosphere could be detected in relation to the pseudomonad group.

Very reliable results emerged in connection with the quantity of spore formers. The natural population of spore forming bacteria was $1 \times 10^3/g$ of leaf mass, and was raised by a power of ten by applying compost extracts. If CASO and rape oil were co-mixed just prior to application, the microbial increase was easier to observe.

In order to clarify the causal connection between elevation of the phyllosphere microflora and U. necator infection reduction, a comparison of the infection frequency in relation to phyllosphere microflora count was undertaken (Figures 1a and 1b). Control vines manifested the lowest germ count $(1.4 \times 10^5 \text{ microorganisms/g})$ fresh leaf mass) as well as the lowest count of aerobic spore formers $(1.65 \times 103 \text{ spore formers/g})$ fresh leaf mass) with a 62% leaf infection. Horse-compost extract sprayings could raise the population level in relation to the total germ count as well as the spore former count to $2.5 \times 105 \text{ or } 9.8 \times 103 \text{ microorgan-}$

isms/g of fresh leaf mass and reduce the *U. necator* infection by ca. 50%.

Figure 1a and 1b: Comparison of *U. necator* leaf infection in comparison to total leaf surface plate count (a) and the aerobic spore former count (b) (values determined from 8 samplings in the July 30-September 9) (source: LANGE and WELTZIEN. unpublished).

Fig 1a. Total Place Count

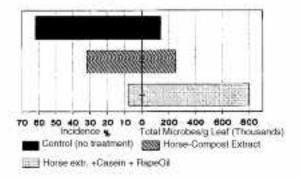
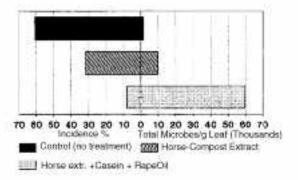


Fig 1b. Total Spore Formers



These results clearly support the significant rise in phyllosphere bacterial populations as a result of sprayings with horse-compost-extract, supplemented with CASO and rape oil, to 8 x 10^5 or 5.9 x 104, and show how it is associated with the greatest reduction of infection to 8% of the control treatment. Thus, the suppressive action of the extract sprayings against mildew can most likely be attributed to enrichment of the phyllospheric populations with regard to the param-



Compost in the vineyard (Source: Woods End Research Lab



Testing kits like this make working with compost easier. (Source: Woods End Research Lab)

eters total plate count and aerobic spore former count.

When spraying to reduce infection by mildew of the actual fruit itself, compost extracts are not likely to be very effective. The primary reason for this is that spray drops are repelled by the waxy surface without wetting or sticking to the berries. In order to obtain a reduction of *Uncinula* infection on the fruit themselves, the use of a microorganism-friendly wetting agent or adhesive is incomparably more important than in the leaf area. According to Sackenheim (personal communication), compost extracts with nutrient doses prior to extraction (1% saccharose + 1% beer yeast) can only reduce berry infection with *U. necator* by 50% when in addition an adhesive (0.1% methyl cellulose) distinctly improves wetting of the inflorescence.

SUMMARY

Grape Powdery Mildew exerts a number of seasonlong effects that act to reduce grape vigor, decrease drought and cold-resistance and impart off-flavors to wine made from infected fruit.

In a number of studies carried out during the late 80's and early 90's in Germany, straight extracts of cattle manure composts and supplemented extracts of horse manure composts have shown excellent promise in both greenhouse and field studies for control of powdery mildew causative agent U. necator. The effects do not appear to be systemic but are antagonistic in nature and correlate with high levels of active microbes on the leaf surface.

Short-term (3 day) extracts of moderately mature composts appear to be the most effective for powdery mildew control. More work will be needed to see the extent to which climatic and compost-type variations influence the results.

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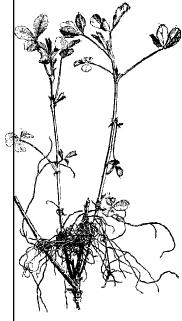
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Cover Crops and Green Manures Number 1 of a Series 7



Al fal fa

(Medicago sativa L.)

A deep-rooted purpleflowered perennial with several stems from a root crown. It is second only to red clover as a forage crop in America. Its chief use is as hay, although it is also pastured. It is sometimes used for silage and serves as a green-manure crop and green feed, frequently being ground into alfalfa meal. In the Far Western States it is considered the most important honey plant. It is one of the oldest crop plants, probably Asiatic in origin, although its home is not certainly known. It is known to have been cultivated in Persia about five centuries before Christ and was first grown in the United States in Georgia in 1736, although it was not until it reached California from Chile in 1850 that its use became widespread in this country. Many special varieties are known, but nearly all that is grown in the United

States is common alfalfa. It is especially adapted to semiarid areas, where it grows on all but extremely alkaline soils; it cannot, however, grow in soils deficient in calcium. Deep, well-drained soils are especially favorable to its growth. It is of greatest value in soil conservation programs when sown with grasses, as in pasture and meadow mixtures, in rotations, or in contour strips. Alfalfa-grass mixtures are playing an important role in rehabilitating slopes retired from cultivation as part of an erosions-control program, especially in the Western States. In Europe it is usually called lucerne.

The available records show that alfalfa is eaten by nearly three times as many species of birds and mammals as any other leguminous plant. The utilization of alfalfa by animals may under

certain conditions damage the crop. This is especially true in irrigated areas where a field of alfalfa affords not only food but also a substratum well loosened by the growth of the alfalfa roots, in which it is easier for rodents to burrow than in adjoining compact soils. In some instances more damage is done by mounds of upturned soil than by the rodents' feeding. Where alfalfa fields are so situated as to create abnormal concentrations of rodents, proper control measures are justifiable. In many cases the use of alfalfa by birds and mammals is incidental and the value of desirable wildlife making chance use of the plants is likely to be greater than that of the alfalfa consumed.

SOURCE: Legumes for Erosion Control and Wildlife Edward H. Graham (WDC; 1941) pp 77-78.