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# Dying-Arm Disease in Grapevines: Diagnosis of Infection with *Eutypa lata* by Metabolite Analysis

NOREEN MAHONEY,† RUSSELL J. MOLYNEUX,*,† LEVERETT R. SMITH,†,‡ THOMAS K. SCHOCH,† PHILIPPE E. ROLSHAUSEN,§ AND W. DOUGLAS GUBLER§

Western Regional Research Center, Agricultural Research Service, U.S. Department of Agriculture, 800 Buchanan Street, Albany, California 94710, and Department of Plant Pathology, University of California, One Shields Avenue, Davis, California 95616

Dying-arm disease in grapevines, produced by infection with the ascomycete *Eutypa lata*, is responsible for major production losses in vineyards. Dieback of the shoots and cordon is believed to be due to acetylenic phenol metabolites produced by the fungus. To identify specific metabolites that could potentially be used for diagnosis of infection, eight *E. lata* isolates were grown in vitro on hot water extracts from grape varieties with various degrees of tolerance to the foliar symptoms of *E. lata* dieback. HPLC analysis showed that eutypinol was consistently produced in large amounts, together with smaller amounts of methyleutypinol and eulatachromene; eutypine, the putative toxin, was produced solely on Sauvignon Blanc extract and then in only barely detectable amounts. When *E. lata* isolates from Cabernet Sauvignon and Merlot were grown on identical media, the amounts of metabolites produced differed significantly between isolates but the pattern of metabolites was quite similar, with eutypinol again predominating. The consistent production of eutypinol indicated that this was the most suitable metabolite for which to analyze in order to diagnose the presence of *E. lata*. Extraction and analysis of grapevine tissues exhibiting symptoms of dieback failed to show the presence of any metabolites. However, when infected cordon sections were placed in water and cultured for 5 days, eutypinol was readily detected in the aqueous solution; metabolites were not produced from uninfected tissue. This provides a method for detection of infected tissue and indicates that the toxic metabolites react at the point of production, disrupting the vascular structure and inhibiting transport of nutrients, rather than being translocated to tissues that exhibit symptoms.

**KEYWORDS:** Grapes; *Vitis vinifera*; *Eutypa lata*; dieback; eutyposis; dying-arm disease; eutypinol

## INTRODUCTION

Dying-arm disease or *Eutypa* dieback is an important perennial canker disease that affects grapevines (*Vitis vinifera*; Vitaceae) worldwide, including the United States, with particular economic impact in the primary wine-producing areas of California. The causative agent, the ascomycete *Eutypa lata* (*1*), enters the plant principally through pruning wounds, leading to necrosis of woody tissues in the vicinity of the point of infection and may subsequently expand into the trunk of the vine. The disease is cryptic, becoming apparent only after bud break as stunting of new shoots, formation of small, deformed, chlorotic leaves, and development of small fruit clusters. The disease is progressive over many years, and failure to control it leads to severe economic losses, primarily as a consequence of decreased longevity of the grapevines. Yield decreases for five vineyards in California growing either Chenin Blanc or French Columbard grapes were estimated to range from 30 to >60%, whereas vineyards over 20 years old had up to 83% yield reduction, relative to their peak production at ~10−12 years of age (*2*). There are significant differences in tolerance to foliar symptoms of infection, with some of the most valuable cultivars, including Cabernet Sauvignon, being particularly sensitive (*3*). The cost to wine grape production alone in California has been estimated to be in excess of $260 million per annum, or ~16% of the gross producer revenue of $1.672 billion for 1999 (*4*).

Phytotoxicity has been attributed to one or more of a number of phenolic metabolites (**Figure 1**), bearing an unusual pentenyne side chain ortho to the hydroxyl group, that have been