

Abstract

Since the late 1990s, a melting decay of 'Red Globe' grapes has occasionally been observed in cold storage facilities in California. Symptoms of the decay included a cracking and dissolution of the epidermis followed by the development of sunken decayed areas. In extreme cases, nearly the entire berry liquefied. The affected berries usually were randomly distributed over the cluster but sometimes berry-to-berry spread seemed to have occurred. The disease has not been observed in the field. Initial symptoms are usually observed after 2 weeks of cold storage (-0.5 to 0°C and 90 to 95% relative humidity). In 2003, melting decay developed in 'Red Globe' grapes in many cold storage facilities in California and in grapes stored for export. Microscopic examination under the skin at the edges of the decayed lesions revealed numerous yeast and bacterial cells. *Bacillus subtilis* (Ehrenberg) Cohn, *Cryptococcus laurentii* (Kufferath) Skinner, and *Aureobasidium pullulans* (de Bary) Arnaud were consistently isolated on potato dextrose agar (PDA) from samples submitted from two cold storage facilities. Certified diagnostic laboratories identified the bacterium and two yeasts. To fulfill Koch's postulates, we performed the following experiments. 'Red Globe' grape berries with the pedicel attached were surface disinfested in a solution of 0.084% sodium hypochlorite, 1.5% ethanol, and 0.005% Tween 20 for 4 min, and washed with sterile deionized water. Grape berries were placed on raised plastic screens in a humid chamber. After the surface of the berries dried, 0.25 ml of a cell suspension (108 cells per ml) of each microorganism was placed on the surface of each berry without wounding. Water (150 to 200 ml) was added to the bottom of the container to create $> 98\%$ relative humidity (measured with a datalogger), and the containers were incubated at 20°C for 7 days or 0.5°C for 6 weeks. With fruit stored at 20°C , the epidermis of all the berries cracked between 24 and 48 h after inoculation. Severe symptoms of melting decay developed 1 week later. With berries incubated at 0.5°C , melting decay symptoms developed after 6 weeks. Symptoms were most severe on berries inoculated with *C. laurentii*. However, when inoculated berries were preincubated at 20°C for 14 h prior to storage at 0.5°C , symptoms developed after 3 weeks. In this case, and when berries were incubated at 20°C , symptom expression was similar among the three pathogens. A number ($>80\%$) of the affected berries disintegrated and liquefied in a pattern similar to that observed in berries from commercial cold storage facilities. The pathogen used to inoculate the grapes was consistently isolated in pure culture from subsequent lesions. Therefore, a postharvest disease in 'Red

Globe' grapes called melting decay can be caused by the bacterium, *B. subtilis*, or two different yeast species. To our knowledge, this is the first report of *B. subtilis* or *C. laurentii* as pathogens of grape berries in the United States and worldwide. *A. pullulans* has been isolated from *Vitis vinifera* (1,2), but is reported for the first time as one of the causal agents of melting decay of 'Red Globe' grapes. All three organisms also caused symptoms on 'Crimson Seedless' grapes, but melting decay has not been reported as a problem in this cultivar.