

Postharvest BTH treatment induces resistance of peach (*Prunus persica* L. cv. Jiubao) fruit to infection by *Penicillium expansum* and enhances activity of fruit defense mechanisms

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Abstract

To investigate how benzo-(1,2,3)-thiadiazole-7-carbothioic acid S-methyl ester (BTH) may affect disease resistance of peach fruit (*Prunus persica* L. cv. Jiubao), harvested fruit were treated with 200 mg/L BTH solution for 5 min immediately after harvest, incubated at 22 °C and 85–95% RH for 60 h, and then inoculated with *Penicillium expansum* at a concentration of 1.2×10^4 conidia per mL. The lesion area and disease-incidence of the BTH-treated fruit were 64.1 and 49.5%, respectively, lower than that of the fruit without BTH-treatment on the 7th day after the inoculation. In the inoculated fruit, the BTH-treatment enhanced the activities of phenylalanine ammonialyase (PAL), polyphenoloxidase (PPO) and peroxidase (POD) as well as the levels of total phenolic compounds and hydrogen peroxide (H₂O₂), which are considered to play important roles in plant disease resistance. Superoxide dismutase (SOD) activity and ascorbic acid (Vc) level in the inoculated fruit were also enhanced by the BTH treatment during the middle and later periods of the inoculation.

The results showed that BTH treatment could significantly enhance the disease resistance of peach fruit after harvest and suggest that postharvest treatment with BTH holds promise as a new technology, substituting for chemical fungicide control of postharvest diseases in fruit.

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1. Introduction

Peach fruit usually have a very short postharvest life. Decay is an important factor, which limits the storage life of peach, and results in appreciable losses at wholesale, retail, and consumer levels.

Control of postharvest diseases of fruit is mostly dependent on controlled atmosphere storage, refrigeration and fungicides (Kader, 1992). Among these, fungicides are widely used to reduce postharvest decay and extend the shelf-life of peach. However, fungicides are becoming less effective because of the development of pathogen resistance, along with consumer concerns about possible risks associated with the use of chemicals (Wilson et al., 1994). Among a number of new strategies being investigated to control postharvest decay without the pollution of the environment and risk to public health, induced resistance in harvested crops is promising (Wilson et al., 1994; Terry and Joyce, 2004).

Systemic acquired resistance (SAR) is an inducible defense mechanism that plays a central role in disease resistance. There is evidence that accumulation of salicylic acid is necessary for the induction of SAR in plants (Yang et al., 1997). In addition, SAR can also be activated by exogenous treatments with chemical inducers such as 2,6-dichloroisonicotinic acid (INA) (Wilson et al., 1994) and benzo-thiadiazole-7-carbothioic acid S-methyl ester (BTH) (Gorlach et al., 1996). Both salicylic acid and INA are moderately good inducers of resistance. However, problems with stability and phytotoxicity have prevented their use as plant protection compounds, whereas, BTH has been developed as a novel crop protection agent, which does not itself have anti-microbial properties, but instead increases crop resistance to disease (Gorlach et al., 1996).

Recent studies have shown the effectiveness of BTH in protecting different plant species against diseases caused by viral, bacterial and fungal pathogens (Gorlach et al., 1996; Benhanou and Belanger, 1998; Cole, 1999). Although BTH is highly effective at inducing enhanced disease resistance, its mode of action and cellular targets are unknown. In addition, most studies have used vegetative plant tissues as the study material and little work has been done to elucidate how BTH may affect disease resistance in horticultural products such as fruit and vegetables. The objective of our study

was to show the effects of BTH in inducing disease resistance of peach fruit after harvest in response to pathogen attack, and on the activity of enzymes or metabolites that might be involved in induced resistance.

2. Materials and methods

2.1. Fruit and BTH treatment

Peach fruit (*Prunus persica* L. cv. Jiubao) were harvested from a commercial orchard at the earliest stage of commercial ripening (green mature) in July of 2002 and 2003, and selected for uniformity of size, ripeness and absence of defects.

Benzo-thiadiazole-7-carbothioic acid S-methyl ester (BTH, 50% wettable granule formulation, Bion[®], Novartis Ltd., Basel, Switzerland) solution was prepared with sterile deionized water plus 0.05% Tween 80. For the BTH treatment, 240 peach fruit were soaked in 200 mg/L BTH solution for 5 min, and then air-dried. The control fruit were soaked in sterile deionized water plus 0.05% Tween 80 for 5 min. After the treatment, both BTH-treated and control fruit were kept in trays covered with plastic film and incubated at 22 °C, 85–95% RH.

2.2. Inoculation and infection

Penicillium expansum was isolated from decayed peach fruit and maintained on potato dextrose agar (PDA). Conidial suspensions of the pathogen were prepared by flooding the 14-day-old culture dishes incubated at 25 °C with sterile distilled water containing 0.05% Tween 80. The spore suspension was adjusted to 1.2×10^4 CFU/mL with sterile distilled water with a haemocytometer.

The inoculations were carried out 60 h after the BTH treatment. Both the BTH-treated and control fruit (80 fruit per treatment, three replicates) were sterilized with 70% ethanol, and then wounded with a syringe at three points (4 mm deep $\times$ 2 mm wide) on the equator of each fruit. Twenty microlitres of the conidial suspension were injected into each wounded site, and the fruit incubated at 22 °C, 85–95% RH. Disease incidence (the percentage of fruit with visible disease development) and lesion diameter on

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