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Abstract: Plant defence inducers (PDIs) constitute a class of bioactive compounds with an emerging potential to be integrated in plant protection strategies. Plant extracts such as grape marc extract (GME) could elicit plant defence responses in different species. Foliar infiltration of GME to tomato and *Arabidopsis* leaves resulted in the appearance of macroscopic symptoms and autofluorescent compounds.

Key words: grape marc, plant defence reactions, integrated control

Introduction

Induced resistance is becoming a widely accepted "green" approach within integrated disease control strategies. Many different substances could elicit plant defence, and there is an increasing demand to search and identify new ones. The polyphenolic rich extract from grapes might offer a new class of phytosanitary bioproduct having not only the PDI activity (Goupil *et al.*, 2012) but also the ability to reduce pesticide photodegradation (Eyrheraguibel *et al.*, 2010). Our previous work provided molecular evidence of GME-PDI activity by monitoring local and systemic upregulation of PR genes (PR1 and PR2) and localised HR-like lesions on tobacco-treated leaves. Therefore it is of crucial importance to determine the potential activity of this botanical extract on a broad range of plants. Herein we show that tomato and *Arabidopsis* responded to this GME as tobacco plants suggesting its potential use as elicitor.

Material and methods

Leaves infiltration GME is a *Vitis vinifera* L. hydroalcoolic extract provided by Grap'Sud as described in Goupil *et al.* (2012). The botanical extract (pH4.3) was applied as an aqueous solution to foliar tissue by infiltration with a plastic seringue. As a negative control, leaves were infiltrated with acidic ultrapure water (pH4.3). Distinct leaf areas were infiltrated (50µl) and examined under bright or UV (at 312 nm) light.

Results and discussion Foliar infiltration of GME to tomato and *Arabidopsis* plants resulted in the appearance of localised HR-like lesions (brown zones). Figure 1 shows the leaf abaxial face of both plant model infiltrated with 0.25% GME. A brown desiccated area appeared within the infiltrated tissue at four days post-infiltration (dpi). Examination of the leaf tissue under UV light revealed that both the infiltrated area and the surrounding zone displayed fluorescence in both species (Figure 1d,l,n,p), suggesting an accumulation of phenolic compound. The rest of the leaf remained unaffected. Recently, the elicitor activity of

GME was shown on tobacco leaves (Goupil *et al*., 2012). GME induced lesions with biochemical changes, such as cell death, local and systemic molecular responses (Goupil *et al*., 2012). The macroscopic changes induced by GME on tomato and *Arabidopsis* leaves might be the result of similar plant defence reactions.

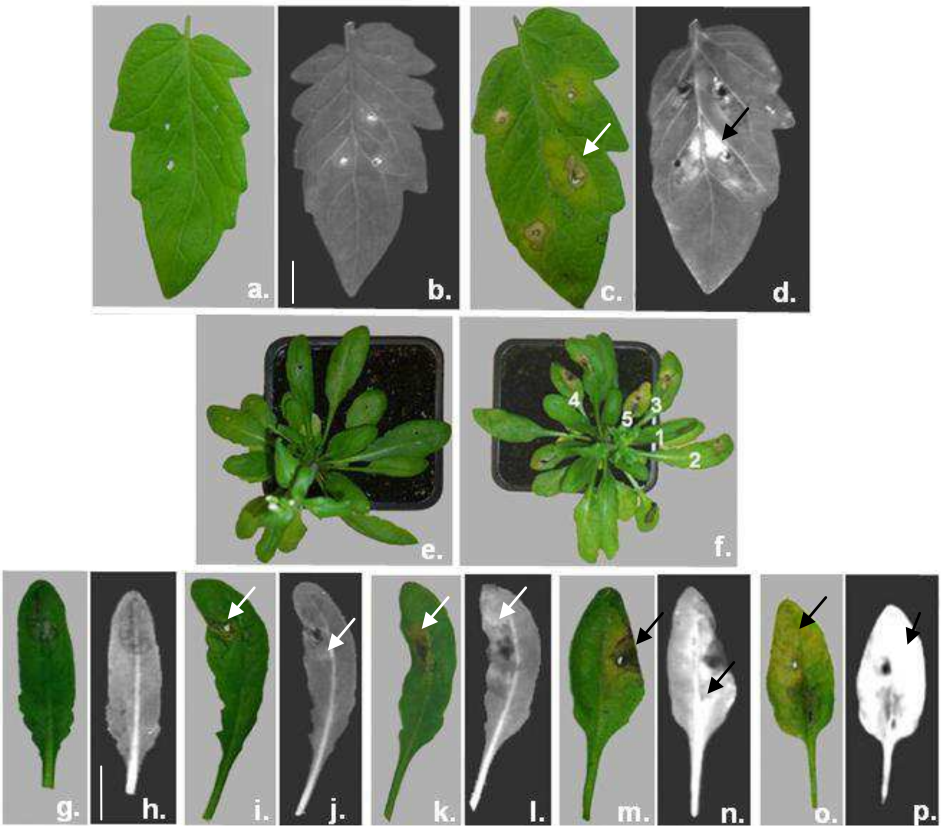

Fig. 1. Macroscopic symptoms induced on tomato (a-d) and *Arabidopsis* (e-p) leaves by acidic water (a,b,e) or 0.25% GME (c,d,f,g-p) infiltration. The leaves were observed under bright light field (a,c,e,f,g,i,k,m,o) or UV light (b,d,h,j,l,n,p) at 4 dpi. Numbers 1-5 in f designate the infiltrated leaves from the apical to the basal position in the plant rosette: leaf 1 (g,h), leaf 2 (i,j), leaf 3 (k,l), leaf 4 (m,n), leaf 5 (o,p). Arrows point necrotic or fluorescent areas. Bars 1 cm.

In *Arabidopsis,* the macroscopic changes induced by GME were also analysed at the whole plant level. At the flowering stage, five rosette leaves were infiltrated with GME and macroscopic symptoms were examined on young and mature leaves. Figure 1f-p shows the necrotic symptoms induced by 0.25% GME at 4 dpi. Younger leaves (Figure 1g,i) displayed faint chlorotic infiltration areas while older leaves (Figure 1n,p) spread out large and strong lesions. Intermediate symptoms were observed on middle-aged leaves (Figure 1k). Fluorescence intensity indicative of phenolics production within infiltration areas was correlated with increasing severity of necrotic lesions (Figure 1h,j,l,n,p). The enhanced capability of a mature leaf to initiate plant defence reactions was demonstrated previously on tobacco. In accordance, *Arabidopsis* older leaves (Figure 1p) displayed almost full fluorescent limb. However, young growing leaves were less reactive to GME elicitor molecules than mature ones (Goupil *et al*., 2012). The data illustrates the importance of plant phenology in defense reaction achievement. Conceivably, expanding tissues having immediate needs of nutrients and energy for primary metabolism, could not achieve optimum processes for both defence reactions and growth (Bolton, 2009) or have not as yet fully developed their defence response.

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