



Synergistic action of serine- and metallo-proteases from *Fusarium oxysporum* f. sp. *lycopersici* cleaves chitin-binding tomato chitinases, reduces their antifungal activity and enhances fungal virulence

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As part of their defence strategy against fungal pathogens, plants secrete chitinases that degrade chitin, the major structural component of fungal cell walls. Some fungi are not sensitive to plant chitinases because they secrete chitin-binding effector proteins that protect their cell wall against these enzymes. However, it is not known how fungal pathogens that lack chitin-binding effectors overcome this plant defence barrier. Here, we investigated the ability of fungal tomato pathogens to cleave chitin-binding domain (CBD)-containing chitinases and its effect on fungal virulence. Four tomato CBD-chitinases were produced in *Pichia pastoris* and incubated with secreted proteins isolated from seven fungal tomato pathogens. Of these, *Fusarium oxysporum* f. sp. *lycopersici*, *Verticillium dahliae* and *Botrytis cinerea* were able to cleave the extracellular tomato chitinases SlChi1 and SlChi13. Cleavage by *F. oxysporum* removed the CBD from the N-terminus, as shown by mass spectrometry, and significantly reduced the chitinase and antifungal activity of both chitinases. Both secreted metallo-protease FoMep1 and serine protease FoSep1 were responsible for this cleavage. Double deletion mutants of FoMep1 and FoSep1 of *F. oxysporum* lacked chitinase cleavage activity on SlChi1 and SlChi13 and showed reduced virulence on tomato. These results demonstrate the importance of plant chitinase cleavage in fungal virulence.

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