

Silencing of FaPG1, a fruit specific polygalacturonase gene, increased strawberry fruit resistance to *Botrytis cinerea*

C. Paniagua¹, C. Sánchez-Raya¹, R. Blanco-Portales², J.A. Mercado¹, E. Palomo-Ríos¹, S. Posé¹

¹Departamento de Botánica y Fisiología Vegetal, Instituto de Hortofruticultura Subtropical y Mediterránea (IHSM-UMA-CSIC). Universidad de Málaga, Málaga, Spain.

²Departamento de Biología Molecular y Bioquímica, Universidad de Córdoba, Córdoba, Spain

Correspondence email: sarapose@uma.es

INTRODUCTION

Plant health is a major target in breeding programs because crops are under constant biotic stress, and climate change is exacerbating pests and disease negative impacts in agriculture. Plant cell walls play crucial defensive roles as protecting barrier, as well as a source of integrity signalling molecules. Plant immunity has evolved a complex multi-layered system which first line of defence is initiated by conserved molecular patterns coming from pathogens, named pathogen-associated molecular patterns or PAMPs, or from their own corrupted cell walls due to pathogen invasion, named damaged-associated molecular patterns or DAMPs. Accumulating evidence from cell wall mutants has unveiled several components and mechanisms of plant innate immunity under biotic stresses. From our cell wall mutant collection in strawberry, transgenic lines with the *FaPG1* gene downregulated was of special interest because *FaPG1* gene encoded a cell wall degrading enzyme with endo-PG activity which releases oligogalacturonic acid (OGA). OGAs are well described for its activity as DAMPs, able to activate plant immune responses against pathogen attacks. **The aim of this work** is to inspect whether the downregulation of *FaPG1* gene could influence the plant innate responses, due to altered quantity and/or patterns of OGAs releases.

MATERIALS

The *FaPG1* transgenic lines has been well studied previously [1-4]. Their main characteristics are a reduction of more than 90% of *FaPG1* gene expression; less polygalacturonase activity; fruit cell walls enriched in pectins, and pectin fractions with longer and more branched structure at nanostructural level. All these features could be related to their better fruit tissue preservation and the firmer fruit phenotype of transgenics in comparison with wild type [1-4].

RESULTS

***FaPG1* line showed better quality (less weight loss and higher bioyield point) and less % of fruit surface infected after 7 days of postharvest**

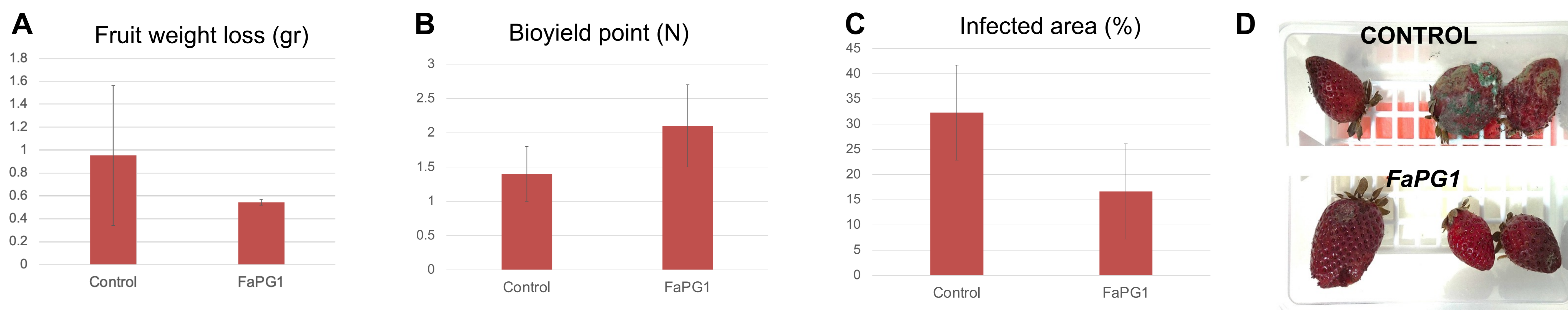


Figure 1. Postharvest behaviour of transgenic *FaPG1* and control fruits. Ripe fruits from *FaPG1* and non transgenic lines were harvest and stored during 4 days at 4°C and 3 days at room temperature, to reproduce a usual postharvest period. Bar graphs correspond with different quality parameters of postharvest assay at final time point (day 7): (A) fruit weight loss during 7 days of experiment; (B) Bioyield point representative of fruit firmness obtained with a puncture test by texturometer; (C) infected area of fruit, evaluating infection degree at surface level; (D) representative image of fruits at postharvest day 7. Postharvest assays were done in triplicate with at least 5 fruits per experiment; data correspond to the mean ± SD.

FaPG1 showed enhance resistance to *Botrytis cinerea*

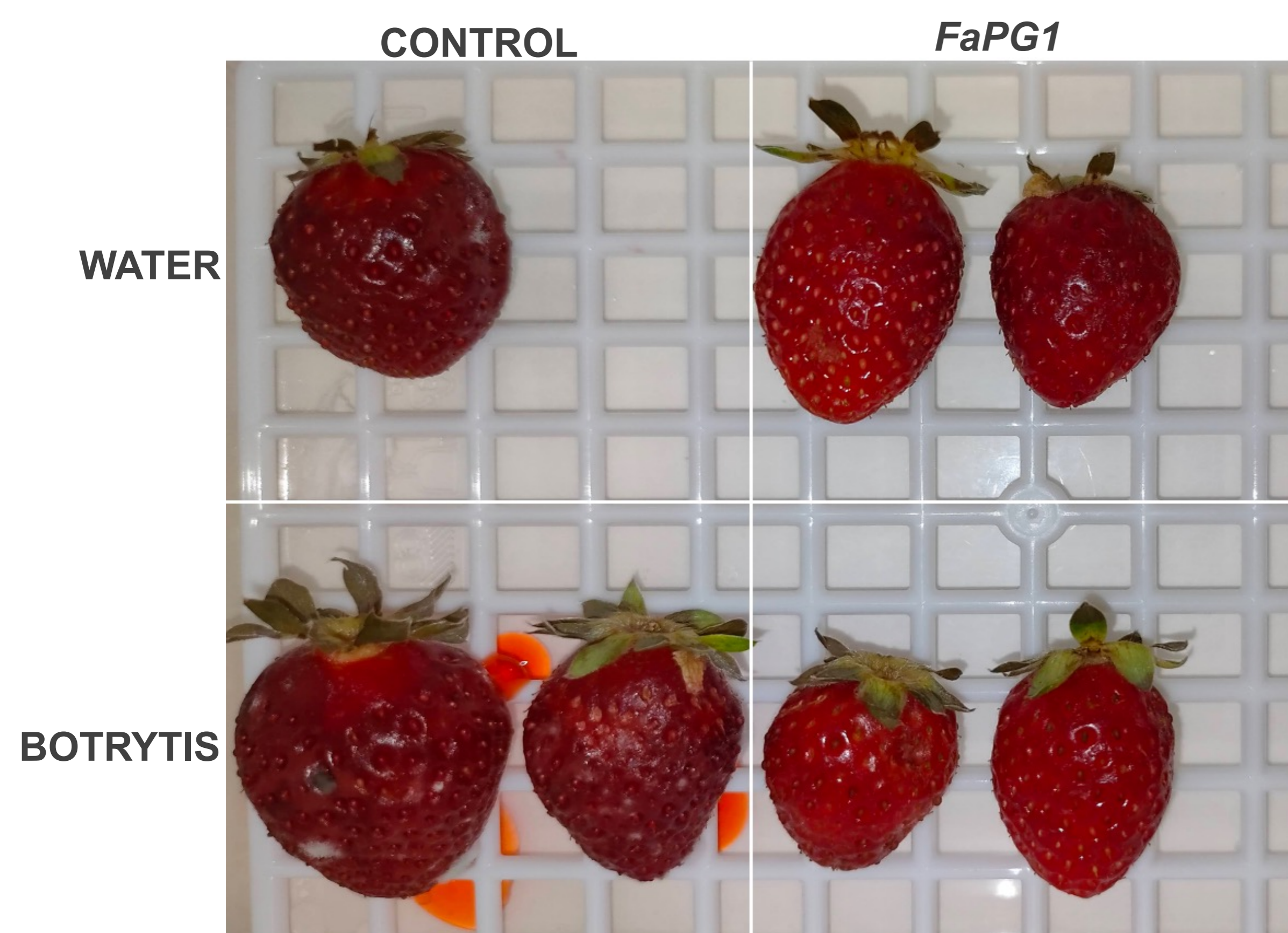


Figure 2. Representative images of *Botrytis cinerea* strawberry assay. *Botrytis cinerea* (B05.10 strain) were grown for 10 days on potato dextrose agar at 25° C. Spores were resuspended in saline solution (NaCl 0.9%) and adjusted to 2.10⁵ conidia/mL using a hemacytometer. Injections with 7 µl of sterilized water (control) or conidia suspension (Botrytis) were inoculated in the fruits and pictures were done at 3 day post-injection.

CONCLUSION

In this work, transgenic fruits showed an enhancing fruit shelf life in comparison with control fruits. Additionally, postharvest and resistance assays showed an increased resistance to fungal infections by *Botrytis cinerea* of *FaPG1* fruits in comparison with non transgenic control.

FUTURE LINES

The next steps will be determinate if the enhanced biotic resistance of this transgenic strawberry line is due to less-degraded cell walls acting as stronger barriers, and/or if DAMPs release alterations are also activating plant natural resistance mechanisms. Obtaining crop varieties armed with better defences (stronger physical barriers and/or active chemical signals) is a potential strategy to enhance plant resistance and reduce losses from biotic attacks in strawberry crops. Additionally, the use of DAMP-based products are being researched and applied as spray foliar treatments and represent a green alternative solutions to fight against pest diseases of crops.

REFERENCES

1. Quesada, et al. (2009). Antisense down-regulation of the *FaPG1* gene reveals an unexpected central role for polygalacturonase in strawberry fruit softening, Plant Physiol, 150, 2, 1022-1032.
2. Posé, et al. (2013). Insights into the effects of polygalacturonase *FaPG1* gene silencing on pectin matrix disassembly, enhanced tissue integrity, and firmness in ripe strawberry fruits. JXB, 64(12), 3803-3815.
3. Posé, et al. (2015). The nanostructural characterization of strawberry pectins in pectate lyase or polygalacturonase silenced fruits elucidates their role in softening. Carb Pol, 132, 134-145.
4. Paniagua et al. (2020). Elucidating the role of polygalacturonase genes in strawberry fruit softening. JXB, 71(22), 7103-7117.