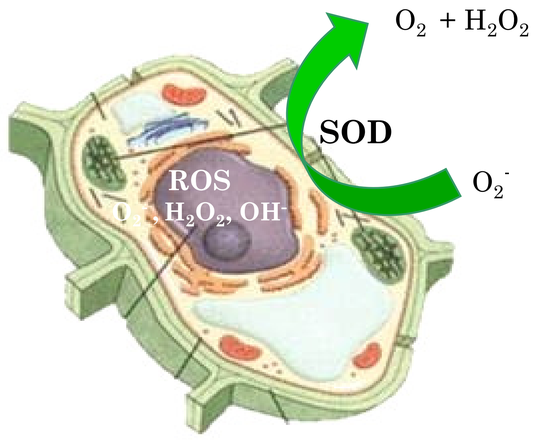
SOD activity and gene expression related to graft compatibility in pear/quince combinations

Irisarri P., Errea P., Pina A.

Unidad de Hortofruticultura, Centro de Investigación y Tecnología Agroalimentaria de Aragón (CITA)

Av. Montañana 930, 50059, Zaragoza, Spain

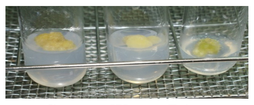


INTRODUCTION

Superoxide dismutase (SOD, EC 1.15.1.1) is an important antioxidant enzyme and constitutes the first level of defence against superoxide radicals in plants. SOD catalyzes the dismutation of O_2^- to H_2O_2 and O_2. According to the cofactors that bind to SOD, there are different forms: manganese SOD (MnSOD), iron SOD (FeSOD) and copper/zinc SOD (CuZnSOD). They are localized in different parts of the cell, mitochondria for MnSOD and chloroplast for FeSOD. CuZnSOD isoforms (CSD) are distributed in different compartments CSD1 is in the cytosol, CSD2 in chloroplasts and CSD3 in peroxisomes. Although exposing plants to stress situations, such as grafting, would trigger the antioxidant defence systems, there are indications that within incompatible rootstock/scion interfaces either the level of reactive oxygen species (ROS) is increased or a less efficient detoxification system is initiated here (Aloni et al., 2008; Nocito et al., 2010). The aim of this study was to evaluate CuZnSOD gene expression of three isoforms (CSD1, CSD2, CSD3) and cytosolic SOD activity in different graft combinations (compatible and incompatible) from pear (Pyrus communis L.) and quince (Cydonia oblonga Mill. Clon BA29).

MATERIAL AND METHODS

DNA and Protein extraction



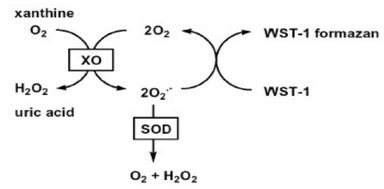
Homografts	Heterografts
Co/Co	Co/BA29 (compatible)
Wi/Wi	Wi/BA29 (incompatible)
BA29/BA29	

Gene Expression

REAL-TIME

Cu-ZnSOD 1	F	GGGAGATGGCCCAACTACTG	111	60
	R	CCAGTTGACATGCAACCGTT		
Cu-ZnSOD 2	F	AGACGATGGGCCTACAACTG	123	60
	R	TTGAAATGTGGTCCTGTGGA		
Cu-ZnSOD 3	F	GCTCTTGGTGATACCTCCAA	122	60
	R	GCAACAACGTTACCCAAATC		

Enzyme Activity



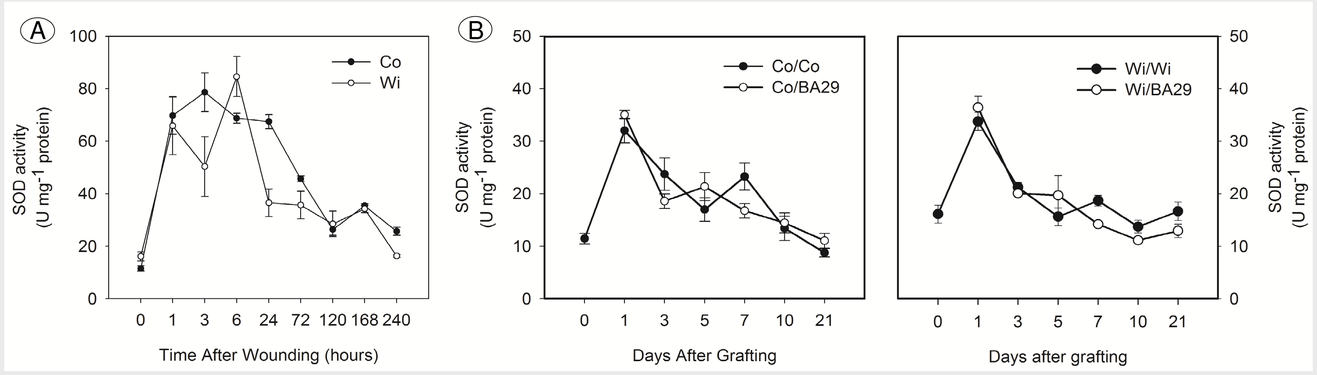


Figure 2. SOD activity. SOD enzymatic activities were significantly higher at 1 (24 h) and 10 (240 h) days after wounding in the compatible cultivar 'Co' than in the incompatible one 'Wi' (A). All combinations expressed the same time-course pattern with the most active SOD activity at 1 DAG, as expected due to the wounding response. After that, antioxidant activity in all combinations decreased compared to 1 DAG, and increased again during a recovery period (B).

RESULTS AND DISCUSSION

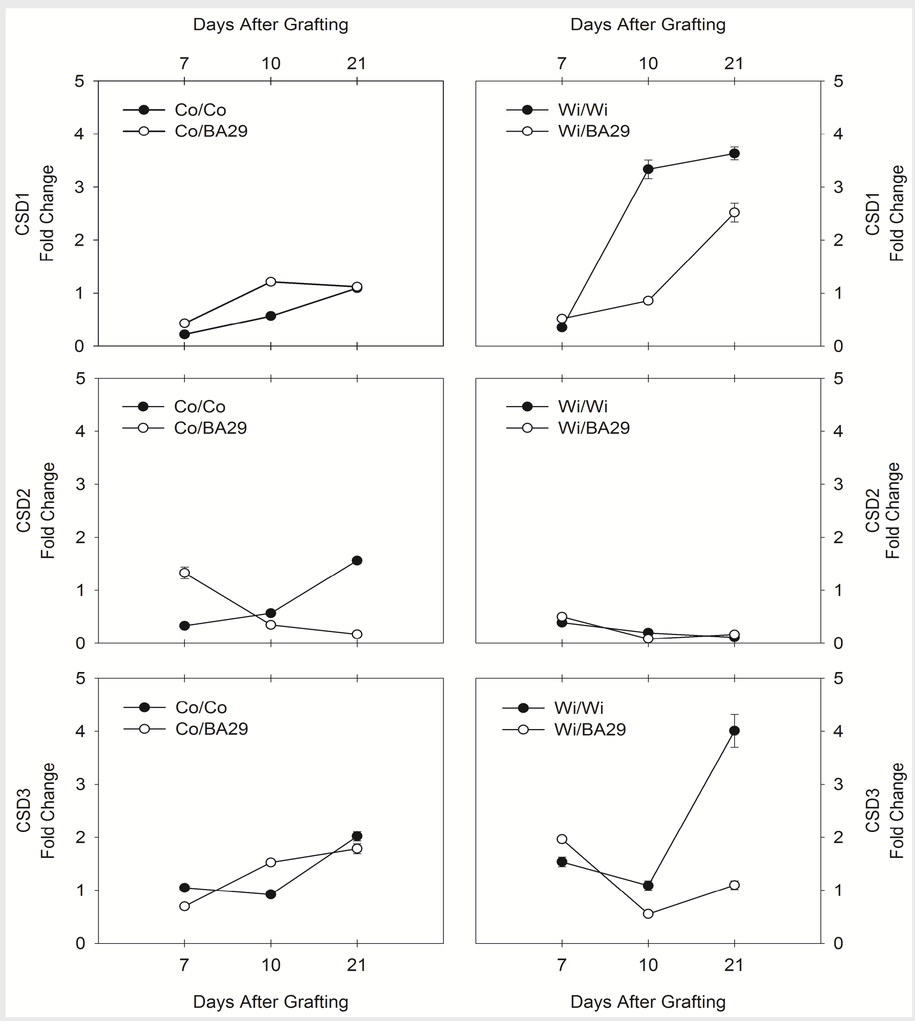


Figure 1. Gene expression. SOD mRNA transcripts were detected in all combinations throughout the graft union development. Among the different combinations, the homograft 'Wi/Wi' showed the higher levels of expression from the second week compared to the others. A 4-fold increase in CSD3 gene expression was observed in the compatible homograft 'Wi/Wi' at 21 DAG compared to the incompatible heterograft 'Wi/Ba29'. It is well documented that an increase in the expression of CSD enhances oxidative stress tolerance under different conditions (Lee et al., 2007)

CONCLUSIONS

Compatible and incompatible graft responses differ in the specificity or abundance of gene transcripts. In this study, we demonstrated that oxidative stress is involved in the grafting incompatibility of *in vitro* pear/quince callus heterografts. The differential expression of genes involved in oxidative stress between the incompatible heterograft ('Wi/Ba29') and their corresponding homografts could be associated with the induction of an oxidative burst at the graft interface.

References

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Acknowledgements

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