幾丁質分解與抗真菌蛋白複合構築之轉基因西瓜抗立枯絲核菌之評估

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摘 要

西瓜為台灣重要農產品之一，也因台灣處於熱帶及亞熱帶，全年氣候皆適合種植，種植面積與種類都很多。西瓜受到真菌的感染很嚴重，Rhizoctonia solani為其中一個有危害性的病源。Rhizoctonia solani為土壤傳播性病原菌，可感染植株幼苗不同部位，造成植物猝倒、根腐、莖爛或萎凋等病徵。在本實驗中送轉
基因Cp-AFP-CHI到西瓜植株中來觀察chitinase和antifungal protein利用結合的方式對於Rhizoctonia solani的抗性。chitinase主要是用來分解真菌細胞壁上的幾丁質，antifungal protein經序列比對類似於植物的防禦素，其構築由中興大學 蕭介夫教授實驗室 陳玉婷博士提供。在本實驗室已成功轉殖出7個轉基因西瓜株系，分別利用聚合?鏈鎖反應和反轉錄聚合?鏈鎖反應分析試驗中，偵測到基因確實併入到genomic DNA中，並證實其mRNA會轉錄成cDNA，表現符合預期之結果。而在瓶內接種真菌的實驗中發現：在接種Rhizoctonia solani之後，非轉基因的植物呈現萎凋、莖爛並且在6天內會完全罹病死亡，而轉基因的西瓜有3個株系抗性較好，在第6天時F27、F47、F49最嚴重的狀態只有葉片萎凋或些微黃化，可以證明比非轉基因的植物有抗性，所以針對此3個轉基因植株再進行西方點漬法的分析，發現蛋白質表現量和接種前不同。於是再進行溫室接種，發現在第12天時，非轉基因西瓜植株發病死亡時，3種轉基因的植株品系僅有些微的病徵，尤其以F47的抗性為最佳，可存活到30天。

關鍵詞 : 抗真菌蛋白、幾丁質分解?、轉基因西瓜、立枯絲核菌、點漬法、病原菌、表現量、蛋白質

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