記錄編號	6334
狀態	NC094FJU00105034
助教查核	
索書號	
學校名稱	輔仁大學
系所名稱	生命科學系
舊系所名稱	
學號	492546094
研究生(中)	劉慈芬
研究生(英)	Liu, Tzu-Fen
論文名稱(中)	Xanthomonas 屬植物病原細菌之檢測及分子類型區分方法之研究
論文名稱(英)	Studies on detection and molecular typing methods of plant pathogenic Xanthomonas spp.
其他題名	
指導教授(中)	李永安
指導教授(英)	Lee, Yung-An
校內全文開放日期	
校外全文	

開放日期	
全文不開放理由	
電子全文送交國圖.	
國圖全文開放日期.	
檔案說明	
電子全文	
學位類別	碩士
畢業學年度	94
出版年	
語文別	中文
關鍵字(中)	植物病原菌 種子 鑑別性培養基 脈衝氏電泳 香菜種子 分子類型區分 插入序列 上菌落數 靈敏度 抗生素 淡綠色 萃取液
關鍵字(英)	plant pathogen seed differential medium PFGE coriander seeds molecular typing insertion sequence taiwan after
摘要(中)	本章的實驗目的為研發 Xanthomonas 屬植物病原細菌的種子檢測法。在研發此屬病原細菌的鑑別性培養基方面，首先以 mTBM 選擇性培養基做測試，並加以改良研發成 Xanthomonas 之鑑別性培養基，命名為 mTBM-42。以 Xanthomonas 屬病原細菌共 8 個種 19 個病原小種作測試，結果顯示在 28℃培養 3-5 天，菌株均呈現淡綠色至深綠色、水亮、上突、菌株周圍有透明與白霧狀兩種暈圈。由植物葉片與種子分離出來的黃色而非 Xanthomonas 菌株，如 Pantoea sp.、Pseudomonas sp.、Erwinia sp.、Arthrobacter sp.，在鑑別性培養基上，菌株型態均為黃色且扁平，除屬於 Arthrobacter sp.菌屬的菌株，周圍只含有透明暈圈外，其餘均無透明與白

霧狀兩種型態的暈圈。將此研發出的鑑別性培養基，應用於罹病甘藍葉片病原菌的分離，在培養 3-4 天即可分離出具明顯特徵的 Xanthomonas 屬病原細菌，可避免選到黃色而非 Xanthomonas 屬的細菌。另外，應用此鑑別性培養基在種子檢測上，以 X. campestris pv. campestris XCC1-1 菌株在此培養基與 LA 培養基上菌落數之比值，顯示不含抗生素的 mTBM-42(A)培養基與 LA 培養基上菌落數之比值為 128%，含抗生素 cephalexin (65 ppm) 與 5-fluorocil (12 ppm) 的 mTBM-42(B)與 LA 培養基上菌落數之比值之回收率為 120%，外加抗生素 cycloheximide mTBM-42(C)與 LA 培養基上菌落數之比值為 109%。以香菜葉枯細菌 X. campestris pv. coriandri NCPPB 1457 在此培養基與 LA 培養基上菌落數之比值，顯示在 mTBM-42(A)其比值為 109%，在 mTBM-42(B)之比值為 101%，在 mTBM-42(C)之比值為 95%。由此結果得知 XCC1-1 或 NCPPB1457 在此選擇性培養基上生長比 LA 培養基好，培養基所加的抗生素也對這兩個菌株只有輕微抑制作用。此實驗同時觀察到這兩個菌株在不含抗生素的培養基上，約 4 天可變綠色，在含抗生素的培養基上，約 5 天可變綠色。將 X. campestris pv. campestris XCC1-1 菌株混合青江菜 (Brassica chinensis) 種子，或 X. campestris pv. coriandri NCPPB 1457 混合香菜種子，再使用 mTBM-42、mTBM-42A 與 mTBM-42B 培養基分離。結果顯示，雖然此培養基不能抑制所有腐生菌生長，但在 28℃培養 3-4 天仍可發現具明顯特徵的 Xanthomonas 屬病原細菌。再利用此鑑別性培養基檢測進口香菜種子之 Xanthomonas 屬細菌。結果顯示只有代號為高雄-1 的種子，可發現到少數具明顯特徵的 Xanthomonas 屬細菌。挑選 2 個代表性菌株 K1-A-27 與 K1-A-37 進行 16S rDNA 序列，結果顯示與 X. campestris 有高達 99 %的相似性，對煙草也會產生過敏反應，證實分離菌株為 Xanthomonas 屬植物病原細菌。再將以上 4 個代表性菌株, K1-A-4、K1-A-9、K1-A-27 與 K1-A-37，與香菜葉片分離菌株 C2-2-1、C4-3、B3-7、B5-5 及 X campestris pv. coriandri NCPPB 1457，以 PFGE 進行分子類型區分分析，結果顯示進口香菜種子 (高雄-1) 所分離之菌株與香菜葉片分離菌株呈現不同的 PFGE pattern。除利用鑑別性培養基檢測外，亦研發 PCR 的檢測法。使用 DNeasy? Tissue Kit 抽取種子萃取液的 total DNA，再以 hrpG-F+hrpG-R 進行 PCR 反應，結果顯示 PCR 靈敏度約為 104-105 cell/ml。 本論文的實驗目的是比較使用 IS-RFLP (IS-restriction fragement length polymorphism analysis) 及 PFGE (pulsed field gel electrophoresis) 兩種方法，在台灣分離之 Xanthomonas campestris pv. campestris (XCC) 菌株的分型之結果。利用 IS1404 (IS3 family)、IS1479 (IS5 family) 以及 ISXac3 做 probe，進行 IS-RFLP 分析。結果顯示使用 IS1404-RFLP 方法，在所測試的 XCC 菌株中有 8 個分子類型。使用 IS1479-RFLP 與 ISXac3-RFLP 方法，也是分成 8 個分子類型，此結果顯示不管使用那種 IS 進行 IS-RFLP 分析，在 XCC 菌株的分型上，所得到的結果是相同的。再將此些菌株的 total DNA，以限制酵素 SpeI 處理後，再以 PFGE 進行分子類型區分分析，也可將其分成 8 個分子類型。進一步使用 PFGE 在香菜 (Coriandrum sativum) 葉枯病原菌株的分型上。因為推測 2000 年夏天在台灣首先發現的病原菌，可能藉由進口種子引入。所以比較在台灣香菜病葉所分離之 14 株疑似的 X. campestris pv. coriandri 菌株，以及最早在印度發現的葉枯病原菌 X.

	campestris pv. coriandri NCPPB 1457 的 PFGE pattern。結果顯示台灣分離的菌株可分成五種 PFGE pattern，而其中一種 type 與 X. campestris pv. coriandriNCPPB 1457 有高度相似性。此結果顯示台灣分離的菌株可能來自包括印度的不同國家，而印度為台灣香菜種子最主要的進口國家。這些菌株進一步使用 hrpG 和 virD4 基因做探針，進行 Southern hybridization。結果顯示大多數的菌株都含有這兩個基因，但有一些菌株則沒有，而含有 hrpG 和 virD4 的菌株呈現多種 RFLP patterns。
摘要(英)	The purpose of this chapter was to develop methods for detection of the plant pathogenic Xanthomonas spp. in plant seeds. The mTBM semi-selective media was chosen to develop a differential medium of Xanthomonas spp., mTBM-42. The nineteen pathovars of eight species of Xanthomonas were tested on mTBM-42. Colonies of Xanthomonas spp. were light green and deep green, glistening, convex with entire margins, and surrounded by a large clear zone and a smaller milky zone after 3-5 days of incubation at 28oC. Some yellow-pigmented non-xanthomonads isolated from plant leaves and seeds, such as the Pantoea sp., Pseudomonas sp., Erwinia sp., Arthrobacter sp., grew on the differential media as yellow, flat, and without clear and milky zones, except Arthrobacter sp., which was surrounded by a large clear zone only. The mTBM-42 differential medium was used to isolate plant pathogenic Xanthomonas spp. from infected cabbage leaves. The characteristic colonies of Xanthomonas sp. were observed after 3-4 days of incubation at 28oC. The medium could differentiate Xanthomonas spp. from yellow-pigmented non-xanthomonads and avoid isolation of non-xanthomonads. Furthermore, the differential medium was used to detect Xanthomonas spp. in plant seeds. The recovery of X. campestris pv. campestris XCC1-1 on mTBM-42 medium was 128%, compared with LA medium. The recovery was 120% on mTBM-42 medium supplemented with cephalexin (65 ppm) and 5-fluorocil (12 ppm) (mTBM-42A), and 109% on mTBM-42A medium with further addition of cycloheximide (75 ppm) (mTBM-42B). The recovery of X. campestris pv. coriandri NCPPB 1457 was 109% on mTBM-42, 101% on mTBM-42 A, and 95% on mTBM-42B. Thus, the addition of antibiotics had a slight inhibitive effect on the growth of Xanthomonas spp. The Xanthomonas spp. tested developed the characteristic colonies in 4 days on mediua without antibiotics and 5 days on mediua with antibiotics. The seeds of Brassica chinensis and coriander were artifically inoculated with X. campestris pv. campestris XCC1-1 and X. campestris pv. coriandri NCPPB 1457, respectively. The seed extracts were plated on mTBM-42, mTBM-42A, and mTBM-42B. Although the media did not eliminate growth of all saprophytes, the characteristic colonies of Xanthomonas spp. could still be observed after 4-5 days of incubation at 28oC. The differential media were further used for the detection of Xanthomonas sp. in imported coriander seeds. A few characteristic colonies of Xanthomonas spp. were found in the coriander seeds of Kaohsiung-1. Two isolates, which K1-A-27 and K1-A-37, were chosen for 16S rDNA sequence analysis. The sequences were highly identical (99%) to that of X. campestris. The hypersensitive reactions were observed in Nicotiana benthamiana incoulated with K1-A-27 and K1-A-37. The

results indicated that the characteristic colonies isolated on the media were plant pathogenic Xanthomonas sp. The molecular types of K1-A-4, K1-A-9, K1-A-27, and K1-A-37 isolates, and strains C2-2-1, C4-3, B3-7, B5-5, NCPPB 1457 of X. campestris pv. coriandri which were isolated from coriander leaves were determined by using pulsed field gel electrophoresis (PFGE). There were two PFGE patterns among isolates from imported seeds of Kaohsiung-1, which were different from the patterns of strains isolated from coriander leaves. PCR assays for seed detection of Xanthomonas spp. was also developed. The total DNA of seed extracts was isolated by DNeasy? Tissue Kit and used in PCR amplification using primer set, hrpG-F/hrpG-R. The sensitivity of PCR assay was approximately 104-105 cell/ml. The purpose of this study is to compare the insertion sequence-restriction fragments length polymorphism (IS-RFLP) and pulsed field gel electrophoresis (PFGE) methods in the molecular typing of Xanthomonas campestris pv. campestris (XCC) strains in Taiwan. Three insertion sequences, IS1404 (IS3 family), IS1479 (IS5 family), and ISXac3, were used as probes for IS-RFLP analyses. There were at least eight molecular types in IS1404-RFLP analysis. The number of molecular types was almost the same by using IS1479-RFLP and ISXac3-RFLP methods, indicating that the results of molecular typing would be the same regardless of what IS used in IS-RFLP analysis. PFGE analysis also obtained the same number of molecular types using SpeI for digestion of total DNAs. PFGE was further used to characterize the suspected strains of coriander (Coriandrum sativum) leaf blight pathogen. The pathogen was first found during the summer of 2000 in Taiwan and might be introduced from imported seeds. PFGE patterns of fourteen isolates of X. campestris pv. coriandri from diseased coriander leaves in Taiwan were compare with that of X. campestris pv. coriandri NCPPB 1457, which was isolated in India where coriander leaf blight first occurred. There were five PFGE pattern types in the strains isolated in Taiwan, and one type is highly similar to that of X. campestris pv. coriandri NCPPB 1457. The results indicated that the strains in Taiwan might be from different foreign countries including India, which is the major exported country of coriander seeds. The strains were further characterized by Southern hybridization using the hrpG and virD4 as probes. The result showed that most strains contained both genes but some did not, and various RFLP patterns were found in strains with hrpG and virD4.

論文目次

目 錄 第一章 Xanthomonas 屬植物病原細菌之種子檢測法之研究 中文摘要--1-1 英文摘要--1-3 前言---1-5 材料與方法---1-13 1.鑑別性培養基之研發---1-13 1-1.鑑別性培養基配方之挑選與改良--1-13 1-2.Xanthomonas 屬病原細菌在鑑別性培養基上之測試-------------------------1-14 1-3.黃色非 Xanthomonas 屬病原細菌在鑑別性培養基上之測試---------------1-14 2.鑑別性培養基之應用-----------------------------

---------------------------------------1-15 2-1.自罹病植物組織分離 Xanthomonas 屬
植物病原菌---------------------------1-15 2-2.自種子分離 Xanthomonas 屬植物
病原菌---------------------------------------1-15 2-2-1.Xanthomonas 屬病原細菌在
鑑別性培養基與 LA 培養基上菌落數 之比值測試---------------------------------
--1-1 2-2-2.Xanthomonas 屬病原細菌在鑑別
性培養基上單一菌落型態之觀察-1-16 2-2-3.種媒病原菌之萃取與使用鑑
別性培養基分離之流程------------------1-17 2-2-4.混合 Xanthomonas 屬病原
細菌之種子以鑑別性培養基之分離測試-1-17 2-2-5.進口香菜種子之檢測---
---1-18 2-2-5-1.香菜種子高雄-1 分離
菌株的鑑定-------------------------------------1-18 2-2-5-2.香菜種子高雄-1 分離
菌株之分子檢測-------------------------------1-18 2-2-5-3.香菜種子高雄-1 分離
菌株分子類型區分----------------------------1-18 3.以 PCR 檢測種子中之種媒
病原菌---1-20 3-1.所使用的引子對之
篩選---1-20 3-2.香菜病葉分離菌
株 Xanthomonas. campestris pv. coriandri Tc3 與種子混 合後之 PCR 敏感度
測試---1-20 4.基礎分生技術--
--1-22 結果------------
---1-26 1.鑑別性
培養基之研發---1-26 1-1.
鑑別性培養基配方之挑選與改良結果---1-26
1-2. Xanthomonas 屬病原細菌在鑑別性培養基上之測試結果------------------
1-29 1-3.黃色非 Xanthomonas 屬病原細菌在鑑別性培養基上之測試結果----
-----1-29 2.鑑別性培養基之應用---
----------1-30 2-1.自罹病植物組織分離 Xanthomonas 屬植物病原菌之結果---
---------------1-30 2-2.自種子分離 Xanthomonas 屬植物病原菌--------------------
------------------1-31 2-2-1.Xanthomonas 屬病原細菌在鑑別性培養基與 LA 培
養基上菌落數 之比值測試---
-------------1-3 2-2-2. Xanthomonas 屬病原細菌在鑑別性培養基上單一菌落
型態之觀察-結 2-2-4.混合 Xanthomonas 屬病原細菌之種子以鑑別性培養
基之分離測試結果 2-2-5.進口香菜種子之檢測結果-------------------------------
--------------------1-33 2-2-5-1.種媒病原菌之萃取與使用鑑別性培養基分離之
結果-------------1-33 2-2-5-2.香菜種子高雄-1 分離菌株的鑑定結果-------------
------------------1-35 2-2-5-3.香菜種子高雄-1 分離菌株之分子檢測結果--------
-----------------1-36 2-2-5-4.香菜種子高雄-1 分離菌株之分子類型區分結果---
----------------1-36 3.以 PCR 檢測種子中之種媒病原菌-----------------------------
-----------------------1-36 3-1.所使用的引子對之篩選結果----------------------------
--------------------------1-36 3-2.香菜病葉分離菌株 Xanthomonas. campestris pv.
coriandri Tc3 與種子混合後合之 PCR 敏感度測試結果----------------------------
---------------------------------1-37 討論--
---------------------------------------1-39 參考文獻--
---1-46 圖表---------------------------------------
---1-51 目 錄 第二章 Xanthomonas 屬
植物病原細菌之分子類型區分方法之研究 中文摘要-----------------------------
---2-1 英文摘要---------------------

	--2-2 前言--------------------- --2-3 材料與方法---- --2-9 1.Xanthomonas 屬植物病原細菌之分子類型區分之研究-------------------------- ---2-9 1-1.菌種的取得、培養條件、儲存的方法------------------------------------- ----------2-9 1-2.IS 種類、引子對設計與選殖-- ---------------2-9 1-3.病原菌的分子類型 (molecular typing)--------------------------- ---------------2-11 1-3-1.IS－RFLP(IS-restriction fragement length polymorphism)- ---------------2-11 1-3-2.脈衝式電泳(pulsed-field gel electrophoresis；PFGE)----- ----------------2-11 2.Xanthomonas 屬植物病原細菌之分子類型區分之應用---- -----------------------2-12 2-1.香菜葉片分離菌株的取得、培養條件、儲存的 方法---------------------------2-12 2-2.香菜病葉分離之菌株鑑定--------------------- -----------------------------------2-13 2-2-1.生理生化測試-------------------------------- ----------------------------------2-13 2-2-2.16S rDNA 基因選殖、定序及比對------ -----------------------------------2-15 2-2-3.對菸草之過敏性測試---------------------- ----------------------------------2-15 2-2-4.分子檢測--------------------------------------- ---------------------------------2-15 2-3.香菜病葉分離之菌株的分子類型 (molecular typing)區分-------------------2-15 2-3-1.脈衝式電泳----------------------- ---2-15 2-3-2.RFLP---------------------------------- ---2-16 2-3-3.PCR-RFLP----------------------------- -------------------------------------2-16 3.基礎分生技術-------------------------------- --2-17 結果-- --2-25 1.Xanthomonas 屬植物病原細 菌之分子類型區分之研究---------------------------2-25 1-3.病原菌的分子類型 (molecular typing)---2-25 1-3-1.IS－RFLP(IS- restriction fragement length polymorphism)---------------2-25 1-3-2.脈衝式電泳 (pulsed-field gel electrophoresis；PFGE)---------------------2-26 2.Xanthomonas 屬 植物病原細菌之分子類型區分之應用---------------------------2-27 2-1.自香菜 細菌性葉枯病的香菜分離菌株---2-27 2-2.香 菜病葉分離之菌株鑑定結果---2-27 2- 2-1.生理生化測試--2-27 2- 2-2.16S rDNA 基因選殖、定序及比對---2-28 2-2-3.對菸草之過敏性測試--2-28 2-2-4.分子檢測---2-28 2-3.香菜病葉分離之菌株的分子類型(molecular typing)區分-------------------2- 29 2-3-1.脈衝式電泳---2- 29 2-3-2.RFLP--2- 30 2-3-3.PCR-RFLP--2-31 討論--- 2-33 參考文獻-- -------2-36 表-- -------------2-41 圖-- -------------------2-58
參考	參考文獻 莊晟榜. 2002. Tween 系列介面活性劑對微生物降解碳氫化合物

文獻	之引響. 私立中原大學化學工程學系碩士學位論文. Andrewes, A. G., Hertzberg, S., Liaaen-Jensen, S., and Starr, M. P. 1973. The Xanthomonas 'carotenoids'-non-carotenoid, brominated, aryl-polyene esters. Acta Chem. Scand. 27:2382-2395. Andrewes, A. G., Jenkins, C. L., Starr, M. P., Shepherd, J., and Hope, H. 1976. Structure of xanthomonadin I, a novel dibrominated arylpolyene pigment produced by the bacterium Xanthomonas juglandis. Tetrahedron Lett. 45:4023-4024. Berg, T., Tesoriero, L., and Hailstones, D. L. 2005. PCR-based detection of Xanthomonas campestris pathovars in Brassica seed. Plant Pathol. 54:416-427. Bradbury, J. F. 1984. Genus Xanthomonas. In Bergey's Manual of Systematic Bacteriology (Krieg, N. R., and Holt, J. G. eds.) 8th ed. vol.1 p. 199-209. The Williams and Wilkins Co., Baltimore. Chet, K. 2002. International Seed Health Initiative-Vegetables. Detection of Xanthomonas axonopodis pv. phaseoli on Bean (Phaseolus vulgaris). Chitarra, L. G., Langerak, C. J., Bergervoet, J. H.W., and van den Bulk, R.W. 2002. Detection of the plant pathogenic bacterium Xanthomonas campestris pv. campestris in seed extracts of Brassica sp. applying fluorescent antibodies and flow cytometry. Cytometry 47:118-126. Claflin, L. E., Vidaver, A. K., and Sasser, M. 1987. MXP, a semi-selective medium for Xanthomonas campestris pv. phaseoli. Phytopathology 77:730-734. Chang, R. J., Ries, S. M., and Pataky, J. K. 1991. Dissemination of Clavibacter michiganensis subs. michiganensis by practices used to produce tomato plants. Phytopathology 81:1276-1281. Clayton, E. E. 1931.Vegetable seed treatment with special reference to the use of hot water and organic mercurials. N. Y. Agric. Exp. Stn. (Geneva), Bull. 183:43. Ghezzi, J. I., and Steck, T. R. 1999. Induction of the viable but nonculturable condition in Xanthomonas campestris pv. campestris in liquid microcosms and sterile soil. FEMS Microbiol. Ecol. 30:203-208. Gitaitis, R., Beaver, R., and Voloudakis, A. 1991. Detection of Clavibacter michiganensis subs. michiganensis in symptomless tomato transplants. Plant Dis. 75:1069-1076. Goszczynsska, T., and Serfontein, J. J. 1998. Milk-Tween agar, a semiselective medium for isolation and differentiation of Pseudomonas syringae pv. syringae, Pseudomonas syringae pv. phaseolicola and Xanthomonas axonopodis pv. phaseoli. J. Microbiol. Methods 32:65-72. Hooper, P., and Dennis, J. 2002. Coriander- overcoming production limitations. RIRDC Publication No 02/147, RIRDC Project No CAG-1A. ISHI-Veg ITG Pepper. International Seed Health Initiative-Vegetables. Detection of Xanthomonas campestris pv. vesicatoria on Pepper (Capsicum annuum). ISHI-Veg ITG Tomato. International Seed Health Initiative-Vegetables. Detection of Xanthomonas campestris pv. vesicatoria on Tomato (Lycopersicon lycopersicum). Jeroen V. B., and Bejo Zaden, B. V. 2002. International Seed Health Initiative-Vegetables. Detection of Xanthomonas campestris pv. carotae on Carrot (Daucus carota). Louws, F. J., Rademaker, J., and de Bruijn, F. 1999. The three Ds of PCR-based genomic analysis of phytobacteria: diversity, detection and disease diagnosis. Annu Rev Phytopathol 37:81-125. Katznelson, H. 1950. The detection of internally-borne bacteria pathogens of beans by a rapid phage-plaque count technique. Science. 112:645-647. Koenraadt, H., van Bilsen, J. G. P. M., and Roberts, S. J. 2005. Comparative test of four semi-

selective agar media for the detection of Xanthomonas campestris pv. campestris in brassica seeds. Seed Sci. Tech. 33:115-125. Lee, Y. A, Liu, Y. H., and Liu, H. L. 2004. First Report of Bacterial Leaf Blight of Coriander Caused by Xanthomonas campestris pv. coriandri in Taiwan. Plant Dis. 88:910. Leite, R. P. JR., Minsavage, G. V., Bonas, U., and Stall, R. E. 1994. Detection and Identification of Phytopathogenic Xanthomonas Strains by Amplification of DNA Sequences Related to the hrp Genes of Xanthomonas campestris pv. vesicatoria. Appl. Environ. Microbiol. 1994:1068-1077. Lundsgaard, T. 1976. A method for detection of Xanthomonas campestris (Pammel) Dowson in brassica seed. Statens Plantetilsyn 21:34-38. McClelland, R. G., and Pinder, A. C. 1994. Detection of low levels of specific Salmonella species by fluorescent antibodies and flow cytometry. J. Appl. Bacteriol. 77:440 – 447. Meng, X. Q., Umesh, K. C., Davis, R. M., and Gilbertson, R. L. 2004. Development of PCR-based assays for detecting Xanthomonas campestris pv. carotae, the carrot bacterial leaf blight pathogen, from different substrates. Plant Dis. 88:1226-1234. Miller, J. H. 1972. Experiments in molecular genetics. Cold Spring Harbor Laboratory, Cold Spring Harbor, N. Y. Muirhead, K. A., Horan, P. K., and Poste, G. 1985. Flow cytometry: present and future. Biotechnology 3:337 – 356. Park, Y. J., Lee, B. M., Ho-Hahn, J., Lee, G. B., and Park, D. S. 2004. Sensitive and specific detection of Xanthomonas campestris pv. campestris by PCR using species-specific primers based on hrpF gene sequences. Microbiol. Res. 159:419- 423. Paveley, N. D., Rennie, W. J., Reeves, J. C., Wray, M. W., Slawson, D. D., Clark, W. S., Cockerell, V., and Mitchell, A. G. 1996. HGCA Research Review No. 34, Home-Grown Cereals Authority, London, UK. Randhawa, P. S., and Schaad, N. W. 1984. Selective isolation of Xanthomonas campestris pv. campestris from crucifer seeds. Phytopathology 74:268-272. Reeves, J. C.1998. Molecular diagnostics for pathogen detection in seed and planting materials. Plant Cell Tissue Organ Cult. 52:33-39. Ribot, E. M., Fitzgerald, C., Kubota, K., Swaminathan, B., and Barrett, T. J. 2001. Rapid pulsed-field gel electrophoresis protocol for subtyping of Campylobacter jejuni. J. Clin. Microbiol. 39:1889 – 1894. Roberts, S. J., Brough, J., Everett, B., and Redstone, S. 2004. Extraction methods for Xanthomonas campestris pv. campestris from brassica seed. Seed Sci. Tech. 32:439-453. Roberts, S. J., and Koenraadt, H. 2003. ISTA-PDC Technical report: Revised method for detection of Xanthomonas campestris pv. campestris in Brassica seed. ISTA Method Validation Reports 1, 1-9. Saettler, A. W., Schaad, N. W., and Roth, D. A. (ed.). 1989. Detection of bacteria in seed and other planting material. American Phytopathological Society, St. Paul, Minn. Sakthivel, N., Mortensen, C. N., and Mathur, S. B. 2001. Detection of Xanthomonas oryzae pv. oryzae in artificially inoculated and naturally infected rice seeds and plants by molecular techniques. Appl. Microbiol. Biotechnol. 56:435-441. Shackleton, D. A. 1962. A method for the detection of Xanthomonas campestris (Pammel 1895) Dowson, 1939, in Brassica seed. Nature 193:78. Shadd, N. W. 1982. Detection of seedborne bacterial plant pathogen. Plant Dis. 10: 885-890. Schaad, N. W. 1989. Detection of Xanthomonas campestris pv. campestris in crucifers. In: Saettler, A. W., Schaad,

N. W., and Roth, D.A., editors. Detection of bacteria in seed and other planting material. St. Paul, MN: The American Phytopathological Society P.68－75. Schaad, N. W., Cheong, S. S., Tamaki, S., Hatziloukas, E., and Panopoulos, N. J. 1995. A combined biological and enzymatic amplification (BIO-PCR) technique to detect Pseudomonas syringae pv. phaseolicola in bean seed extracts. Phytopathology 85:243－ 248. Shadd, N. W., and Donaldson, R. 1980. Comparison of two methods detection of Xanthomonas campestris infected crucifer seed. Seed Sci. Tech. 8:383-392. Schaad, N.W., and Franken, A. A. J. M. 1996. ISTA Handbook on Seed Health Testing Working Sheet No 50 (2nd Ed): Xanthomonas campestris pv. campestris. ISTA, Zurich. Schaad, N. W., and Kendrick. R. 1975. A qualitative method for detecting Xanthomonas campestris in crucifer seed. Phytopathology 65:1034-1036. Schaad, N. W., and Stall, R. E. 1988. Xanthomonas. In Laboratory Guide for identification of plant pathogenic bacteria. 2nd ed. (Schaad, N. W. ed.): 81-94. The American Phytopathological Society. St. Paul. Minnesota. Srinivasan, M. C., Neergaard, P., and Mathur, S. B. 1971. A technique for Xanthomonas campestris in routine seed health testing of crucifers. Seed Sci. Tech. 1:853-859. Srinivasan, M. C., Patel, M. K., and Thirumalachar, M. J. 1961. A bacterial blight disease of coriander. Proc. Indian Acad. Sci. Sect. B 53:298-301. Sutton, M. D., and Wallen, V. R. 1970. Epidemiological and ecological relations of Xanthomonas phaseoli and X. phaseoli var. fuscan on bean in southwestern Ontario 1961-1968. Can. J. Bot. 48:1329-1334. Van Vuurde, J. W. L., Kastelein, P., and Van der Wolf, J. M. 1995. Immuno- fluorescence colony-staining (IFC) as a concept for bacterial and ecological research. EPPO Bull. 25:157－162. Vauterin, L., Hoste, B., Yang, P., Alvarez, A., Kersters, K., and Swings, J. 1993. Taxonomy of the genus Xanthomonas. In Xanthomonas (Swings, J. G., and Civerolo, E. L. eds):157-192. Chapman and Hall Inc. Boundary Row, London. Wakimoto, S. 1954. The determination the presence of Xanthomonas oryzae phage technique. Sci. Bull. Fac. Kyushu Univ. 14：495-498. Walcott, R. R. 2003. Managing seedborne disease in greenhouse transplant production. ASHS Publication Horticultural Technology Vol. 13, No. 1. Yuen, G. Y., Alvarez, A. M., Benedict, A. A., and Trotter, K. J. 1987. Use of monoclonal antibodies to monitor the dissemination of Xanthomonas campestris pv. campestris. Phytopathology 77:366-370. 參考文獻 徐世典, 張東柱, 張清安, 蔡進來, 蔡東纂, 安寶貞, 林長平, 林益昇, 高清文, 郭克忠, 曾國欽, 曾顯雄, 黃振文, 黃德昌, 黃錫東, 陳殿義, 楊秀珠, 詹富智, 蔣慕琰, 鄧汀欽, 謝文瑞, 謝煥儒, 與 顏志恆. 2002. 台灣植物病害名彙. 中華民國植物病理學會. 邱淑萍. 1996. 黃原桿菌插入序列 IS1403 與 IS1404 之選殖、特性分析與分布之研究. 輔仁大學生命科學研究所碩士論文. 劉雅惠. 2003. 十字花科黑腐病菌 virD4 同源基因功能分析、存在情形及其應用. 輔仁大學生命科學研究所碩士論文. 陳冠蓓. 2004. 白色海芋軟腐病之病原菌、分子類型區分、及 pectate lyase 基因的表現及應用. 私立輔仁大學生物學研究所碩士論文. Berthier, Y., Thierry, D., Lemattre, M., and Guesdon, J. L. 1994. Isolation of an insertion sequence (IS1051) from Xanthomonas campestris pv. dieffenbachiae with potential use for strain identification and characterization. Appl. Environ. Microbiol. 60: 377-384.

Bradbury, J. F. 1984. Genus Xanthomonas. In Bergey's Manual of Systematic Bacteriology (Krieg, N. R., and Holt, J. G. eds.) 8th ed. vol.1 p. 199-209. The Williams and Wilkins Co., Baltimore. Calos, M. P., and Miller, J. H. 1980. Transposable elements. Cell 20: 579-595. Charlier, D., Piette, J., and Glansdorff, N. 1982. IS3 can function as a mobile promoter in E. coli. Nucleic Acids Res. 10: 5935-5948. Chiu, S. P., Chang, C. M., Chen, M. Y., and Lee, Y. A. 1996. Genetic diversity among strains of Xanthomonas campestris pv. campestris in Taiwan. Plant Pathol. Bull. 5: 211-212 (Abstract). Collins, C. M., and Gutman, D. M. 1992. Insertional inactivation of an Escherichia coli urease gene by IS3411. J. Bacteriol. 174: 883-888. Comai, L., and Kosuge, T. 1983. Transposabe element that causes mutations in a plant pathogenic Pseudomonas. J. Bacteriol. 154: 1162-1167. Finney, M. 1993. Pulsed-field gel electrophoresis, p. 2.5.9 – 2.5.17. In F. M. Ausubel, R. Brent, R. E. Kingston, D. D. Moore, J. G. Seidman, J. A. Smith, and K. Struhl (ed.), Current protocols in molecular biology, vol. 1. Current Protocols, Greene-Wiley, New York. Galas, D. J., and Chandler, M. 1989. Bacterial insertion sequences. In Mobile DNA. D. E. Berg and M. M. Howe (ed.) : 109-162. American Society for Microbiology, Washington, D. C. Green, L., Miller, R. D., Dykhuizen, D. E., and Hartl, D. L. 1984. Distribution of DNA insertion element IS5 in natural isolates of Escherichia coli. Proc. Natl. Acad. USA 81: 4500-4504. Hanahan, D. 1983. Studies on transformation of Escherichia coli with plasmid. J. Mol. Biol. 166:557-580. Jaurin, B., and Normark, S. 1983. Insertion of IS2 creates a novel amp C promoter in Escherichia coli. Cell 32: 809-816. Johnson, J. M., Weagant, S. D., Jinneman, K. C., and Bryant, J. L. 1995. Use of pulsed field gel electrophoresis for epidemiologic study of Escherichia coli O157:H7 during a food-borne outbreak. Appl. Environ. Microbiol. 61: 2806- 2808. Kearney, B., and Staskawicz, B. J. 1990. Characterization of IS476 and its role in bacterial spot disease of tomato and pepper. J. Bacteriol. 172: 143-148. Krause U., Thomson-Carter, F. M., and Pennington, T. H. 1996. Molecular epidemiology of Escherichia coli O157: H7 by pulsed-field gel electrophoresis and comparison with that by bacteriophage typing. J. Clin. Microbiol. 34: 959-961. Lee, Y. A., and Chiu, S. P. 1995. A repetitive sequence widely distributed in Xanthomonas campestris is an insertion sequence belonging to IS3 family. Plant Pathol. Bull. 4: 201 (Abstract). Lee, Y. A., and Chiu, S. P. 1998. IS1403 and IS1404: analysis and distribution of two new insertion sequences in Xanthomonas campestris. Bot. Bull. Acad. Sin. 39: 231-239. Lee, Y. A., Liu, Y. H., and Liu, H. L. 2004. First Report of Bacterial Leaf Blight of Coriander Caused by Xanthomonas campestris pv. coriandri in Taiwan. Plant Dis. 88: 910. Leyns, F., De Cleene, M., Swings, J. – G., and De Ley, J. 1984. The host range of the genus Xanthomonas. Bot. Rev. 50: 308-356. Lipuma, J. J. 1998. Molecular tools for epidemiologic study of infectious disease. Pediatr. Infect. Dis. J. 17:667-675. Mark, M. T., and Noah, A. R. 2001. Optimal estimationof transpositionrates of insertion sequences for molecular epidemiology. African Journal of Biotechnology 2:710-713. Maslow, J. N., Slutsky, A. M., and Arbeit, R. D. 1993. Application of pulsed-field gel electrophoresis to molecular epidemiology, p. 563 – 572. In D. H. Persing, T. F. Smith, F. C. Tenover, and T. J.

White (ed.), Diagnostic molecular microbiology: principles and applications. American Society for Microbiology, Washington, D.C. Maslow, J. N., and Mulligan, M. E. 1996. Epidemiologic typing systems. Infect. Control Hosp. Epidemiol. 17: 595-604. Miller, J. H. 1972. Experiments in molecular genetics. Cold Spring Harbor Laboratory, Cold Spring Harbor, N. Y. Ribot, E. M., Fitzgerald, C., Kubota, K., Swaminathan, B., and Barrett, T. J. 2001. Rapid pulsed-field gel electrophoresis protocol for subtyping of Campylobacter jejuni. J. Clin. Microbiol. 39:1889－1894. Rubens, C. E., Heggen, L. M., and Kuypers, J. M. 1989. IS861, a group B Streptococcal insertion sequence related to IS150 ans IS3 of Escherichia coli. J. Bacteriol. 171: 5531-5535. Sawyer, S. A., Dykhuizen, D. E., Dubose, R. F., Green, L., Mutangadura-Mhlanga, T., Wolczyk, D. F., and Hartl, D. L. 1987. Distribution and abundance of insertion sequences among natural isolates of Escherichia coli. Genetics 115: 51-63. Srinivasan, M. C., Patel, M. K., and Thirumalachar, M. J. 1961. A bacterial blight disease of coriander. Proc. Indian Acad. Sci. Sect. B 53:298-301. Steffens, L., Franke, S., Nickel, S., Schwarzkopf, A., Flemmig, T. F., and Karch, H. 1994. DNA-fingerprinting of Eikenella corrodens by pulsed-field gel electrophoresis. Oral Microbiol. Immunol. 9:95-98. Struelens, M. J. 1998. Molecular epidemiologic typing systems of bacterial pathogens: Current Issues and Perpectives. Mem Inst Oswaldo Cruz, Rio de Janeiro. 93:581- 585. Tanaka, M. M., and Rosenberg, N. A. 2001. Optimal estimationof transpositionrates of insertion sequences for molecular epidemiology. Afr. J. Biotechnol. 2:710-713. Tenover, F. C., Arbeit, R. D., Goering, R. V., Mickelsen, P. A., Murray, B. E., Persing, D. H., and Swaminathan, B. 1995. Interpreting chromosomal DNA restriction patterns produced by pulsed-field gel electrophoresis: criteria for bacterial strain typing. J. Clin. Microbiol. 33:2233－2239. Tenover, F. C., Arbeit, R. D., and Goering, R.V, the Molecular Typing Working Group of the Society for Healthcare Epidemiology of America. 1997. How to select and interpret molecular typing methods for epidemiological studies of bacterial infections: a review for healthcare epidemiologists. Infect. control hosp. epidemiol. 18:426-439. Tu, J., Wang, H. R., Chang, S. F., Charng, Y. C., Lurz, R., Dobrinski, B., and Wu, W. C. 1989. Transposable elements of Xanthomonas campestris pv. citri originating from indigenous plasmid. Mol. Gen. Genet. 217: 505-510. Vanden Mooter, M., and Swings, J. 1990. Numerical analysis of 295 phenotypic features of 266 Xanthomonas strains and related strains and an improved taxonomy of the genus. Int. J. Syst. Bacteriol. 40: 348-369. Vauterin, L., Hoste, B., Kersters, K., and Swings, J. 1995 Reclassification of Xanthomonas. Int. J. Syst. Bacteriol. 45: 472-489. Vauterin L., Swings, J., Kersters, S., Gillis, M., Mew, T. W., Schroth, M. N., Palleroni, N. J., Hildebrand, D. C., Stead, D. E., Civerolo, E. L., Hayward, A. C., Maraite, H., Stall, R. E., Vidaver, A. K., and Bradbury, J. F. 1990. Towards an improved taxonomy of Xanthomonas. Int. J. Syst. Bacteriol. 40: 312-316. Wood, M. S., Byrne, A., and Lessie, T. G. 1991. IS406 and IS407, two gene-activating insertion sequences from Pseudomonas cepacia. Gene 105: 101-105. 限制酵素： New England Biolabs 2005-2006 p.277 Restriction Digest Tool - http://www.tigr.org/tigr-

	scripts/CMR2/restrict_display.pl
論文頁數	204
附註	
全文點閱次數	
資料建置時間	
轉檔日期	
全文檔存取記錄	
異動記錄	M admin Y2008.M7.D3 23:18 61.59.161.35