

乳品病原性微生物生物晶片之開發

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摘要

乳品是營養價值很高的食品，市面上有許多的乳品深受消費者的喜愛。然而，乳牛成長的區域到榨乳與製成商品的工廠，皆可能有多種人畜共通且與環境汙染相關的病原性微生物存在，而這些乳品微生物所造成的經濟損失很大；新鮮度又是乳品類講求的重點，從製程到上市的時間非常短，因此發展快速準確的鑑定方法亦是非常重要的。本研究的目的在於針對容易造成乳品質害之微生物Brucella spp.、Acinetobacter spp.、Pseudomonas spp.、Aeromonas spp.、Enterobacter spp.、Moraxella catarrhalis和Pasteurella multocida進行PCR引子組與DNA探針的設計，初步完成可同時檢測數種影響乳品質之病原性與環境微生物屬的生物晶片，並且沒有交叉雜合的反應出現。未來只要將欲偵測的特異性探針加入即可擴增鑑定之菌屬。另外，食品中毒案件發生的原因很多，但其形成和危害常與食因性病原微生物息息相關。其中常見的包含有Salmonella屬、Staphylococcus aureus、Listeria monocytogenes及Escherichia coli等菌株。食品中毒案件之中通常具有一種以上的病原菌存在，普遍又因環境存在相關之病原菌間接汙染到食品本身。因此本研究測試了經由人工方式所污染的食品，在PCR靈敏度試驗方面，未經預培養直接檢測下，訊號約可達到 $N \times 10^3 \sim 10^4$ cfu/ml，並利用multiplex PCR搭配本研究所建立的檢測晶片離型進行雜合實驗。依照圖譜結果顯示，可成功區分出Listeria monocytogenes、Staphylococcus aureus、Streptococcus agalactiae、Enterobacter sakazakii、Escherichia coli O157:H7、Vibrio parahaemolyticus、Salmonella spp.以及Pseudomonas fluorescens，即使在其他非目標菌株的存在之下，不論是對於單一或是混和菌株的試驗中，都可以得到不受干擾的特異性訊號，此一生物晶片系統的建立，可用於食品上對於常見之病原菌的例行檢測，達到快速又準確的檢測結果。

關鍵詞：乳品、病原性微生物、生物晶片

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