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The optimization of conditions in gold nanoparticles assisted multiplex PCR 優化納米金粒子所輔助的聚合酶連鎖反應

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The Optimization of Conditions in Gold Nanoparticles Assisted Multiplex PCR



GOLD PRIZE AWARD

Polymerase chain reaction (PCR) is a molecular technique in which a segment of DNA is replicated to make millions of copies. PCR is commonly used in various applications such as the diagnosis and monitoring of genetic diseases, the identification of specific bacteria and viruses, and in studies of the functions of targeted segments. However, there are limitations to the PCR process, as the sample source may contain PCR inhibitors or unspecific/degrading DNA segments.

This project focuses on developing a new protocol of duplex PCR that uses optimised AuNPs-assisted conditions. A series of PCR conditions are tested, and the effects of AuNPs on the performance of duplex PCR are evaluated in terms of efficiency and specificity.

The tests find that gold nanoparticles (AuNPs) in concentrations of 0.001 nM can significantly improve the efficiency of PCR. Measurements of the intensity of the amplification band (as revealed by electrophoresis) show that specific products are significantly increased (by 93%) after as few as 20 amplification cycles, with the use of DNA templates as low as 500 fg/ul. However, no significant improvement in the reaction's specificity or sensitivity is demonstrated in this study. Another finding is that under certain conditions, AuNPs can provide both enhancing and inhibitory effects on the specificity, sensitivity and efficiency of PCR.

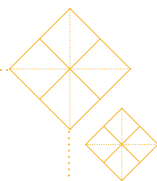
This project provides useful information for developing a new duplex PCR protocol for the rapid detection of pathogens in food and in human specimens.

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優化納米金粒子所輔助的 聚合酶連鎖反應



金獎

聚合酶連鎖反應 (PCR) 屬於分子生物技術，能利用僅僅一節段 DNA 複製出數以百萬計複本。PCR 廣泛應用於不同領域，包括診斷及監察遺傳疾病、辨別特定細菌與病毒、研究目標 DNA 節段的功能等等。然而，由於樣本源可能含有 PCR 抑制因子或不明 / 會自然降解的 DNA 節段，因此這項技術目前仍有局限。

本項目旨在開發一套以優化金納米粒子為輔助條件的全新雙重 PCR 協定，過程中測試了一系列 PCR 條件，並精確衡量了雙重 PCR 在效能與特異性方面的表現。

測試發現，金納米粒子濃度只需達到 0.001 個納米，便能顯著提升 PCR 的效能。從擴增帶數的強度讀數 (利用電泳顯示) 可見，只需使用低至 500 fg/ul 的 DNA 樣板，在少至 20 個擴增循環後，特定產物便會明顯增長 (增幅 93%)，不過反應特異性與敏感度並沒有顯著提升。此外，研究的另一發現是，在特定條件下，金納米粒子能夠同時提升並抑制 PCR 的特異性、敏感度及效能。

本項目的發現，將有助開發全新的雙重 PCR 協定，從而應用於快速檢測食物及人體樣本的病原體。

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