新城病磁珠化診斷試劑之研製

製劑研究組
江 翰　助理研究員

摘要

現今的新城病快速診斷試劑，大多以定性為主。然其敏感性或其專一性，有時會因檢體個別差異造成判定上的不一致性。有鑑於此，本研究分別採用免疫磁珠化抗體-抗原親和性及基因增幅技術 (genomic amplification techniques ; GAT) 兩種策略進行新城病磁珠化診斷試劑的研製，以縮減病原檢測的空窗期，亦可減少偽陽性及操作程序繁鎖的缺點。本實驗分別採用免疫磁珠化抗體-抗原親和性、免疫共沈澱技術 (Co-immunoprecipitation ; Co-IP) 及基因增幅技術(GAT ; genomic amplification techniques)等策略進行新城病磁珠化診斷試劑的研製進行探討，增進其敏感度並減少試劑在判讀方面的不一致性。因其主要抗原性區間為 Np gene, HN gene 及 F gene。其中 F gene 的變異性較高，故以 HN-F 及 Np-HN 複合性抗原決定位融合蛋白作為其抗原。以原核表現系統進行融合蛋白產製，均須經重新折疊後才能成為可溶性抗原。微量生物標記檢測技術之開發，一直是醫檢研究的重點之一。利用磁性奈米粒子作為待測生物標記的標示物，並使用高溫超導磁性免疫分析儀，透過磁減量檢測技術之超高靈敏度檢測平台。結果顯示，磁減量生醫檢測平台的靈敏度，與酵素免疫分析法相較下，高出至少 10 倍。而磁性酵素免疫分析法靈敏度亦至少較酵素免疫分析法至少高出 5 倍，在時間上若不計入磁珠化抗體接合的時間，約可提升 15％的時效。此表示磁珠化試劑，具有作為早期檢測之應用潛力。

Developement the Newcastle Disease immuno- magnetic bead diagnostic reagent

Han Chiang

Abstract

The Newcastle disease diagnostic reagent major in identified the viral antigen or it antibody properties had bias for its specificity and sensitivity what due to difference specimens or components. Because of these issues we used two stratagems to develop and study the bead-linked magnetized diagnostic reagent productivity by immuno-magnetic bead methods and genomic amplification technique that not only reduce the latency of NDV diagnosis and avoid the complicated procedure but also enhance the accuracy and specificity.This study adopted the affinity immunomagnetized antibody and antigen, co-immunoprecipitation, and genoamplification techniques aim to produce a Newcastle disease magnet bead diagnostic reagent for improving and enhancing the diagnostic specificity and sensibility. Since the major antigenicity regions are located on Np , HN and F genes, and the F gene is highly variation, we constructed both HN-F and Np-HN fusion proteins as antigens that conversely expressed in the inclusion body forms by prokaryote express system and should be refolded to become soluble proteins. Until now, we have carried out the expression, purification, stabilizing test and preparation of hybridoma of HN-F protein. Development of the micro biological markers examination technology of tracing has been the medicine examines one of research key points. Using magnetic nanoparticles as biomarkers tested marker and use the high-temperature superconducting magnetic immunoassay analyzer, through the reduction of detection of ultra-high magnetic sensitivity of detection platforms. According the result, that the magnetic platform of reducing the sensitivity of biomedical testing, and enzyme immunoassay for comparison, at least 10 times higher. The magnetic sensitivity of ELISA has at least compared with enzyme immunoassay for at least 5 times higher, if not included in the bead antibody bonding time is saved about 15％ of the time. The bead reagent has potential applications while detection the early stage of NDV infection.