

建立牛結節疹野外毒及疫苗毒之分型即時聚合酶連鎖反應

疫學研究組

丁履紉 副研究員

摘要

牛結節疹 (LSD) 對牛隻健康及經濟影響甚鉅，WOAH 視為應通報的疾病。LSD 減毒活毒疫苗具有良好保護力，能有效控制疾病蔓延，缺點是可能產生輕微或全身皮膚反應，與野外病毒感染病徵無法分別。因此，本實驗針對野外病毒株與疫苗病毒株 ankyrin repeat protein 基因 71 個鹼基缺損差異處，設計各別專一型的特異性引子與探針，開發成可檢驗疫苗毒 (Vac) 和疫苗重組野外病毒 (WV) 的分型雙重即時聚合酶連鎖反應 (DIVA dual qPCR)。經驗證 qPCR-Vac 及 qPCR-WV 檢測能力與 WOAH 推薦痘病毒屬不分型 qPCR-capri 之吻合度 κ 值分別為 0.92 及 0.97，表示一致性皆極高。合成陽性標準寡核苷酸以 qPCR-Vac 及 qPCR-WV 檢測，敏感度皆可達 10 個拷貝數。因此，新開發的套組為可靠的工具，搭配於疫苗接種後期間，即時診斷動物感染野外毒與否，俾利防疫措施依據。

Development of dual DIVA real-time PCR to differentiate between virulent and vaccine strains of lumpy skin disease virus

Lu-Jen Ting

Abstract

Lumpy skin disease (LSD) is a notifiable disease of World Organisation for Animal Health (WOAH) due to its significant health and economic impact on cattle. LSD attenuated live vaccines have satisfactory protection and can effectively control the spread of the disease. However, in some cases the vaccine strain may cause mild or systemic skin reactions, which are indistinguishable from the infection of virulent field strain. In this study, it was found that the virulent virus carries 71 bases less than the vaccine strain in the ankyrin repeat protein gene and this feature allows to design specific primers and probes for each specific DIVA dual real-time polymerase chain reaction, to differentiate vaccine strain (by qPCR-Vac) from **vaccine-like recombinant LSDVs** (by qPCR-WV). It was verified that the detection ability of qPCR-Vac and qPCR-WV with the WOAH recommended capripoxvirus qPCR (qPCR-capri) was 0.92 and 0.97, respectively, indicating that the agreement were both high. The synthetic positive control oligonucleotide was detected by qPCR-Vac and qPCR-WV, both with sensitivity up to 10 copies. Therefore, the newly developed DIVA qPCRs are reliable to check clinical signs timely and to allow authorities to control the disease.