豬瘟抗體 ELISA 檢測方法之建立

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摘要

豬瘟為高傳染性及高致病性之豬隻重要病毒性疾病,為免疫情爆 發嚴重影響國內養豬產業,我國以 LPC 疫苗免疫來防範豬瘟的發生。 測定豬瘟抗體力價,可評估疫苗保護效力亦可藉由小豬移行抗體高低 來調整豬瘟免疫適期。豬瘟抗體力價測定是以血清中和試驗為主,傳 統中和試驗檢測需經培養細胞、螢光抗體染色及肉眼判讀等繁瑣步 驟,耗時三天。替代方法 ELISA 抗體檢測則操作步驟單純,容易上 手,僅需半天時間。為提升豬瘟抗體檢測效能,本所與高雄醫學大學 共同合作以哺乳類細胞株量產豬瘟病毒 E2 醣蛋白, 開發豬瘟病毒 E2 抗體 ELISA 檢測方法。相較於昆蟲細胞及酵母菌表現系統,哺乳類細 胞是最高等表現系統,具有最佳的轉譯後修飾,保有最好的抗原性, 可提高抗體檢測之準確度。為進一步了解自製豬瘟抗體 ELISA 原型 套組與中和抗體間之相關性,本試驗比較不同血清中和抗體力價與間 接型(indirect)及阻斷型(blocking) ELISA 檢測之相關性,結果顯 示相關係數 r 值分別為 0.911 及 0.877。

Development of an ELISA for detection of antibodies against classical swine fever

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Abstract

Classical swine fever (CSF) is a highly contagious and often fatal viral disease of pigs. To avoid outbreaks of the disease and affecting the pig industry, the LPC vaccine was used to prevention of classical swine fever in swine herds in Taiwan. Classical swine fever antibody titers can be used to assess the immune responses and vaccine efficacy; also to adjust the appropriate vaccine program by means of detecting the maternal antibody titer in piglets. Serum neutralization test (SNT) is a major method for measurement of anti-CSF antibody titer. Traditional SNT is a time-consuming and heavy labor job, including cell culture, fluorescent antibody staining and visual interpretation, which takes 3 days. Otherwise, it is a simple procedure and easy operation to detect anti-CSF antibody by ELISA. To enhance the efficiency of classical swine fever antibody detection, we cooperated with the Kaohsiung Medical University using a mammalian cell line for high level of production of glycoprotein E2 of CSF virus and constituted an anti-E2 antibody diagnostic ELISA. Compared to insect cells and yeast expression system, mammalian system is the most advanced protein expression system. It has the best post-translational modifications of proteins, retention of the best antigenicity, and improved the binding of anti-E2 antibody. To further understand the correlation between the ELISA and the SNT, we used different neutralizing antibody titers of pig sera to analyze the homemade indirect ELISA and blocking ELISA. The results showed that the r value of correlation coefficient is 0.911 and 0.877 respectively.