Fig. 6. Western blot analysis of Tir and EspA expression in wild type and rpoS mutants. Cultures were grown aerobically at 37°C in LB media supplemented with 44mM NaHCO3 to OD600=1.5 or in DMEM media in 5% CO2 (two known LEE-induction conditions). Cell pellets were resuspended in SDS loading buffer and boiled for 5 min. Resultant cell extracts were resolved on 10% SDS-PAGE gel. Proteins were transferred to a PVDF membrane by electrophoresis, followed by incubation of the membrane with anti-Tir or anti-EspA specific antibody. Signals were detected using ECL solution and Hyperfilm-ECL film (Amersham).