牛痘病毒蛋白複合物在病毒進入細胞過程中扮演之角色

Investigation of components of vaccinia viral protein complex involved in virus entry

中文摘要

英文摘要

Vaccinia virus is large DNA virus with an envelope that mediates viral fusion with host membrane during virus entry process. Vaccinia mature virus contains multiple envelope protein that are known to play a role in virus attachment. Viral H3, A27 and D8 proteins bound to cell surface glycosaminoglycans and viral A26 protein bound to an extracellular matrix protein laminin. In addition, A26 and A27 protein forms a protein complex on mature virions through disulfide bonds formed between Cys441/442 of A26 protein and Cys71/72 of A27 protein.

The proposal continued the above study and we have generated a mutant vaccinia virus containing a C441/442A double mutant A26 protein in order to address the importance of this disulfide bond in virus biology. The result showed that, in the absence of the intermolecular disulfide bond, A26 protein remained bound to A27 protein and was incorporated into mature virions suggesting that disulfide bond formation between A26 and A27 protein is not essential for A26-A27 complex formation.

In addition to mediating virion attachment to cells, viral A27 was shown to interact with A17 protein to mediate cell fusion. In order to delineate how these viral envelope proteins mediate membrane fusion we established a lentiviral expression system to express viral A17 and A27 protein, individually or in combinations. HeLa cells transduced with lentiviruses expressing A27 protein was isolated and enriched for a high level of A27 protein. This A27-expressing HeLa cell clone, A27-HeLa, complemented the genetic defect of A27 mutant virus in cell cultures, validating the lentiviral expression approach. When we tested the cell fusion activity of viral A17 and A27 proteins using the above expression system although A17 and A27 form a complex in cells no obvious cell fusion was detected. Immunofluorescence analysis showed that both A17 and A27 proteins were cytoplasmic with no cell surface expression. The results thus suggest that viral envelope proteins contain different structures such that they are recognized and processed differently from cellular proteins in cells.