Charaterization of white spot syndrome virus (WSSV)structural protein VP11(ORF394)

熊慧叡、張雲祥

E-mail: 321850@mail.dyu.edu.tw

ABSTRACT

White spot syndrome virus (WSSV) is a large enveloped virus. The WSSV viral particle consists of three structural layers that are surrounding its core DNA: an outer envelope, a tegument and a nucleocapsid. This study characterized a WSSV structural protein, VP11 (WSSV-T1 ORF394, GenBank accession no. AF440570). Immuno electromicroscopic analysis and Western blot hybridization of the intact viral particles and the separated viral components, the results showed that VP11 is an envelope protein. Membrane topology predition demonstrated that VP11 is a type transmembrane protein with a highly hydrophobic transmembrane domain at its N-terminal, and a C-terminal exposed on the virion surface. Immunofluorescence assay performed on VP11 transfected Sf9 cells demonstrated a similar result. However, trypsin digestion analysis of virions gave a controversial conclusion. Binary Co-immunopresipitation assays performed between VP11 and other a major WSSV structural proteins, respectively, VP11 was found to interact with both VP51A and itself. Yeast two-hybrid assay revealed that VP11 possessed autoactivation ability. We hypothesized that VP11 might act as a transcriptional factor. VP11 thus might play an important role in the replication of WSSV, not only in viral particle assembly but also in regulating at viral gene expression.

Keywords : white spot syndrome virus (WSSV)、VP11、structural protein、protein interaction、autoactivation

Table of Contents

表2. 構築於細胞中表現蛋白質載體引子66 表3. 構築蛋白質表現載體引子67 表4. 構築於酵母菌中表現蛋白質載體引子68 表5. 共免疫沉澱分析蛋白質交互作用統整表69

REFERENCES

1.周宗錄，2007。蝦白點症病毒結構性蛋白VP51A（ORF294）特性分析。私立大葉大學分子生物科技學系碩士論文。 2.李媛婷，2008。白點症病毒結構性蛋白質VP51A（ORF294）與其它結構蛋白質之交互作用研究。私立大葉大學分子生物科技學系碩士論文。 3.Campadelli-Fiume, G., M. Amasio, E. Avitabile, A. Cerretani, and C. Forghieri, T. Gianni, and L. Menotti. 2007. The multipartite system that mediates entry of herpes simplex virus into the cell. Rev Med Virol. 17:313-326. 4.Chang, Y. S., C. F. Lo, S. E. Peng, K. F. Liu, C. H. Wang, and G. H. Kou. 2002. White spot syndrome virus (WSSV) PCR-positive Artemia cysts yield PCR-negative nauplii that fail to transmit WSSV when fed to shrimp postlarvae. Dis. Aquat. Org. 49(1):1-10. 5.Chang, Y. S., W. J. Liu, C. C. Lee, T. L. Chou, Y. T. Lee, T. S. Wu, J. Y. Huang, W. T. Huang, T. L. Lee, G. H. Kou, H. J. Wang, and C. F. Lo. 2010. A 3D Model of the Membrane Protein Complex Formed by the White Spot Syndrome Virus Structural Proteins. PLoS One. 5(5):e10718. 6.Chang, Y. S., W. J. Liu, T. L. Chou, Y. T. Lee, T. L. Lee, W. T. Huang, G. H. Kou, and C. F. Lo. 2008. Characterization of white spot syndrome virus envelope protein VP51A and its interaction with viral tegument protein VP26. J. Virol. 82(24):12555-12564. 7.Chazal, N., and D. Gerlier. 2003. Virus entry, assembly, budding, and membrane rafts. Microbiol. Mol Biol Rev. 67:226-237. 8.Chen, L. L., C. F. Lo, Y. L. Chiu, C. F. Chang, and G. H. Kou. 2000. Natural and experimental infection of white spot syndrome virus (WSSV) in benthic larvae of mud crab Scylla serrata. Dis. Aquat. Org. 40:157-161. 9.Chen, L. L., H. C. Wang, C. J. Huang, S. E. Peng, Y. G. Chen, S. J. Lin, W. Y. Chen, C. F. Dai, H. T. Yu, C. H. Wang, C. F. Lo, and G. H. Kou. 2002a. Transcrip-tional analysis of the DNA polymerase gene of shrimp white spot syndrome virus. Virology 301:136-147. 10.Chen, L. L., J. H. Leu, C. J. Huang, C. M. Chou, S. M. Chen, C. H. Wang, C. F. Lo, and G. H. Kou. 2002b. Identification of a nucleocapsid protein (VP35) gene of shrimp white spot syndrome virus and characterization of the motif important for targeting VP35 to the nuclei of transfected insect cells. Virology 293:44-53. 11.Chen, W. Y., K. C. Ho, J. H. Leu, K. F. Liu, H. C. Wang, G. H. Kou, and C. F. Lo. 2008. WSSV infection activates STAT in shrimp. 32(10):1142-1150. 12.Chen, W. Y., K. C. Ho, J. H. Leu, K. F. Liu, H. C. Wang, G. H. Kou, and C. F. Lo. 2008. WSSV infection activates STST in shrimp. Dev Comp Immunol. 32(10):1142-1150. 13.Davis, S., B. Bozon, and S. Laroche. 2003. How necessary is the activation of the immediate early gene zif268 in synaptic plasticity and learning? Behav Brain Res 142(1-2):17-30. 14.Flegel, T. W. 1997. Major viral diseases of black tiger prawn (Penaeus monodon) in Thailand. World J. Microbiol. Biotechnol. 13:433-442. 15.Inouye, K., S. Miwa, N. Oseko, H. Nakano, and T. Kimura. 1994. Mass mortali-ties of cultured kuruma shrimp, Penaeus japonicus, in Japan in 1993: electron mi-croscopic evidence of the causative virus. Fish Pathol. 29:149-158. (In Japanese.) 16.Kang, S. T., J. H. Leu, H. C. Wang, L. L. Chen, G. H. Kou, and C. F. Lo. 2009. Polycistronic mRNAs and internal ribosome entry site elements (IRES) are widely used by white spot syndrome virus (WSSV) structural protein genes. Virology. 387:353-363. 17.Li, L. J., J. F. Yuan, C. A. Cai, W. G. Gu, and Z. L. Shi. 2006. Multiple envelope proteins are involved in white spot syndrome virus (WSSV) infection in crayfish. Arch Virol. 151:1309 – 1317. 18.Li, Z., Q. Lin, J. Chen, J. L. Wu, T. K. Lim, S. S. Loh, X. Tang, and C. L. Hew. 2007. Shotgun identification of the structural proteome of shrimp white spot syn-drome virus and iTRAQ differentiation of envelope and nucleocapsid subprote-omes. Mol Cell Proteomics. 6:1609-1620. 19.Liu, W. J., Y. S. Chang, H. C. Wang, J. H. Leu, G. H. Kou, and C. F. Lo. 2008. Transactivation, dimerization, and DNA-binding activity of white spot syndrome virus immediate-early protein IE1. J Virol. 82(22):11362-11373. 20.Lo, C. F., and G. H. Kou. 1998. Virus-associated white spot syndrome of shrimp in Taiwan: a review. Fish Pathol. 33:365-371. 21.Lo, C. F., C. H. Ho, S. E. Peng, C. H. Chen, H. C. Hsu, Y. L. Chiu, Y. T. Chen, C. F. Chang, K. F. Liu, M. S. Su, C. H. Wang, and G. H. Kou. (1996a). Infection of white spot syndrome associated virus (WSBV) in cultured and wild-caught shrimps, crabs and other arthropods. Dis. Aquat. Org. 27:215-225. 22.Lo, C. F., C. H. Ho, S. E. Peng, C. H. Chen, H. E. Hsu, Y. L. Chiu, C. F. Chang, K. F. Liu, M. S. Su, C. H. Wang, and G. H. Kou. 1996b. White spot syndrome as-sociated virus (WSBV) detected in cultured and captured shrimp, crabs and other arthropods. Dis. Aquat. Org. 27:215-225. 23.Lo, C. F., J. H. Leu, C. H. Ho, C. H. Chen, S. E. Peng, Y. T. Chen, C. M. Chou, P. Y. Yeh, C. J. Huang, H. Y. Chou, C. H. Wang, and G. H. Kou. 1996c. Detection of baculovirus associated with white spot syndrome (WSBV) in penaeid shrimps using polymerase chain reaction. Dis. Aquat. Org. 25:133-141. 24.Mettenleiter, T. C. 2004. Budding events in herpesvirus morphogenesis. Virus Res. 106:167 – 180. 25.Mettenleiter, T. C., B. G. Klupp, and H. Granzow. 2006. Herpesvirus assembly: a tale of two membranes. Curr Opin Microbiol. 9:423-429. 26.Nakano, H., H. Koube, S. Umezawa, K. Momoyama, M. Hiraoka, K. Inouye, and N. Oseko. 1994. Mass mortalities of cultured kuruma shrimp, Penaeus japonicus, in Japan in 1993: epizootiological survey and infection trails. Fish Pathol. 29:135-139. 27.Rajc?鴨i, J. 2003. Molecular mechanisms of virus spread and virion components as tools of virulence. A review. Acta Microbiol Immunol Hung. 50:407-431. 28.Reske, A, G. Pollara, C. Krummenacher, B. M. Chain, and D. R. Katz. 2007. Un-derstanding HSV-1 entry glycoproteins. Rev Med Virol. 17:205-215. 29.Tsai, J. M., H. C. Wang, J. H. Leu, A. H. Wang, Y. Zhuang, P. J. Walker, G. H. Kou, and C. F. Lo. 2006. Identification of the nucleocapsid, tegument, and envelope proteins of the shrimp white spot syndrome virus virion. J Virol. 80:3021-3029. 30.Tsai, J. M., H. C. Wang, J. H. Leu, H. H. Hsiao, A. J. Wang, G. H. Kou. and C. F. Lo. 2004. Genomic and proteomic analysis of thirty-nine structural proteins of shrimp white spot syndrome virus. J Virol. 78:11360-11370. 31.van Hulten, M. C. W., J. Witteveldt, S. Peters, N. Kloosterboer, R. Tarchini, M. Fiers, H. Sandbrink, R. K. Lankhorst, and J. M. Vlak. 2001. The white spot syn-drome virus DNA genome sequence. Virology 286:7-22. 32.Vlak, J. M., J. R. Bonami, T. W. Flegel, G. H. Kou, D. V. Lightner, C. F. Lo, P. C. Loh, and P. W. Walker. (2005). Nimaviridae. In: VIIIth Report of the International Committee on Taxonomy of Viruses.

(Fauquet, C. M.., M. A. Mayo, J. Maniloff, U. Desselberger, and L. A. Ball.) Elservier. 32:187-192. 33.Wang, C. H., C. F. Lo, J. H. Leu, C. M. Chou, P. Y. Yeh, H. Y. Chou, M. C. Tung, C. F. Chang, M. S. Su, and G. H. Kou. 1995. Purification and genomic analysis of baculovirus associated with white spot syndrome (WSBV) of Panaeus monodon. Dis. Aquat. Org. 23:239-242. 34.Wang, C. H., C. F. Lo, J. H. Leu, C. M. Chou, P. Y. Yeh, H. Y. Chou, M. C. Tung, C. F. Chang, M. S. Su, and G. H. Kou. 1995. Purification and genomic analysis of baculovirus associated with white spot syndrome (WSBV) of Penaeus monodon. Dis. Aquat. Org. 23:239-242. 35.Wongteerasupaya, C., J. E. Vickers, S. Sriurairatana, G. L. Nash, A. Akarajamorn, V. Boosaeng, S. Panyim, A. Tassanakajon, B. Withyachumnarnkul, and T. W. Fle-gel. 1995. A non-occluded, systemic baculovirus that occurs in the cells of ecto-dermal and mesodermal origin and causes high mortality in the black tiger prawn Penaeus monodon. Dis. Aquat. Org. 21:69-77. 36.Wu, W., L. Wang, and X. Zhang. 2005. Identification of white spot syndrome vi-rus (WSSV) envelope proteins involved in shrimp infection. Virology 332:578-583. 37.Xie, X., and F. Yang. 2006. White spot syndrome virus VP24 interacts with VP28 and is involved in virus infection. J. Gen. Virol. 87:1903-1908. 38.Xie, X., L. Xu, and F. Yang. 2006. Proteomic analysis of the major envelope and nucleocapsid proteins of white spot syndrome virus. J Virol. 80:10615-10623. 39.Yang, F., J. He, X. Lin, Q. Li, D. Pan, X. Zhang, and X. Xu. 2001. Complete ge-nome sequence of the shrimp white spot bacilliform virus. J. Virol. 75:11811-11820. 40.Zhang, J. S., S. L. Dong, Y. W. Dong, X. L. Tian, and C. Q. Hou. 2008. Bioassay evidence for the transmission of WSSV by the harpacticoid Nitora sp. J Invertebr Pathol. 97(1):33-39. 41.Zhang, X., C. Huang, X. Tang, Y. Zhuang, and C. L. Hew. 2004. Identification of structural proteins from shrimp white spot syndrome virus (WSSV) by 2DE-MS. Proteins. 55:229-235.