

Shrimp possesses a conserved apoptosis mechanism by which the caspase-3 activity is modulated by HtrA2 and IAP.

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Abstract

Shrimp has a defense mechanism for suppressing viral replication by promoting apoptosis of infected cells. Recently, *Pmcaspase* and *PmIAP* have been found in *Penaeus monodon*. The interaction of *PmIAP* with *Pmcaspase* leads to the reduction in apoptosis in insect cells. In several organisms, apoptosis can be activated by the binding of HtrA2 proteins to IAP proteins. We have shown for the first time that shrimp, both *Penaeus monodon* and *Litopenaeus vannamei* also possesses HtrA2. The mature form of PmHtrA2 was expressed and used for production of polyclonal antibody specific to HtrA2. To investigate the expression of PmHtrA2 in shrimp, the crude proteins from several shrimp tissues were imaged by Western blot using the polyclonal antibody specific to the recombinant PmHtrA2. The expression of the 47-kDa immature PmHtrA2 protein could be detected in shrimp lysates from the gills and the muscles. We also showed that PmHtrA2 was able to interact with PmIAP and the binding of the two proteins was mediated by the BIR2 domain of PmIAP. In addition, the BIR2 of PmIAP was shown to be able to inhibit Pm caspase activity. The inhibitory effect of the BIR2 domain on Pm caspase was impaired under the presence of the IBM peptide of PmHtrA2, implying a role for PmHtrA2 in apoptosis activation. These results suggested that *P. monodon* possesses a conserved mechanism by which the caspase-3 activity is modulated by HtrA2 and IAP, as previously seen in insects and mammals. When determining the role of LvHtrA2 in WSSV infection, it was found that LvHtrA2 transcription was early up-regulated in the WSSV-infected shrimp at 8h post-infection (p.i.) and expression still remained high at 48 h p.i.. It also demonstrated that dsRNA specific to LvHtrA2 reduced the cumulative mortality in the WSSV-infected shrimp compared with the control group. Additionally, depletion of the LvHtrA2 transcripts reduced expression levels for caspase-3 (Cap-3) gene in shrimp. This result could suggest that LvHtrA2 may involve in apoptosis mediated mortality rather than providing immune protection during WSSV infection.