

1.1 Introduction

Antheraea mylitta cytoplasmic polyhedrosis virus (AmCPV) is one of the most widespread pathogens of Indian non-mulberry silkworm, *A. mylitta*. Almost 20% crop is damaged annually due to this virus attack. A large number of CPV-infected *A. mylitta* larvae develop chronic diarrhea that eventually leads to a condition known as “Grasserie” and the death of the larvae (Jolly et al., 1974).

CPV belongs to the genus *Cypovirus* and family *Reoviridae* (Mertens et al., 2005, Payne and Mertens 1983). CPV infects the midgut of the wide range of insects belonging to the order Diptera, Hymenoptera and Lepidoptera (Bellonick and Mori 1998). Viral infection is often characterized by the production of large number of occlusion body called polyhedra in the cytoplasm of infected cells. Like other members of *Reoviridae* family, CPV genome is also composed of 10 double stranded RNA segments (S1-10) (Payne and Mertens, 1983). A small 11 segment (S11) has been reported in some cases such as *Trychoplusia ni* cytoplasmic polyhedrosis virus (TnCPV-15) (Rao et al., 2000) and BmCPV (Arella et al., 1988). Each dsRNA segment is composed of a plus strand mRNA and its complementary minus strand in an end to end base pair configuration except for a protruding 5' cap on the plus strand.

Among the viruses of the family *Reoviridae*, complete nucleotide sequences of double stranded RNA genome have been reported for the members of the genera *Orthoreovirus*, *Rotavirus*, *Orbivirus*, *Phytoreovirus*, *Coltivirus*, *Seadornavirus* and putative members of *Fijivirus* (Attoui et al., 2005a, 2005b, Cowled et al., 2009, Duncan et al., 1999, Estes et al., 1989, Nakashima et al., 1996, Suzuki, 1995). Blue tongue virus which is a member of genus *Orbivirus* within the family *Reoviridae* has been characterized in details and it has been shown that it has seven structural proteins (VP1-VP7) organized into an outer capsid and an inner capsid (Commonly known as a “core”) containing the ten dsRNA segments. The largest genome segment (L1) encoded VP1 protein which is an RNA dependent RNA polymerase (Boyce et al., 2004, Roy, 2008, Urakawa et al., 1989). Complete nucleotide sequence of type 1 *Dendrolymus punctatus* CPV (DpCPV) (Zhao et al., 2003a 2003b), type-15 TnCPV (49), type-1 BmCPV (Hgiwara et al., 1998a, 1998b, 2000, 2001, 2002, Ikeda et al., 1998) and *Choristoneura occidentalis* CPV (Co-CPV) (Graham et al., 2008) have been reported and deposited in the GenBank. In case of the BmCPV-1, segments 1, 3, 4, 6, 7 and 10 encode structural proteins VP1, VP2, VP3, VP4, VP5 and polyhedrin, respectively, whereas segment 5, 8, and 9 encodes non structural protein, p101, p44 and NS5, respectively (Hgiwara et al., 1998a, 1998b, 2000, 2001, 2002, Ikeda et al., 1998). In case of DpCPV segment 1, 3, 4, 6, 7 and 10 encode viral structural proteins, VP1, VP2, VP3, VP4, VP5, and polyhedrin

respectively while segments 2, 5, encode nonstructural proteins, RNA dependent RNA polymerase and P101, respectively. Segments 8 and 9 encode other nonstructural proteins (Zhao et al., 2003a, 2003b). The sequence analysis revealed that S2 of type1 LdCPV and S1 of type 14 LdCPV codes for a putative viral RdRp (Rao et al., 2003).

The Indian non-mulberry Saturniidae silkworm *A. mylitta* is wild in nature and produces exotic variety of silk called tasar. Being wild in nature, AmCPV infects a major part of this silkworm crop and thereby reduces the yield of tasar silk (Jolly et al., 1974).

The structure and genome of AmCPV have been characterized by electron microscopy and gel electrophoresis which reveal that it is similar to that of a type- 4 CPV (Qanungo et al., 2000) and consists of 11 ds RNA molecules. Molecular and biochemical analysis also showed that the genome segment 6, 7, 8, and 10 of AmCPV encodes viral structural proteins, P68, P61, P60 and polyhedrin respectively, while segment 9 encodes a nonstructural protein, NSP38, having RNA binding property and segment 11 has no open reading frame to encode any protein (Chavali et al., 2007, 2008, Jangam et al., 2006, Qanungo et al., 2002, Sinha-Dutta et al., 2005). The other genome segments of this virus (segments 1-5, 11) have not been characterized at molecular level.

Since, about 8 millions of people in India make their livings on sericulture, development of specific anti-viral compound is very much necessary. This is possible by understanding the function of each genome segment in the viral life cycle or replication process.

The work embodied in this thesis describes molecular characterization (cloning, sequencing and expression) of genome segment 2 (S2) of AmCPV and biochemical analysis of S2 encoded protein and shows that it encodes viral RNA polymerase to transcribe viral RNA. Bioinformatics, Phylogenetic and computational modeling as well as mutational analysis of AmCPV RdRp also shows that it is very similar to other viral RdRps with its own characteristics features and may act as a target site for the development of effective anti-viral compound(s) for the improvement of tasar sericulture in India.