

# Iteradensovirus from the Monarch Butterfly, *Danaus plexippus plexippus*.

Qian Yu, Peter Tijssen

► **To cite this version:**

Qian Yu, Peter Tijssen. Iteradensovirus from the Monarch Butterfly, *Danaus plexippus plexippus*. Genome Announcements, American Society for Microbiology, 2014, 2 (2), pp.1-2. <10.1128/genomeA.00321-14>. <pasteur-01146107>

**HAL Id: pasteur-01146107**

**<https://hal-riip.archives-ouvertes.fr/pasteur-01146107>**

Submitted on 27 Apr 2015

**HAL** is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.



# Iteradensovirus from the Monarch Butterfly, *Danaus plexippus plexippus*

Qian Yu, Peter Tijssen

INRS, Institut Armand-Frappier, Université du Québec, Laval, Quebec, Canada

**The 5,006-nucleotide (nt)-long genome of a new virus from monarch butterfly pupae was cloned and sequenced. It was flanked by inverted terminal repeats (ITRs) of 239 nt with 163-nt hairpins. The monosense genome with three open reading frames is typical of the genus *Iteradensovirus* in the subfamily *Densovirinae* of the family *Parvoviridae*.**

Received 25 March 2014 Accepted 3 April 2014 Published 17 April 2014

Citation Yu Q, Tijssen P. 2014. Iteradensovirus from the monarch butterfly, *Danaus plexippus plexippus*. *Genome Announc.* 2(2):e00321-14. doi:10.1128/genomeA.00321-14.

Copyright © 2014 Yu and Tijssen. This is an open-access article distributed under the terms of the [Creative Commons Attribution 3.0 Unported license](https://creativecommons.org/licenses/by/4.0/).

Address correspondence to Peter Tijssen, peter.tijssen@iaf.inrs.ca.

Monarch butterflies (*Danaus plexippus plexippus*) migrate from eastern and central North America for overwintering in Mexico. Migration of this emblematic butterfly has been in rapid decline in recent years, prompting the presidents of the United States and Mexico and the Prime Minister of Canada to discuss this problem during a meeting in February 2014. Several factors may be responsible for this trend. The cool and relatively moist high mountain habitats of Oyamel fir forests are ideal for both the firs and the butterflies. The forest canopy and the clustering of the monarchs protect them against freezing (1). Severe logging and climate change threaten these forests, and a massive reforestation effort is under way to reverse this trend. Second, the extensive use of genetically modified herbicide-resistant soybeans and corn may be reducing the number of larval host plants, milkweeds, especially in their main habitat in the Corn Belt (2–4), encouraging the suggestion of a milkweed corridor. However, this has been disputed elsewhere (5). Third, pathogens such as bacteria, parasites, and viruses may affect monarch populations (6–8).

Virus was partially purified from three infected pupae obtained from a butterfly farm in Granby (Quebec, Canada) by the method described for *Galleria mellonella* densovirus (9) and visualized by electron microscopy. A preliminary genome characterization was obtained with the sequence-independent single-primer amplification (SISPA) method (10–12), showing two SpeI restriction sites in a preliminary 4.7-kb sequence. Viral DNA was then blunt ended by a mixture of Klenow large-fragment and T4 DNA polymerase, digested with SpeI, and cloned into EcoRV and SpeI sites in the pBluescriptSK II(-) vector, yielding clones with 3.4-kb inserts and clones with 1.5-kb inserts. Sequences of several complete clones, obtained in both directions with Sanger's method (10, 11), were identical except for the flip-flop sequences in the hairpins. The sequence between the two SpeI sites was obtained after PCR amplification with gene-specific primers.

The *D. plexippus plexippus* iteradensovirus (DppIDV) genome contained the typical inverted terminal repeats (ITRs) of members of the *Iteradensovirus* genus (*Bombyx mori* densovirus 1 [BmDENV-1], *Casphalia extranea* densovirus [CeDENV], *Sibine fusca* densovirus [SfDENV], *Papilio polyxenes* densovirus [PpDENV], and *Dendrolimus*

*punctatus* densovirus [DpDENV]) (10, 11, 13–15). The 239-nucleotide (nt) ITRs with 163-nt terminal J-shaped hairpins were about 90% conserved with those of the other iteradensoviruses. The overall sequence was about 86% identical to CeDENV, about 84% identical to SfDENV and BmDENV, about 78% identical to PpDENV, and about 71% identical to DpDENV.

Similar to other iteradensoviruses, the DppIDV monosense genome contained three intronless genes with essentially identical positions and sizes. The largest, open reading frame 1 (ORF1) (nt 360 to 2618), had a coding capacity of 752 amino acids (aa) and the typical nucleoside triphosphatase (NTPase) motif for NS1. ORF2 (nt 2677 to 4710), with the phospholipase A2 motif, typical for parvovirus VP, had a coding capacity of 677 aa. ORF3, with a 451-aa coding capacity (nt 487 to 1842) corresponded to NS2 and overlapped NS1 at its N terminus. As a comparison, NS1 is aa 753 to 775, NS2 is aa 451 to 455, and VP is aa 668 to 681 for the other iteradensoviruses.

**Nucleotide sequence accession number.** The GenBank accession no. for DppIDV is [KF963252](https://www.ncbi.nlm.nih.gov/nuclot/KF963252).

## ACKNOWLEDGMENTS

This work was supported by the Natural Sciences and Engineering Research Council of Canada grant to P.T. Q.Y. acknowledges support from a scholarship from the People's Republic of China and tuition waivers from INRS-IAF.

## REFERENCES

- Anderson JB, Brower LP. 1996. Freeze-protection of overwintering monarch butterflies in Mexico: critical role of the forest as a blanket and an umbrella. *Ecol. Entomol.* 21:107–116. [http://dx.doi.org/10.1111/j.1365-2311.1996.tb01177.x](https://doi.org/10.1111/j.1365-2311.1996.tb01177.x).
- Knight A, Brower LP. 2009. The influence of eastern North American autumnal migrant monarch butterflies (*Danaus plexippus* L.) on continuously breeding resident monarch populations in southern Florida. *J. Chem. Ecol.* 35:816–823. [http://dx.doi.org/10.1007/s10886-009-9655-z](https://doi.org/10.1007/s10886-009-9655-z).
- Brower LP, Fink LS, Walford P. 2006. Fueling the fall migration of the monarch butterfly. *Integr. Comp. Biol.* 46:1123–1142. [http://dx.doi.org/10.1093/icb/icl029](https://doi.org/10.1093/icb/icl029).
- Malcolm SB, Cockrell BJ, Brower LP. 1989. Cardenolide fingerprint of

- monarch butterflies reared on common milkweed, *Asclepias syriaca* L. J. Chem. Ecol. 15:819–853.
5. Niiler E. 1999. GM corn poses little threat to monarch. Nat. Biotechnol. 17:1154. <http://dx.doi.org/10.1038/70691>.
  6. Arnott HJ, Smith KM, Fullilove SL. 1968. Ultrastructure of a cytoplasmic polyhedrosis virus affecting the monarch butterfly, *Danaus plexippus*. I. Development of virus and normal polyhedra in the larva. J. Ultrastruct. Res. 24:479–507. [http://dx.doi.org/10.1016/S0022-5320\(68\)80050-4](http://dx.doi.org/10.1016/S0022-5320(68)80050-4).
  7. Bartel RA, Oberhauser KS, De Roode JC, Altizer SM. 2011. Monarch butterfly migration and parasite transmission in eastern North America. Ecology 92:342–351. <http://dx.doi.org/10.1890/10-0489.1>.
  8. de Roode JC, de Castillejo CL, Faits T, Alizon S. 2011. Virulence evolution in response to anti-infection resistance: toxic food plants can select for virulent parasites of monarch butterflies. J. Evol. Biol. 24: 712–722. <http://dx.doi.org/10.1111/j.1420-9101.2010.02213.x>.
  9. Tijssen P, Li Y, El-Far M, Szelei J, Letarte M, Zádori Z. 2003. Organization and expression strategy of the ambisense genome of denonucleosis virus of *Galleria mellonella*. J. Virol. 77:10357–10365. <http://dx.doi.org/10.1128/JVI.77.19.10357-10365.2003>.
  10. Yu Q, Fédière G, Abd-Alla A, Bergoin M, Tijssen P. 2012. Iteravirus-like genome organization of a densovirus from *Sibine fusca* stoll. J. Virol. 86: 8897–8898. <http://dx.doi.org/10.1128/JVI.01267-12>.
  11. Yu Q, Hajek AE, Bergoin M, Tijssen P. 2012. *Papilio polyxenes* densovirus has an iteravirus-like genome organization. J. Virol. 86:9534–9535. <http://dx.doi.org/10.1128/JVI.01368-12>.
  12. Allander T, Emerson SU, Engle RE, Purcell RH, Bukh J. 2001. A virus discovery method incorporating DNase treatment and its application to the identification of two bovine parvovirus species. Proc. Natl. Acad. Sci. U. S. A. 98:11609–11614. <http://dx.doi.org/10.1073/pnas.211424698>.
  13. Fédière G, Li Y, Zádori Z, Szelei J, Tijssen P. 2002. Genome organization of *Casphalia extranea* densovirus, a new iteravirus. Virology 292:299–308. <http://dx.doi.org/10.1006/viro.2001.1257>.
  14. Li Y, Zádori Z, Bando H, Dubuc R, Fédière G, Szelei J, Tijssen P. 2001. Genome organization of the densovirus from *Bombyx mori* (BmDENV-1) and enzyme activity of its capsid. J. Gen. Virol. 82:2821–2825.
  15. Wang J, Zhang J, Jiang H, Liu C, Yi F, Hu Y. 2005. Nucleotide sequence and genomic organization of a newly isolated densovirus infecting *Dendrolimus punctatus*. J. Gen. Virol. 86:2169–2173. <http://dx.doi.org/10.1099/vir.0.80898-0>.