## Genetic characterization of Yug Bogdanovac virus

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**Abstract** We present pyrosequencing data and phylogenetic analysis for the full genome of Yug Bogdanovac virus (YBV), a member of the Vesicular stomatitis virus serogroup of the Rhabdoviridae isolated from a pool of *Phlebotomus perfiliewi* sandflies collected in Serbia in 1976. YBV shows very low nucleotide identities to other members of the Vesicular stomatitis virus serogroup and does not contain a reading frame for C'/C proteins.

**Keywords** Vesiculovirus · Yug Bogdanovac virus · Chandipura virus · Isfahan virus · 454 Pyrosequencing

Yug Bogdanovac virus (YBV) was isolated in 1976 from a pool of 200 unengorged female *Phlebotomus perfiliewi* sandflies collected in Serbia. Electron microscopy and serological analysis (complement fixation test, immuno-fluorescence, plaque reduction neutralization assay) placed it into the Vesicular stomatitis virus (VSV) serogroup of the Rhabdoviridae. Antibodies against YBV were found in humans (6/274 tested) [1] and domestic animals [1–3] but the role of YBV as a human pathogen is unclear. An

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antigenic relationship to Chandipura virus (CHDV) isolated in India and West Africa [4-7] and to Isfahan virus (ISFV) isolated in Iran and Turkmenia [8, 9], two members of the VSV serogroup implicated as causes of febrile and neurological diseases (CHDV) in humans [10-12] was described. In order to determine the genome sequence of this European vesiculovirus, YBV was passaged thrice in Vero B4 cells and RNA extraction was performed as described [13]. In order to cover the termini, a self-complimentary 3'-FLAC adapter and a 5'-RACE adapter were ligated to the genome prior to pyrosequencing [14]. A MID-barcoded Roche/454 Rapid Library was produced from 300 ng adapter-ligated viral genomic RNA following reverse transcription at 65 °C for 30 min (Transcriptor (Roche). 82 % of 11,970 reads were YBV genome specific (coverage 294-fold). The 11,202 nucleotide -ssRNA genome shows the typical genomic organization of vesiculoviruses including 3'-leader followed by 5 structural genes, and 5'-trailer sequence (Fig. 1a, GenBank JF911700). The full-length genome of YBV virus shows very low nucleotide identities to the genomes of CHDV (55.7 %), ISFV (55.3 %, [15]), VSIV (52.8 %), and of rabies virus (41.0 %) (Fig. 1b). Nevertheless, the high bootstrap values of the phylogenetic analysis confirm serological grouping of YBV within the genus Vesiculovirus of rhabdoviruses (Fig. 1b). In YBV, the C'/C proteins, two small highly basic, non-structural proteins encoded in a second ORF within the P gene of most vesiculoviruses [16, 17] are absent as in Alagoas virus (VSAV) [18]. Both 3'-leader and 5'-positive-sense antigenomic trailer of YBV are highly complementary to each other in the first 32 nucleotides (Fig. 1c, [19]). The genomic data presented here will help to design a YBV-specific RT-PCR which could be used to monitor YBV activity in phlebotomine sandflies in the Balkans.

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**Fig. 1 a** YBV genome: The non-segmented negative strand 11.2 kb RNA genome sequentially encodes 5 transcription units for the nucleoprotein (N), phosphoprotein (P), matrix protein (M), glycoprotein (G), and the RNA-dependent RNA-Polymerase (L) each followed by a conserved polyadenylation signal (PA) and a short intergenic region. The amino acid sizes of the putative encoded proteins are indicated. **b** Neighbor-joining phylogenetic analysis of full genome nucleotide sequences using ClustalW and a 1,000-fold bootstrap

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## References

- A. Gligic, R. Tesh, Z. Miscevic, A. Travassos da Roas, V. Zikovic, Mikrobiologija 20, 97–105 (1983)
- J. Vesenjak-Hirjan, V. Punda-Polic, M. Dobe, J. Hyg. Epidemiol. Microbiol. Immunol. 35, 129–140 (1991)
- A. Gligic, Z. Miscevic, Acta Veterinaria-Beograd 44, 319–322 (1994)
- V. Dhanda, F.M. Rodrigue, S.N. Ghosh, Indian J. Med. Res. 58, 179 (1970)

approach rooted to the sequence of RABV (FJ712195) and collapsed VSV subtree. Bootstrap values are given in *percent*. YBV (JF911700), CHDV (GU212856, GU212858), ISFV (AJ810084), VSV (EF197793, EU849003, AF473864, AF473865, AF473866, J02428, NC\_001560 (all VSIV)), EU373657 (COCV-Ind2), EU373658 (VSAV-Ind3), and RABV (FJ712195). c Alignment of the 3'-leader (YBV 3' Le) and the positive-sense complement of the 5'-trailer (YBV 5' TrC) region of YBV. Residues that match the consensus are *shaded gray* 

- D. Fontenille, M. Traorelamizana, J. Trouillet, A. Leclerc, M. Mondo, Y. Ba, J.P. Digoutte, H.G.Z. Zeller, Am. J. Trop. Med. Hyg. 50, 570–574 (1994)
- Y. Ba, J. Trouillet, J. Thonnon, D. Fontenille, Bull. Soc. Pathol. Exot. 92, 131–135 (1999)
- G.E. Kemp, O.R. Causey, H.W. Setzer, D.L. Moore, J. Wildl. Dis. 10, 279 (1974)
- R.B. Tesh, S. Saidi, E. Javadian, P. Loh, A. Nadim, Am. J. Trop. Med. Hyg. 26, 299–306 (1977)
- S.Y. Gaidamovich, L.M. Altukhova, V.R. Obukhova, E.N. Ponirovsky, V.G. Sadykov, G.A. Klisenko, N.A. Sveshnikova, Y.M. Kasymov, Vopr. Virusol. 25, 618–620 (1980)
- 10. T.J. John, Lancet 364, 2175 (2004)
- 11. M. Van Ranst, Lancet 364, 821-822 (2004)
- S. Basak, A. Mondal, S. Polley, S. Mukhopadhyay, D. Chattopadhyay, Biosci. Rep. 27, 275–298 (2007)
- M. Dilcher, L. Hasib, M. Lechner, N. Wieseke, M. Middendorf, M. Marz, A. Koch, M. Spiegel, G. Dobler, F.T. Hufert, M. Weidmann, Virology 423, 68–76 (2012)
- 14. Z. Hubalek, J. Halouzka, Arch. Virol. 84(3-4), 175-180 (1985)
- 15. A.C. Marriott, Arch. Virol. 150, 671-680 (2005)
- E. Kretzschmar, R. Peluso, M.J. Schnell, M.A. Whitt, J.K. Rose, Virology 216, 309–316 (1996)
- 17. C.F. Spiropoulou, S.T. Nichol, J. Virol. 67, 3103-3110 (1993)
- S.J. Pauszek, R. Allende, L.L. Rodriguez, Arch. Virol. 153, 1353–1357 (2008)
- 19. S.P.J. Whelan, G.W. Wertz, J. Virol. 73, 297-306 (1999)