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Blackcurrant-Associated Rhabdovirus: New Pathogen for Blackcurrants in the Baltic Sea Region

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Abstract : Newly discovered viruses provide novel knowledge for basic phytovirus research, serve as tools for biotechnology and can be helpful in identification of epidemic outbreaks. Blackcurrant-associated rhabdovirus (BCaRV) have been discovered in USA germplasm collection samples from Russia and France. As it was reported in one accession originating from France it is unclear whether the material was already infected when it entered in the USA or it became infected while in collection in the USA. Due to that BCaRV was definite as non-EU viruses. According to ICTV classification BCaRV is representative of Blackcurrant betanucleorhabdovirus specie in genus Betanucleorhabdovirus (family Rhabdoviridae). Nevertheless, BCaRV impact on the host, transmission mechanisms and vectors are still unknown. In RNA-seq data pool from Ribes plants resistance gene study by high throughput sequencing (HTS) we observed differences between sample group gene transcript heat maps. Additional analysis of the whole data pool (total 393660492 of 150 bp long read pairs) by rnaSPAdes v 3.13.1 resulted into 14424 bases long contig with an average coverage of 684x with shared 99.5% identity to the previously reported first complete genome of BCaRV (MF543022.1) using EMBOSS Needle. This finding proved BCaRV presence in EU and indicated that it might be relevant pathogen. In this study leaf tissue from twelve asymptomatic blackcurrant cv. Mara Eglite plants (negatively tested for blackcurrant reversion virus (BRV)) from Dobele, Latvia (56°36'31.9"N, 23°18'13.6"E) was collected and used for total RNA isolation with RNeasy Plant Mini Kit with minor modifications, followed by plant rRNA removal by a RiboMinus Plant Kit for RNA-Seq. HTS libraries were prepared using MGI Easy RNA Directional Library Prep Set for 16 reactions to obtain 150 bp pair-end reads. Libraries were pooled, circularized and cleaned and sequenced on DNBSEQ-G400 using PE150 flow cell. Additionally, all samples were tested by RT-PCR, and amplicons were directly sequenced by Sanger-based method. The contig representing the genome of BCaRV isolate Mara Eglite was deposited at European Nucleotide Archive under accession number OU015520. Those findings indicate a second evidence on the presence of this particular virus in the EU and further research on BCaRV prevalence in Ribes from other geographical areas should be performed. As there are no information on BCaRV impact on the host this should be investigated, regarding the fact that mixed infections with BRV and nucleorhabdoviruses are reported.

Keywords : BCaRV, Betanucleorhabdovirus, Ribes, RNA-seq
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