

ANÁLISE DO VIROMA POR HTS REVELA INFECÇÃO MÚLTIPLA EM PLANTA DE VIDEIRA

ANALYSIS OF VIROME BY HIGH-THROUGHPUT SEQUENCING REVEALED MULTIPLE INFECTION IN A SINGLE GRAPEVINE PLANT

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Resumo:

Grapevines (*Vitis* sp.) can host up to 86 virus species, some of which affect plant vigor, production and fruit quality. In 2019, cv. BRS Bibiana, a hybrid vine showing mild leafroll symptoms, collected in an experimental field in Canoinhas-SC, Brazil, was investigated for viruses. Ribosomal-depleted total RNA (TNA) from this sample was used to prepare a sequencing library generated with TruSeq Stranded Total RNA with Ribo-Zero Plant kit (Illumina, USA), then subjected to high-throughput sequencing (HTS) on Illumina HiSeq X Ten platform. The HTS yielded 45,419,722 total reads, which were quality trimmed with FastQ and Trim Galore softwares. *De novo* assembled contigs built by SPAdes (rnaSPAdes) software generated 256,894 contigs, some of which, after QUAST (MetaQUAST) analysis, corresponded to virus/viroid genomes that were identified by comparison with viral/viroidal reference sequences available in the NCBI database. In total, four RNA viruses/viroids, common grapevine pathogens, were identified in the BlastN/BlastX analyses. The following obtained sequences were deposited in GenBank: grapevine rupestris stem pitting-associated virus (GRSPaV, *Foveavirus*, *Betaflexiviridae*), GRSPaV-BIB-BR isolate, 8,679 nucleotides (nt), GenBank accession code MW357717; grapevine virus B (GVB, *Vitivirus*, *Betaflexiviridae*), GVB-BIB-BR isolate, 7,599 nt, MW357718; grapevine yellow speckle viroid 1 (GYSVd-1, *Apscaviroid*, *Pospiviroidae*), GYSVd1-BIB-BR isolate, 366 nt, MW357719, and hop stunt viroid (HSVd, *Hostuviroid*, *Pospiviroidae*), HSVd-BIB-BR isolate, 297 nt, MW357720. The highest pairwise nt sequence identities between these Brazilian and homologous isolates (with query cover 99-100%) showed 99.39% nt identity with Brazilian GRSPaV NUB1-BR isolate (MK804766); 97.81% nt identity with Brazilian GVB ISA-BR isolate (KX790785); 99.73% nt identity with Nigerian GYSVd1 UDSV1-4 isolate (MF576400), and 100% nt identity with Canadian and Chinese HSVd BacSB11_5310, BacMF3_243, BacMF3_783, and chi100Y-02-4-5 isolates (MT769774, MT769768, MT769769, AB219944), indicating high homologies among compared isolates. To confirm the HTS results, fresh TNA extract from original source was screened by conventional or RT-qPCR using specific primers and/or probes. The results demonstrate the complexity of grapevine viral diseases and show that mixed virus/viroid infection of grapevine can be considered the rule, since this condition represents the majority of cases.

Palavras-chave: Vine; HTS; NGS; Virus; Viroid

Apoio

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