Molecular detection and genome analysis of circoviruses of European eel (Anguilla anguilla) from Lake Balaton

Balatoni angolnákból (Anguilla anguilla) kimutatott circovírusok molekuláris elemzése

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Circoviruses are small, non-enveloped viruses with a circular single-stranded (ss) DNA genome, which ranges from about 1.3-2.3 kb in size. The genome contains at least two open reading frames (ORF) arranged on different strands, these encode the replication (Rep) and capsid (Cap) proteins.

Before the last decade, circoviruses were known only from birds and swine, but recently, due to the different metagenomic methods and PCR based diagnostic tests, numerous circovirus-related sequences were described from environmental samples, invertebrates and also from lower vertebrates (amphibian and fish).

According to the increasing number of the detected piscine circoviruses, a monitoring program (Bioclimate) is carried out for screening circoviruses in Hungarian freshwater fishes. Samples were taken regularly also from Lake Balaton. In March 2014, fifteen living European eels (*Anguilla anguilla*) and two razorfish (*Pelecus cultratus*) were caught among other species at the Sió-floodgate sampling site. After euthanasia, tissue samples were collected from the gills, liver, spleen, kidney and intestine. The presence of circoviral DNA was detected by a broad-spectrum nested PCR, targeting the Rep gene. In case of positive results, rolling circle amplification and inverse nested PCR reactions were carried out to amplify the remaining part of the circular genomes.

Six out of the fifteen eels, and one razorfish sample were found to be positive for circoviral DNA. Analysis of these fragments revealed the presence of three different circovirus-related rep-like sequences. One of them (representing with 3 samples) was identical with the previously described eel circovirus (EeCV) by Doszpoly et al. (2014). The whole genome amplification of these 3 samples was also successful. Comparing the partial rep-like sequences, two other samples were similar with 96% nucleotide identity to the above mentioned ones, and were identical to the rep-like sequence originating from razorfish. The 6th circovirus positive sample showed only 50% similarity to the EeCV, but it was found to be identical to the rep-like sequences of roaches (*Rutilus rutilus*) caught from Lake Balaton, recently.

Circoviruses were described as host-specific or narrow host range microorganisms. To the best of our knowledge, this is the first report about the detection of the same circovirus related sequences in various, distantly related fishes and the occurrence of different rep-like sequences in the same fish species.

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