RECENT OUTBREAKS OF CARP EDEMA VIRUS DISEASE IN SERBIA

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INTRODUCTION

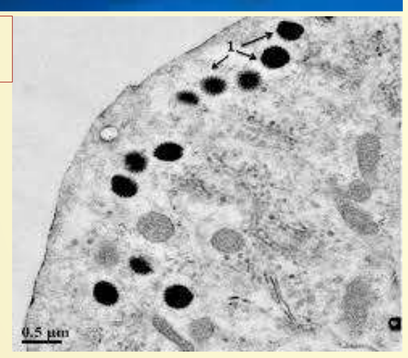
Common carp (*Cyprinus carpio*) is one of the most important fish in Serbian aquaculture, with annual production of approximately 11.000 tons.In order to maintain and intensify the production, one of the main goals is to prevent occurence and spreading of the diseases, which could limit the sustainability of fish production. With that goal, annual control of listed fish diseases, namely KHV and SVCV has been done. But, the occurence and spread of new disease which may significantly influence on health of carp is disturbing fact which requires attention of all parts in production chain.

Carp edema virus disease (CEVD) is emerging disease considered to be a potential risk for the carp aquaculture and for global food security, with new data about its spread, economic and biological properties being published rapidly over the last few years.

The first outbreak of disease caused by carp edema virus (CEV) was reported in 1974 in Japan, and for long time this disease was detected exclusively in ornamental koi carp, but recently it was confirmed as a causative agent of disease in koi and common carp in Europe, USA and many Asian countries.

In Europe, CEV was detected in the following countries: United Kingdom, Austria, Germany, Czech Republic, Slovakia, Hungary, Poland, Netherlands, France, Italy, Switzerland, Belgium, Serbia and Croatia.

CEVD in koi carp.
Photo: Surachai Pikulkaew



MATERIAL and METHODS

Carp were collected from two neighbouring carp farms in the north-western part of Serbia. The carp were sampled for disease diagnosis because the fish exhibited lethargy and anorexia, which eventually led to mortality. External examination was performed and sampling for further investigation was performed. After gill clipping and skin scraping, the presence of external parasite infection was inspected with an optical microscope.

For bacterial culture, the organ surface was sterilized using a heated blade and punctured to perform parenchymal swab sampling. Bacterial cultures of the gills, liver, spleen, and kidney were established for 48 h using tryptic soy agar plates at 20°C. The gill and kidney tissue were sampled for virus isolation, DNA extraction and real-time polymerase chain reaction (PCR).

Samples were tested for CEV according to PCR and real-time PCR protocols described by Way et al. (2017). For detection of CEV p4a DNA, a probe-based qPCR assay developed by the Centre for Environment, Fisheries and Aquaculture Science (CEFAS) in Weymouth, UK, was performed using the primers CEFAS_qF: AGTTTTGTAKATTGTAGCATTTCC, CEFAS_qR: GATTCCTCAAGGAGTTDCAGTAAA and the double-labelled probe [FAM]-AGAGTTTGTTTCTTGCCATACAAACT-[BHQ1]. The amplification program included an initial denaturation at 95°C for 10 min, followed by 40 cycles of denaturation at 95°C for 30 s and annealing at 60°C for 30 s.

DISCUSSION

These outbreaks further confirm the spread of CEV and the need for practitioners to be vigilant for outbreaks of this disease. Fish may remain potential carriers of this pathogen, and strict biosecurity measures should be enforced for any carp farm that has had a confirmed CEV outbreak.

To prevent further spreading of the disease, it is very important to introduce CEV testing before fish movement. To avoid further transmission of the virus to common carp populations in Serbia, testing of CEV should become part of fish disease surveillance programs.

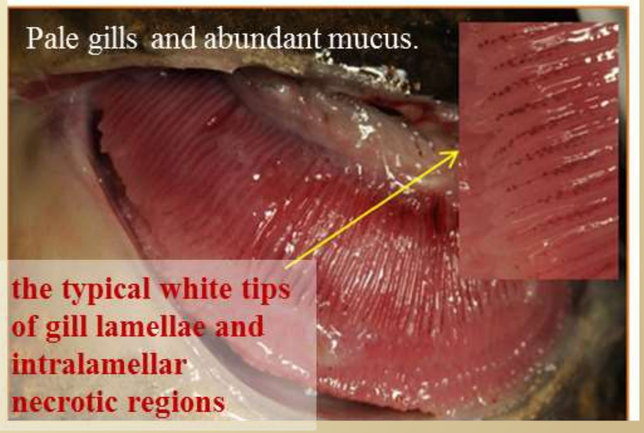
Fish health service should be aware of the presence of CEV in Serbia which may result in high losses in carp aquaculture. Action should be taken also to prevent transmission of CEV to carp populations in open waters.

RESULTS

Mortality started with clinical signs of hypoxia while fish swam slowly without escaping reactions. The gills were pale and covered with a thick mucus layer. In advanced cases, the lesions in the gills turned into a necrotizing form. A moderate to high amount of opportunistic freshwater bacteria were isolated from gills and skin of diseased fish. All carp were found negative for CyHV-3, but all fish tested positive for CEV by polymerase chain reaction (PCR) and qPCR.

Figure 1. CEVD in carp - sleepy behavior - infected fish become lethargic and unresponsive

Figure 2. CEVD in carp - The virus causes severe damage to gill lamellae, leading to hypoxia and mortality



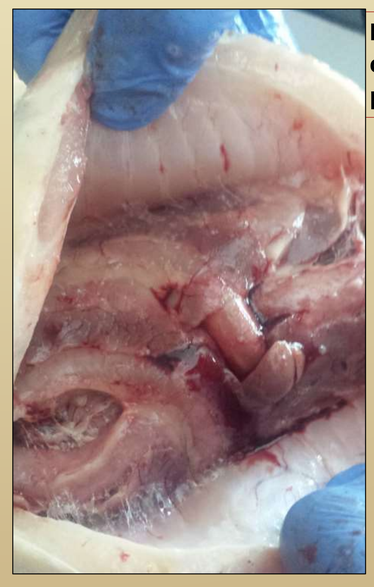


Figure 3. CEVD in carp – Fibrinous peritonitis

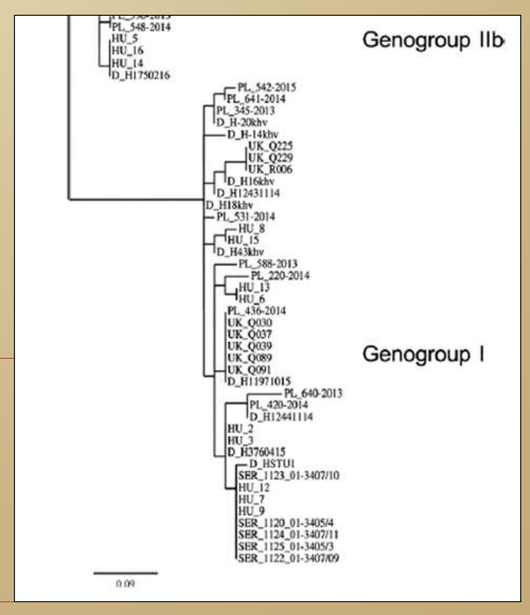


Figure 4. Phylogenetic analysis of the nucleotide sequence encoding the fragment of CEV P4a core protein.

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