

## DETECTION OF A NEW VARIANT KHV USING A GENERIC CYPRINID HERPESVIRUS PCR

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## The cyprinid herpes viruses

- Cyprinid herpes virus-1 (carp pox)
- Cyprinid herpes virus-2 (Goldfish herpesvirus)
- Cyprinid herpes virus-3 (koi herpesvirus)

Cefas routinely screens all unexplained mortalities of carp for KHV and carp pox, and all unexplained mortalities of goldfish for the goldfish herpes virus.

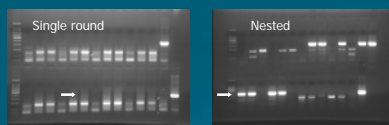
TK primer

Generic CyHV DNA polymerase primer set

[Isolation of a Cyprinid Herpesvirus (Cyhv-2) From Goldfish *Carassius Auratus* in the UK. K.R. Jeffery, K. Bateman, A. Bayley, S.W. Feist, J. Hulland, C. Longshaw, D. Stone, G. Woolford and K. Way\*. J.Fish Dis. In press]



## Discovery of a new KHV variant



- 7 samples (4 sites) positive for KHV by the nested CyHV pol PCR assay
- All samples negative by the TK PCR assay
- All samples negative by tissue culture



## Variant shares 94% nucleotide identity in the published KHV major capsid protein gene

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Query 1: 10079 10166 10253 10340 10427 10514 10601 10688 10775 10862 10949 11036 11123 11210 11297
|b|101 109228 10079 10166 10253 10340 10427 10514 10601 10688 10775 10862 10949 11036 11123 11210 11297

Query 63: 10079 10166 10253 10340 10427 10514 10601 10688 10775 10862 10949 11036 11123 11210 11297
|b|101 109228 10079 10166 10253 10340 10427 10514 10601 10688 10775 10862 10949 11036 11123 11210 11297

Query 121: 10079 10166 10253 10340 10427 10514 10601 10688 10775 10862 10949 11036 11123 11210 11297
|b|101 109228 10079 10166 10253 10340 10427 10514 10601 10688 10775 10862 10949 11036 11123 11210 11297

Query 181: 10079 10166 10253 10340 10427 10514 10601 10688 10775 10862 10949 11036 11123 11210 11297
|b|101 109228 10079 10166 10253 10340 10427 10514 10601 10688 10775 10862 10949 11036 11123 11210 11297

Query 241: 10079 10166 10253 10340 10427 10514 10601 10688 10775 10862 10949 11036 11123 11210 11297
|b|101 109228 10079 10166 10253 10340 10427 10514 10601 10688 10775 10862 10949 11036 11123 11210 11297

Query 301: 10079 10166 10253 10340 10427 10514 10601 10688 10775 10862 10949 11036 11123 11210 11297
|b|101 109228 10079 10166 10253 10340 10427 10514 10601 10688 10775 10862 10949 11036 11123 11210 11297

Query 361: 10079 10166 10253 10340 10427 10514 10601 10688 10775 10862 10949 11036 11123 11210 11297
|b|101 109228 10079 10166 10253 10340 10427 10514 10601 10688 10775 10862 10949 11036 11123 11210 11297

Query 421: 10079 10166 10253 10340 10427 10514 10601 10688 10775 10862 10949 11036 11123 11210 11297
|b|101 109228 10079 10166 10253 10340 10427 10514 10601 10688 10775 10862 10949 11036 11123 11210 11297
    
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## Divergence between KHV and the variant strain

	% Nucleotide Identity	% Amino Acid identity (similarity)
DNA polymerase Gene	96	98 (99)
Intercapsomeric triplex protein gene	97	93 (98)
Major capsid protein gene	94	98 (99)



## What next?

- Further evidence for a variant of KHV  
Sequence data for additional KHV genes using degenerate primers in a nested PCR

- Require information on replication, virus load and distribution of the of the new variant in the host

Develop an alternative more specific PCR assay based on the Major capsid protein, DNA polymerase and Intercapsomeric triplex protein gene sequences

*In situ* hybridisation

Tissue culture

- Require more information on the distribution of the virus in the UK and Europe

Circulate new primer sequences to European NRLs as soon as possible

