## VBD Molecular Epidemiology Report Form (2009-11-02) 066 - ARB-RES-FOR-066

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Date sample received at IAH: <b>2<sup>nd</sup> November 2009</b>	Diagnostic Report date: Results sent by email To
IAH -ISIS/STARS sample number: A89/09 sample 12 and	Philip Ivens 4/11/09
A89/09 sample 18	Diagnostic Report sent to: Not reported through
Sender Ref: Philip Ivens	Stars
Date collected: 22 <sup>nd</sup> October 2009	Date cleared for general release:
Species: Equine	
Orbivirus Ref Collection No: GAM2009/01 & GAM2009/02	Sequencing Report date: 08/02/2010
Date received for sequencing: 29/01/2009	Prepared by: Kasia Bankowska /Sushila Maan
Material used: A89/09 sample 12 KC2 cell culture	Checked by: Sushila Maan/Peter Mertens
A89/09 sample 18 KC1 cell culture	Serotype: AHSV-9
Genome segment sequenced: Seg-2	No. of nt compared: 1776
Method used: RT-PCR	
Primers used: AHSV-9 seg-2 specific internal primers	
Region and No. Nt determined: 625-2400bp (1776nt)	
Gene length: 3202 nt	
No. of ambiguities: 1	

## Comments / conclusions:

The virus isolate was initially identified as AHSV-9 by the Non-vesicular reference laboratory, Institute for Animal Health, by conventional RT-PCR using type specific primers targeting Seg-2 (Sailleau et al 2000).

Subsequent sequence analysis by the Arbovirus Molecular Research Group (AMRG) confirms virus type (as AHSV-9) in both samples investigated. The Seg-2 nucleotide sequence of the Gambian samples A89/09 12 and 18 (<u>GAM2009/01</u> & <u>GAM2009/02</u> respectively) in the fragments analysed (625-2400pb) showed 100% identity to the AHSV-9 South African reference strain (PAKrrah/09), except for one ambiguous base at position 2303. The similarity of this Seg-2 sequence to that of the reference strain, which was originally isolated in Pakistan, indicates that Seg-2 of the test virus has a recent common ancestry with the reference of AHSV-9, and presumably with the vaccine strain that derived from it. However, no Seg-2 sequence data was available from the AHSV-9 vaccine strain itself for further comparison.

Most Closely Related Viruses					
In order of similarity to target sequence	Virus type / name	Reference collection number / [Accession number]	No. nt compared / segment length	% Identity	
1	Gambian sample 2009	GAM2009/01 A89/09-12	1776/3202	100%	
2	Gambian sample 2009	<u>GAM2009/02</u> A89/09-18	1776/3202	100%	
3	AHSV-9 reference strain	PAKrrah/09	1776/3202	100%	
Relationships to most closely related Reference Virus Strain (PAKrrah/09)					
In order of similarity to target sequence	Virus type / name	Reference collection number / [Accession number]	No. nt Compared / segment length	% Identity	
1	AHSV-9 reference strain	PAKrrah/09	1776/3202	100%	

Report prepared by: Kasia Bankowska, Sushila Maan, Carrie Batten, Chris Oura & Peter Mertens, 08 February 2010

If further information is required concerning these results please contact the Nonvesicular reference laboratories, Institute for Animal Health, Pirbright, UK.