

Molecular detection and pathotyping of APMV-1

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● Sutton et al., 2018 – a case of failed detection

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19RS-750-vir96 -Pigeon VI.1.1 TTCTACAATGGAGCTAATGAACATTCTTCAATATCTAGCGGGAAAGTTGATTGGACAATCCGTGGTTTCCTATGATGAAGATACTTC!  
FW .....  
Probe MGB .....  
Probe MGB2 .....  
RV .....
```

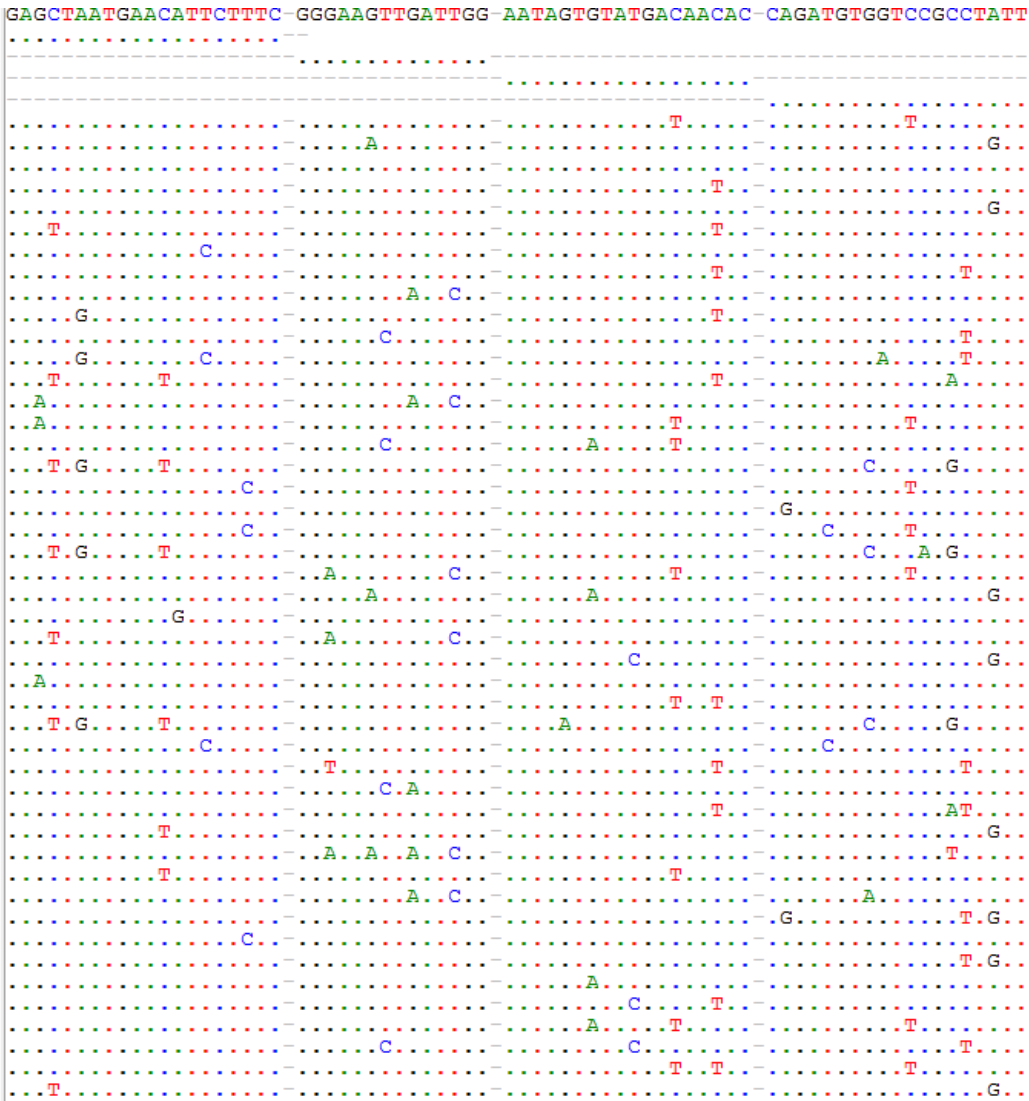
```
19RS-750-vir96 -Pigeon VI.1.1 TATAAAGAATGATGCTATAATAGTGATGATAACACACGAAATTGGATTAGCGAAGCCCAGAATTCAGATGTAGTTCGCTTATTCC!  
FW .....  
Probe MGB .....  
Probe MGB2 .....C.....  
RV .....G..C..C.....
```

- Virus 19RS-750-VIR96 belonging to genotype VI.1.1. was undetectable using Sutton et al., 2018
- L gene specific primers were used to amplified the region targeted by the assay
- Sequences analysis showed three mismatches in the reverse primer binding region
- The three mismatches identified were sufficient to yield a false negative result

Sutton et al., 2018– in silico evaluation

Primers and probes concatamere
 FW
 Probe MGB
 Probe MGB2
 RV

EF026579	representative	42	sequences
JN400897	representative	31	sequences
AY262106	representative	31	sequences
KX822746	representative	30	sequences
JN400896	representative	18	sequences
AY562986	representative	16	sequences
KX791184	representative	15	sequences
GU978777	representative	14	sequences
KP001164	representative	14	sequences
FJ986192	representative	13	sequences
JX401403	representative	11	sequences
KP089979	representative	10	sequences
GQ288379	representative	10	sequences
AY935489	representative	9	sequences
FJ766531	representative	9	sequences
KT760568	representative	9	sequences
AB534205	representative	8	sequences
AJ880277	representative	7	sequences
JF343538	representative	6	sequences
GQ338311	representative	6	sequences
JN688862	representative	4	sequences
KM374059	representative	3	sequences
KJ600786	representative	3	sequences
KX761866	representative	3	sequences
KC152049	representative	3	sequences
KU295455	representative	3	sequences
KJ607169	representative	3	sequences
JX974435	representative	3	sequences
HQ008337	representative	3	sequences
KR074406	representative	2	sequences
KJ769262	representative	2	sequences
AY562985	representative	2	sequences
KC987036	representative	2	sequences
KC542902	representative	2	sequences
AY935499	representative	2	sequences
KY171995	representative	2	sequences
AY935492	representative	2	sequences
KU140419	representative	2	sequences
AB853928	representative	2	sequences
KC542897	representative	2	sequences
AY505060	representative	2	sequences
AY845400	representative	2	sequences
KX247376	representative	2	sequences
JX401404	representative	2	sequences
JX901119	representative	2	sequences
JN599167	representative	2	sequences



● AMPV-1 pathotyping

Protocols	Applied by	Conventional/ real-time
Aldous et al., 2003	2	conventional
APHA SOP VI587	3	real-time
APHA VI.448	2	conventional
Collins et al., 1993	1	conventional
Creelan et al., 2002	7	conventional
De Battisti et al., 2012	1	conventional
Fuller et al., 2009	3	real-time
Gohm et al., 2000	2	conventional
Kant et al., 1997	4	conventional
Kho et al., 2000	2	conventional
Moharam et al., 2019	1	real-time
Oberdorfer et al., 1994	1	conventional
Pchelkina et al., 2013	1	conventional
Sabre et al., 2017	1	real-time
Seal et al., 1995	3	conventional
Wise et al., 2004	2	real-time
In house	2	conventional

● Open discussion topics

- Does APMV-1 pathotyping usually rely on one molecular method or more molecular assays are commonly employed?
- After APMV-1 detection do you proceed in parallel with virus isolation?
- Shall we consider to adopt a lineage specific qRT-PCR?
- Shall we consider a COST project to join forces and promote sharing of viruses/samples and trainings on AI and ND?