

Supplementary material to:

Title: **Host barriers limit viral spread in a spillover host: a study of deformed wing virus in the bumble bee *Bombus terrestris***

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- **Table S1** Colony screening results for six common honey bee viruses
- **Table S2** Raw data on samples and PCR results underpinning Figures 1, 2 and 3 (main manuscript).
- **Table S3** Results of capillary electrophoresis to identify one-step RT-PCR products of haemolymph from bumble bees and honey bees fed DWV-B
- **Figure S1** Electropherograms of one-step RT-PCR products of haemolymph from bumble bees and honey bees fed DWV-B
- **Table S4** Screening pollen viruses

Table S1 Colony screening results of experimental *A. mellifera* and *B. terrestris* colonies for six common honey bee viruses, undertaken by real-time PCR. Samples were run in technical duplicates. N/A = negative. Honey bee beta-actin served as a quality control internal 'reference' for RNA extraction and cDNA synthesis.

Colony	Actin	DWV-A	DWV-B	BQCV	SBV	SBPV	CBPV
<i>A. mellifera</i> R25	27.26 -	N/A N/A	37.35 N/A	39.35 37.79	N/A N/A	N/A N/A	N/A N/A
<i>B. terrestris</i> Colony 1	23.46 23.04	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A
<i>B. terrestris</i> Colony 2	23.38 23.17	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A
<i>B. terrestris</i> Colony 3	22.78 22.66	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A
<i>B. terrestris</i> Colony 4	23.59 23.49	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A
<i>B. terrestris</i> Colony 5	23.48 23.50	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A
<i>B. terrestris</i> Colony 6	19.42 19.53	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A
<i>B. terrestris</i> Colony 7	21.59 21.47	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A
<i>B. terrestris</i> Colony 8	24.67 24.31	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A

The Excel file is available at: [link article doi](#)

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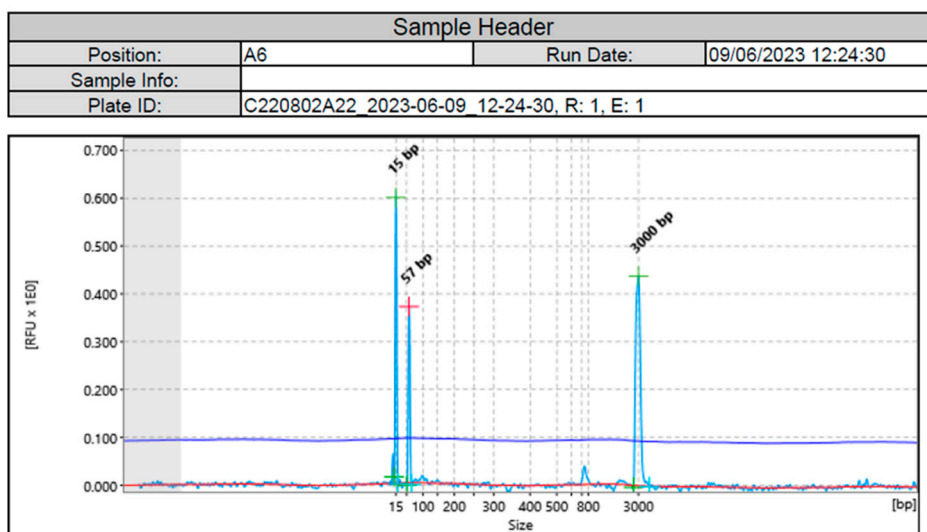


Fig. S1a Haemolymph of a control-fed *B. terrestris* worker (lane A6)

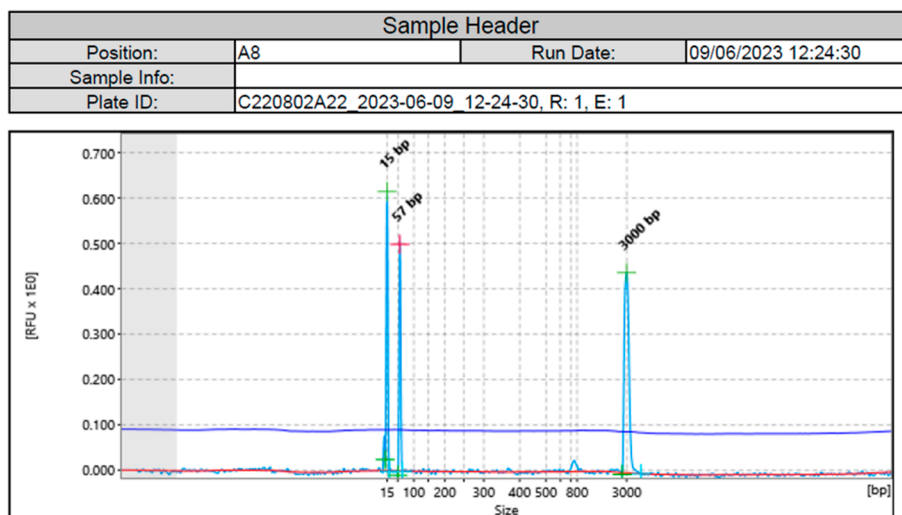


Fig. S1b Haemolymph of a control-fed *A. mellifera* worker (lane A8)

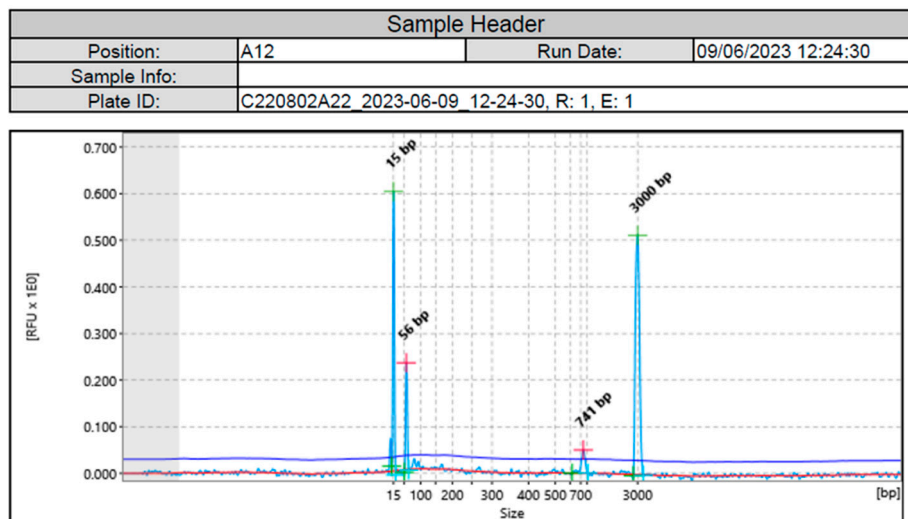


Fig. S1c Haemolymph of a DWV-B-fed *B. terrestris* worker (lane A12)

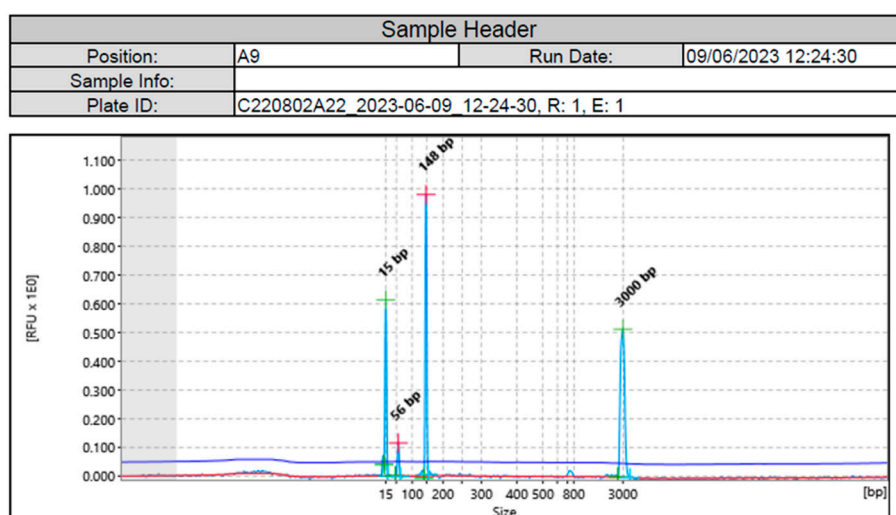


Fig. S1d Haemolymph of a DWV-B-fed *A. mellifera* worker (lane A9)

Figure S1 Electropherograms of PCR products using haemolymph as template, collected from control-fed *B. terrestris* (**1a**), and *A. mellifera* (**1b**) workers and DWV-B-fed *B. terrestris* (**1c**) and *A. mellifera* (**1d**) workers (data presented in Table S3). All samples show a PCR product with a length of 56-57 bp when tested for the presence of DWV-B, likely due to primer dimerization, indicated through the short fragment length. DWV-B was detected only in DWV-B-fed *A. mellifera* (**1d**: a product length of 148 bp), while no DWV-B was detected in the hemolymph of a DWV-B-fed *B. terrestris* (**1c**).

Table S4 Results (Cq value) of honey bee pollen screening for DWV-A, DWV-B and BQCV by RT-PCR. Commercially available honey bee pollen regularly carries bee viruses. Shown are two different pollen batches; gamma irradiated pollen from Great Britain and non-irradiated pollen from Austria. The latter was used to feed *Bombus terrestris* colonies in the laboratory as a source of bumble bees for our experiments.

Pollen sample	DWV-A	DWV-B	BQCV
gamma irradiated			
pollen sample a	N/A	N/A	N/A
(technical duplicate of a)	N/A	N/A	N/A
pollen sample b	N/A	N/A	N/A
(technical duplicate of b)	N/A	N/A	N/A
pollen sample c	N/A	39.10	38.02
(technical duplicate of c)	N/A	37.93	38.31
pollen sample d	N/A	N/A	N/A
(technical duplicate of d)	N/A	N/A	N/A
non-irradiated pollen			
pollen sample 1	37.01	N/A	23.78
pollen sample 2	37.02	N/A	28.75
pollen sample 3	39.20	N/A	30.04
pollen sample 4	37.13	38.62	30.04