Table S1: Characteristics of viruses and viroid isolates

Virus / viroid	Isolate (original name)	Provided by	Plants used for propagation and detection
PepMV-EU	NIB V 135 (0206)	Scientia Terrae[c]	*Datura stramonium*
PepMV-Ch2[a]	NIB V 131 (2206/06A1)	Scientia Terrae[c]	*Datura stramonium*
PepMV-Ch2[b]	NIB V 147 (PCH06/104; 22-9-2008)	PRI[d]	*Lycopersicon esculentum* cv. Moneymaker
PVY^{NTN}	NIB V 001	NIB[e]	*Nicotiana tabacum* cv. White Burley
PSTVd	NIB V 095 (16/may/06)	FERA[f]	*Lycopersicon esculentum* cv. Moneymaker

[a]isolate used for survival experiment;
[b]isolate used for water-mediated transmission experiments;
[c]isolates provided by Dr. Inge Hanssen, Scientia Terrae Research Institute, Belgium;
[d]isolate provided by Dr. Rene van der Vlugt, Plant Research International, The Netherlands;
[e]isolate maintained in tissue culture of *Solanum tuberosum* cv. Pentland squire at NIB, Slovenia;
[f]isolate provided by Dr. Neil Boonham, Food and Environment Research Agency, UK.

Table S2: Overview of the experimental hydroponic systems

expt	Virus/viroid isolate	Inoculated plants (no. of inoculated plants)	Bait plants (no. of bait plants)	Duration of irrigation with infected nutrient solution	Test plants mechanically inoculated with nutrient solution
PepMV-tomato	PepMV-Ch2	*Lycopersicon esculentum* cv. Moneymaker (6)	*Lycopersicon esculentum* cv. Moneymaker (6)	8.5.2010–12.14.2010	*Nicotiana glutinosa*, *Lycopersicon esculentum* cv. Moneymaker, *Datura stramonium*
PepMV+PVY-tomato	PepMV-Ch2 and PVY^{NTN}	*Lycopersicon esculentum* cv. Moneymaker (6)	*Lycopersicon esculentum* cv. Moneymaker (6)	8.5.2010–12.14.2010	*Lycopersicon esculentum* cv. Moneymaker, *Nicotiana tabacum* cv. White Burley
PVY-potato	PVY^{NTN}	*Solanum tuberosum* cv. Igor – plants grown from tissue culture (6)	*Solanum tuberosum* cv. Igor – plants grown from tissue culture (6)	2.4.2011–6.15.2011	*Nicotiana tabacum* cv. White Burley
PSTVd-tomato	PSTVd	*Lycopersicon esculentum* cv. Moneymaker (6)	*Lycopersicon esculentum* cv. Moneymaker (6)	2.2.2011–6.13.2011	*Lycopersicon esculentum* cv. Moneymaker
PSTVd-potato	PSTVd	*Lycopersicon esculentum* cv. Moneymaker (6)	*Solanum tuberosum* cvs. Hermes (4), Donald (4), Nicola (4) – seed tubers	7.18.2012–11.20.2012	/

Table S3: RT-qPCR versus biological assay for the detection of PepMV-EU, PVY^{NTN} and PSTVd

inoculum dilutions[a]	PepMV-EU		PVY^{NTN}		PSTVd	
	detection[b]	infectivity[c]	detection[b]	infectivity[c]	detection[b]	infectivity[c]
10^{-1}x	+ (16)	+	+ (12)	+	+ (21)	+
10^{-2}x	+ (18)	+	+ (13)	+	+ (24)	+
10^{-3}x	+ (22)	+	+ (18)	+	+ (28)	+
10^{-4}x	+ (25)	+	+ (22)	+	+ (31)	-
10^{-5}x	+ (28)	-	+ (25)	+	+ (33)	-
10^{-6}x	+ (32)	-	+ (29)	+	+ (34)	-
10^{-7}x	+ (35)	-	+ (32)	-	+ (36)	-
10^{-8}x	-	-	+ (35)	-	+ (39)	-
10^{-9}x	nt	nt	-	-	-	-
10^{-10}x	nt	nt	-	-	-	-
Water control	-	-	-	-	-	-

Abbreviations: +, positive; -, negative; nt, not tested.

[a]Ten-fold serial dilutions were made by diluting purified PepMV-EU particles (Gutierrez-Aguirre., I., N. Mehle, D. Delić, K. Gruden, R. Mumford, and M. Ravnikar. J. Virol. Methods 162:46-55, 2009) in buffer for mechanical inoculation. In the case of PVY^{NTN} and PSTVd macerated tissue from infected *Nicotiana tabacum* cv. White Burley or *Lycopersicon esculentum* cv. Moneymaker, respectively, was used for ten-fold serial water dilutions. The plant tissue was always macerated in Bioreba bags with synthetic intermediate layer used for filtration;

[b]detection of virus/viroid in inoculum by RT-qPCR (in bracket the average Cq values are given);

[c]the infectivity of each dilution was monitored by mechanical inoculation of 2 - 4 test plants (*Nicotiana glutinosa* for PepMV-EU, *Nicotiana tabacum* cv. White Burley for PVY^{NTN}, and *Lycopersicon esculentum* cv. Moneymaker for PSTVd) and observation of symptoms development together with molecular (PepMV-EU and PSTVd) or serological (PVY^{NTN}) analysis, two (PVY^{NTN}), four (PepMV-EU) or five (PSTVd) weeks after mechanical inoculation.

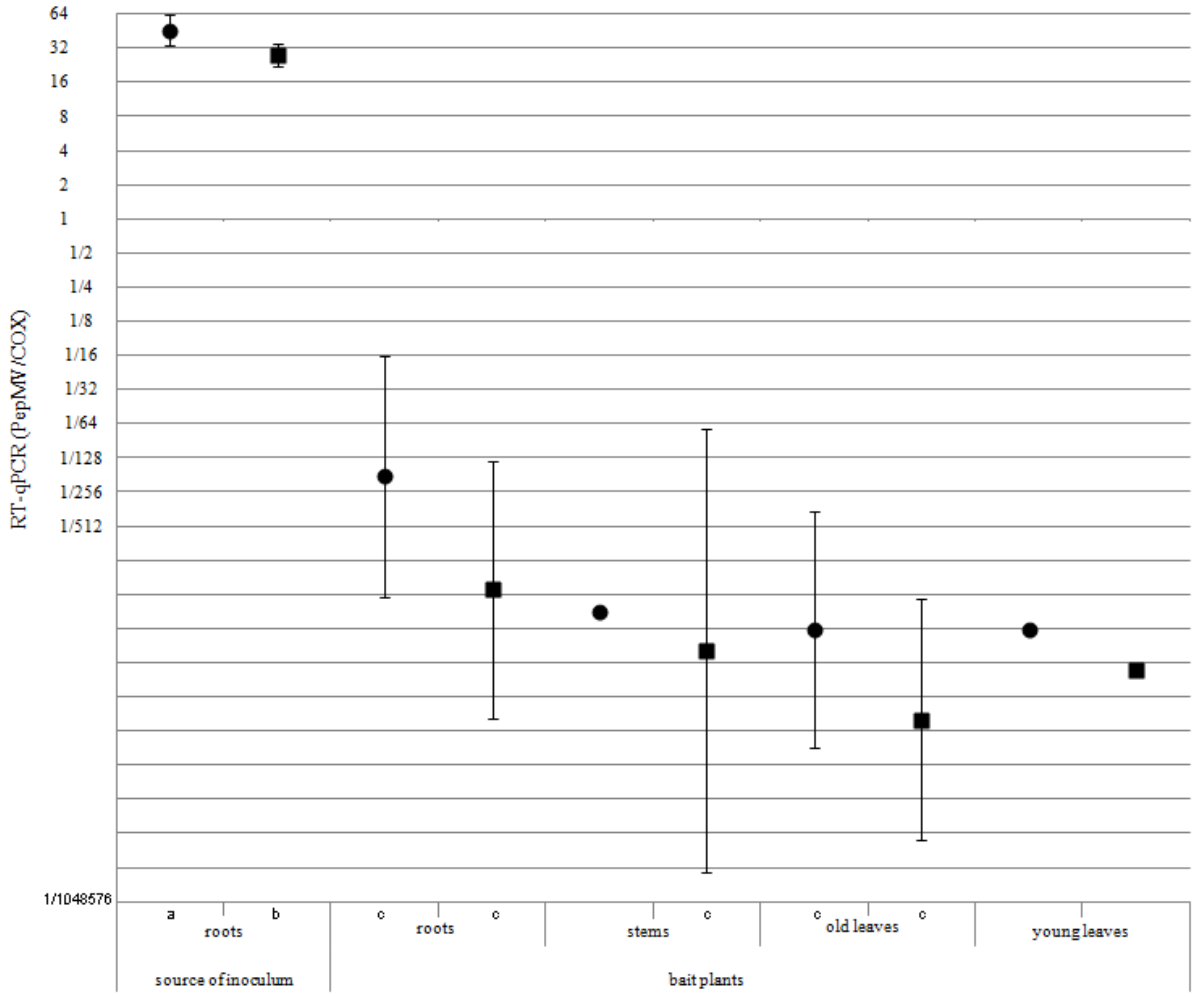

Figure S1: The amount of PepMV-Ch2 RNA relative to COX in positive samples (inoculum and different organs from bait plants) is shown for both PepMV-tomato (circles) and PepMV+PVY-tomato (squares) experimental hydroponic systems 134 days after initiation the irrigation with infested nutrient sollution. The average relative viral RNA amount and standard errors (for the cases where RNA was detected in more than four samples) are presented. Different letters (a, b and c) mean significant differences ($p<0.05$).
The presented values were obtained by calculating the antilogarithm of the subtraction between the Cq values obtained with PepMV assay and the Cq values obtained with COX assay. The stable expression of COX in different plant samples and organs (Baebler, Š., H. Krečič-Stres, A. Rotter, P. Kogovšek, K. Cankar, E.J. Kok, K. Gruden, M. Kovač, J. Žel, M. Pompe-Novak, and M. Ravnikar. Mol. Plant Pathol. 10:263-275, 2009; Hren, M., P. Nikolić, A. Rotter, A. Blejec, N. Terrier, M. Ravnikar, M. Dermastia, and K. Gruden. BMC Genomics 10. Online http://www.biomedcentral.com/1471-2164/10/460, 2009), allows comparing the amounts of viral RNA between samples.

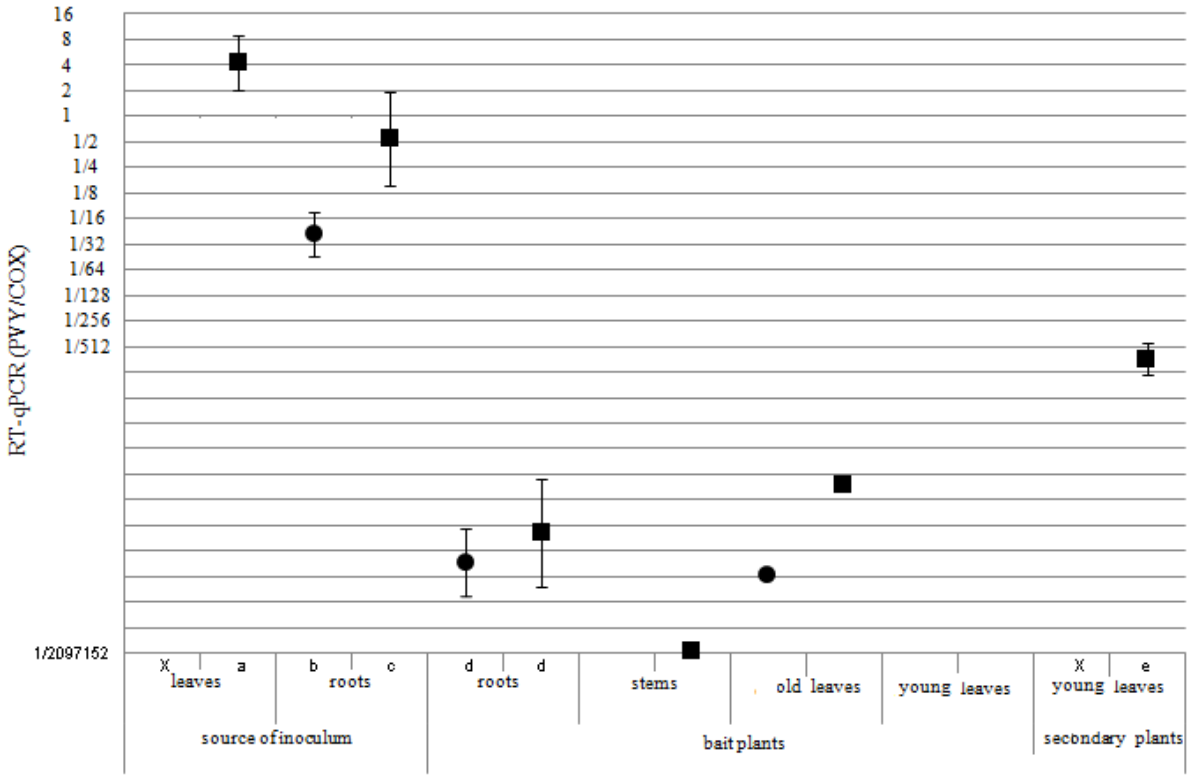

Figure S2: The amount of PVY^{NTN} RNA relative to COX in positive samples (inoculum and different organs from bait plants) is shown for both PepMV+PVY-tomato (circles) and PVY-potato (squares) experimental hydroponic systems 134/ 131 days after initiation the irrigation with infested nutrient sollution. Secondary plants are plants grown from tubers obtained from bait plants. The average relative viral RNA amount and standard errors (for the cases where RNA was detected in more than three samples) are presented. The absence of spots indicates that no virus was detected. Different letters (a, b, c, d and e) mean significant differences ($p<0.05$). Mark X means not tested.

The presented values were obtained by calculating the antilogarithm of the subtraction between the Cq values obtained with PVY assay and the Cq values obtained with COX assay. The stable expression of COX in different plant samples and organs (Baebler, Š., H. Krečič-Stres, A. Rotter, P. Kogovšek, K. Cankar, E.J. Kok, K. Gruden, M. Kovač, J. Žel, M. Pompe-Novak, and M. Ravnikar. Mol. Plant Pathol. 10:263-275, 2009; Hren, M., P. Nikolić, A. Rotter, A. Blejec, N. Terrier, M. Ravnikar, M. Dermastia, and K. Gruden. BMC Genomics 10. Online http://www.biomedcentral.com/1471-2164/10/460, 2009), allows comparing the amounts of viral RNA between samples.

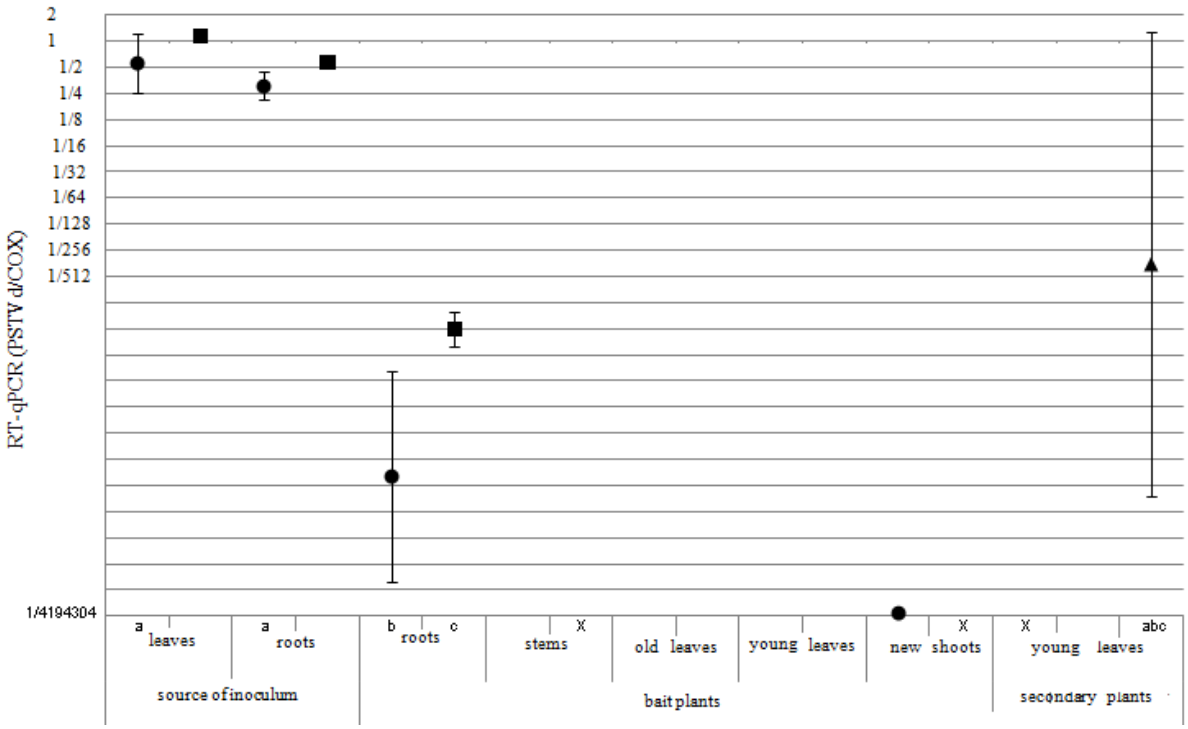

Figure S3: The amount of PSTVd RNA relative to COX in positive samples (inoculum and different organs from bait plants) is shown for both PSTVd-tomato (circles) and PSTVd-potato (squares) experimental hydroponic systems 141/ 125 days after initiation the irrigation with infested nutrient sollution. In the most right side the amount of PSTVd RNA relative to COX is shown in young leaves of secondary plants (those grown from tubers obtained from bait plants) from the experiment of inoculum injection into the substrate (triangle). The average relative viroid RNA amount and standard errors (for the cases where RNA was detected in more than three samples) are presented. The absence of spots indicates that no viroid was detected. Different letters (a, b and c) mean significant differences ($p<0.05$). Mark X means not tested.

The presented values were obtained by calculating the antilogarithm of the subtraction between the Cq values obtained with PSTVd assay and the Cq values obtained with COX assay. The stable expression of COX in different plant samples and organs (Baebler, Š., H. Krečič-Stres, A. Rotter, P. Kogovšek, K. Cankar, E.J. Kok, K. Gruden, M. Kovač, J. Žel, M. Pompe-Novak, and M. Ravnikar. Mol. Plant Pathol. 10:263-275, 2009; Hren, M., P. Nikolić, A. Rotter, A. Blejec, N. Terrier, M. Ravnikar, M. Dermastia, and K. Gruden. BMC Genomics 10. Online http://www.biomedcentral.com/1471-2164/10/460, 2009), allows comparing the amounts of viroid's RNA between samples.