

Title: Identifying Putative Resistance Genes for Barley Yellow Dwarf Virus-PAV in Wheat and Barley

Table S1. List of the primers used in this study.

(sequences, amplicon sizes, GenBank (NCBI database) accession numbers and efficiencies of all primers used in qPCR experiments with samples of *Hordeum vulgare* and *Triticum aestivum* (2 pairs designed per gene of interest). Primers for reference genes and BYDV-PAV standard [17] are also included.)

ID	Target Gene	GenBank	species	Sequence (5'->3')		position	Amplicon size (bp)	Efficiency (%)	References
NBS	rga S-9203: resistance gene of the NBS-LRR class	AJ507095.1	<i>Hordeum vulgare</i>	1F	CACCACCGAGAGAATAGAACCA	17-38	170	110	
				1R	TCCTTGAGAAACGCCACCT	186-168			
				2F	TGTTGGGTTTGGGGGTCT	652-669	172	110	
				2R	GTCAGTATGGGTGCTTGTGG	823-803			
	NBS-LRR resistance gene homologue from TcLr35	DQ205351.2	<i>Triticum aestivum</i>	1F	TGGTCGGTTTTGAGGACGAG	726-745	113	94	
				1R	AAGCGTTGTTTTTCCTGCCC	838-819			
				2F	AGGGTTGCAGTCTTGGTGAG	484-423	150	103	
				2R	CAAGTCAGTCGGTAGGCGAG	553-534			
CC-NBS-LRR	disease resistance protein RGA2-like	AK371901.1	<i>Hordeum vulgare</i>	1F	GCAGTGTTTCCGGGTGACTA	1539-1558	145	105	
				1R	AGCATCGCTCGACAAGTTCA	1683-1664			
				2F	ACGCCAAATGGTACAGCTCA	2321-2340	71	110	
				2R	TGAACCCACTAGCATCGCTC	2391-2372			
	coiled-coil nucleotide-binding leucine-rich	MW656173.1	<i>Triticum aestivum</i>	1F	CCACCTCCTCTCGTACTCCA	11402-11421	85	106	

	repeat (CC-NB-LRR) immune receptor (Lr13) gene			1R	ACTGGTATGCTGTTCTGCCC	11486- 11467			In this study
				2F	TTGCTGGCGTGATTGACTCT	17656- 17675	163	93	
				2R	GTTGAAGGTAGGGTGCCGAA	17818- 17799			
Rec Kin	LRR receptor-like serine/threonine- protein kinase At3g47570	AK364948.1	<i>Hordeum vulgare</i>	1F	TTTACTGCGGTGGGAGCATT	632-651	88	108	
				1R	CTTGGTGGGCATCACCTCTT	719-700			
				2F	TGTTGCTGCTCCAGTTTCA	1569- 1588	89	110	
				2R	TTCTTTCACCACATCGCCCA	1657- 1638			
	LRR receptor-like kinase	GU084176.1	<i>Triticum aestivum</i>	1F	GCTTCTTCTGCTGGCGTTTC	125-144	154	98	
				1R	GCTTCGGTTCAGTTCCTCA	278-259			
				2F	TAACGACAGCGAAAACCCCA	2642- 2661	95	107	
				2R	TGCCGAAATCCCCCAAATGA	2736- 2717			
Cas Kin	Casein kinase	MT363976.1	<i>Hordeum vulgare</i>	1F	ACTACAAAGGCCGTCAAGGG	617-636	71	101	
				1R	AACATCCCAAAGGCTAGGGC	687-668			
				2F	AAGATGGATGGCGTGAGTCC	313-332	148	102	
				2R	TGCCAACATAAACCTGCCCA	460-441			
	Casein kinase-like protein	AF479055.1	<i>Triticum aestivum</i>	1F	AACATCTGCTTTCCCGAGCC	255-274	84	100	
				1R	GCCGCCCTTCCATAGCATA	338-320			
				2F	GCAGTCGTCATGTCCCGAA	58-76	81	105	
				2R	ATGGATGCCCTCACTTGGAT	138-119			
Pro Kin	Protein kinase	AJ495779.1	<i>Hordeum vulgare</i>	1F	TGGAATCGTGATGTGGGAGC	565-584	103	105	
				1R	CAGAGGCCGTAAGGTGTTGT	667-648			
				2F	CTAAGAGATCCCCAACGCCC	395-414	89	104	
				2R	CGCACTCCACCAGAGATCAG	483-464			
	Protein kinase	MH551224.1		1F	TTCAGCCATGTCAAAGGCCA	50-69	108	103	

			<i>Triticum aestivum</i>	1R	TAGTCATCCTGCTCACCCCA	157-138		
				2F	GTCTTGGCTGCATGTTTGCT	691-710	186	104
				2R	TTCTGCTGTGCCTTCCAACA	876-857		
Pro Phos	Protein Phosphatase 2B Regulatory Subunit (Calcineurin Regulatory Subunit)	BM816073.1	<i>Hordeum vulgare</i>	1F	GAAGAAGCCGGGTACACACA	525-544	124	99
				1R	ATCCACATTCGCCGTTCACT	648-629		
				2F	CTGGTAGAGGTTTTGCGGGA	447-466	99	99
				2R	CTGTGTGTACCCGGCTTCTT	545-526		
	Serine/threonine- protein phosphatase PP2A-1 catalytic subunit (PP2Ac-1)	EF101900	<i>Triticum aestivum</i>	1F	CGTGTCCAAGAGGTTCCACA	611-630	98	105
				1R	CCAGCACCACGAGGAGAAAT	708-689		
				2F	CAGCCCTGGTGGAATCTGAA	525-544	169	103
				2R	GAAATGCCCCAACCACATCG	693-674		
Myb	transcription factor GAMYB	AK251726.1	<i>Hordeum vulgare</i>	1F	GGGGACACTTCGTCTCATCC	1729-1748	138	110
				1R	GAAAACAGGCTTGCACTCCG	1866-1847		
				2F	GAGTGCAAGCCTGTTTTCGG	1849-1868	115	110
				2R	TGGAGGTTCCGTGAGGAACT	1963-1944		
	R2R3-MYB transcription factor (RIM1)	KU864997.1	<i>Triticum aestivum</i>	1F	ACAGCTTTTGGTCGGAGACA	655-674	120	97
				1R	AGTCCATCTCGTCGTTGCTG	774-755		
				2F	GCCATGCTCGGCAATAGATG	345-364	97	101
				2R	GCTTCTTGAGGTGTGTGTGC	441-422		
Gras	GRAS-domain transcription factor	AK371946.1	<i>Hordeum vulgare</i>	1F	CCTTCTTCATGCCCTCTCG	924-943	117	109
				1R	GGTGGTTTCGGGTGAGGTAG	1040-1021		
				2F	TCCCATGCTGTTCCACGAAG	811-830	91	107
				2R	GTGCTGGTGCTGATACTGGT	901-882		
	GRAS transcription factor SHRCD	KT809308.1	<i>Triticum aestivum</i>	1F	GGCATGTTCCCCTCCGAC	213-230	70	102
				1R	GGTCCTTCCACCCGAGCTT	282-264		
				2F	GGCGCAGTCCGGGTTC	146-161	85	108

				2R	GTCGGAGGGGAACATGCC	230-213				
Mads	MADS-box transcription factor (type II subfamily)	AK370732.1	<i>Hordeum vulgare</i>	1F	TATTTGAAGATGGGGCGGGG	171-190	132	97		
				1R	GGCATCGCAGAGAATACCCA	302-283				
				2F	CGGCAGTTGATGGGACAAGA	501-520	163	107		
				2R	CTTGGTGGACAAGACTCCCC	663-644				
	MADS box transcription factor	AB084577.1	<i>Triticum aestivum</i>	1F	CAACAGCGTGAAAGCAACCA	304-323	97	108		
				1R	CTGGTAGTGCTGGGCATTGA	400-381				
				2F	GGAGGGAAGGCTGGACAAAG	508-527	156	102		
				2R	TGTTGCCCCCTTTCAGTCTC	663-644				
Reference Gene				Sequence (5'->3')			Amplicon size (bp)	Efficiency %		
TubB	beta-tubulin			F	CAAGGAGGTGGACGAGCAGATG		84	90	[24]	
				R	GACTTGACGTTGTTGGGGATCCA					
GAPDH	Glyceraldehyde-3-phosphate dehydrogenase			F	TGTCCATGCCATGACTGCAA		105	101		
				R	CCAGTGCTGCTTGAATGATG					
Calibration gene		species		Sequence (5'->3')			position	Amplicon size (bp)	Efficiency %	
PVinterF	Coat protein	Luteovirus, Barley yellow dwarf virus		F	GTTGAGTTTAAGTCACACGC		3182- 3201	294	99	[24]
YanRA				R	TGTTGAGGAGTCTACCTATTTG		3475- 3454			[29]

Table S2. Expression profile showing the fold change mean values, SEM and TTest.

(A. Barley 10 dai; B. Barley 30 dai; C. Wheat 10 dai; D. Wheat 30 dai)

A Target gene		Graciosa 10 dai		Travira 10 dai		Wbon 10 dai		Wysor 10 dai		V08:3 10 dai		V13:8 10 dai	
		Control	Infected	Control	Infected	Control	Infected	Control	Infected	Control	Infected	Control	Infected
<i>NBS</i>	AVERAGE	1.000	3.843	1.000	1.230	1.000	0.767	1.000	0.874	1.000	0.855	1.000	1.787
	SEM	0.182	0.734	0.112	0.186	0.269	0.461	0.157	0.173	0.069	0.010	0.268	0.621
	TTEST		0.020		0.348		0.665		0.635		0.203		0.309
<i>CC-NBS-LRR</i>	AVERAGE	1.000	5.845	1.000	2.883	1.000	0.773	1.000	0.857	1.000	1.483	1.000	2.387
	SEM	0.116	0.691	0.068	0.717	0.298	0.200	0.131	0.190	0.097	0.169	0.224	0.804
	TTEST		0.002		0.059		0.621		0.561		0.073		0.172
<i>Receptor-like Kinase</i>	AVERAGE	1.000	7.021	1.000	3.266	1.000	0.815	1.000	0.472	1.000	1.089	1.000	2.166
	SEM	0.133	2.019	0.159	0.720	0.370	0.303	0.105	0.129	0.095	0.098	0.545	0.793
	TTEST		0.041		0.037		0.749		0.050		0.578		0.292
<i>Casein Kinase</i>	AVERAGE	1.000	1.723	1.000	2.181	1.000	0.442	1.000	1.158	1.000	1.641	1.000	0.969
	SEM	0.211	0.277	0.290	0.356	0.163	0.184	0.017	0.269	0.148	0.446	0.360	0.133
	TTEST		0.106		0.062		0.113		0.589		0.193		0.940
<i>Protein Phosphatase</i>	AVERAGE	1.000	1.268	1.000	1.664	1.000	0.510	1.000	0.877	1.000	0.949	1.000	0.754
	SEM	0.144	0.180	0.131	0.236	0.150	0.021	0.014	0.177	0.151	0.136	0.125	0.083
	TTEST		0.309		0.070		0.085		0.424		0.831		0.177
<i>Protein Kinase</i>	AVERAGE	1.000	2.769	1.000	1.481	1.000	0.475	1.000	1.027	1.000	1.232	1.000	1.099
	SEM	0.203	0.124	0.033	0.036	0.177	0.142	0.010	0.685	0.069	0.182	0.208	0.293
	TTEST		0.002		0.001		0.129		0.962		0.250		0.797
<i>MYB TF</i>	AVERAGE	1.000	4.540	1.000	0.852	1.000	0.704	1.000	0.549	1.000	0.723	1.000	0.850
	SEM	0.266	0.592	0.175	0.176	0.014	0.070	0.121	0.041	0.129	0.018	0.084	0.012
	TTEST		0.004		0.583		0.013		0.066		0.195		0.152
<i>GRAS TF</i>	AVERAGE	1.000	2.510	1.000	1.258	1.000	1.038	1.000	1.062	1.000	1.080	1.000	0.995
	SEM		0.013	0.098	0.083	0.140	0.136	0.036	0.710	0.060	0.150	0.494	0.219
	TTEST		0.001		0.116		0.866		0.914		0.598		0.993
<i>MADS-box TF</i>	AVERAGE	1.000	4.540	1.000	2.458	1.000	0.739	1.000	1.764	1.000	1.444	1.000	1.832
	SEM	0.109	0.592	0.095	0.024	0.261	0.196	0.100	1.296	0.083	0.367	0.254	0.665
	TTEST		0.888		0.0001		0.528		0.490		0.228		0.307

B Target gene		Graciosa 30 dai		Travira 30 dai		Wbon 30 dai		Wysor 30 dai		V08:3 30 dai		V13:8 30 dai	
		Control	Infected	Control	Infected	Control	Infected	Control	Infected	Control	Infected	Control	Infected
NBS	AVERAGE	1.000	3.314	1.000	2.082	1.000	1.623	1.000	1.208	1.000	0.898	1.000	0.563
	SEM	0.010	0.835	0.243	0.170	0.073	0.392	0.113	0.222	0.136	0.156	0.065	0.075
	TTEST		0.050		0.022		0.194		0.450		0.648		0.012
CC-NBS-LRR	AVERAGE	1.000	2.857	1.000	1.781	1.000	1.413	1.000	1.439	1.000	0.529	1.000	0.436
	SEM	0.165	0.324	0.152	0.414	0.181	0.230	0.111	0.103	0.295	0.133	0.035	0.084
	TTEST		0.007		0.152		0.231		0.044		0.219		0.003
Receptor-like Kinase	AVERAGE	1.000	10.429	1.000	2.677	1.000	0.814	1.000	0.402	1.000	0.204	1.000	0.347
	SEM	0.052	0.964	0.248	0.449	0.170	0.093	0.154	0.122	0.291	0.026	0.096	0.048
	TTEST		0.001		0.031		0.392		0.038		0.053		0.004
Casein Kinase	AVERAGE	1.000	2.621	1.000	1.908	1.000	1.953	1.000	1.129	1.000	0.248	1.000	1.012
	SEM	0.078	0.179	0.203	0.092	0.053	0.246	0.182	0.072	0.062	0.019	0.113	0.195
	TTEST		0.001		0.015		0.019		0.546		0.064		0.960
Protein Phosphatase	AVERAGE	1.000	1.504	1.000	1.109	1.000	1.291	1.000	1.348	1.000	1.418	1.000	0.702
	SEM	0.169	0.103	0.180	0.206	0.105	0.072	0.093	0.201	0.062	0.061	0.087	0.059
	TTEST		0.063		0.711		0.083		0.191		0.009		0.047
Protein Kinase	AVERAGE	1.000	1.775	1.000	1.252	1.000	1.538	1.000	1.524	1.000	1.488	1.000	0.878
	SEM	0.076	0.212	0.119	0.057	0.118	0.209	0.274	0.148	0.149	0.034	0.019	0.084
	TTEST		0.026		0.129		0.088		0.168		0.033		0.228
MYB TF	AVERAGE	1.000	2.106	1.000	1.498	1.000	1.105	1.000	1.038	1.000	0.598	1.000	0.849
	SEM	0.152	0.261	0.206	0.313	0.097	0.098	0.260	0.222	0.099	0.046	0.195	0.140
	TTEST		0.022		0.255		0.489		0.917		0.021		0.563
GRAS TF	AVERAGE	1.000	0.786	1.000	1.208	1.000	1.242	1.000	1.437	1.000	0.476	1.000	0.273
	SEM	0.229	0.039	0.232	0.166	0.071	0.208	0.485	0.371	0.298	0.056	0.159	0.056
	TTEST		0.408		0.507		0.334		0.514		0.159		0.013
MADS-box TF	AVERAGE	1.000	0.569	1.000	1.652	1.000	1.347	1.000	0.658	1.000	0.136	1.000	0.305
	SEM	0.243	0.047	0.339	0.233	0.250	0.537	0.632	0.169	0.408	0.019	0.167	0.155
	TTEST		0.157		0.188		0.590		0.629		0.102		0.169

C Target gene		Semper 10 dai		SGS27-02_10 dai		Tobak 10 dai		Sparta 10 dai		Elan 10 dai		PSR 3628_10 dai	
		Control	Infected	Control	Infected	Control	Infected	Control	Infected	Control	Infected	Control	Infected
NBS	AVERAGE	1.000	31.864	1.000	18.342	1.000	7.319	1.000	5.890	1.000	2.098	1.000	1.887
	SEM	0.119	2.684	0.262	3.064	0.575	1.150	0.229	1.782	0.392	1.276	0.444	0.491
	TTEST		0.0003		0.030		0.008		0.053		0.457		0.251
CC-NBS-LRR	AVERAGE	1.000	13.875	1.000	2.478	1.000	10.377	1.000	4.617	1.000	2.542	1.000	0.765
	SEM	0.736	2.846	0.687	1.226	0.072	2.151	0.324	1.770	0.164	0.702	0.676	0.213
	TTEST		0.012		0.352		0.010		0.115		0.124		0.757
Receptor-like Kinase	AVERAGE	1.000	20.345	1.000	13.595	1.000	2.385	1.000	0.519	1.001	0.689	1.000	0.396
	SEM	0.354	2.165	0.353	4.307	0.210	0.518	0.137	0.091	0.222	0.153	0.204	0.042
	TTEST		0.001		0.043		0.061		0.043		0.177		0.044
Casein Kinase	AVERAGE	1.000	6.161	1.000	1.811	1.000	4.165	1.000	1.652	1.000	2.649	1.000	0.949
	SEM	0.037	1.907	0.071	0.290	0.003	0.398	0.106	0.108	0.575	0.101	0.483	0.196
	TTEST		0.054		0.053		0.002		0.012		0.048		0.927
Protein Phosphatase	AVERAGE	1.000	7.951	1.000	1.438	1.000	1.660	1.000	1.775	1.000	1.860	1.000	0.941
	SEM	0.256	0.992	0.141	0.166	0.076	0.019	0.264	0.305	0.144	0.445	0.470	0.001
	TTEST		0.002		0.115		0.007		0.127		0.140		0.906
Protein Kinase	AVERAGE	1.000	0.397	1.000	1.444	1.000	1.918	1.000	0.849	1.000	1.454	1.000	1.012
	SEM	0.070	0.063	0.143	0.062	0.045	0.321	0.099	0.130	0.335	0.439	0.405	0.056
	TTEST		0.003		0.047		0.034		0.407		0.457		0.978
MYB TF	AVERAGE	1.000	21.074	1.000	1.264	1.000	2.987	1.000	0.672	1.000	0.631	1.000	1.361
	SEM	0.054	2.078	0.060	0.326	0.230	0.083	0.093	0.148	0.173	0.081	0.175	0.282
	TTEST		0.001		0.470		0.007		0.135		0.125		0.338
GRAS TF	AVERAGE	1.000	19.726	1.000	1.208	1.000	5.585	1.000	0.914	1.000	0.535	1.000	1.258
	SEM	0.049	1.612	0.206	0.382	0.035	0.560	0.133	0.042	0.288	0.144	0.435	0.333
	TTEST		0.0003		0.656		0.002		0.568		0.222		0.663
MADS-box TF	AVERAGE	1.000	7.112	1.000	1.385	1.000	2.039	1.000	0.750	1.000	0.933	1.000	0.844
	SEM	0.303	4.337	0.035	0.195	0.043	0.627	0.232	0.243	0.438	0.317	0.378	0.046
	TTEST		0.233		0.124		0.114		0.499		0.907		0.702

D Target gene		Semper 30 dai		SGS27-02 _ 30 dai		Tobak 30 dai		Sparta 30 dai		Elan 30 dai		PSR 3628 _ 30 dai	
		Control	Infected	Control	Infected	Control	Infected	Control	Infected	Control	Infected	Control	Infected
NBS	AVERAGE	1.000	30.701	1.000	1.822	1.000	1.143	1.000	0.524	1.000	4.629	1.000	0.310
	SEM	0.092	8.088	0.184	0.684	0.298	0.282	0.494	0.090	0.241	1.226	0.233	0.034
	TTEST		0.016		0.425		0.707		0.397		0.044		0.043
CC-NBS-LRR	AVERAGE	1.000	5.949	1.000	1.092	1.000	0.258	1.000	0.891	1.000	0.839	1.000	0.443
	SEM	0.019	0.899	0.095	0.163	0.044	0.022	0.072	0.038	0.104	0.133	0.136	0.030
	TTEST		0.005		0.861		0.0001		0.251		0.193		0.013
Receptor-like Kinase	AVERAGE	1.000	9.030	1.000	0.625	1.000	2.638	1.000	0.603	1.000	0.685	1.000	1.152
	SEM	0.034	0.868	0.135	0.105	0.159	0.936	0.024	0.106	0.137	0.081	0.130	0.282
	TTEST		0.001		0.094		0.160		0.011		0.120		0.650
Casein Kinase	AVERAGE	1.000	0.905	1.000	0.922	1.000	1.538	1.000	0.702	1.000	0.916	1.000	0.962
	SEM	0.018	0.158	0.179	0.132	0.034	0.227	0.149	0.088	0.110	0.051	0.125	0.262
	TTEST		0.550		0.743		0.079		0.160		0.529		0.901
Protein Phosphatase	AVERAGE	1.000	0.868	1.000	0.815	1.000	0.462	1.000	0.769	1.000	0.856	1.000	0.639
	SEM	0.002	0.467	0.208	0.102	0.003	0.070	0.304	0.089	0.028	0.009	0.102	0.088
	TTEST		0.729		0.468		0.002		0.506		0.008		0.055
Protein Kinase	AVERAGE	1.000	0.326	1.000	0.747	1.000	1.735	1.000	0.576	1.000	0.822	1.000	0.484
	SEM	0.008	0.045	0.147	0.113	0.071	0.222	0.159	0.026	0.101	0.068	0.155	0.027
	TTEST		0.0001		0.244		0.034		0.058		0.214		0.025
MYB TF	AVERAGE	1.000	2.617	1.000	0.534	1.000	0.475	1.000	0.706	1.000	0.595	1.000	2.195
	SEM	0.089	0.035	0.172	0.161	0.057	0.069	0.020	0.098	0.036	0.073	0.077	0.539
	TTEST		0.001		0.119		0.004		0.043		0.008		0.093
GRAS TF	AVERAGE	1.000	4.910	1.000	1.300	1.000	0.469	1.000	0.704	1.000	1.319	1.000	2.463
	SEM	0.012	1.614	0.330	0.071	0.155	0.117	0.135	0.245	0.085	0.073	0.037	0.530
	TTEST		0.047		0.423		0.052		0.349		0.047		0.051
MADS-box TF	AVERAGE	1.000	1.674	1.000	1.891	1.000	1.765	1.000	0.938	1.000	1.664	1.000	2.064
	SEM	0.060	0.617	0.270	0.352	0.086	0.224	0.080	0.294	0.171	0.121	0.029	0.735
	TTEST		0.244		0.115		0.033		0.716		0.034		0.221

Figure S1. BYDV-PAV titre for all barley and wheat samples with multifactorial ANOVA values.

