

**Supplemental Figure 1. Pseudogenization of Zea mays NRPF1 locus.** Domesticated Zea mays ssp mays and its wild progenitor Z. mays ssp parviglumis (teosinte) both possess a full length NRPE1 paralog but lack an intact NRPF1 coding sequence. In Z. mays ssp mays variety B73, NRPE1 has been misannotated as two independent genes, shown in dark blue and purple. The coding region of NRPF1 is truncated shortly after the G domain and contains a transposable element in intron 4. The teosinte NRPF1 is also truncated after the G domain, but lacks the transposable element found within the Z. mays ssp mays locus.



**Supplemental Figure 2. Pol V subunit paralogs are expressed in rice.** Expression analysis from BAR Rice eFP (<u>http://bar.utoronto.ca/efprice/cgi-bin/efpWeb.cgi</u>) for rice *NRPE1*, *NRPF1*, *NRPD/E2*, and *NRPF2*.

	Z. marina	IHQLRS	SFN	DLI	ΥSÇ	V I	EN N	ΓK	D٦	( G 1	LΡ	ΙA	KF	SVIQFEY-E	VDRPES
	S. polyrhiza	LSQLRN	TYN	ΕIL	DSC	V I	EN H	LΚ	G]	K	ΙP	VΙ	NF	SIVLFEYGL	DDGSNA
_	P. equestris	LSKPRS	SYR	EGQ	DLÇ	VI	EDL	LΚ	II	K	FΡ	ΙT	DF	FVIQFQYRV	KDDIDI
ù	A. officinalis	VPQLRM	SYD	ELL	GLÇ	VI	KD	L N	RI	K	I P	ΙV	AF	SIIQLAY-E	LDEARA
R	M. acuminata	LNQLRS	RRD	ERA	ΕLÇ	$\mathbf{v}$	2 <b>N</b> H	LR	sI	<b>1 K</b> I	) P	ΙV	KF	SLVQLEYVD	DEGVNS
z	E. oleifera	LDQLRS	K Y N	ELV	ELÇ	V I	EN H	LΚ	SI	<b>K</b> 1	LΡ	ΙV	NF	SIIQFEYGE	DGANS
	P. dactylifera	LHQLRS	K Y N	ELV	ELÇ	VI	<b>E N</b> H	LΚ	SI	<b>K</b> 1	ΓD	ΙV	NF	SVIQFEYGE	DGANS
	A. comosus	LDKTRY	TQN	QFV	ЕMF	۱V	<b>E N</b> H	LΚ	SI	ιĸς	<b>2</b> P	ΙV	EF	SVIQFDYGD	DDRVDI
	S. angustifolia	LEQSRC	SKS	QFV	ЕMF	VI	EN N	FΚ	S]	ιĸς	<b>2</b> Q	ΙS	DF	SIMQFKYME	DNEMDS
	B. distachyon	LEQSRC	SKS	QFV	ЕMF	۱V	<b>) N</b> N	LΚ	D	γKζ	<b>2</b> Q	ΙS	DF	SIMQLKYNE	DDATDI
	B. stacei	LEQSRS	SKS	QFV	ΕVF	۱VI	<b>) N</b> N	LΚ	D	γKζ	2 L	ΙS	DF	SIMQLKYNE	DDATDI
~	H. vulgare	LEQSRC	SNS	QSV	EVF	VI	) N N	LΚ	D	γKÇ	<b>2</b> Q	ΙΑ	DF	SIMQLKYKE	EDDTDE
Ж	T. aestivum	LEQSRC	SSS	QSV	ΕTF	۱VI	<b>) N</b> N	LΚ	D	γKζ	<b>2</b> Q	ΙΑ	DF	SIMQLKYKE	EDD <b>T</b> DB
R	A. tauschii	LEQSRC	SSS	QSV	ΕTF	۱V	DN N	LΚ	D	γKÇ	<b>2</b> Q	ΙΑ	DF	SIMQLKYKE	EDDTDE
2	O. sativa	LEQSRF	AEN	QVV	EMF	۱V	DN N	LΚ	D]	ΙKÇ	<b>2</b> Q	IS	DF	SIIQLNYTE	DDALDE
eae	E. tef	LEQSRC	SKT	QFV	EMF	R V I	EN N	LΚ	R 🕻	γKÇ	<b>2</b> Q	IS	DF	SIIQLKYGE	GDDTDF
ac.	S. italica	LEQSRF	SKS	QIA	EMF	l V I	EN N	ΓK	G١	7Κζ	<b>2</b> Q	ΙS	EF	SIIQLKYKE	DDETDE
ö	S. viridis	LEQSRF	SKS	QIA	EMF	l V I	EN N	LΚ	G١	7Κζ	<b>2</b> Q	IS	EF	SIIQLKYKE	DDETDE
_	P. hallii	LEQSRF	SKS	QFV	EMF	۱V	EN N	ΓK	SI	ΓKÇ	<b>2</b> Q	ΙS	DF	SIIQLKYKE	DDETDH
	P. virgatum	LDQSRF	SKS	QFV	EMF	۱V	EN N	ΓK	S	7Κζ	<b>2</b> Q	IS	DF	SIIQFKYKE	DDETDE
	S. bicolor	LEQSRC	STS	QFV	ELF	۱V	EN N	ΓK	s	7Κζ	<b>2</b> Q	IS	DY	SIIQLKYGE	DDEADS
	Z. mays	LEQSRC	STS	QFV	EFF	۱V	EN N	ΓK	ΝN	7 K Ç	<b>2</b> Q	IS	DS	SIIQLKYGE	DDETDS
	S. angustifolia	LGRFR-								1	ΕP	IV	DF	SIVQF	- G S T D Z
	B. distachyon	LDKFR-								1	N P	IV	DF	SVVKF	-DSPNS
_	B. stacei	LDKFR-								I	D P	ΙV	DF	LVVKF	-DSTNS
Ē	H. vulgare	LDKLR-								F	ΚP	ΙV	DL	SIIKF	-DLTNS
R	T. aestivum	LDKLR-								F	ΚP	ΙV	DF	SVVKF	-DSTDS
Z	A. tauschii	LDKLR-								F	ΚP	ΙV	DF	SVVKF	-DSRNS
ae	O. sativa	LNKYR-								I	ΕP	ΙM	DF	LVLQF	-GSRDI
S	E. tef	LNKFR-								B	EL	ΙV	DF	SVLQF	-DSRVV
09	S. italica	LDELR-								B	ES	ΤV	DF	SIVQE	-DSTDV
Ъ	S. viridis	LDELR-								B	E S	ΤV	DF	SIVQP	-DSTDV
	P. hallii	LCELR-								3	r s	ΤL	DF	STVKF	-DSTNV
	P. virgatum	LDEYR-								I	E S	IV	DF	LIVQF	- C S T N Z
	S. bicolor	IDKFS-								F	ΧS	ΙV	DF	SIVQF	-DSTNV

**Supplementary Figure 3. Structural differences between NRPE1 and NRPF1.** Examples of two sequences that are present in all NRPE1 sequences, but are absent in all NRPF1 sequences.



#### Supplementary Figure 4. Duplication of NRPB5/D5, but not NRPE5.

(A) A maximum likelihood phylogenetic tree of *NRPB5/D5* shows a duplication event early in monocot evolution. The exact placement of this duplication is unclear, but there are two highly supported clades (100% bootstrap support, colored green and brown) of Poaceae sequences, suggesting retention of multiple paralogs throughout the grass family. (B) The *NRPE5* maximum likelihood phylogenetic tree closely mirrors the species tree, indicating there has not been a duplication of the Pol V fifth subunit. Branches will 100% bootstrap support are marked with an asterisk. Lineage-specific duplications were collapsed into a single terminal branch.



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**Supplementary Figure 5. Multiple paralogs of NRPB/D/E9 in grasses.** (A) Neighbor-joining tree and (B) alignment of monocot NRPB/D/E9 coding sequences demonstrates there are three groups of sequences in Poaceae (A, B, C). Colored residues disagree with the consensus.



Supplemental Figure 6. Expression patterns of AGO4 paralogs in rice and maize. BAR Rice eFP (<u>http://bar.utoronto.ca/efprice/cgi-bin/efpWeb.cgi</u>) and maize eFP (<u>http://bar.utoronto.ca/efp\_maize/cgi-bin/efpWeb.cgi</u>) expression data demonstrate that OsAGO4a and OsAGO4b are expressed throughout rice development. OsAGO15 might be more narrowly expressed, however we were unable to detect transcripts using RT-PCR in floral tissue. ZmAGO4a/119, ZmAGO4b/104, and ZmAGO15/105 are widely expressed.



# Supplemental Figure 7. *SPT5*, but not *SPT5L*, transcription elongation factors are duplicated in grasses.

Maximum likelihood phylogenetic trees of the conserved domains in *SPT5* (A) and *SPT5L* (B) families demonstrate that *SPT5* duplicated at the divergence of Poaceae. The paralogous clades are highlighted in green and brown. *SPT5L*, the paralog associated with Pol V in Arabidopsis, does not show a similar duplication. Branches will 100% bootstap support are marked with an asterisk. Lineage-specific duplications were collapsed into a single terminal branch.



Supplemental Figure 8. *DRD1* and *CLSY3/4*-type SWI/SNF nucleosome remodeling proteins are duplicated in grasses.

We assessed orthologs of the DRD1-type (including *DRD1*, *CLSY1/2*, and *CLSY3/4* clades) SWI/SNF nucleosome remodeling proteins and discovered duplications of *CLSY3/4* and *DRD1* in grasses. SWI/SNF proteins were recovered from monocot and selected dicot genomes from Phytozome. Reciprocal BLAST and phylogenetic trees were used to classify these proteins and eliminate non-DRD1-type SWI/SNF proteins. (A) An unrooted neighbor-joining tree of selected peptide sequences demonstrates that all sequences are unambiguously assigned to one of three subgroups. Support values from 100 bootstrap replicates. (B-D) Coding sequences were recovered and used to build maximum likelihood phylogenetic trees of the conserved helicase domains of each SWI/SNF subclass. Branches with support values <50 were collapsed and branches with 100% support are marked with an asterisk. The *CLSY1/2* tree (B) recapitulates the expected topology with a few lineage-specific duplications. The *CLSY3/4* (C) and *DRD1* (D) trees identify deeper duplications in the monocot grouping. For both trees placement of the duplication is somewhat unresolved, probably due to errors in annotation, however the additional paralogs (*CLSY5* or *DRD1L*) are only recovered in grass species, suggesting these duplications occurred at the divergence of Poaceae.

## Supplemental Table 1. List of gene sequences used in this study

Gene	Species	Gene ID
NRPE1	A. comosus	Aco023234
NRPE1	A. tauschii	F775_02057
NRPE1	A. officinalis	evm.model.AsparagusV1 06.200
NRPE1	B. distachyon	Bradi4q45065
NRPE1	B. stacei	Brast05G298800
NRPE1	E. oleifera	Unannotated, FGENESH predicted coding sequence
NRPE1	E. tef	snap masked-scaffold3807-abinit-gene-1.26-mRNA-1
NRPF1	H vulgare	MI OC 62016
NRPE1	M acuminata	GSMUA Achr3T05690/ GSMUA Achr3T05700/ GSMUA Achr3T05710
	$\cap$ sativa	
	D. dactylifora	DDK 30-200500
	P. oguostris	$PDR_{3030023210003}$
	P. equesins	Pebol A00210
	P. Ildilli D. virginaturga	
	P. Virgaturii	Pavil. Gauli 147
NRPE1	S. angustitolia	Unannotated, FGENESH predicted coding sequence
NRPE1	S. bicolor	Sobic.004G043750
NRPE1	S. Italica	Seita.1G086100
NRPE1	S. polyrhiza	Spipo2G0033800
NRPE1	S. viridis	Sevir.7G147000
NRPE1	T. aestivum	Traes_6DS_43BCD40B9
NRPE1	Z. mays	GRMZM2G153797 + GRMZM5G851807
NRPE1	Z. marina	Zosma75g00810
Gene	Species	Gene ID
NRPF1	A. tauschii	F775_23577
NRPF1	B. distachyon	Bradi2g61683
NRPF1	B. stacei	Brast01G012100
NRPF1	E. tef	snap_masked-scaffold3548-abinit-gene-3.18-mRNA-1
NRPF1	H. vulgare	MLOC_54009
NRPF1	O. sativa	LOC Os01g73430
NRPF1	P. hallii	Pahal.103212
NRPF1	P. virgatum	Pavir.Aa03252
NRPF1	S. angustifolia	Unannotated, FGENESH predicted coding sequence
NRPF1	S. bicolor	Sobic.003G439200
NRPF1	S. italica	Seita.7G138600
NRPF1	S. viridis	Sevir.1G084000
NRPF1	T. aestivum	Traes 3AL E77A5F03A
NRPF1	Z. mays	GRMZM2G470305
Gene	Species	Gene ID
NRPD/E2	A. comosus	Aco009438
NRPD/E2	A. tauschii	F775_01648
NRPD/E2	A. officinllis	evm.model.AsparagusV1_03.117
NRPD/E2	B. distachyon	Bradi5g23540
NRPD/E2	B. stacei	Brast09G228900
NRPD/E2	E. oleifera	Unannotated, FGENESH predicted coding sequence
NRPD/E2	E. tef	maker-scaffold4272-snap-gene-0.34-mRNA-1
NRPD/E2	H. vulgare	HORVU2Hr1G115600
NRPD/E2	M. acuminata	GSMUA Achr8T17010 001
NRPD/E2	O, sativa	LOC Os04q54840
NRPD/F2	P. dactvlifera	PDK_30s897181L002
NRPD/F2	P. equestris	PEQU 35605
NRPD/F2	P hallii	Pahal G02501
NRPD/F?	P viroatum	Pavir Gb00384
NRPD/F2	S angustifolia	Unannotated EGENESH predicted coding sequence
	J. anguoniona	charmetated, recreter produced boding boquened
NRPD/E2	S. bicolor	Sobic.001G448100

NRPD/E2	S. italica	Seita.7G258200
NRPD/E2	S. polyrhiza	Spipo15G0003500
NRPD/E2	S. viridis	Sevir.7G269600
NRPD/E2	T. aestivum	Traes_2DL_4FD242B37
NRPD/E2	Z. mays	GRMZM2G054225_T01
NRPD/E2	Z. marina	Zosma248g00150
		<b>v</b>
Gene	Species	Gene ID
NRPF2	A. tauschii	F775 00990
NRPF2	B. distachvon	Bradi3g15852
NRPF2	B. stacei	Brast03G026500
NRPF2	E. tef	maker-scaffold17955-snap-gene-0.28-mRNA-1
NRPF2	H. vulgare	HORVU7Hr1G076850
NRPF2	O. sativa	LOC Os08g07480
NRPF2	P. hallii	Pahal.F01121
NRPF2	P. virgatum	Pavir.Fb00476
NRPF2	S. bicolor	Sobic.007G056900
NRPF2	S. italica	Seita.6G047400
NRPF2	S. viridis	Sevir.6G044700
NRPF2	T. aestivum	Traes_7DL_222F95DC4
NRPF2	Z. mays	GRMZM2G133512_T01
Gene	Species	Gene ID
NRPB/D5	A. coerulea	Aqcoe7G260800
NRPB/D5a	A. comosus	Aco018959
NRPB/D5b	A. comosus	Aco024268
NRPB/D5	A. thaliana	AT3G22320
NRPB/D5	A. trichopoda	evm_27.model.AmTr_v1.0_scaffold00068.11
NRPB/D5a	B. distachyon	Bradi2g52777
NRPB/D5b	B. distachyon	Bradi5g13967
NRPB/D5a	E. oleifera	
NRPB/D30	E. Olellera	maker coeffeld 1202 augustus gans 2.17 mDNA 1
NRPD/D3 NDDD/D5		maker-scarou izgz-augustus-gene-z. 17-mkina-i mrpa05214.1 v1.0 bybrid
NRPB/D52	N acuminata	GSMUA Achrilin randomT09620 001
NRPR/D5h	M. acuminata	GSMUA Achr2T00920_001
NRPB/D5a	0 sativa	LOC Os01059140
NRPB/D5b	O. sativa	LOC Os04g41040
NRPB/D5a	P. dactvlifera	cds10971
NRPB/D5b	P. dactylifera	cds22816
NRPB/D5a	P. hallii	Pahal.E01240
NRPB/D5b	P. hallii	Pahal.G00686
NRPB/D5	P. trichocarpa	Potri.006G161600
NRPB/D5	S. angustifolia	STRANG_00084297-RA
NRPB/D5a	S. bicolor	Sobic.003G328700
NRPB/D5b	S. bicolor	Sobic.006G127500
NRPB/D5a	S. Italica	Seita.5G352900
NRPB/D5b	S. Italica	Seita. /G144900
NKPB/D5C	J. Italica	Selia.30031100
NRFD/DO NDDD/D5a	S. pulyITIIZa	Spip0030040300
NRDR/DSA	S. VIIIUIS S. Viridia	Sevir 7G15350000
NRPR/D50	S. viriais S. viridis	Sevir 5G050500
NRPR/D52	J. VIIIUIS	Traes 3B 37BBD6C41
NRPR/D5h	T aestivum	Traes 2BL 1FD85D348
NRPB/D5	T. cacao	Thecc1EG037836t1
NRPB/D5	V. vinifera	GSVIVT01015349001
NRPB/D5	Z. marina	Zosma440g00110
		-

NRPB/D5	Z. mays	GRMZM2G099183
Gene	Species	Gene ID
NRPE5	A. coerulea	Agcoe2G036200
NRPE5	A. comosus	Agcoe2G036200
NRPE5	A. thaliana	AT3G57080
NRPE5	A. trichopoda	evm 27.model.AmTr v1.0 scaffold00116.10
NRPE5	B. distachvon	Bradi4g26550
NRPE5	F. vesca	mrna21892.1-v1.0-hvbrid
NRPE5	M. acuminata	GSMUA Achr9T19340 001
NRPE5	O. sativa	LOC Os12q04510
NRPE5	P. dactylifera	PDK 30s65509269L001
NRPE5	P. equestris	PEQU 23089
NRPE5	P. hallii	Pahal.H00047
NRPE5	P. trichocarpa	Potri.001G025500
NRPE5	P. virgatum	Pavir.Ha01857
NRPE5	S. angustifolia	STRANG_00052800-RA
NRPE5	S. bicolor	Sobic.005G030600
NRPE5	S. italica	Seita.8G027200
NRPE5	S. viridis	Sevir.7G328000
NRPE5	V. vinifera	GSVIVT01033595001
NRPE5	Z. marina	Zosma192g00210
NRPE5	Z. mays	GRMZM2G162697
	-	
Gene	Species	Gene ID
NRPB/D/E9a	A. comosus	AcoU01788
NRPB/D/E9b	A. comosus	Aco027798
NRPB/D/E9C	A. comosus	Acou09500
NRPB/D/E9	A. officinalis	evm.model.AsparagusV1_07.1236
NRPB/D/E9a	B. distachyon	Bradi4g04820
NRPB/D/E9b	B. distachyon	Bradi1g/8509
NRPB/D/E9c	B. distachyon	Bradi1g20820
NRPB/D/E9a	B. stacell	Brast10G053700
NRPB/D/E9b	B. stacell	Brast02G002000
NRPB/D/E9C	B. stacell	Brast06G047100
NRPB/D/E9a	M. acuminata	GSMUA Achr10G02200 001
NRPB/D/E90	M. acuminate	GSMUA Achr/105620.001
NRPB/D/E9C	M. acuminata	GSMUA ACNI4G24050 001
NRPB/D/E9a	O. sativa	LOC Os12g37370
NRPB/D/E90	O. sativa	
NRPD/D/E90	O. Saliva D. dootyliforo	LOC OS07942395 ede/202
NRPD/D/E9a	P. dactylilera	CUS4303
NRFD/D/E90 NDDR/D/E00	P. Uaciyiliela D. oquostris	
NRFD/D/E9a NDDD/D/E0b	P. equestris	PEQU_29040 DECIL 21522
NRFD/D/E90 NRPR/D/E0a	P. equesins D. ballii	PEQU_21525 Pabal 10/1586
NRFD/D/L9a NRPR/D/E0b	P. nallii D. hallii	Pahal C01606
NRPD/D/E90	P. nallii D. hallii	Pahal 80/703
NRPD/D/E90	P. nami D. virgatum	Pallal.004700 Pavir Ca02562
NRPR/D/E9a	P virgatum	Pavir. 116609
NRPR/D/E90	P virgatum	Pavir Ba00418
NRPR/D/E92	S angustifolia	STRANG 00026340-RA
NRPR/D/E9a	S angustifolia	STRANG 00027772-RA
NRPR/D/E90	S. angustifolio	STRANG 00003454-RA
	S hicolor	Sobie 008G128900
NRPR/D/EQA	S hicolor	Sobic 009G165600
NRPR/D/Far	S hicolor	Sobic 002G374200
NRPR/D/F02	S italic	Seita 3G335700
	O. nano	

NRPB/D/E9b	S. italic	Seita.7G044300
NRPB/D/E9c	S. italic	Seita.2G388500
NRPB/D/E9	S. polyrhiza	Spipo2G0057100
NRPB/D/E9a	S. viridis	Sevir.3G350200
NRPB/D/E9b	S. viridis	Sevir.7G049300
NRPB/D/E9c	S. viridis	Sevir.2G399000
NRPB/D/E9	Z. marina	Zosma25g01500
NRPB/D/E9a	Z. mays	GRMZM2G046061
NRPB/D/E9b	Z. mays	GRMZM2G023028
NRPB/D/E9c	Z. mays	GRMZM5G898768
Gene	Species	Gene ID
AGO4	A. comosus	Aco017860
AGO4a	A. tauschii	F775_27134
AGO4b	A. tauschii	F775_27714
AGO4	A. officinalis	evm.model.AsparagusV1_07.1132
AGO4a	B. distachyon	Bradi2g10370
AGO4b	B. distachyon	Bradi2g14147
AGO4a	B. stacei	Brast01G298200
AGO4b	B. stacei	Brast08G001100
AGO4	E. oleifera	Unannotated, FGENESH predicted coding sequence
AGO4a	E. tef	maker-scaffold315-augustus-gene-0.27-mRNA-1
AGO4b	E. tef	snap_masked-scaffold1886-abinit-gene-0.21-mRNA-1
AGO4a	H. vulgare	HORVU3Hr1G038830
AGO4b	H. vulgare	HORVU1Hr1G095300
AGO4	M. acuminata	GSMUA_Achr9T05150
AGO4a	O. sativa	LOC_Os01g16870
AGO4b	O. sativa	LOC_Os04g06770
AGO4	P. dactylifera	cds36720
AGO4	P. equestris	PEQU_18408
AGO4a	P. hallii	Pahal.E03348
AGO4b	P. hallii	Pahal.C01666
AGO4a	P. virgatum	Pavir.Eb01138
AGO4b	P. virgatum	Pavir.J00737
AGO4a	S. angustifolia	Unannotated, FGENESH predicted coding sequence
AGO4b	S. angustifolia	Unannotated, FGENESH predicted coding sequence
AGO4a	S. bicolor	Sobic.003G129500
AGO4b	S. bicolor	Sobic.009G259900
AGO4a	S. italica	Seita.5G043300
AGO4b	S. italica	Seita.3G117800
AGO4	S. polyrhiza	Spipo3G0017600
AGO4a	S. viridis	Sevir.5G040900
AGO4b	S. viridis	Sevir.3G120100
AGO4a	T. aestivum	TRIAE_CS42_3AS_TGACv1_211331_AA0688710
AGO4b	T. aestivum	TRIAE_CS42_1DL_TGACv1_062840_AA0220790
AGO4a/119	Z. mays	GRMZM2G589579
AGO4b/104	Z. mays	GRMZM2G141818
Gene	Species	Gene ID
AGO15	A. tauschii	F775_02440
AGO15	B. distachyon	Bradi2g10360
10015	B. stacei	Brast01G298300
AGU15		
AGO15 AGO15	E. tef	maker-scaffold1490-snap-gene-1.50-mRNA-1
AGO15 AGO15 AGO15	E. tef H. vulgare	maker-scaffold1490-snap-gene-1.50-mRNA-1 HORVU7Hr1G107770
AGO15 AGO15 AGO15 AGO15	E. tef H. vulgare O. sativa	maker-scatfold1490-snap-gene-1.50-mRNA-1 HORVU7Hr1G107770 LOC_Os01g16860
AG015 AG015 AG015 AG015 AG015	E. tef H. vulgare O. sativa P. hallii	maker-scaffold1490-snap-gene-1.50-mRNA-1 HORVU7Hr1G107770 LOC_Os01g16860 Pahal.E03353
AG015 AG015 AG015 AG015 AG015 AG015	E. tef H. vulgare O. sativa P. hallii P. virgatum	maker-scatfold1490-snap-gene-1.50-mRNA-1 HORVU7Hr1G107770 LOC_Os01g16860 Pahal.E03353 Pavir.Eb01136

AGO15	S. italica	Unannotated, FGENESH predicted coding sequence
AGO15	S. viridis	Sevir.5G041600
AGO15/105	Z. mays	GRMZM2G469673/GRMZM2G048335/GRMZM2G089743
Gene	Species	Accession
SPT5	A. thaliana	AT2G34210
SPT5	A. trichopoda	evm_27.model.AmTr_v1.0_scaffold00002.351
SPT5	A. comosus	Aco002639.1
SPT5	A. officinalis	evm.model.AsparagusV1_05.376
SPT5a	B. distachyon	Bradi1g46230.3.p
SPT5b	B. distachyon	Bradi3g57590.2.p
SPT5a	B. stacei	Brast07G070000.1
SPT5b	B. stacei	Brast04G034700.1
SPT5	F. vesca	gene30070v1.0hybrid
SPT5	M. acuminata	GSMUA_Achr3P08080_001
SPT5a	O. sativa	LOC_Os06g10620.1
SPT5b	O. sativa	LOC_Os02g53230.1
SPT5a	P. hallii	Pahal.D02854.1
SPT5b	P. hallii	Pahal.G02677.1
SPT5	P. trichocarpa	Potri.009G124200.1
SPT5	P. dactylifera	LOC103718524
SPT5a	P. virgatum	PavirDb02038.1
SPT5b	P. virgatum	Pavir.Aa00338.1
SP15a	S. angustitolia	STRANG 00000456RA
SPT5a	S. italica	Seita.4G084400.1.p
SP15b	S. italica	Seita.1G335500.1
SP15	S. polyrhiza	Spipo10G0038700
SP15a	S. viridis	Sevir.4G083700.1
SP15b	S. viridis	Sevir.1G342100.1
SP15	I. cacao	
SP15	Z. marına	Zosma52g01330.1
SP15a	Z. mays	GRMZM2G142072_P01
SP15a	Z. mays	GRMZM2G171488_102
Gene	Species	Accession
SP15L	A. thaliana	A 15G04290.1
SPISL	A. trichopoda	
SPISL	A. COMOSUS	AC0005272.1
SPISL	B. distacriyon	B1a012g20517.2 Broat08C068200.2
SPIJL		DIdSU00000000.2
SPISE	r. vesca M. ocuminata	CSMUA Achr7D16000 001
SPTSL	$\cap$ sativa	$OC \Omega = 0.5 \times 130 \times 100 \times 1000 \times 100 \times 10$
SPT5I	D. saliva D. hallii	Pabal C02/15 2
SPT5I	P trichocarna	Potri 008G032800 1
SPT5I	P trichocarpa	Potri 010G228800 1
SPT5I	P virgatum	Pavir Ca01318
SPT5I	S italica	Seita 3G186400 1
SPT5I	S nolvrhiza	Spino3G0088300
SPT5I	S viridis	Sevir 3G191100 1
SPT5I	T cacao	Thecc1EG043328t1
SPT5I	V vinifera	GSVIVT01016203001
SPT5L	Z. mays	GRMZM2G375222 P01
SPT5L	Z. marina	Zosma159g00420.1
Gene	Species	Accession
CLSY1	A. comosus	Aco011099
CLSY1	A. thaliana	AT3G42670

Gene	Species	Accession
CLSY2	A. thaliana	AT5G20420
CLSY1	B. distachyon	Bradi1g16720
CLSY1	B. stacei	Brast06G002700
CLSY1	F. vesca	aene21045-v1.0-hvbrid
CLSY1	M. acuminata	GSMUA Achr8G01860 001
CLSY1	O. sativa	
CLSY1	P hallii	Pahal .101776
CLSY1a	P trichocarna	Potri 008G073500
CLSY1b	P trichocarpa	Potri 010G183800
CLSV1a	P virgatum	Pavir Ba00302
CLSV1h	P virgatum	Pavir Bb03632
CLSY1	S angustifolia	STRANG 00023332-RA
CLSY1a	S italica	Seita 2G442300
CLSV1b	S italica	Seita 96461000
CLSY1	S viridis	Sevir 2G454400
CLSY1	T cacao	Thecc1EG042431
CLSY1	V vinifera	GSVIVG01032746001
CLSY1	7 mays	GRMZM2G108166
	<u> </u>	
Gene	Species	Accession
CLSY3	A. comosus	LG08:1256212112567155
CLSY3	A. officinalis	evm.model.AsparagusV1 04.2656
CLSY3	A. thaliana	AT1G05490
CLSY4	A. thaliana	AT3G24340
CLSY3	F. vesca	gene10657-v1.0-hybrid
CLSY3	F. vesca	gene10658-v1.0-hybrid
CLSY3	F. vesca	gene23334-v1.0-hybrid
CLSY3	M. acuminata	GSMUA Achr7G06840 001
CLSY3	O. sativa	LOC Os05a32610
CLSY3	P. hallii	Pahal.F01921
CLSY3	P. trichocarpa	Potri.014G050200
CLSY3a	P. virgatum	Pavir.J01490
CLSY3b	P. virgatum	Pavir.Cb01678
CLSY3	S. angustifolia	STRANG 00015789-RA
CLSY3	S. italica	Seita.3G254300
CLSY3	S. viridis	Sevir.3G260900
CLSY3	S. polyrhiza	Spipo5G0053000
CLSY3	T. cacao	Thecc1EG036301
CLSY3	V. vinifera	GSVIVG01013277001
CLSY3	Z. marina	Zosma89g01000.1
CLSY3/RMR1	Z. mavs	GRMZM2G154946
CLSY5	B. distachyon	Bradi3q50300
CLSY5	B. distachyon	Bradi2g26500
CLSY5	B. distachvon	Bradi2g43501
CLSY5	B. stacei	Brast01G217700
CLSY5	B. stacei	Brast04G120300
CLSY5	B. stacei	Brast08G132400
CLSY5	O. sativa	LOC Os02q43460
CLSY5	P. hallii	Pahal.A02764
CLSY5a	P. virgatum	Pavir.Aa01193
CLSY5b	P. virgatum	Pavir.Ab02445
CLSY5a	S. anaustifolia	STRANG 00083573-RA
CLSY5b	S. angustifolia	STRANG 00018510-RA
CLSY5c	S. angustifolia	STRANG 00037404-RA
CLSY5d	S. anaustifolia	STRANG 00042628-RA
CLSY5e	S. angustifolia	STRANG 00038294-RA
CLSY5	S. viridis	Sevir.1G259000
	-	

CLSY5/RMR1 -like	Z. mays	GRMZM2G178435
Gene	Species	Accession
DRD1	A. comosus	Aco017413
DRD1	A. thaliana	AT2G16390.1
DRD1a	B. distachyon	Bradi2g21450
DRD1b	B. distachyon	Bradi3g19890
DRD1L	B. distachyon	Bradi1g74070
DRD1	B. stacei	Brast03G065000
DRD1a	B. stacei	Brast08G078200
DRD1b	B. stacei	Brast03G077600
DRD1L	B. stacei	Brast02G053000
DRD1	F. vesca	gene21858-v1.0-hybrid
DRD1	O. sativa	LOC_Os08g14610
DRD1a	O. sativa	LOC_Os07g25390
DRD1b	O. sativa	LOC Os06g14440
DRD1L	O. sativa	LOC_Os03g06920
DRD1	P. hallii	Pahal.B01415
DRD1L	P. hallii	Pahal.100535
DRD1a	P. tricocarpa	Potri.004G159000
DRD1b	P. trichocarpa	Potri.009G120700
DRD1a	P. virgatum	Pavir.Bb00923
DRD1b	P. virgatum	Pavir.J23343
DRD1a	S. angustifolia	STRANG_00017656-RA
DRD1b	S. angustifolia	STRANG_00048730-RA
DRD1c	S. angustifolia	STRANG_00048722-RA
DRD1L	S. angustifolia	STRANG_00017091-RA
DRD1a	S. italica	Seita.6G109100
DRD1b	S. Italica	Seita.9G302200
DRD1L	S. italica	Seita.9G529000
DRD1L	S. viridis	Sevir.9G533600
DRD1	S. polyrhiza	Spipo9G0003300
	I. cacao	
DRD1	V. vinitera	GSVIVG01024225001
DRD1a	∠. mays	GRIMZM2G5/4858
DRD1b	∠. mays	GRMZM2G393742

## Sup. Table 2. Reverse transcriptase PCR primers

Primer	Sequence
OsE1 F	GGCCAAAGAAACAATCCTGGTAG
OsE1 R	CTCACTAGTGGTATCAACTGCAGTAGACTGTGT
OsF1 RT F	GCGAGTTGTGCTTTTACACCAGT
OsF1 R	CTCACTAGTTTTGAAGTACTTTCTGCAGAATGAATCT
OsDE2 RT F	GAGAAAGGATGACTTTGCCAGAC
OsDE2 RT R	GGTGATCGAACCTGTCTTAGAGTA
OsF2 RT F	GGAAGCAAACATGACTTTGCCAG
OsF2 RT R	CGAGCGAGTCTGTCTTAGAGTAA

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#### Sup. Table 3. NRPE1 and NRPF1 CTD characteristics

	length <sup>1</sup>	Ago hooks
NRPE1, Non-Poaceae		
Ananas comosus	431	9
Asparagas officinalis	539	8
Elaeis oleiferia	291	5
Musa acuminata	375	6
Phalaenopsis equestris	471	8
Phoenix dactylifera	313	7
Spirodela polyrhiza	246	4
Zostera marina	707	23
NRPE1, Poaceae		
Aegilops tauschii	444	15
Brachypodium distachyon	513	15
Brachypodium stacei	496	15
Eragrostis tef	595	14
Oryza sativa	541	13
Panicum hallii	506	13
Panicum virgatum	526	15
Streptochaeta angustifolia	585	16
Seteria italica	541	16
Seteria viridis	541	15
Sorghum bicolor	538	14
Triticum aestivum	468	16
Zea mays	468	9
NRPF1, Poaceae		
Aegilops tauschii	314	5
Brachypodium distachyon	265	2
Brachypodium stacei	288	2
Hordeum vulgare	324	3
Oryza sativa	249	0
Panicum virgatum	241	2
Streptochaeta angustifolia	189	1
Seteria italica	242	1
Seteria viridis	242	1
Sorghum bicolor	276	2
Triticum aestivum	324	4

## Sup. Table 4. Evidence of positive selection following the duplication of *NRPE1*.

NRPE1	Position <sup>1</sup>	BEB score	Amino acid <sup>1</sup>	Ancestral / F1 amino acid <sup>1</sup>	Surface (S) / Interior (I)	Feature/Structure
High confidence (sites on branch)	513	0.66	М	F/Y	Surface	Pore
Lower confidence (sites in clade)	1049	0.990**	G	Р	Interior	Jaw

NRPF1	Position <sup>1</sup>	BEB score	Amino acid <sup>1</sup>	Ancestral / E1 amino	Surface (S) /Interior (I)	Structure / Feature in template model
				acid <sup>1</sup>		
High confidence	539	0.878	К	Р	Surface	Pore / Near RBP 8 surface
(sites on branch)						
	503	0.873	К	V/G	Surface	Pore
	742	0.845	K	V/A	Surface	Funnel-Bridge helix / RBP2 interaction site
	304	0.823	F	E/D	Surface	Clamp-Rudder
	349	0.768	E	S	Surface	Active site pocket
	1184	0.734	А	С	Surface	Clamp
	223	0.609	S	Q/K	Surface	Clamp / RPB2 interaction site
	825	0.597	1	Ν	Surface	Foot
	1034	0.59	А	К	Surface	Cleft
	899	0.554	Н	S/P	Surface	Jaw / Near RPB2 surface
	367	0.534	А	K/N	Surface	Dock
Lower confidence	226	0.985*	С	Р	Surface	Clamp / Near RPB2 surface
(sites in clade)						
	234	0.998**	V	V/I	Surface	Clamp
	235	0.991**	S	Р	Surface	Clamp-Lid
	237	0.980*	V	l/F	Surface	Clamp-Lid
	240	0.999**	G	G	Surface	Clamp-Lid / Near RPB2 surface
	243	0.976*	F	1	Surface	Clamp-Lid
	244	0.955*	S	М	Surface	Clamp-Lid
	254	0.966*	R	К	Surface	Clamp
	259	0.976*	K	K	Surface	Clamp
	270	0.972*	S	S	Surface	Clamp
	301	0.984*	D	D	Surface	Clamp-Rudder / Near RPB2 surface
	309	0.954*	Т	S	Surface	Clamp-Rudder
	319	0.995**	Т	Τ	Surface	Clamp

341	0.976*	G	G	Surface	Active site pocket
360	0.999**	К	Q/R	Surface	Dock
368	0.980*	Q	R	Surface	Dock
371	0.994**	D	E	Surface	Dock
382	0.991**	Т	R/K	Surface	Dock
395	0.987*	К		Surface	Dock / Indel relative to NRPE1
396	0.996**	D		Surface	Dock / Indel relative to NRPE1
399	0.977*	Ν	S	Surface	Dock / Near Rbp2 surface
402	0.995**	Н	H/Y	Surface	Dock
420	0.991**	1	I/V	Surface	Active site pocket
440	0.987*	Q	Υ	Surface	Active site pocket
496	0.974*	G	G	Interior	Pore
521	0.969*	R	K	Surface	Pore
532	0.957*	S	V/L	Interior	Pore
556	0.992**	Т	А	Interior	Pore
557	0.993**	K	L	Surface	Pore
558	0.998**	S	Р	Surface	Pore
678	0.999**	Р	L	Surface	Funnel
696	0.993**	L	F	Surface	Funnel / Near RPB2 surface
718	0.989*	Т	L	Surface	Funnel / Near RPB2 surface
739	0.966*	E	S/T	Surface	Funnel-Bridge helix / Near RPB2 surface
755	0.976*	S	Т	Surface	Funnel-Bridge helix / Near RPB2 surface
843	0.983*	А	N	Surface	Cleft / Near RPB5 surface
861	0.999**	Ν	K	Surface	Cleft
862	0.973*	E	K	Surface	Cleft
889	0.961*	K	E	Surface	Jaw
921	0.993**	S	E/D	Surface	Jaw
929	1.000**	Т	V	Surface	Jaw
940	0.999**	М	L	Interior	Jaw
949	0.977*	F	L/F	Surface	Jaw
977	0.963*	G	D/E	Surface	Jaw
987	0.964*	Т	L/V	Surface	Jaw / Near RPB9 surface
988	0.985*	E	E	Surface	Jaw / Near RPB9 surface
993	0.987*	F	I/V	Surface	Jaw / Near RPB9 surface
1025	0.991**	S	А	Surface	Cleft / Near RPB9 surface
1054	0.978*	S	Ν	Interior	Cleft
1057	0.989*	А	A	Interior	Cleft
1078	0.991**	Μ	1	Surface	Cleft / Near RPB5 surface
1087	0.957*	Q	E/D	Surface	Cleft / Near RPB5 surface
1093	0.999**	S	С	Surface	Cleft / Near RPB5 surface
1099	0.998**	Т	V	Surface	Cleft

1128	0.965*	Q	К	Surface	Cleft / Near RPB5 surface
1174	0.954*		Μ	Surface	Clamp / Near RPB5 surface
1201	0.999**	Ν	H	Surface	Clamp

<sup>1</sup> Relative to *O. sativa* paralogs

NRPF2	Position <sup>1</sup>	BEB	Amino acid <sup>1</sup>	Ancestral /	Surface (S) /	Feature/Structure
		Score		acid <sup>1</sup>		
High confidence				4014		
(sites on branch)	747	0.989*	М	Y	Surface	Rbp2 domain 6 / Near RBP1 surface
	614	0.902	E	S	Surface	Rbp2 domain 4 / Near RPB9 surface
	299	0.864	G	Y	Surface	Rbp2 domain 2
	741	0.853	Т	F	Surface	Rbp2 domain 6
	682	0.815	Ν	E	Surface	Rbp2 domain 5
	139	0.568	V	S	Surface	Protrusion
	383	0.51	Μ	L	Interior	Rbp2 domain 2
Lower confidence						
(sites in clade)	75	0.951*	R	N/D	Surface	Protrusion
	99	0.998**	F	F	Interior	Protrusion
	102	0.988*	L	L	Interior	Protrusion
	105	0.964*	V	V	Interior	Protrusion
	133	0.951*	E	E	Surface	Protrusion
	138	0.984*	W	W	Surface	Protrusion
	146	0.975*	V	E	Surface	Protrusion
	208	0.967*	К	R	Surface	Protrusion
	223	0.959*	L	L	Surface	Protrusion
	227	0.998**	E	D	Interior	Protrusion
	261	0.997**	S	K/R	Surface	Rbp2 domain 2
	274	0.990**	R	K	Surface	Rbp2 domain 2
	279	0.972*	S	Y	Surface	Rbp2 domain 2
	283	0.990**	Q	E/D	Interior	Rbp2 domain 2
	441	1.000**	Μ	R	Surface	
	442	1.000**	Н	Н	Surface	
	444	1.000**	Q	E	Surface	
	457	0.998**	G	G/S	Surface	
	473	0.997**	V	I/V	Interior	
	515	0.989*	Μ	L	Interior	Rbp2 domain 3
	522	0.999**	V	V	Surface	Rbp2 domain 3
	525	0.999**	А	А	Surface	Rbp2 domain 3
	527	0.999**	К	K	Surface	Rbp2 domain 3 / Near RPB1 surface
	599	1.000**	L	Р	Surface	
	611	0.999**	D	D	Surface	Rbp2 domain 4 / Near RPB9 surface
	639	0.989*	Q	Н	Surface	Rbp2 domain 4 / Near RPB1+RPB9 surfaces
	658	0.972*		1/1/	Surface	Rbp2 domain 4

Sup. Table 5. Evidence of positive selection following the duplication of *NRPD/E2*.

6	64	0.996**	Ν	K	Surface	
6	670	0.998**	Ν	G	Surface	
6	699	1.000**	Y	W	Surface	Rbp2 domain 5 / Near RBP1 surface
7	743	0.999**	К	R	Surface	Rbp2 domain 6 / Near RBP1 surface
7	752	0.998**		Н	Interior	Rbp2 domain 6
7	761	0.986*	Т	Т	Surface	Rbp2 domain 6
7	75	0.974*	L	L	Surface	Rbp2 domain 6
8	300	0.952*	S	R	Surface	Rbp2 domain 6
8	319	0.993**	I	V	Surface	Rbp2 domain 6 / Near RBP1 surface
8	333	0.991**	L	М	Interior	Rbp2 domain 6
8	346	0.998**	Q	E	Surface	Rbp2 domain 6
8	371	0.989*	Ν	D	Surface	Rbp2 domain 6
8	387	0.998**	I	D	Surface	Rbp2 domain 6
9	916	0.997**	I	I	Surface	Rbp2 domain 6
9	927	0.954*	E	Q	Surface	Rbp2 domain 6
9	938	0.999**	Т	K	Surface	Rbp2 domain 6
9	942	0.959*	F	V	Interior	Rbp2 domain 6
9	948	0.989*	Т	V	Surface	Rbp2 domain 6
1	073	1.000**	Н	I	Surface	Rbp2 domain 6
1	134	0.998**	V	F	Surface	Rbp2 domain 7 – Zinc finger / Near RBP1 surface
1	154	1.000**	R	R	Surface	Rbp2 domain 7 – Zinc finger
1	177	1.000**	R	Ν	Surface	Rbp2 domain 7 – Zinc finger / Near RBP1 surface
1	181	0.992**	V		Surface	Rbp2 domain 7 / Near RBP1 surface
1	196	1.000**	S	S	Surface	Rbp2 domain 7 / Near RBP1 surface
1	207	0.984*	L	V	Surface	Rbp2 domain 7 / Near RBP1 surface

<sup>1</sup> Relative to *O. sativa* paralog