

## Supplementary information for

### **A meiosis-specific Spt5 homolog involved in non-coding transcription**

Julita Gruchota, Cyril Denby Wilkes, Olivier Arnaiz, Linda Sperling  
and Jacek K. Nowak

**Supplementary Figures**

**Supplementary Tables**

**Supplementary References**

# Supplementary Figures

**A**

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Pt_Spt5m      1  -----
Pt_Spt5v      1  -----
Tt_Spt5       1  -----
Ot_Spt5-g19   1  -----
Ot_Spt5-g12   1  MICNLIKLCFDIQIKQEEGAQPQQNQKIKSKKQQQPKDEDESESDSEQLKRRMRGKKRLSINDYLDDVEDDQQA-----
At_SPT5-1     1  -----
At_SPT5-2     1  -----
At_KTF1       1  -----
Dm_Spt5       1  -----
Hs_Spt5       1  -----
Tb_Spt5       1  -----
Sc_Spt5       1  -----MSDNSDTNVMQDHD-----QQFADPVVVPQSTDTKDENT
Pf_Spt5       1  -----
  
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Pt_Spt5m      1  -----
Pt_Spt5v      1  -----MSQKNDDYRS
Tt_Spt5       1  -----MSDD-----FLEEDNNSYNY
Ot_Spt5-g19   1  -----
Ot_Spt5-g12   73 ADTN YARKGSKTLSKEFDKKLNTVDDKAPTKDSQLLKKRSKPQTS TSELEQRVKKEYQRNQRNDDSDAQMKKENE-CNS
At_SPT5-1     1  -----MSQY-----S-----DDD SHEDD-SEM
At_SPT5-2     1  -----MPRS-----RDEDELDGDYEALDL-EEE
At_KTF1       1  -----
Dm_Spt5       1  -----MSSEV-----SNMSDSGSEDGSI SNKSQRSAR-SKS
Hs_Spt5       1  -----MSSED-----SNFSEED-----
Tb_Spt5       1  -----MS-----
Sc_Spt5       36 SDKD TVDSGNVTTT ESTE-RAES-TSNIPPLDGE EKEAKSE PQP DN AETAATEQVS--S-SNGPATDDA QATLN-TDS
Pf_Spt5       1  -----
  
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Pt_Spt5m      1  -----
Pt_Spt5v      11 DQD-----DKSQSS-----I SIE L P D S S S E Q K Q S K S K-----
Tt_Spt5       17 DDE-----E-----D G S Y G E V S N A K R K-----
Ot_Spt5-g19   1  -----MKKEVRYE D H Y D E E D V S Y E E V K N K R S--K Q T K--
Ot_Spt5-g12   152 DT E G G I I N T K S N Q A A K S Q E A K Q S S Q L N K Q P V K-----D E N L K K R R E A--K Q D D--
At_SPT5-1     18 E D E E-----E D E Y E P S S R K G R S--G K K R G R S N S D-----
At_SPT5-2     24 E E D E-----E E E E E G R G G G S--R R K R G R S N F I D D Y A E E D S Q E E D--D E E Y G S S R G--
At_KTF1       1  -----MDR-----K G K G K Q V-----A G S D S Y G G Q R K R N S V E F--
Dm_Spt5       32 R S R S R G S R G S R S V S R S R S R S Q S G S R S S E S P Q R R D N R G K S D E S-G E E E E P P G E I--D S E Y D E E N--
Hs_Spt5       15-----S E S S D E E A E V D E E R S A A G S E-K E E P E D E E E--E E E Y D E E E E--
Tb_Spt5       4  -----L G V K E L A-----D L L D H E P M-----D L V V P S D P S E V G S Y D E V Y K K E Q Q E R E E S R K H R
Sc_Spt5       110 S E A N E I V K K E E G S D E R K R P R E E D T N S D C-----D T K D E G D N K E D D D E D D D--D D D D E D D--
Pf_Spt5       1  -----
  
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Pt_Spt5m      1  -----M K K T K C Q F I E Q E A S E Y S S E E D D D P-----K F E I N R K E-----A E R Q-
Pt_Spt5v      42 -----K L L K K Q N Q K K T R-L N K Q M F I T E A S E T S D N E S D D N-S--V G E I T K G-
Tt_Spt5       35 -----G--K T L K Q N K K K K-Q K N Q F E F E I A E S N G E E E E V N Q--E K R L R N Q-
Ot_Spt5-g19   35 -----K D K R S S K K O K I D-A R M F V E V E A E E A S I L S D D L--D B E N E K I F R T Q--Q--
Ot_Spt5-g12   201 -----K L Q R L K N K Q K Q K K-A S E F E E E A C E G D D E E E D D--E K P N R K I K E S E F Y T E E Q L R V K T Q R L D K N F
At_SPT5-1     47 -----S D G R G S K K K S S-G S A F I D W E V E V D D D V E D D D D V D V E D G K Q L K F-G-----D F S L C F
At_SPT5-2     75 -----G K G A A S K R K K P S-A S I F I D R E A H Q V D D E E E E-----D-----E A E D D F
At_KTF1       29 -----R D E G L R I K K K N P E-V L Q F F E S A E V Y Y G G S S D E D--D D G L-----G F
Dm_Spt5       99 -----D D H P R K K K K K E F-F G G F I I D E A E V D D V D E D D E W--E E G A N E I G I V G--N E
Hs_Spt5       60 -----E D D D R P P K K P R-H G G F I L D E A V D D Y E D E D Q W--E D G A E D I L E K E--E I E A S N
Tb_Spt5       55 R K R S R H G A K S R I K H S G G K S D G V N P E V R S K F V I A A E S G E T-E S D D G-----E-
Sc_Spt5       165 -----D E A P T K R R R-Q E R N R F L D E A E V S D D E D E D E--E--D S E L V R E--G F
Pf_Spt5       1  -----
  
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Pt_Spt5m      39 -----L K E-----I Q S R R A Q A K-E-----R-----L N K R L Q E-----L M E Q D--
Pt_Spt5v      85 -----K Q E M Y N E L A-----L R K K H R-D
Tt_Spt5       77 -----G D A N L Y-----R Q R K A R-D
Ot_Spt5-g19   80 -----H E S Q Y Y T N D Q-----L R R Q Q G-I
Ot_Spt5-g12   265 -----L D N M M K R Y D N V K S Q G D E G E E D Y D E D E A G Y Y S E Q D E E E D Q R H R R K E E K K D Q P L Y N R D R--D F I Q K Y Q E A Y E K H A L K T H
At_SPT5-1     99 I V S G E A D-----L P N E D S D H R R Q Y Y Q R G F-----H P H-----E-
At_SPT5-2     114 I V D N G T D-----L P D E R G D R R-----Y E R R F-----L P R D E N D--E-
At_KTF1       70 -----L N D M E D-----E P E V E S S K A G K-----G E-----K G K S-S F V-
Dm_Spt5       145 -----I D E L G P-----T A R D I E I R R R G T--N-----L W D T Q K E--D-
Hs_Spt5       110 -----I D N V V-----L D E D R S G A R R L Q--N-----L W R D Q R E--E-
Tb_Spt5       102 V V G S D E D-----S E S D V Y G G P M A-----M E E G G K R H I F-
Sc_Spt5       208 I T H G D D-----E D-D E A S--A P G A R R D D R-----L R Q L D Q-
Pf_Spt5       1  -----
  
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**Acidic region**

Pt Spt5m 65 -----AID-----ETQN-----NSEYSSNVIDQ-----  
 Pt Spt5v 103 -----VVKQLEFRYQDGDQDDIQIGDD-----  
 Tt Spt5 90 -----MSEVIHLEQRGDQTPPMDD-----  
 Ot Spt5-g19 98 -----NMNML-----QSMEE-----RYRDTVQDDNEFEDEDDQOGLIGE-----  
 Ot Spt5-g12 343 GREGLTGQSQLQNSRKARESSDPIKLIANKQKLRDVKVHQKQVKKEVKDENDADTDDQVKMEVDE-----EQ-----  
 At SPT5-1 128 -----DVD-----ELKLR-TLERLSTKY-----AKD-----  
 At SPT5-2 143 -----DVE-----DLERR-QERFSSRH-----HE-----  
 At KTF1 97 -----FPKEEDLNEEFDRIMEFRYKPGSGFLR-----  
 Dm Spt5 172 -----EIE-----EYLRKKYADESIAR-----HFG-----  
 Hs Spt5 136 -----ELG-----EYMKKYAKSSVGET-----VYG-----  
 Tb Spt5 130 -----LEGEEDMTAEVVARATEFRYRSGRKKANRTEA-----  
 Sc Spt5 236 -----D-----LNKTSERLAQRALAKELRURY-----GRSSSK-----  
 Pf Spt5 1 -----

Pt Spt5m 83 -----DDEDQINDDDDTLKGKPKNDPKWRFKCGRSILEVQRLEISNLP-----KNSHIVSVFVTPNVKGF-----  
 Pt Spt5v 124 VDKS-----EDEIEKPGI-----RDPKFWRVSKKCKGKEQEAATSMEKHNH-----LDITNPEEIVSVFALKFBA-----  
 Tt Spt5 111 LDFDDID-MEHMPENIRKYGHOQRDQNTIEVSPVCKTKREKVVICLNKLAQ-----LNPEQNYGIYSISYVPSVESL-----  
 Ot Spt5-g19 132 -----YEGDDMIDKELPSVNDPFLWQVVRGGERQATLQMLNKSLEDF-----ARFGKHSLISVCTCKVEGF-----  
 Ot Spt5-g12 413 -----ARHSLTQQEVASHGQLPSIMDSKLWKNICWGMVQLVQQLRKAEDF-----LNHEKPFMIESFNCDKTQCC-----  
 At SPT5-1 148 -----DYELLDVNDVDDQALLPSVDPKLVKCAIGREREMAVCLMOKIYD-----FGSEPKIRSAIALDHQAVY-----  
 At SPT5-2 162 -----EYDEEATEVEQQALLPSVDPKLVWVKCAIGREREMAVCLMOKIFD-----FGADLQIRSVVALDHLKNF-----  
 At KTF1 125 -----YADDIKDAEMDALPPTSPDPKWKVKCAIGRERRSVFCMLHKEVEL-----RKIGTKLEIISVFSVDHVKGF-----  
 Dm Spt5 193 -----DGGHEMSDEITQOTLLPGLDPNLMWVKCAIGEEATATLMLRKYITY-----LNTDDPQIKSILAPEVGVKGY-----  
 Hs Spt5 157 -----GSEPLSDDITQQQLLPGVDPNLMWVKCAIGEEATATSLMRKELAY-----QPTDDPQIKSIVVAPPHVKGY-----  
 Tb Spt5 162 LLSSGIPKGLSSLRVASHLLPQDTPDKLVAVKCAIPRMAVVARLVNKKCYAFRIGRNYEKVLDIGKITSVFCFDHVKEY-----  
 Sc Spt5 263 -----QYRAAQDGYVPRFLLPSVDTATWGVVCPGKEELIRKLLKKKFNLD-----RAMGKKKIKLISFORDNYTCR-----  
 Pf Spt5 1 -----MAGKLEAVRVTHGQPEETAKLQYSKVRT-----YNEPIYALAPSRVKGY-----

Pt Spt5m 148 IEFESSEFENDVKEFMRSIM--YG---QPIYIQPECDSSLLEKK-N---NNQVQGVVRFKHNKNY---GKDLGKVRVN-----  
 Pt Spt5v 186 IITIANFEQHVMRATEGTTIVRQ---QPPELVESEQCPNIFKPA---EVEQDPEEGQWVRVQHNLY---SGDLARVQVD-----  
 Tt Spt5 183 IYIEGVNKRAVINFIDKYPDCNC---NKIELVNIIEFQAFEDQKQESYNAEVGOVVRSEKARNSDYINDIGOHFIFK-----  
 Ot Spt5-g19 198 IYIEAHKFIHVKEAIVGLSVILCGK---CLLQKEEMPGLYQNDK-QT---NLQTHQWVRPQGLY---GGDLGLVEA---  
 Ot Spt5-g12 482 IYIEAMNMSHVKSIHQMTGIYK---QKIDMIPYKEMTQLLKVCS-EINETTLQAHQWVRVKNK-PY---AGDLGFVEMIE-----  
 At SPT5-1 214 IYIEADMEAHVKEAIGVRNIYA-NQKILVPIKEMTAVLSVES-KA---DLSRDSWVRVRLG-LY---KGDLAQVVDV-----  
 At SPT5-2 227 IYIEADKEAHVKEAIGVRNIYA-NQKILVPIEMTDVLSVES-KA---DLSRDTWVRVRLG-LY---KGDLAQVVDV-----  
 At KTF1 194 IYIEADKEHVDVLEACKSLVGLIYA---TRMVLPKAETPNLLIVQK-KT---KKVSEGTWVRVKNK-KY---KGDLAQVAVS-----  
 Dm Spt5 262 IYIEAMKQTHVKTCDNNGNRMGKWKQEVPIKEMTDVLSVES-KA---GLKVKQWVRVRLG-LY---KDDIAQVDYVD-----  
 Hs Spt5 225 IYIEAMKQTHVKTATEGVNRLGYNVQVPIKEMTDVLSVES-KA---ANLKKPKSWVRVRLG-LY---KDDIAQVDYVD-----  
 Tb Spt5 242 IYIEASHKAFVENAINGLVGLER---SNISLVNPSSELMQMEHRP-SE---DKRVGSEVRLRR-QY---KDDIAQVAVD-----  
 Sc Spt5 335 IYIEAPKQSVLEKFCNGVPIIYI---SQKLLIPYQELPLLKPNK-SD-DALAEEGSEVRLKRG-LY---KGDLAQVDQIS-----  
 Pf Spt5 46 IYIEAPKQSVLEAIGRIRHARG---VLPGEVPEKLEHFEKPK-AV---SGLEPGDLVEVITAG-PA---KQOKAKVVKLD-----

Pt Spt5m 216 LIONFAIKVIKRNKQ-----GQKEPIS-----WORTR-----  
 Pt Spt5v 259 QEERKLKIKVPRQKLLMKSLEEENKKNKSKNNANADQSRFQMKEEGEPPIEDNEPFFORGSKFKFLKRAKYLQEQR-----  
 Tt Spt5 261 KN-NRVVVKLIPRIE---DMRKKYEQVKNNSKKR---NQDSFVQVGDGAE---DDQ-ANKKGGKFPYN-----  
 Ot Spt5-g19 267 TVDDVYLRLLIPRIE---DLSKQD-----LNGKGRDD-----KAKN-----  
 Ot Spt5-g12 555 G-GDRALVVKLIPRIE---RVVTQNELGONLELY-----HK---A-----WOKKE-----  
 At SPT5-1 286 NVKRVTVKLIIPRIE---DLQALAN-----KLEGTE-----NVKR-----  
 At SPT5-2 299 NVQRVTVKLIIPRIE---DLQALAS-----KLDGEE-----VSKK-----  
 At KTF1 265 DTNNALVVKLIIPRIE---DIQALTO-----KYGGGV-----  
 Dm Spt5 335 LAQNOVHVKLIIPRIE---DYTRVRG-----ALRTTATES-----DDSKR-----  
 Hs Spt5 298 PSQNTISLKNIPRIE---DYDRIKA-----RMSLKD-----WFAKR-----  
 Tb Spt5 313 SASREVTVKVPREE---DLYGKT-----  
 Sc Spt5 407 ENNLEVMKIVPREE---DYGKFD-----EIDPTT-----QQRKS-----  
 Pf Spt5 117 ESDEVVQETDAVPIPVTIKGDYVRLIS-----KIQKEE-----

Pt Spt5m 244 -----QQTNQIETTNOSEFIYSDIEKEY-----KCAKIDGFNLLRCPLKNEH-----  
 Pt Spt5v 339 KYIRGPKPIPIQMTKKSINDNDNEPESQFFY-TTLEDEWTSAKK-----DGEEIILPVHQLVIG-----  
 Tt Spt5 319 KFGNQPVVPTRTVDEIL---KTRPEQOFSLDEYEKDWSECYKAV---SEE-YRPNESTNLEFQHCQVYKIFNCKDLIFN-----  
 Ot Spt5-g19 300 -----FSNQRPBORIFNKSLPQYS---LEHRQIPALGNKTFVYKQYLL-KNGFLYKFPSPKQIQID-----  
 Ot Spt5-g12 591 -----FGGVAGQNVFORLFPQNVNECT--K-DRFEPELO-KNFFIWKEQMF-RNGFLYFEEKINKLID-----  
 At SPT5-1 317 -----KAF-APPFRFMNIDEARELHIRVEHRRDPMTG-DYFNIGGMF-KDGFHYKQVSTKSTAAQ-----  
 At SPT5-2 330 -----KAF-VPPFRFMNIDEARELHIRVEHRRDPMTG-DYFNIGGMF-KDGFHYKQVSTKSTAAQ-----  
 At KTF1 292 -----TVQKQTPAERLISSEIEFRPLIQVRRDRDTG-ITFEHLDSLML-KDGFHYKQVSTKSTAAQ-----  
 Dm Spt5 370 -----KKKRRPAAKPEDEAARATG---GEVHSDG-DFLLEFCNRYR-KDGFHYKQVSTKSTAAQ-----  
 Hs Spt5 330 -----KKFKRPPQRLFDAEKRSIG---GDVASDG-DFLIFEGNRYR-RKGFHYKQVSTKSTAAQ-----  
 Tb Spt5 333 -----CNKPEVRRPQRLVFNLAAGAHN---R---GEMYAWGDLFDGEGMLHLSVLSQVSISS-----  
 Sc Spt5 438 -----RRPTFAHRAEPQLFNPTMALRVDQ---ANLYKRD-DRHFTYKNEDE-IDGFLYKQVSTKSTAAQ-----  
 Pf Spt5 -----

**KOW 2**

Pt Spt5m 287 --NITITDEELQMFDPDVRKIIVQ-----Q-AKREILRRVDVQFKEGQKVRITIGEDDINKGPKFKIMHIFD  
Pt Spt5v 397 --NIKPTVEELQYFYPDVQDYRLQLQK-----LHSSLKQVVEQKSQIQIGDYITLTHDQAK--SSRYKVSQQLQ  
Tt Spt5 393 --EQLKSEELKQYFPHLFLDILNQN-----G-RFLKMANNQTTFVVGDPVLDNTLQK----GKIISLQK  
Ot Spt5-g19 359 --NVPTPEEVOQFQTSYMKSSK-DDINQI--DEITGEELIRKTFMGGSSDINKGDKIRVVKGDIN--GLYGVQVVTIEG  
Ot Spt5-g12 651 --KYBPRLAEMKRFQKDQHTSTDYLSQDQEDWIMDDATVMKTIKNDGLQQLVGDREVEVINKNQG--GKKGITLIRIDN  
At SPT5-1 376 --NVPTPELELEREKRPNENGE--IDFV--DES-----TLFANRKKGHFMKGDAAVIVTKGDLK--NLKGWIEKRVDE  
At SPT5-2 389 --NVPTPELELEREKRPNENGE--GDFG--GLS-----TLFANRKKGHFMKGDAAVIVTKGDLK--NLKGWIEKRVDE  
At KTF1 354 --GVPTPELELEREKRPNENGE--IDFV--DES-----TLFANRKKGHFMKGDAAVIVTKGDLK--NLKGWIEKRVDE  
Dm Spt5 425 --GVPTPELELEREKRPNENGE--IDFV--DES-----TLFANRKKGHFMKGDAAVIVTKGDLK--NLKGWIEKRVDE  
Hs Spt5 385 --GVPTPELELEREKRPNENGE--IDFV--DES-----TLFANRKKGHFMKGDAAVIVTKGDLK--NLKGWIEKRVDE  
Tb Spt5 386 VKMEKPTVEELAAEFSSDLNRV-R-----EAASHFASNGRGSAGLRIGDMVRVIVSGQLR--ETVGTIENIFL  
Sc Spt5 497 --NIOPTVEELAREGSKGAVDLTSSVS--QS-----IKKAAAKVTEFQPGDRLEVLNCEQ--GSKGIVTETTK  
Pf Spt5 -----

**KOW 3**

Pt Spt5m 352 DQM-I--EELM----CKKNREYTYLVHASEIRLAKLYQEAQKVIDGPHKGVGVIIICIKQGVVIVSNQHGTFKVPHSV--  
Pt Spt5v 462 DENKLIISKTVKNNKNNKRNNEYKDISEAKLAFKLYQQVSVISGPNLGLSGTILKMDLTAQISFEA---GRIDALI  
Tt Spt5 452 ETARVLIKK--KK---LSIEEYPIKQIVKYFEDGSRVKVIVSGTSEGITGTIVISTKGDVCEVFDN---NNTIEVRT  
Ot Spt5-g19 432 SDV----LFK---PTIEGFEDDLRIPMDFVVKHFEPGQVRFVIDGKFKGTGIVVSSSETQFANVALTQ--NNEIRIFA  
Ot Spt5-g12 727 EEF----ILT---KTLDKPYELKQSEVIKIFECGSESVRVIAGTHSGESGITATAEKHAVLMDGATSENKILL  
At SPT5-1 439 ENV----LIR---SEMKDLPNPILAVNGRELCKYFEPGNEFVKVIVSGIHEGEGTGIIVKVDQHMLIILSDT--TKEHCKVL  
At SPT5-2 452 ENV----LIR---SEVKGLPDPILAVNERELCKYFEPGNEFVKVIVSGIHEGEGTGIIVKVDQHMLIILSDT--TKEHCKVL  
At KTF1 392 -----L---PT-----CR--EGCKGEGSGGCKGEGSGGCKG--EGSR-----G  
Dm Spt5 487 TMT----TVM---PKHQDLKDFLIFKASELRKYKTDHARVLAGRYEGTGLIIRVEPTRVIVVSDL--TNEIEVLP  
Hs Spt5 446 NKI----TLM---PKHDLKDMLEFFPAQLKFKMGDHFVKVIVGRFEGTGLIIRVEENFVILFSDL--TMEHCKVL  
Tb Spt5 450 DTNTVALSCR---VPRGETIKLRVETPLCVKHEFTEGTHVVIDGCVHAGESGTIVVVALGDVHVFSDRATATREVVRA  
Sc Spt5 560 DIA-----T---IKINGFTTPELFPISLIRKIFEPGQHVTVINGEHQGLAGLVIMVEQGVVTFMSTQ--TSREITITA  
Pf Spt5 -----

Pt Spt5m 424 ---QFGHKNFQ-----TDNSLVKFG-----NNDFOICGVIIQKQL--QSAVV  
Pt Spt5v 539 SDLQSQKNVIKKVDIENPDAGAGQKPGKRNLDLVKFG-----LIDNDIGGILNLSN--NEVSV  
Tt Spt5 521 KDLAITGEAS--QDAEKIVVNESSDRNLGLKDKLVKLT-----GF-NNVGLILDLSK--DYIKI  
Ot Spt5-g19 502 NSLKLKSEIDQCVLGTGY-IDKKN-FSKYSANLIMYN-----TKYVGVIIQKQL--QSAVV  
Ot Spt5-g12 799 SNLKSKEDEMEHV-KRDY-IQKSVIEIKYNAGEMIMYQSTHCSQLSTSSSSCVNASSTVSGSICGYIQNP--DYIKV  
At SPT5-1 509 DHVAKSAEVTKGVTKI-----GDYELHDLVLLS-----DFSFGVILKLS--EAFQI  
At SPT5-2 510 DHVVESSVTTGVTKI-----GDYELHDLVLLS-----DFSFGVILKLS--EAFQI  
At KTF1 423 ---GKG---EGS---SDF---KSESSYELYNLVCFSS-----RKDFGIIVGVDDKDGKGYKV  
Dm Spt5 557 RDLQLCSVATGVDCI-----GQHWGDLVQLD-----SQNVGVIVRLER--BNFHV  
Hs Spt5 516 RDLQLCSVATGVDCI-----GQHWGDLVQLD-----PQTVGVIVRLER--TFQV  
Tb Spt5 526 DDCHRSNLVGSF-----GHTSGSILKLFDLVMLPD-----SSSVGVVRLER--NDVGV  
Sc Spt5 628 NNLSKSID-----TTATSSVALHDLVELS-----AKNVACLHQAGH--DIFKV  
Pf Spt5 -----

**KOW 4**

Pt Spt5m 460 LDINNVEQVIRNEKEE---ILINGVELNEQGETFRINDNVIIH---SGQY-ANKFGLVKKHCINGKLLFNHNF--  
Pt Spt5v 597 LDLTNQIKNINKLATER---NSINIRNNVKNMYGNDTRQDLVVIIH---LGFY-KNNKATVLLHYEDYLFENG-KFDN  
Tt Spt5 576 LDQNGKIKNVSSFSIN--TKIDIRKYIKNAEGNNTNNSNIIIK---QGQY-SGYQCOVILHYKSLFLENP-KFRD  
Ot Spt5-g19 556 INNEDIQNLKLSDIN--KKIDFRKRTTIDSHRNTLYADNVVRIH---NGKY-KGKQGVVKYIITKTLFLWDK-EFYQ  
Ot Spt5-g12 875 INSHNQVMVQOASIT--KKIFPRRTFAIDRNHTIASSEVRIY---DGSQNLGKCEVKGFKQDVLVYLLKGLVQ  
At SPT5-1 554 LKGVDPSSVSVKAS-EIKYKIWKINVDRYKVVAVDVRVIV---EGPS-KGKQGPVQYKGVLFTHDR--HLE  
At SPT5-2 555 LKGVDPDRPEVALVKLR-EIKCKLEKINVDRYKVVAVDVRVIV---EGPS-KGKQGPVQYKGVLFTHDR--HLE  
At KTF1 466 LKEGIDGPVVVTVGKREMONGPFDKFTAILLNKQOISNDVVRIS---KGPS-EGKQGPVQYKGVLFTHDR--HLE  
Dm Spt5 602 LGMNKCKIECKPTALH---KRKENRHTVAIDADQNOIRRDVVKVM---EGPH-AGRSGETKHLIYRSIAFLHCR-MYTE  
Hs Spt5 561 LNMVGVVTVRHQAVT---KKKDNFAVAIDSEQNNIHVDLVKVI---LQPH-SGREGETRHLFRSFAFLHCK-KLVE  
Tb Spt5 572 LTDRMTRYSTTOIK---PVLGFRQTTDQLANMIRGSEVITQNDSSPYHLDGQTRVEQVENVITLFRVK-TVKE  
Sc Spt5 670 IDETCKVSTITKGSILSK-INTARAVSSVDANGNEIKGTTIVEK---VG---SRREGQVLYIQQQEIVVSK-KIVE  
Pf Spt5 -----

Pt Spt5m 527 PYQIIEENANNCKLVFSKQN---KP-----DEGANT-----ISQMPN-----KQN  
Pt Spt5v 668 TQGVILEKANNCGLVSSSKP-----DEGANT-----ISQMPN-----E  
Tt Spt5 647 TYTVE--NINNVSLQTTNPVIQR--GEA---KIAYDMKYGK-----QDNLKP-----D  
Ot Spt5-g19 628 SNGIFVENARNVVLGGDELM-KQNGCA-----VANSNRR  
Ot Spt5-g12 949 TNGIYSILTRNVLAGQDFV-KNVQENHKGFIL-----GQDRKQ  
At SPT5-1 627 HTGFICTRCSSCLAGG---NFRTP-----ALVPPSPRRFQRADMGYNPAGGRHQGGRGR  
At SPT5-2 628 HAFICAKCTSCIVVGGSSS-GANRNCGDLSRYGNFKAP-----APVPPSPGRFQRGGGYNNSGRRHGGGR-R  
At KTF1 540 NGGYFCCKSQSCEKVKLFTEESNEKTCGFDGTAFFDFVSSPK-----SPVHPSPEKEWQPRERYNS--SNQGDI  
Dm Spt5 673 NGGFVCKTRHQLAGGST-TVSLACIVVGGLGFMSPRIQ-----SPVHPSGGG---GARGGARGGRGFV-T  
Hs Spt5 632 NGGFVCKTRHQLAGGST-P-RDVTNFTVGGFAPMSPRIS-----SPVHPSAGG-QRGGFGSPGGGGGMSRGR-R  
Tb Spt5 647 NSGIVAVDASCVILGGRTTKQIPAKQLPTVNRNPHNASRADLSVNPFRMTSEDW---A  
Sc Spt5 741 NAGIVVNPNSVAVASKDN---MLSNKMDLSKMNBEIISK-----MGFPSSKTFQ---PIQSRG  
Pf Spt5 -----

**KOW 5** \*

Pt Spt5m 552 DQMCITGQICQLKIKGOMQGYRGGQIVQIKSGYLIVQISANNTKVKVSEKDLLI-----  
Pt Spt5v 702 DWKNLRGQMMVIRKQOMQSYRGMVQEVTSRVATIQLSAKNIVVKVPL-ECIKSESSNSH-----LQVGNTPQHHPGM  
Tt Spt5 688 DMKQYVGRQMRIIGGKYKGFQCTVTDIRNDQIKVEINSKFTVVFIPKCDIATDKGQTE-----LEYGTPSYNAQS  
Ot Spt5-g19 662 FRDVAWAKENIEITKGEWKGYRGRVCRADLRQAIVELSSKCRKIPLEISLKEVDVVGKSGTTRGD---MTYGGQT---VY  
Ot Spt5-g12 989 KDRKILRSYVATTAGEYKGLKGRVIFADELICKVEILAKDKVQLPRGMVME--IRDPTKPMEI---RDLGVVEP---MS  
At SPT5-1 681 GDDHLVGTIVYKIRLGPFKGYSGRVLEVKLKLVRVELEAKIVTGKHFERKAISDM-----TDN-----  
At SPT5-2 698 GDDSLLGTTVYKIRLGPFKGYSGRVVEVKGNVVELEMKIVTVD---RGAISDNVA-----TTE-----  
At KTF1 605 GSTYSIGQKLRIRVGPLKGYLCRVIALRYSDVTVKLDLSQHKIFTVKSEHTAEVVRDRN--TVLST-----SGDAG  
Dm Spt5 737 RDRELLGKTKIKISGGPYKCAVGIYKDATESTARVELHTISCOTISVDRNHIAIVGVGTGKEG--SV---STYGTTPARTPGY  
Hs Spt5 701 RDNELLGQTVRISQGPYKGYIGVVKDATESTARVELHSTCQTSVDRQRITTVGSRPPGG--MT---STYGTTP---MY  
Tb Spt5 705 GNSEWYEMDTE-----  
Sc Spt5 796 GREVALGKTVRIRSEGYKQQLGIVKDVNGDKATVELHSHKXKHITDKHKITYYNREGGEGITYDELVNRRCNVEP  
Pf Spt5 -----

\* \*

Pt Spt5m -----  
Pt Spt5v 773 S--TR-----VWDDIEGVQQSAMRGGYQSPFIYQTPMEND-----  
Tt Spt5 760 IYQ-----PNNHENINSPSY-YAN---N-----  
Ot Spt5-g19 735 E---AGKTPMQYNTESYYPHSIHWGA-NQSPGYGTDYDYNNGMSIGFSRAGSEHYENKPEQTPRNQNVV--KHE--  
Ot Spt5-g12 1060 FDD-AQKRDMLQEDD-----EMMIDTTAR-QQNMMLQQNFANQLKDYDCFGGEAQIATP  
At SPT5-1 739 VVAIPQYN-MGSQTPMHPSTRPLHPC-MTPMRHSGATPIIDGMRTPMRGR--AWNBYMMSPPRDNWE  
At SPT5-2 754 FRDTSRYS-MGSETPMHPSTRPLHPY-MTPMRDSGATPIIDGMRTPMRDR--AWNBYTMSPPRDNWE  
At KTF1 672 ---TGSFQPFQM---LGI-----ESSTGDWALGAGTSSEGGNWNIGGPTSDSHSLNIERN---MVQL  
Dm Spt5 812 GAQTPSYTAAGSKTPLVGSQIPENWDT-DTRTPYGTMTPSIDGSMTPR-HG---AWDETANTTPANN-D--FDYSLE-EP  
Hs Spt5 772 GSOIPMYG-SGSRTPMYGSOIPLQDG-SRTPHYGSQIPLIDGSRTPAQSG---AWDPNNNTPSIAEEE--YEYAFDDEP  
Tb Spt5 -----  
Sc Spt5 870 -----QAR-MG---PSYVSAAPRNMATGGIAAGAAATSSGLSGGTPGWSSFDGC-----KTPAVNAHG-----GSG  
Pf Spt5 -----

Pt Spt5m -----  
Pt Spt5v -----  
Tt Spt5 -----  
Ot Spt5-g19 -----  
Ot Spt5-g12 1112 RGSDDKDWGDDQNLGGLHREGRDQISQNF-----DCDSQFDLDAAD-----  
At SPT5-1 803 DGNPGSWGTSPTSP-----YEAATPGSDIGSSTPGRSSYRDAGTPINNG  
At SPT5-2 818 DGNPGSWGTSPTSP-----QYQPGSPPSR-----AYEAPTPGSGVASTPG--GSYSDAGTPRDHG  
At KTF1 726 CREKNPWGGSKPTSD-----VSPTVADD--NTSAWANAANKPASASDQPGGNPWGKTP--ASEAGTVSGWG  
Dm Spt5 883 SPSPG-----YNPSTPGY-----QMTSQFAPQTPGTL--YGSDRSYSP-----NPSPPSP-----  
Hs Spt5 845 TPSPQAYGGTPNPQTPGYDPDSS-PQVNPQYNPQTPGTPAMYNTD-QFSPYAAPSPQGSVQPSPPSQ-SYHQV-APSPAG  
Tb Spt5 -----  
Sc Spt5 927 GGGVSSWGGAS-----TWG  
Pf Spt5 -----

Pt Spt5m -----  
Pt Spt5v -----  
Tt Spt5 -----  
Ot Spt5-g19 -----  
Ot Spt5-g12 -----  
At SPT5-1 844 FVYYLL---CLNANA---PSPMTPSSTSYLPTIPGGQA-----MTPGTDLDVMSLDIVNIF---QFTDYVSL-----  
At SPT5-2 868 SAYANAPSPYLPSTP---GQPMTSSASYLPGIPGGQP-----MTPGTGLDVMSFVIGGDAEAWFMPDILVDIHKAGE  
At KTF1 791 --DT-----SASNVEASSWEKQ--GA-----STSNVADLGSWGTHGGSSGGNKQD-  
Dm Spt5 926 -----APSPYPVGYM--NTPSPSTYSPNIPGGIPQSPYNPQTPCASLD-----SSMGDWCTTDIEVRIHT-HD  
Hs Spt5 921 YQNTHPSPASYHPTPSPMAYQASPSPPVGYSPMIPGAPSPGGYNPHTPESGIE-----QNSSDWVTTDIQVKVRDYL  
Tb Spt5 -----  
Sc Spt5 941 -----  
Pf Spt5 -----

Pt Spt5m -----  
Pt Spt5v -----  
Tt Spt5 -----  
Ot Spt5-g19 -----  
Ot Spt5-g12 -----  
At SPT5-1 902 -----FFLCGHHQDGSVVALGHRGEGE-----TIRATQNKVSLVCPKNE-----RVKILGGK  
At SPT5-2 938 DTD---VGVIDVSDGTCKVSLGSSGEGD---TIMALPSELEIIPPRKSD---RVKIVGGQ  
At KTF1 832 -----EDSVWGKLEBASESQKKESSWGKGGSDGESSWGNKDGNSASKKDGVSWGQDKGSDESCKGSAWSN  
Dm Spt5 986 DTDLVGQTGIIRTVSNGVCSVFLRQEDRSV-----SIVSEHLAPVLPNGD-----EFKIIYGD  
Hs Spt5 994 DTQVVGQTGVIRSVTPGGMCSVYLKDKSEKVV-----SISSEHLEPIPTPKNN-----KVKVILGE  
Tb Spt5 -----  
Sc Spt5 941 -----GQGNGGA-----  
Pf Spt5 -----

Pt\_Spt5m -----  
 Pt\_Spt5v -----  
 Tt\_Spt5 -----  
 Ot\_Spt5-g19 -----  
 Ot\_Spt5-g12 -----  
 At\_SPT5-1 951 YCGSTAKVIGEDGQDGIVLDESLDIKILKTTILAKLVHE-----  
 At\_SPT5-2 990 YRGSTGKCLIGIDGSDGIVLIDNLDVKILDLALLAKFVQP-----  
 At\_KTF1 902 QCGDFGSGKKKDGSSGWNKSAEDSNANSKGV---PDWGQPNDGSSWGKKGDGAASWGKKDDGG--SWG-----KKD---  
 Dm\_Spt5 1040 DRESVGRVLSKDGDFVFCR--INEEIKLLPINFLLCKMKSID-----  
 Hs\_Spt5 1048 DREATGVLLSIDGEDGIVLMDLDEQLKILNTRFLGKLEA-----  
 Tb\_Spt5 -----  
 Sc\_Spt5 948 -----SAWGGAGGGASAWGGQGTGATSTWGGASAWGNKSSWG-----  
 Pf\_Spt5 -----

Pt\_Spt5m -----  
 Pt\_Spt5v -----  
 Tt\_Spt5 -----  
 Ot\_Spt5-g19 -----  
 Ot\_Spt5-g12 -----  
 At\_SPT5-1 -----  
 At\_SPT5-2 -----  
 At\_KTF1 968 -----DGNKDDGGSSWGKKDDGQKDDGGSSWEKKFDGGSSWGKKDDGGSSWGKKDDGGSLWGKKDDGGSSWGKEDDGG-----  
 Dm\_Spt5 -----  
 Hs\_Spt5 -----  
 Tb\_Spt5 -----  
 Sc\_Spt5 985 GASTWASGGESNGAMSTWGGTGDR-----SAYGGASTWGGNNNN-----  
 Pf\_Spt5 -----

Pt\_Spt5m -----  
 Pt\_Spt5v -----  
 Tt\_Spt5 -----  
 Ot\_Spt5-g19 -----  
 Ot\_Spt5-g12 -----  
 At\_SPT5-1 -----  
 At\_SPT5-2 -----  
 At\_KTF1 1041 SLWGKKDDGESSWGKKDDGESSWGKKDDGGSSWGKKDEGGYSEQTFDRGGRGFGGRRGGRRGRDQFGRGSSFGNSEDP-----  
 Dm\_Spt5 -----  
 Hs\_Spt5 -----  
 Tb\_Spt5 -----  
 Sc\_Spt5 1024 -----  
 Pf\_Spt5 -----

Pt\_Spt5m -----  
 Pt\_Spt5v -----  
 Tt\_Spt5 -----  
 Ot\_Spt5-g19 -----  
 Ot\_Spt5-g12 -----  
 At\_SPT5-1 -----  
 At\_SPT5-2 -----  
 At\_KTF1 1121 APWSKPSGGSSWGKQDGDGGSSWGKENDAGGGSSWGKQDNGVGSWGKQNDG--SGGGSSWGKQNDAGGGSSWGKQDSG-----  
 Dm\_Spt5 -----  
 Hs\_Spt5 -----  
 Tb\_Spt5 -----  
 Sc\_Spt5 1024 -----KSTRDGGASAWGNQDDGNRSAWNNQGNKSNYGGNSTWGGH-----  
 Pf\_Spt5 -----

Pt\_Spt5m -----  
 Pt\_Spt5v -----  
 Tt\_Spt5 -----  
 Ot\_Spt5-g19 -----  
 Ot\_Spt5-g12 -----  
 At\_SPT5-1 -----  
 At\_SPT5-2 -----  
 At\_KTF1 1199 GDGSSWGKQDGGGDSGS AWGKQNNNTSGGSSWGKQSDAGGGSSWGKQDGGGGSSWGKQDGGGGSGSAWGKQNETSNGSSW-----  
 Dm\_Spt5 -----  
 Hs\_Spt5 -----  
 Tb\_Spt5 -----  
 Sc\_Spt5 -----  
 Pf\_Spt5 -----

Pt\_Spt5m -----  
 Pt\_Spt5v -----  
 Tt\_Spt5 -----  
 Ot\_Spt5-g19 -----  
 Ot\_Spt5-g12 -----  
 At\_SPT5-1 -----  
 At\_SPT5-2 -----  
 At\_KTF1 1279 GKQNDSGGGSSWGKQDGGGGSSWGKQNDGGGGSSWGKQDGGSSKPNNEHSGGGRGFGERGGGGFRGGRNQSGRGGRSF  
 Dm\_Spt5 -----  
 Hs\_Spt5 -----  
 Tb\_Spt5 -----  
 Sc\_Spt5 -----  
 Pf\_Spt5 -----

Pt\_Spt5m -----  
 Pt\_Spt5v -----  
 Tt\_Spt5 -----  
 Ot\_Spt5-g19 -----  
 Ot\_Spt5-g12 -----  
 At\_SPT5-1 -----  
 At\_SPT5-2 -----  
 At\_KTF1 1359 DGGRSSWKTNDQENTWKSQSGGSDWKKGWGEDSNNSKPSGSSAGGCAGNWPSWDTNSKKTNDKPGDDSKSAWGTSSND  
 Dm\_Spt5 -----  
 Hs\_Spt5 -----  
 Tb\_Spt5 -----  
 Sc\_Spt5 -----  
 Pf\_Spt5 -----

Pt\_Spt5m -----  
 Pt\_Spt5v -----  
 Tt\_Spt5 -----  
 Ot\_Spt5-g19 -----  
 Ot\_Spt5-g12 -----  
 At\_SPT5-1 -----  
 At\_SPT5-2 -----  
 At\_KTF1 1439 QVNTDNNNDSWNKKPNNDVGTSGEADNAWGGKTNAVAPSPSGSAAWGTGDKKKTGW  
 Dm\_Spt5 -----  
 Hs\_Spt5 -----  
 Tb\_Spt5 -----  
 Sc\_Spt5 -----  
 Pf\_Spt5 -----

**B**

P.tetra_Spt5m	1	-----MRKRTKCFIEQEASEGYSSEE-DDDPKF
P.biaur_Spt5m	1	-----MRKRTKCFIEQEASEGYSSEE-DDDPKF
P.sexau_Spt5m	1	-----MRKRTKCFIEQEASEGYSSEEVD-DDDPKF
P.caudat_Spt5m	1	-----MRKKMRNMFIEEQASEGYSSQEEYEEQIQ
P.tetra_Spt5v	1	MSQKNDDYRSDDQDKSQSSIDSDIEDIPDSSESEDFKQKSKSKLLKKQKNQKKTFLNKQKFDTEASETSDNESLIDN----
P.biaur_Spt5v	1	MSAKNDDYRSDDQDKSQSSIDSDIEDIPDSSESEDFKFKTKNKKLLKKQKNQKKTFLNKQKFDTEASETE-NESEID----
P.sexau_Spt5v	1	MSAKNDDYRSDDQDKSQSSIDSDMGDIQDSSESMDFKQTKNKKLLKKQKNQKKTFLNKQKFDTEASETSENSEDENRFD
P.caudat_Spt5v	1	MSANNDDYRSDDDM-SQSSITSFEDDEE--FSSQDFVATKNKKDLKKQKNLKKKLTQKQFIEEASET-----

<b>Acidic region</b>		
P.tetra_Spt5m	29	-----EINRKEAERQLKEIQSRAQAKERENKRLQELMEQDAIDETQNNSEYSSNVIDQDDDEQDNDDDDDTLKGGPKLN
P.biaur_Spt5m	30	-----EINRKEAERQLKEIQSRAKAKERENKRLQELMEQDAIDETENNSNTSRNENQDNDQDDDDDDDESLKGGPKLN
P.sexau_Spt5m	29	-----EINRKEAERQLKEIQSRAKAKERENKRLQELMEQDAIDETANSSDKSKNENNERDYDQDDDDDTSTKGGPKLN
P.caudat_Spt5m	29	-----KVEDKKLDKHYQEMLVRQKKEKERIKQFEELIRNEVIGNTSSSSSHE-----QES-----YDNDPTEGPRLE
P.tetra_Spt5v	77	--SVGEITKGGKQEQMYNELALARKHHRDVV-----KQLEERY-----CDGDQDIQIGDDVKSSEDELEKPGLR
P.biaur_Spt5v	76	--VGGEITKGGKQEQIYKEIDLARKHTRNVV-----EITQORY-----CDGDININ--VEEQRSDDDELEKPGLR
P.sexau_Spt5v	81	EVVKTEITKKNQEQEYKAMDLRKYNKNIV-----EDLENRY-----CNGEGLN--VEEVKTDDELEKPGLR
P.caudat_Spt5v	66	---VGEISKKRDEIYDTNLLRRRQNNVV-----TEENRY-----KDGDLVV--DEAQTDDDDQVEPPGWR

P.tetra_Spt5m	104	DPKVVRFKCGRSILEVQ--R-LEISNLLP--KNSPIVSVF TPNVKGFIFFBSFEFEDVKEFVRSI---MYGQPIYIQP
P.biaur_Spt5m	105	DPKVVRFKCGRSICEVQ--R-LEISNLLP--KNSPIVSVF TPNVKGYIFFBSYFEKDVKELKMSI---MYGQPIYIQP
P.sexau_Spt5m	104	DPKIWRFKCGRSIFEVQ--R-LLDLSNQLP--KNSPIVSVF TPNVKGHIFFBSYFEKDVKEFVRSI---MYGQPIYIQP
P.caudat_Spt5m	94	DPKVVRFKCGRSKVVVE--L-LQQIVKLP--ENCKIVS FENPNLKLIFFEAYFEQDVKTIKLDI---MYGQPTYIEP
P.tetra_Spt5v	138	DPKFWRVSCNKGKEQEAFTSI FKHNHLLDT-NPLEIVSVFAKKFPAIFIEANFEQHVMRAGEGITVIRQCPELVBVS
P.biaur_Spt5v	135	DPKFWRVSCNKGKEQEAFTSIFKHNHLLDT-NPLEIVSVFAKKFPAIFIEANFEQHVMRAGEGITVIRQSPPELVBVS
P.sexau_Spt5v	142	DPKFWRVSCNKGKEQEAFTSIFKHNHLLDT-NPLEIVSVFAKKFPAIFVEAYFEQHVMAAGEGITVIRQVPELVBVS
P.caudat_Spt5v	125	DPKFWRVSCNKGKEHEAVTSIFRSHLLDTEFLEIVSVFTKKFPCVIFFEAYFETHVIKALEGAIIVRQTPPELVEP

P.tetra_Spt5m	176	EDCDSLLEIKK--NNNIQVGQVRFKHNKNGKDLGVLVRLNLIQNFAIKVLRKNGQGK-----
P.biaur_Spt5m	177	EDCDSLLEIKK--NNNIQVGQVRFKHNKNGKDLGVLRLNLMQNFALIKVLRKNGQGK-----
P.sexau_Spt5m	176	EDCYSLLEIKK--NNNIQVGQVRFKHNKNGKDLGVLVRLNLIQNFAIKVLRKNGQGK-----
P.caudat_Spt5m	166	EKCPSLLEVQR--SINIQLGQVVRNRHRTYSGDLRVMRNRITNITLKVLRKNGQGK-----
P.tetra_Spt5v	217	EQCPNLFKPAEVEQIDIEEGQVVRVROHNIYSGDLARVMQVDOERKLIKLVPRKLLMKSLEEEENKKNKSKNNAN
P.biaur_Spt5v	214	EQCPNLFKPAEVEQIDIEEGQVVRVROHNIYSGDLARVMQVDOERKLIKLVPRKLLMKSLEEEENKKNKSKNNNSN
P.sexau_Spt5v	221	EQCPNLFKPPVEVEQIDIEEGQVVRVROHNIYSGDLARVMSVDOERKLIKLVPRKLLMKSLEEEENKKNKSKNNAN
P.caudat_Spt5v	205	ETCLOLFKPAEVEQVDIEEGSVWRVROHNIYQGLARVMSVDOERKLIKLVPRKLLMKSLEEENKKNKSKK----N

<b>Linker 1</b>		
P.tetra_Spt5m	235	-----EP-----ISWQRTROQTNQIETTQSEFFIYSDIEKENK
P.biaur_Spt5m	236	-----EP-----ISWQRSROQTNQLETKNESEFFIYSDIEKENK
P.sexau_Spt5m	235	-----EP-----ISWQRTROQTSIIIKNDSFFIYSDIEKENK
P.caudat_Spt5m	225	-----EP-----ISWORENEGEINFSKEQNSEFFIYSDIEKENK
P.tetra_Spt5v	297	ADQSRFQMKKEEGEPPEDNEPFQSGSKFRLKRAKYLQEQRYIRGPKIPIQMTKKSINDNDDEPTSOFFYITIRDEMT
P.biaur_Spt5v	294	ADQSRFQMKKEEGEPPEDNEPFQSGSKFRLKRAKYLQEQRYIRGPKIPIQMTKKSINDNDDEPTSOFFYITIRDEMT
P.sexau_Spt5v	301	ADQSRFQMKKEEGEPPEDNEPFQSGSKFRLKRAKYLQEQRYIRGPKIPIQMTKKSINDNDDEPTSOFFYITIRDEMT
P.caudat_Spt5v	282	TDQIRFQMKKEEGEPLVE--ETVQKSGKFMRLKRAKYLQEQRYIRGPKIIVQIQKKQINDNDDEPTSOFFYSSIRDEMT

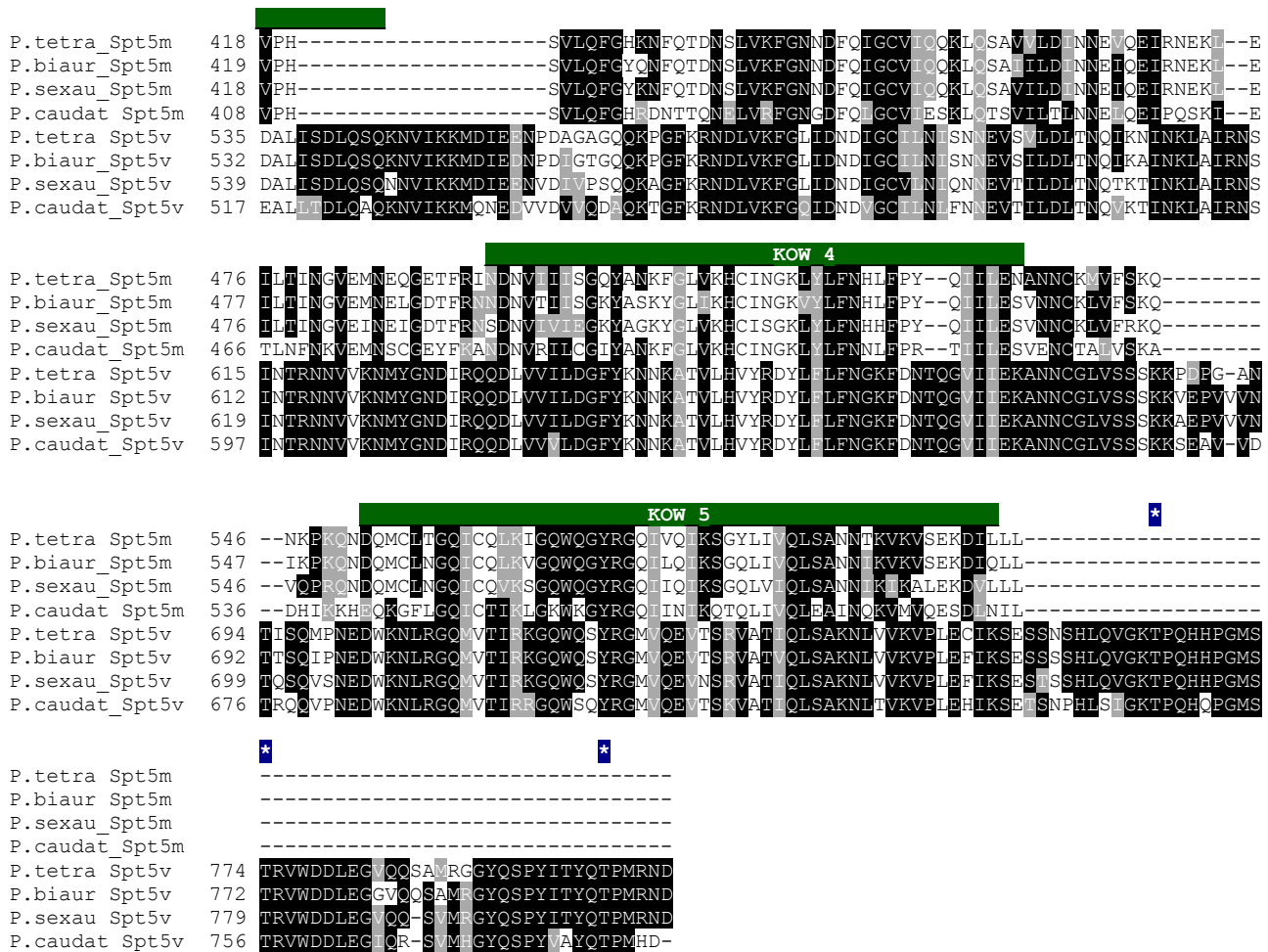
  

<b>KOW 1</b>		
P.tetra_Spt5m	268	CAKIDGFNLLKCPKNI-EHNTITITDEELOMFDPVDRKILVQAKREILRRVDVQ--FKEGQKVRLLIGEDDLNKGPKF
P.biaur_Spt5m	269	CSIIDGFNLLKCPKNI-EHNTITITDEELOMFDPVDRKILVQAKREILRRVDVQ--FKEGQKVRLLIGEDDLNKGPKF
P.sexau_Spt5m	268	CAKIDGFNLLKCPKNI-EHNTITITDEELOMFDPVDRKILVQAKREILRRVDVQ--FKEGQKVRLLIGEDDLNKGPKF
P.caudat_Spt5m	258	CAKIDGFALLKCNVKNIDHNITITDEELOMFDPVDRKILVQAKREILRRVDVQ--FKEGQKVRLLIGEDDMNSGPEQF
P.tetra_Spt5v	377	SAKKDGFELITLVPVHQLTGNIKPTVEILQYFDPVQYRVLIIQKLHSSLKQVVEQKSKIQIGDYITITN--QDQTKPSRY
P.biaur_Spt5v	374	SAKKDGFELITLVPVHQLTGNIKPTVEILQYFDPVQYRVLIIQKLHSSLKQVVEQKSKIQIGDYITITN--QDQTKPSRY
P.sexau_Spt5v	381	SAKKDGFELINLVPVHQLTGNIKPTVEILQYFDPVQYRVLIIQKLHSSLKQVVEQKSKIQIGDYITITN--QDQTKPSRY
P.caudat_Spt5v	360	SAKKDGFELITLVPVHQLKGNVKNITTEELOFDPVDSYRQILQKMYSQLKQVVDQKQKIQVGDYITITD--QDQYRSKAF

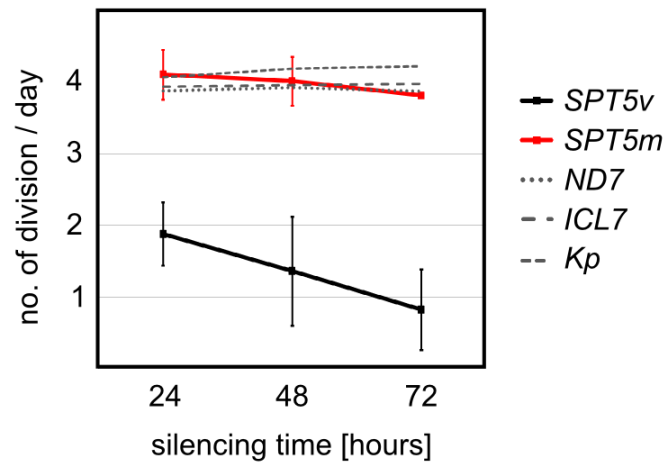
  

<b>KOW 2</b>			<b>KOW 3</b>		
P.tetra_Spt5m	345	KIMKIFDDQMIELM-----CKKDNREYTYLVHASETRLAFKLYQEAQKVIDGPHKGDVGVIIICIKQYVVLSNQHGTFK			
P.biaur_Spt5m	346	QIIKIFDDQMIELM-----CKKDNREYTYLVHASETRLAFKLYQEAQKVIDGPHKGDVGVIIICIKQYVVLSNQHGTFK			
P.sexau_Spt5m	345	KIIKIFDDQMIELM-----CKKDNREYTYLVHASETRLAFKLYQEAQKVIDGPHKGDVGVIIICIKQYVVLSNQHGTFK			
P.caudat_Spt5m	335	KIMKIFDDNMELM-----CKKDKREYTYLVHASETRLAFKLYQEAQKVIDGPHKGDVGVIIICIKQYVVISTYGSYK			
P.tetra_Spt5v	455	KVSQLQDENKLIIVTKTVKNKNNNEKRYEYKQDISAKLAFKLYQVSVISGNTGLSGTIIKDDDLTAQISTEAGRIV			
P.biaur_Spt5v	452	KVAQIILQDENKLIIVTKTVKNKNNNEKRYEYKQDISAKLAFKLYQVSVISGNTGLSGTIIKDDDLTAQISTEAGRIV			
P.sexau_Spt5v	459	KVAQIILQDENKLIIVTKTVKNKNNNEKRYEYKQDISAKLAFKLYQVSVISGNTGLSGTIIKDDDLTAQISTEAGRIV			
P.caudat_Spt5v	438	KVVQMLPDENKLIIVTKNVKDKS-GEVANYEYKQDISAKLAFKLYQVSVISGNTGLSGTIIKDDDKTAQISTEAGRIV			

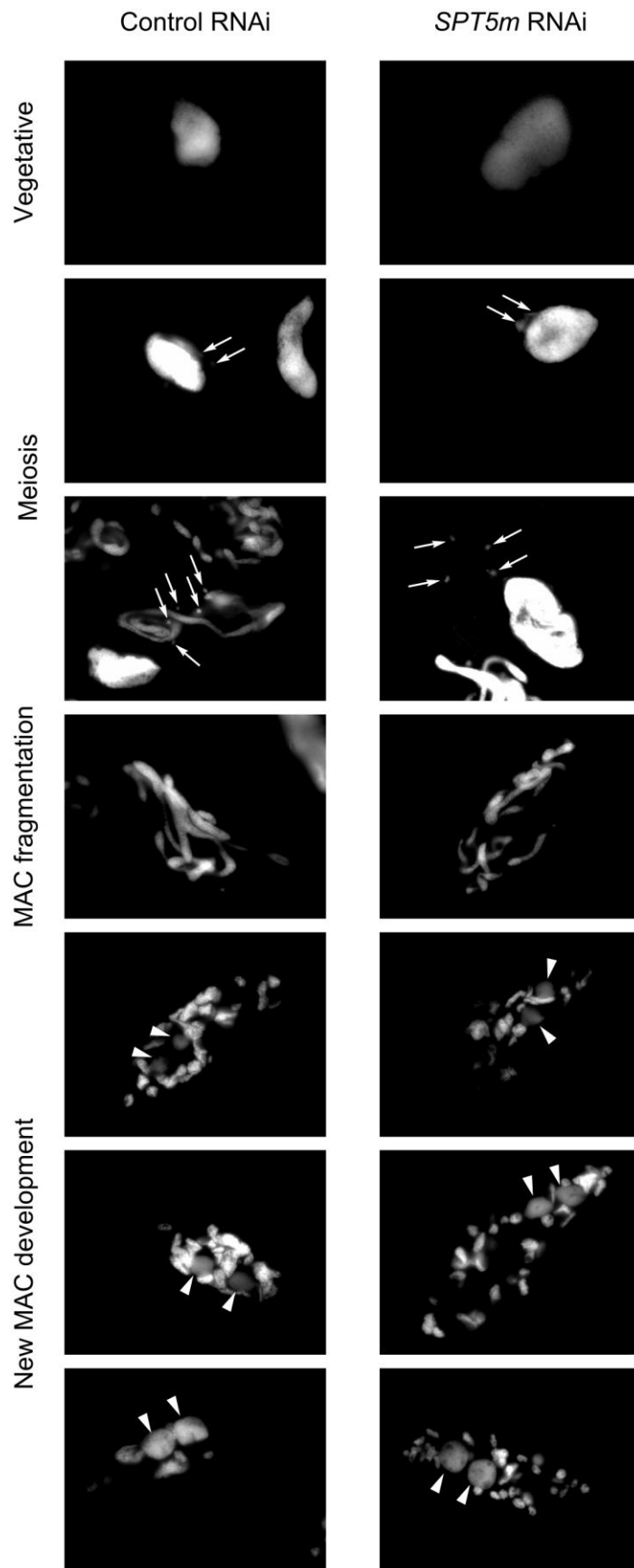




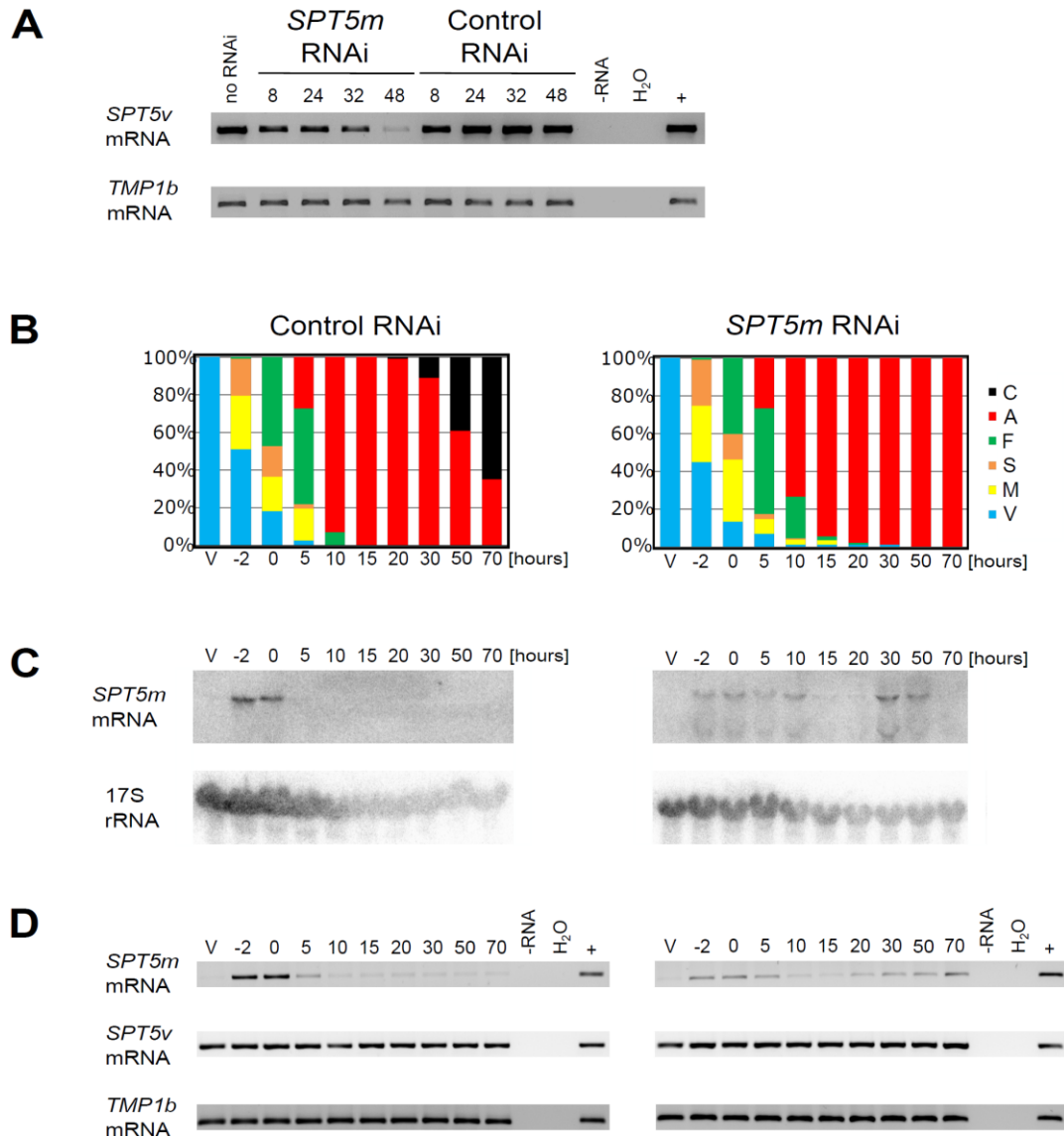
**Figure S1. Alignment and conservation of predicted structural domains for *P. tetraurelia* Spt5 proteins.** A. Complete protein sequences from different organisms were aligned as described in the legend of Figure 1. The structural annotation above the alignment is based on (65) and (23). The Glu residue important for interaction with Spt4 (66) is conserved in both *Paramecium* Spt5 proteins and is highlighted in red. Abbreviations and accession numbers are as follows: Pt, *Paramecium tetraurelia*, Spt5v: GSPATG00013468001, Spt5m: GSPATG00023145001; Tt, *Tetrahymena thermophila*, XP\_976854.1; Ot, *Oxytricha trifallax* Spt5-g19: EJY83850.1, Spt5-g12: Contig19613.0.g12(protein) (OxyDB); Tb, *Trypanosoma brucei*, Q7YUT3; Hs, *Homo sapiens*, NP\_001124296.1; At, *Arabidopsis thaliana*, KOW1: AED90724.1, SPT5-1: NP\_180968.2, Spt5-2: NP\_192575.2; Dm, *Drosophila melanogaster*, NP\_652610.1; Sc, *Saccharomyces cerevisiae*, NP\_013703.1; Pf, *Pyrococcus furiosus*, Q8TZK1.1. B. Alignment of Spt5m and Spt5v proteins from four *Paramecium* species.



**Figure S2. Silencing of *SPT5v* and *SPT5m* during vegetative growth.** The average division rate observed upon RNAi against *SPT5v*, *SPT5m* and control non-essential genes *ND7* and *ICL7* or without silencing (*Kp*) is shown. The data summarizes results obtained for more than 36 cell lines (*SPT5v*: n=93; *SPT5m*: n=36; *ND7*: n=36; *ICL7*: n=57; *Kp*: n=87). Vertical bars show standard deviation. Cells grown in *SPT5m* silencing medium grew as control cells, while for *SPT5v*-RNAi we observed slow-growth phenotype.

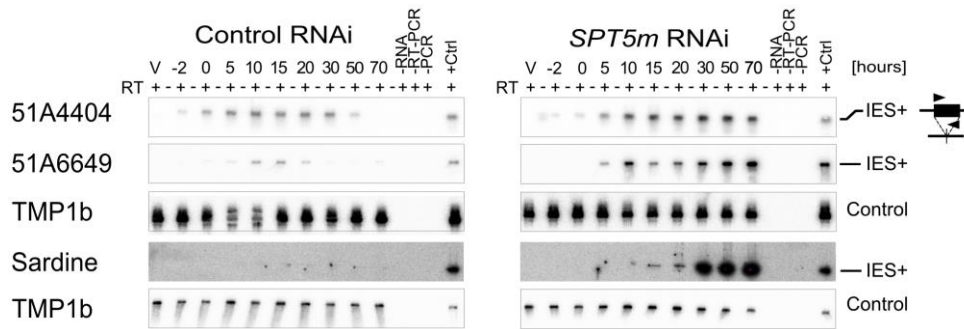


**Figure S3. Cytological observation of DAPI-stained cells silenced for *SPT5m* or the control gene (*ND7*). Arrows point to micronuclei, arrowheads indicate new macronuclei.**

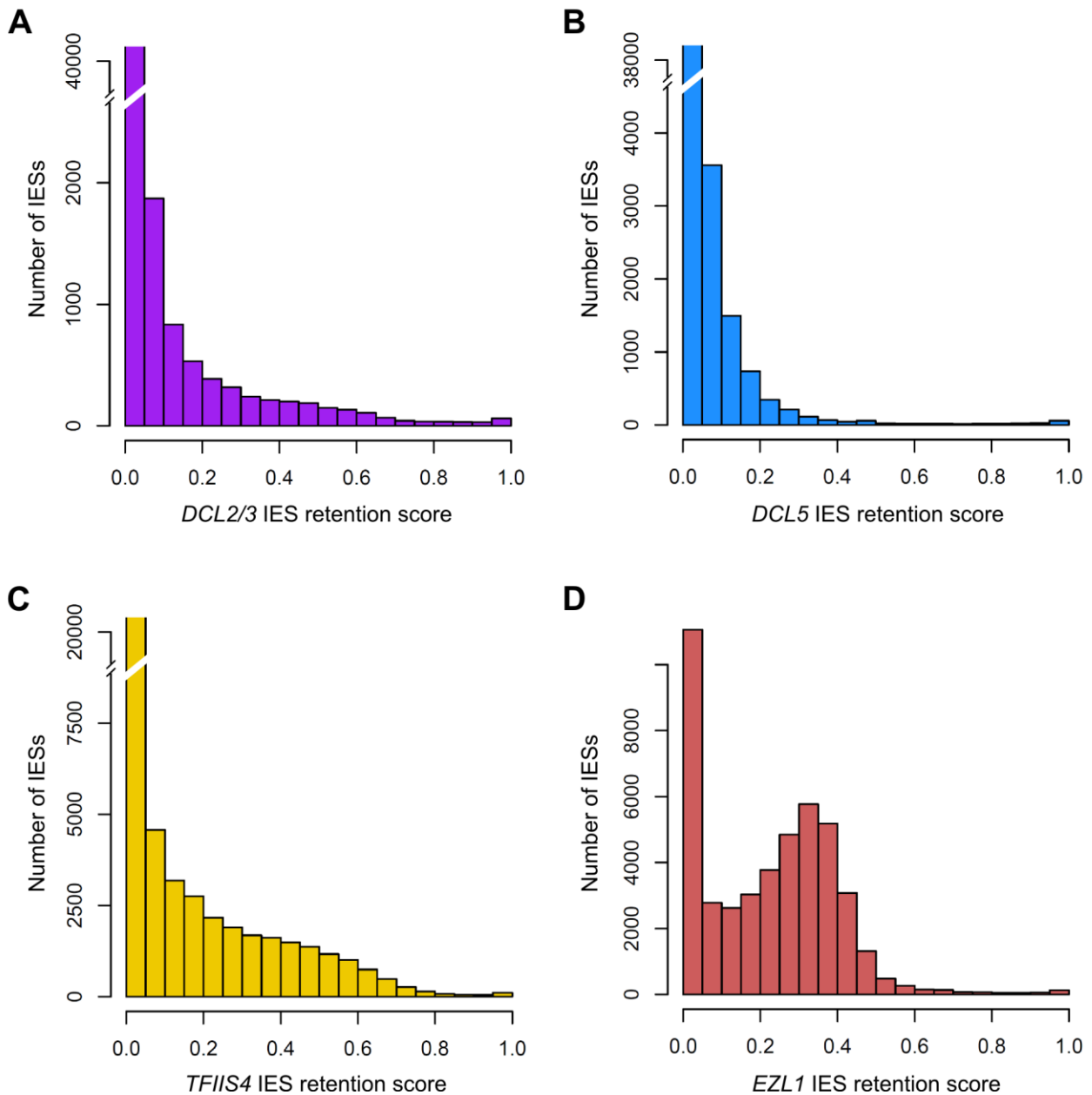


**Figure S4. Evaluation of silencing efficiency for *SPT5v* and *SPT5m*-RNAi.** A. RT-PCR analysis of the expression level of *SPT5v* after silencing of this gene. RNA samples were obtained from cultures without RNAi treatment (noRNAi) and after 8, 24, 32 and 48 hours of silencing from *SPT5v*-silenced and control RNAi cultures (*ND7* RNAi). Reverse transcription was performed using oligo dT. In order not to saturate the PCR reactions, only 25 PCR cycles were performed. Control RT-PCR obtained for the constitutively expressed *TMP1b* gene encoding a trichocyst matrix protein are shown below. A large decrease of *SPT5v* mRNA level is visible after 32-48 hours. B. Histograms show the progression of autogamy in strain 51mt7  $\Delta A$ . As a control we used cells silenced for the unrelated *ND7* gene. For each time-point (V: vegetative culture; -2: meiosis and early MAC fragmentation; 0: around 50% of cells with fragmented MAC; 5 to 70: 5 to 70 hours following time 0, respectively), cells were stained with DAPI to visualize old and new MACs. V: vegetative parental MAC; M: meiosis; S: skein formation; F: fragmented old MAC but no detectable developing new MACs; A: fragmented old MAC + 2 visible anlagen, C: post-karyonidal cells. C. Northern blot validation of *SPT5m*

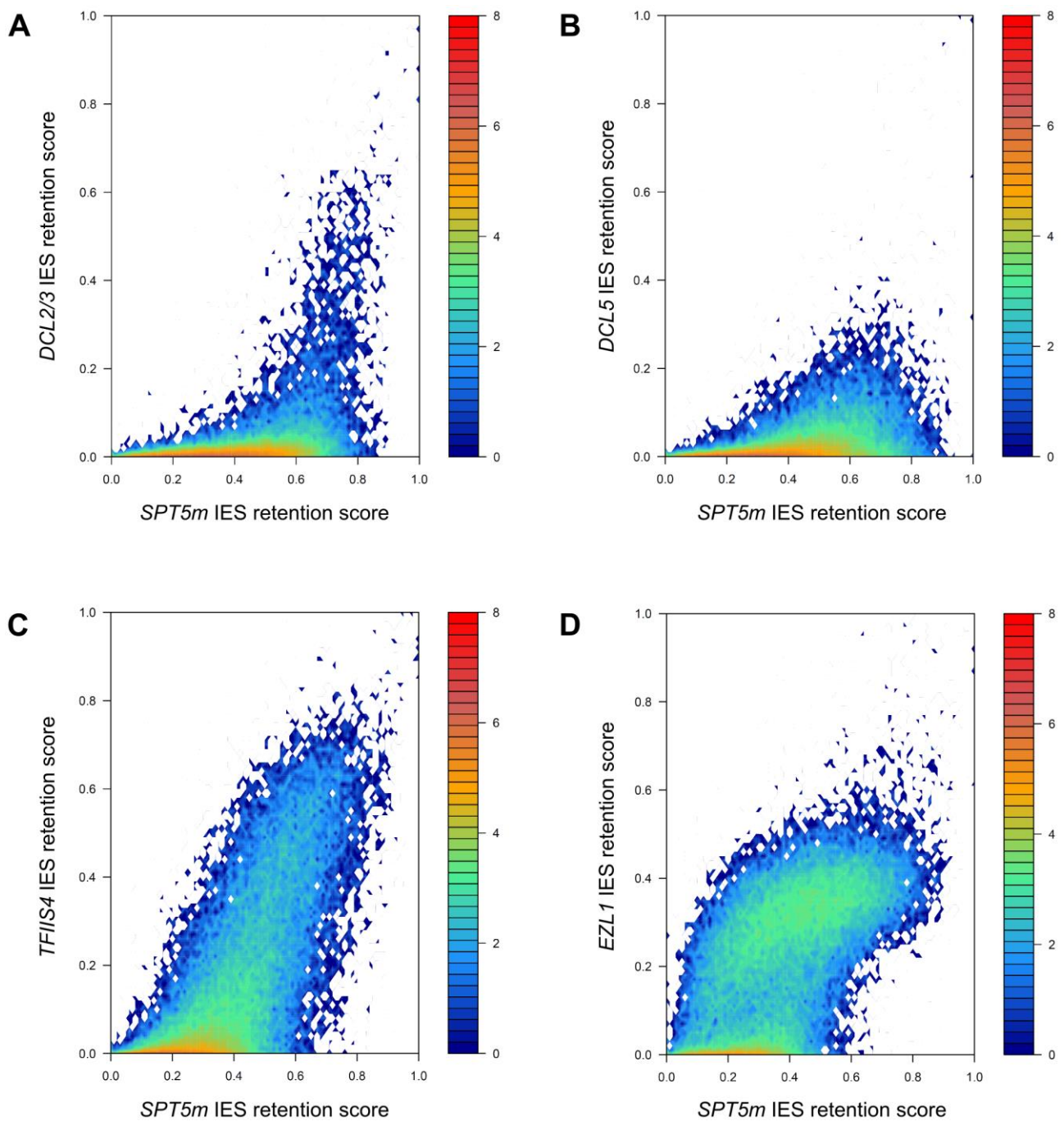
silencing. The blot was hybridized with an *SPT5m* probe and, subsequently, with a 17S rRNA probe as an RNA loading control. Additional band and smear visible in *SPT5m*-RNAi panel most probably correspond to some degradation products of *SPT5m* mRNA that comigrates with 18s and 28s rRNA (compare with Figure S3 in (41)) . Appearance of *SPT5m* mRNA signal in late time points corresponds most probably to unusual late expression of *SPT5m* due to weak silencing at that time (siRNA level decreases with time). D. RT-PCR analysis of expression level of *SPT5m* and *SPT5v* after silencing of *SPT5m*. RNA samples were treated as described in panel A. *SPT5m* expression is lower after RNAi than in the control, while *SPT5v* mRNA level is not altered, as expected



**Figure S5. Detection of transcriptional activity of the new MAC in *SPT5m*-silenced cells during autogamy.** RT-PCR and Southern blot detection of IES-containing transcripts (IES+) as well as transposon-derived transcripts in a control culture (cells silenced for *ND7* gene expression) and in *SPT5m*-silenced cells. Autogamy stages are described in detail in **Figure S4B**: V – vegetative cells, -2 – cells during meiosis/early MAC fragmentation, 0 to 70 – autogamy stages in hours. PCR primers were located within tested IESs: 51A4404, 51A6649 (34) and a conserved region of the *Sardine* transposon common to eight copies of this element (27) (primers within ORF1 of the transposon, SardinUp: GAACACCTCCTATCCTTTAATCATC, SardinLo: GACCTAATCGCAATCATTATGGATTTC). Control RT-PCR obtained for the constitutively expressed *TMP1b* gene encoding a trichocyst matrix protein are shown below. Early appearance and accumulation of IES+ signal toward the end of autogamy was observed in *SPT5m* silencing for all tested loci, while in control cells IES+ products appeared only at earlier time-points. Results obtained for *SPT5m* were similar to those observed when genome rearrangements were totally blocked by *PGM*-RNAi (34). These results suggest that *SPT5m*-RNAi inhibits IES excision and elimination of *Sardine* transposons, which would result in amplification of non-rearranged DNA and, in consequence, increase of IES+ transcript production.

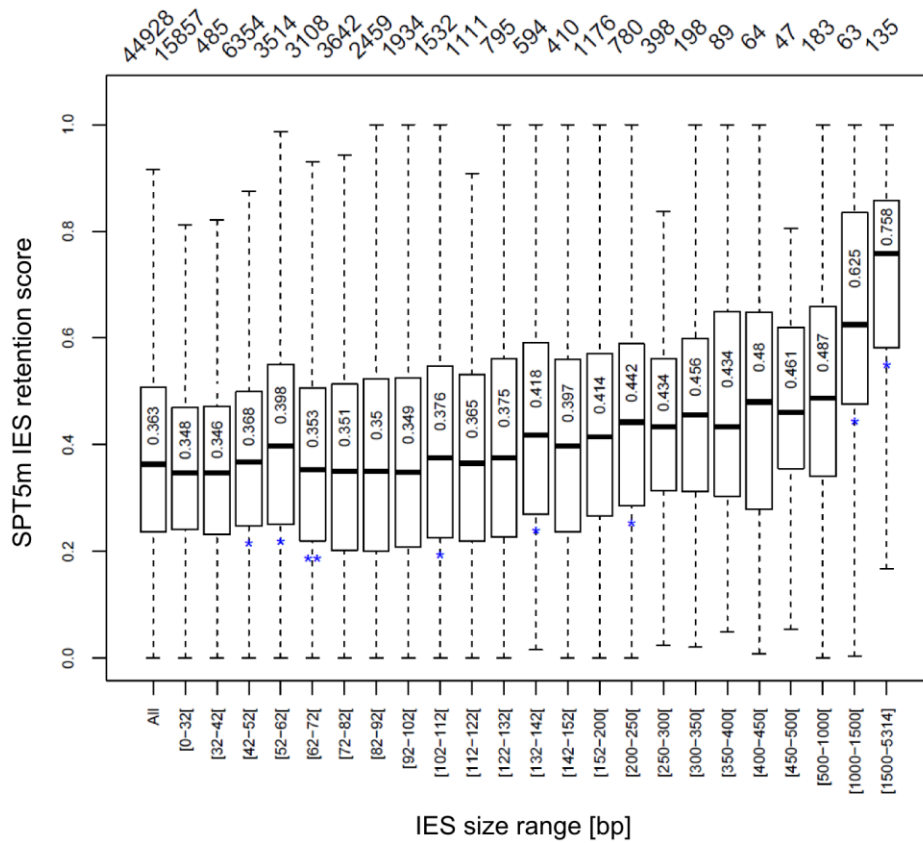


**Figure S6. Analysis of IES excision in cells silenced for expression of *DCL2/3*, *DCL5*, *TFIS4* and *EZL1*.** Retention score distribution determined by re-sequencing of DNA extracted from a cell fraction enriched in new MACs, after silencing of each factor. IES retention scores for silencing of *DCL2/3*, *DCL5*, *TFIS4* or *EZL1* were determined using published data (34, 37, 38). A. *DCL/DCL3*-KD retention scores; B. *DCL5*-KD retention scores; C. *TFIS4*-KD retention scores; D. *EZL1*-KD retention scores.



**Figure S7. Relation between *SPT5m*-KD retention scores and *DCL2/3*, *TFIIS4*, *EZL1* and *DCL5* retention scores.** Heatmaps show the relation between *SPT5m* retention scores for all IESs and retention scores obtained for other factors. The color represents the number of IESs (in log<sub>2</sub> scale) according to the legend on the right. *SPT5m*-KD retention scores were compared to: A. *DCL/DCL3*-KD retention scores; B. *DCL5*-KD retention scores; C. *TFIIS4*-KD retention scores; D. *EZL1*-KD retention scores.





**Figure S8. Relation between IES size and *SPT5m* IES retention score.** The box plot displays the *SPT5m* IES retention score distribution for each group of IESs corresponding to a peak in the periodic IES size distribution (27). The median retention score (horizontal line inside the box) and the first (top of box) and third (bottom of box) quartiles are shown. Blue stars indicate that the retention score distribution of a given group is significantly different from the retention score distribution of the previous group according to a Mann-Whitney test.

## Supplementary Tables

Targeted gene	<i>SPT5m</i>	<i>ND7</i>	<i>ICL7</i>	none *
% wild type	1%	94%	97%	90%
% sick	2%	3%	0%	5%
% death	98%	3%	3%	5%
Total cells	480	288	192	240
No. of experiments	10	6	4	5

\* Control grown in standard *K. pneumoniae* medium.

### Table S1. *SPT5m* silencing in autogamy.

Survival test of post-autogamous cells submitted to RNAi against *SPT5m* gene and control non-essential genes - *ICL7* and *ND7*. For each condition, the number of replicate experiments is indicated in the last line.

Targeted gene	<i>SPT5m</i>	<i>ND7</i>	none *
% wild type	3%	100%	97%
% sick	8%	0%	0%
% death	89%	0%	3%
Total cells	36	34	38

\* *K. pneumoniae*

### Table S2. *SPT5m* silencing in conjugation.

Survival test of post-conjugation cells submitted to RNAi against *SPT5m* gene and control non-essential *ND7* gene.

Sample	Gene	23 nt	25 nt (scnRNA)	26-29 nt (iesRNA)	25 nt /23 nt	26-29 nt /23 nt
Early	<i>ND7</i>	19.03	361.27	4.23	<b>18.98</b>	0.22
	<i>SPT5m</i>	148.22	66.61	1.90	<b>0.45</b>	0.01
	<i>DCL2/3</i>	100.25	14.34	1.58	<b>0.14</b>	0.02
Late	<i>ND7</i>	63.70	83.51	238.04	1.31	<b>3.74</b>
	<i>SPT5m</i>	199.53	39.83	156.35	0.20	<b>0.78</b>
	<i>DCL2/3</i>	171.14	23.80	122.34	0.14	<b>0.71</b>

**Table S3. Reduction in scnRNA after *SPT5m* RNAi.**

The sRNA-seq Illumina sequencing samples for RNAi of the indicated gene(s) (first two columns) were mapped and normalized as described in Materials and Methods. The table presents the normalized counts for 23 nt sRNAs, 25 nt sRNAs presumed to be scnRNAs and the sum of the counts for 26- 29 nt sRNAs, presumed to be iesRNAs (next three columns). Finally, count ratios are presented in the last two columns, first of the scnRNA count divided by the 23 nt count and then of the iesRNA count divided by the 23 nt count. Striking differences in the ratios between *ND7* RNAi and *SPT5m* or *DCL2/DCL3* RNAi (boldface type) were found at early and late times for the scnRNA and iesRNA ratios, respectively.

	<i>DCL2/3</i> - sensitive IESs	<i>DCL5</i> - sensitive IESs	<i>TFIIS4</i> - sensitive IESs	<i>EZL1</i> - sensitive IESs	<i>SPT5m</i> - sensitive IESs	All IESs
Number of IESs	3 020	2 475	20 524	30 892	41 380	44 928
Average IES length [nt]	208	54	94	94	78	79
Number of <i>SPT5m</i> -sensitive IESs	2 999	2 452	20 366	29 996	41 380	41 380
Percentage of <i>SPT5m</i> -sensitive IESs	99%	99%	99%	97%	100%	92%
Average <i>SPT5m</i> retention score	0.68	0.61	0.50	0.43	0.40	0.38

**Table S4. Analysis of IES retention shows a gradation of effects after RNAi of different factors.**

IES retention was determined for each sequencing sample as described in Materials and Methods, and significantly retained IESs according to the statistical test were considered to be sensitive to the factor that had been depleted by RNAi. The table provides the number, average length and average retention scores for the sensitive IESs, and shows that the IESs sensitive to each of the factors represents a subset of the *SPT5m*-sensitive IESs, the *DCL2/3*-sensitive IESs being the largest and most highly retained after *SPT5m*-RNAi.

Sample	Acc. no.	Reference
sRNA sequencing, <i>SPT5m</i> KD, T0 timepoint	SAMN04413729	This study
sRNA sequencing, <i>SPT5m</i> KD, T15 timepoint	SAMN04413730	This study
sRNA sequencing, <i>ND7</i> KD, T0 timepoint	SAMN04413731	This study
sRNA sequencing, <i>ND7</i> KD, T15 timepoint	SAMN04413732	This study
sRNA sequencing, <i>DCL2/3</i> KD, early timepoint	SRR907876	(30)
sRNA sequencing, <i>DCL2/3</i> KD, late timepoint	SRR907877	(30)
sRNA sequencing, control, early timepoint	SRR907874	(30)
sRNA sequencing, control, late timepoint	SRR907875	(30)
DNA from developing MAC, <i>SPT5m</i> KD	SAMN04358097	This study
DNA from developing MAC, control	ERX466735	(37)
DNA from developing MAC, <i>PGM</i> KD	ERA137444	(27)
DNA from developing MAC, <i>DCL2/3</i> KD	SRR2015146	(30)
DNA from developing MAC, <i>DCL5</i> KD	SRR2015148	(30)
DNA from developing MAC, <i>TFIIS4</i> KD	SRX710531	(34)
DNA from developing MAC, <i>EZL1</i> KD	ERX466734	(37)

**Table S5. List of all sequencing samples with their accession numbers.**

### Supplementary References

64. Guo,M., Xu,F., Yamada,J., Egelhofer,T., Gao,Y., Hartzog,G.A., Teng,M. and Niu,L. (2008) Core structure of the yeast *spt4-spt5* complex: a conserved module for regulation of transcription elongation. *Structure*, 16, 1649–1658.
65. Martinez-Rucobo,F.W., Sainsbury,S., Cheung,A.C. and Cramer,P. (2011) Architecture of the RNA polymerase-Spt4/5 complex and basis of universal transcription processivity. *EMBO J*, 30, 1302–1310.