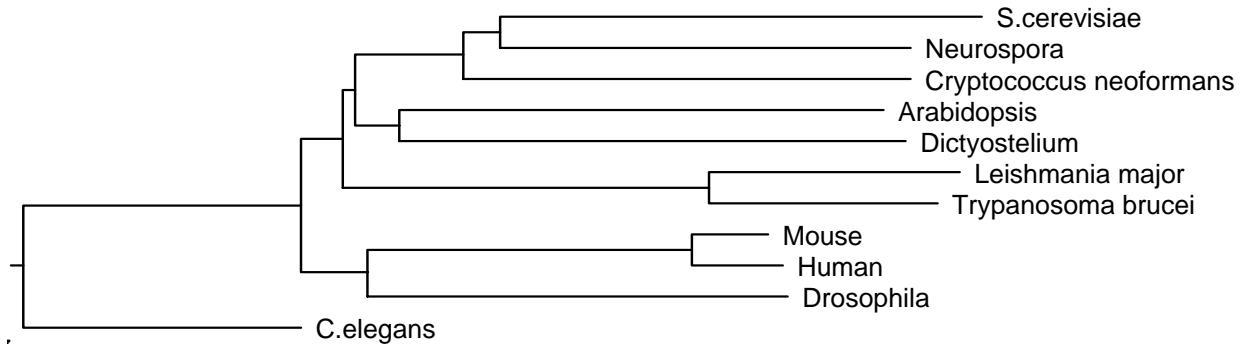


Figure S1.

Evolutionary tree depicting relationships amongst SPLs



An evolutionary tree amongst eukaryotic SPLs was constructed using the ClustalW algorithm as implemented in the DNAstar LaserGene package. Sequences used were Human (CAA09590), Mouse (AAH26135), *Drosophila melanogaster* (CAC10531), *Leishmania major* (AY770983) , *Trypanosoma brucei* (XP\_826696), *Neurospora crassa* (XP\_327047), *Saccharomyces cerevisiae* (AAB64470), *Cryptococcus neoformans* (EAL17929), *Caenorhabditis elegans* (AAD44756), *Arabidopsis thaliana* (AAM44962) and *Dictyostelium discoideum* (AAP37027).

The percent amino acid identity amongst these species may be found in the matrix below

Percent Identity												
1	2	3	4	5	6	7	8	9	10	11		
39.0	38.2	38.2	37.5	37.0	34.7	37.5	35.4	63.4	37.9	1	Leishmania major	
48.0	84.5	41.2	41.4	38.0	40.0	40.2	37.9	38.8	2	Mouse		
47.6	39.4	40.2	39.6	38.7	39.0	39.1	39.2	3	Drosophila			
43.1	42.2	36.1	38.5	39.6	37.7	38.8	4	Human				
37.3	36.3	42.3	40.5	38.6	40.8	5	Arabidopsis					
37.6	37.7	38.8	36.3	37.3	6	C.elegans						
37.8	45.5	36.3	42.5	7	38.4	38.7	38.9	8	S.cerevisiae			
36.6	46.4	37.7	40.4	9	36.6	46.4	46.4	9	Dictyostelium			
37.7	10	38.9	38.9	10	37.7	38.9	38.9	10	Neurospora			
	11										Trypanosoma brucei	
											Cryptococcus neoformans	

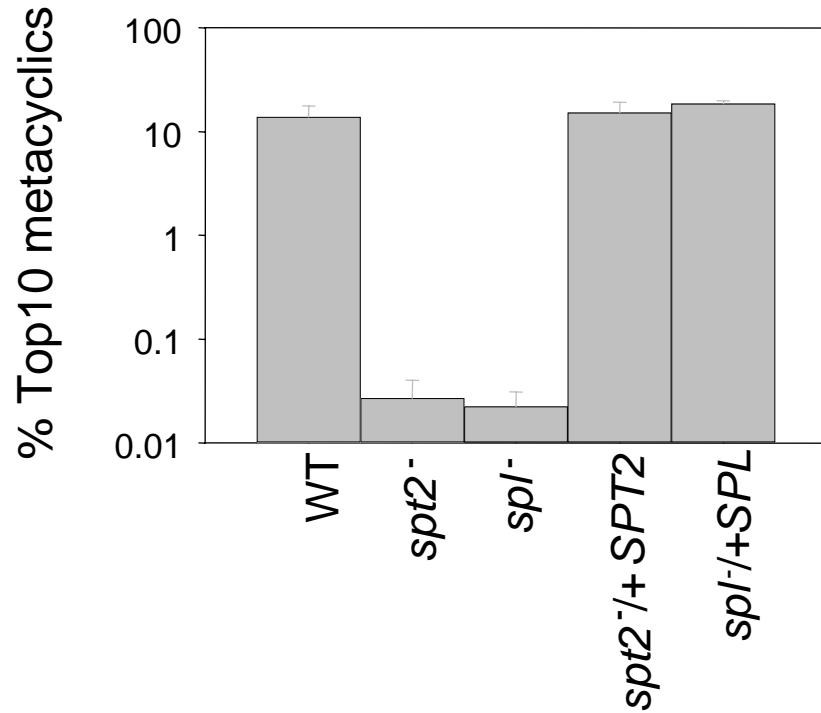
## Figure S2.

### Alignment of *Leishmania* SPL with eukaryotic SPLS

1 -----MTTICQVLNERLKDKSPTQIIVITLSSVVAARIAVNCFRDGR--- Lm  
 1 -----MPGTDLKLKDFFEPYLEILESYSTKAKNYVNGYCTKYEPWQLIAWSVLCTLLIVWVYELIFQPES-- Mm  
 1 -----MPSTDLLMLKAFFEPYLEILELEVYSTKAKNYVNGHCTKYEPWQLIAWSVVWTLLIVWGYEFVFQPES-- Hs  
 1 -----MDSVKHTTEIIVDLTKMHMINDRLSRYDPVVLVLAFFGTLVYTKVVHLYRKSEDPM Ce  
 1 MSGVSNKTVSINGWYGMPIMHLLREGDFAQFAMILTINELKIAIHGYLRNTPWYNMLKDYLFWIFCYKLISNFFYLLKVYG Sc  
 43 -----LAKRSYQAAWRGIRTLAAPP-IIRKEVKVVSVVKMPSKGKGEFKALALPEKSREAEVLQLVTQLHHDDLS- Lm  
 66 -----LWSRFKKKKLFLKIRKMPFIGRKIEQQVSKAKKDLVKNMPFLKVDKDVKTLPAQGMGTAEVLERLKEYSSMD- Mm  
 66 -----LWSRFKKKKCFKLTRKMPIIGRKIQDKLNKTKDDISKNMSFLKVDKEYVKALPSQGLSSSAVLEKLKEYSSMD- Hs  
 59 -----ILKRMGAYVFSLLRKLPAVRDKIEKELAAEKPKLIESIHKDDDKQFISTLPIAQLSQDSIMELAKKYEDYN- Ce  
 81 PVRЛАVRTYEHSSRRLFRWLDSPLRGTEKEVTKVQKSIEDELIRSDSQLMNPQLPSNGIPQDDVIEELNLNDLIP Sc  
 113 ---YEKG-[F]SGAVYHG-GRSHTAFINDVMAIFQWSNPLHS[DI]FGATRKMEAEIVSMVLHMYNGHLLPDAGGVVTSGGTE Lm  
 138 -GSWQEG-KASGAVYNG-EPKLTEL[VQAYGEFTWSNPLH]PDI[FPGLRKLEAEIVRMTCSLFN]GGPDS-CG-CVTSGGTE Mm  
 138 -AFWQEG-RASGTVYSG-EEKLTLLVKAYGDFAWSNPLH[PDI[FPGLRKIEAEIVRIACSLF]N]GGPDS-CG-CVTSGGTE Hs  
 131 -TFNIDGGRVSGAVYTDRAEHINLLGKIYEKYAFSNPLH[PDPVFP]GARKMEAEELIRMVNLNYPEDS-SG-SVTSGGTE Ce  
 161 HTQWKEG-KVSGAVYHG-GDDLIHLQTIAYEKYCVANQ[LHPDVEPAVRKMESEVVSMVLRFNAPSDTGCG-TTTSGGTE Sc  
 188 [SILMALKAYRDWGRMTRGIEHPSVVAAPITIHPFDKGAEYFCIDLIKVPVLVTTGCVDPKEMEYI]RYDTIAVAA[SAPNF] Lm  
 213 SILMACKAYRDLALEK-GIKTP[EIVAPESAHA]AFDKAAHYFGMKIVRVALK-KNMEVDVQAMKRAISRNNTAMLVCSTPQF Mm  
 213 SILMACKAYRDLAFEK-GIKTP[EIVAPQSAHA]AFNKAASYFGMKIVRVPLT-KMMEVDVGRAMRRASI[RNTAMLVCSTPQF] Hs  
 208 SIIMACFSYRNRAHSL-GIEHPVILACKTAHAAFDKAALCGMRLRHVPVD-SDNRVDSLKEMERLIDSNSVCMLVG[SAPNF] Ce  
 238 SLLLACLSAKMYALHHRGITEPEIIAPVTAHAGFDKAAYYFGMKLHRV[ELDPTTYQV]DLGKVKKFINKNTILLVG[SAPNF] Sc  
 268 PHGVIDPIEEISEMAYKHNIGM[HVDCCCLGGFIMP]FLEKTRGR-PAPVVDFRNR[GVT]SISC[DTH]KYGYAP[KGT]STV[MYRSKE] Lm  
 291 PHGVMDPVPEVAKLAVRYKIPLHVDA[CLGGFLIV]FMEKAGYPLEKFDFRVRKGVT[SISADTH]KYGYAP[KGS]S[VVMYSNEK] Mm  
 291 PHGVIDPVPEVAKLAVKYKIPLHVDA[CLGGFLIV]FMEKAGYPLEHFDFRVRKGVT[SISADTH]KYGYAP[KGS]SSLVLYSDKK Hs  
 286 PSGTIDPIPEIAKLGKKYGYIPVHVDA[CLGGFMIP]FMNDAGY-LIPVFDFRNP[GVT]SISC[DTH]KYGCTPKGS[SIVMYSRSKE] Ce  
 318 PHGIADDIEGLGKIAQKYKLPLHVDSLCLGSFIVSFMEKAGYKNLPLDFRVP[GVT]SISC[DTH]KYGFAP[KGS]S[VIMYRNSD] Sc  
 347 LRSFQ[FSCVAEW[P]PGGMY[CSP]AVSGSK[P]GNVIAGAWAAMVRMGMEGYVDCCHK[VTTRETMTREL]SK-LPYIRIIGEPAAS Lm  
 371 YRTYQFFVGADWQGGVYAS[PSIAGSRPGGIIAACWAALMHFGENGYVEATKQII]IKTARFLKSELEN-IKNIFIFGDPQLS Mm  
 371 YRNYQFFVDTDWQGGIYAS[PSIAGSRPGGISAACWAALMHFGENGYVEATKQII]IKTARFLKSELEN-IKGIFVFGNPQLS Hs  
 365 LHHFQYFSVADWCGGIYAT[PSIAGSRAGANTAVA]ATLLSFGRDEYVRRCAQIVKHTRMLAEKIEK-IKWIKPYGKSDVS Ce  
 398 LRMHQYYVNPWTGGLYGSPTLAGSRPGAI[VVG]CWTATMVNMGENGYIESCQE[VGAAMFKKKYIQENIPDLNIMGNPRYS] Sc  
 426 VFAFTSNVID[FRLGDDLKLRGWVNLNTLQFPPGLQFSV]LLQTPPAV[TARFLSDVKEIGDILFAESEKLIADGKKPVLGE Lm  
 450 VIALGSNDFDIYRLSNMMSAKGWNFNYLQFPPSIHFCITLVHTRKRAV[AIQFLKDIRES-----]VTQIMKNPKAKT-T Mm  
 450 VIALGSRDFDIYRLSNLMTAKGWNLNQLQFPPSIHFCITLLHARKRAV[AIQFLKDIRES-----]VTQIMKNPKAKT-T Hs  
 444 LVAFSGNGVNIYEVSDKMMKL[GWNLNTLQNPAAIHICL]TINQANE[VNAFAVDLEKI-----CEELAAKGEQKAD-S Ce  
 478 VISFSSKTLN[HELSDRSLKKGWHFNALQK]PVALHMAFRLSA-HV[VDEICDILRTT-----]VQELKSESNSKPSPD Sc  
 506 SSGTLYGT[AQRVPDRTIMQDVLRFLNTY]YQGXX Lm  
 521 GMGAIYGM[AQATIDRKLV]AEI[S]VFLDCYTTDPVTQGNQMNGSPKPR Mm  
 521 GMGAIYGM[AQTTVDRNMAELSSVFLDSLY]STD[V]TQGSQMNGSPKPH Hs  
 516 GMAAMYGM[AQV]PKSVVDEVIALYIDATYSAPPSTSNCe  
 549 GT[SAL]YGV[AGSVKTAGVADKLIVGFLDALY]KLGPGETATK Sc

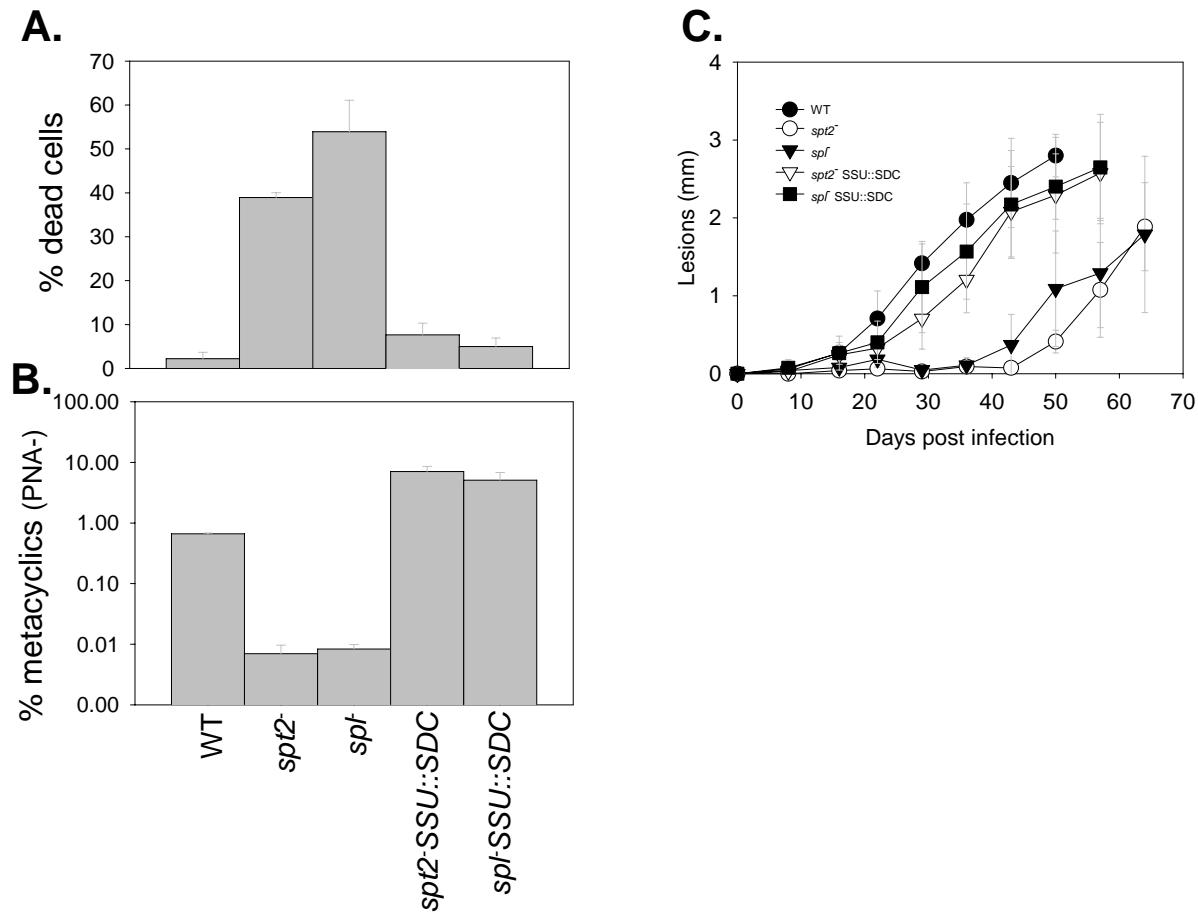
Residues show to be essential when mutated in other eukaryotic SPLs are indicated by black triangles. A putative pyridoxal phosphate binding region is boxed. Residues identical in all 5 sequences are shaded and boxed (for specific sequence information see the legend to Fig. S1).

Fig. S3



**Fig. S3.** *SpI*<sup>-</sup> and *spt2*<sup>-</sup> mutants are defective in metacyclogenesis. Promastigotes were grown to late stationary phase (three days after reaching maximum density) and the percentage of metacyclics was determined using the density centrifugation method and represented as “Top10 metacyclics”.

Fig. S4



**Fig. S4.** Overexpression of serine decarboxylase (SDC) restores defects in *spt2*- and *spl*- mutants. WT, *spt2*<sup>-</sup>, *spl*<sup>-</sup>, *spt2*<sup>-</sup> SSU::SDC, and *spl*<sup>-</sup> SSU::SDC promastigotes were grown in M199 medium as described in *Materials and Methods*. Three days after entry into stationary phase, the percentage of dead cells (propidium iodide-positive) was determined by flow cytometry (**A**), and the percentage of metacyclcs was determined using the peanut agglutination method (**B**). Stationary phase promastigotes were used to infect BALB/c mice at  $1.0 \times 10^6$  cells/mouse and the progression of lesion is shown in (**C**). Error bars represent standard deviations.

Table S1. EtN rescues the defects of *spl*<sup>-</sup> and *spt2*<sup>-</sup> mutants.

	Density (#/ml)	% PI +	% Top 10
WT control	2.8 x 10 <sup>7</sup>	5.12	13.8
WT + EtN	2.9 x 10 <sup>7</sup>	4.24	10.0
<i>spt2</i> <sup>-</sup> control	7.4 x 10 <sup>6</sup>	38.0	0.32
<i>spt2</i> <sup>-</sup> + EtN	2.8 x 10 <sup>7</sup>	11.6	19.5
<i>spl</i> <sup>-</sup> control	7.3 x 10 <sup>6</sup>	49.5	0.29
<i>spl</i> <sup>-</sup> + EtN	2.6 x 10 <sup>7</sup>	7.82	17.4

Promastigotes were grown to late stationary phase (three days after reaching maximum density) in the absence or presence of 0.5 mM EtN. Culture density was determined using a Coulter Counter; percentage of dead cells was determined by flow cytometry of parasites stained with propidium iodide (PI + %); and the percentage of metacyclics was determined using the density centrifugation method and represented as “Top10 metacyclics”.