

A

<i>Pf SufS</i>	-----MLRGPRCLYI-----	YLFFV---FLPFSFCYIRNNNDNR	30
<i>Bs SufS</i>	-----	-----	0
<i>Ec SufS</i>	-----	-----	0
<i>At cNif1</i>	-----MEGVAMK-----	LP---SFPNA---ISIGHRS	21
<i>Pf IscS</i>	MKFLQIIKHL-KL-QNKKNALDNFVNCRTEHISNINKLFLNNFSSTKEHSEHGQVKHEN	-----	58
<i>Bs YrvO</i>	-----	-----	0
<i>Ec IscS</i>	-----	-----	0
<i>At mNif1</i>	MASKVIS---ATIRRRTLTKPHGTFSRCR-----	YL---STA---AA-----	32
<i>Sc Nfs1</i>	MLKSTATRSITRLSQVYNVPAATYRACLVSRR-----	FY---SPP---AAGVKLDDN	46
 <i>Pf SufS</i>	FVY -----	IVKSIRKGPNIK-----LR-----	47
<i>Bs SufS</i>	-----	-----	0
<i>Ec SufS</i>	-----	-----	0
<i>At cNif1</i>	FSR -----	VRCSSSLSV---CSAA-----	37
<i>Pf IscS</i>	FLNSTLKYEENSQNGSTNNLKNGKYNMYVSEG NVNINEEKYKDNNNISSNNTQYNNNNSSNS	-----	118
<i>Bs YrvO</i>	-----	-----	0
<i>Ec IscS</i>	-----	-----	0
<i>At mNif1</i>	-----	-----	32
<i>Sc Nfs1</i>	FS -----	LETHTDIQA-----AAKAQASARASAS	70
 <i>Pf SufS</i>	-LTKDEKPNIDNHIIDYFKNVREHF PFFKE--	-NKSЛИYFDSAATTHKPSCVIEKMSEFY	103
<i>Bs SufS</i>	-----MNITDIREQF PILHQVNGHDLVYLD	SAATSQKPRAVIETLDKYY	45
<i>Ec SufS</i>	-----MIFSVDKVRADF PVLSREVNGLPLAYLDSAA	SAQKPSQVIDAEAEFY	47
<i>At cNif1</i>	-AASSATISTDSESVS LGHRVRKDFRILHQEVNGSKLVYLD	SAATSQKPAAVLDALQNYY	96
<i>Pf IscS</i>	GSLNDEGPL-WKEHID-----	DVNENKKKKMNRFYLDSQATTMIDPRVLDKMLPYM	169
<i>Bs YrvO</i>	-----MERIYLDHAATSPMDERVLEQMIPH	-----	26
<i>Ec IscS</i>	-----MKLPIYLDYSATT PVDPRVAEKMMQFM	-----	27
<i>At mNif1</i>	---ATEV---NYEDE-----	SIMMKGVRISGRPLYLDMQATT PIDPRVFDAMNASQ	77
<i>Sc Nfs1</i>	GTTPDAVVASGSTAMS-----	HAYQENTFGTRPIYLDMQATTPTDPRVLDMLKFY	122
 <i>Pf SufS</i>	--KKENSNIHRGIYKL SHNATNNYEKV RETIKEYINCEKNDN IIFTNGSTYGLNVVCKMM	-----	161
<i>Bs SufS</i>	--NQYNSNVH RGVHTLGTRATDG YEGAREK VRKF INAKSMAE IIFTKGTTTSLNMVALSY	-----	103
<i>Ec SufS</i>	--RHGYAAV H RGIHTL SAQATEK MENV RKRASL FINARS AEEELV FVRGTTEGINLVANSW	-----	105
<i>At cNif1</i>	--EFYNSNVH RGIH YLSAKATDEF E LARKV ARFINAS D SREIV FTRNATEAINLVAYS W	-----	154
<i>Pf IscS</i>	--TYIYGN AHSRNHF FGWESE KAVE DARTNLLNLINGKNNKE IIFTSGATESNNLALIGI	-----	227
<i>Bs YrvO</i>	--SGSFGNPSS-IHSFGRESRKWVDEARAQIAEIG-AAEQE IIFTSGGT EADNL AIMGT	-----	82
<i>Ec IscS</i>	TMDGTFGNPASRSHRF GWQAE EAVDIARNQIA DLVG-ADPREIVFTSGATESDNLAIKGA	-----	86
<i>At mNif1</i>	--IHEYGNPHSR THLY GWEA ENA VENARN QVAKLIE-ASPKEIVFVGATEANNMAVKGV	-----	134
<i>Sc Nfs1</i>	--TGLYGNPHSNT HSYGWETNT AVENARAHVAKMIN-ADPK E IIFTSGATESNNMVLKGV	-----	179
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<i>Pf SufS</i>	IEEI--IKKEEDEIYLSYMEHH SNTI PWQEYINKEKKGRIKYVPLNKSGYINIKKLISNM	-----	219
<i>Bs SufS</i>	ARAN--L KPGDEV VITYMEHH HANI IPWQQAVKA-TGATLKYIPLQEDGTISLEDVRET	-----	159
<i>Ec SufS</i>	GNSN--VRAGDN III ISQMEHH HANI VPWQMLCAR-VGAELRVIPLNPDGTIQLETLPTLF	-----	161
<i>At cNif1</i>	GLSN--L KPGDEV ILTVAE HHSCIVPWQIVSQK-TGAVLKFVTLNEDEV PDI NK LRELI	-----	210
<i>Pf IscS</i>	CTYYNKL NKQKNH IIITSQIEHKCILQTCRFLQ-T-KGF EVTYLKPDTNGLV KLD DIKNSI	-----	285
<i>Bs YrvO</i>	ALARKD--LGRHII ITKIEHH AVLHTCEKLE-G-DGF DITYL DV DQNGR VS A KVKE AL	-----	137
<i>Ec IscS</i>	ANFYQK--KGK HIIITSKTEHKAVL DTCRQLE-R-EGF EVTYLAPQRNGIIDLKE LEA AM	-----	141
<i>At mNif1</i>	MH FYKD--TKKHVITTQTEHKCVL DSCRHLQ-Q-EGF EVTYLPVKT DGLV DLEM L REAI	-----	189
<i>Sc Nfs1</i>	PRFYKK--TKKHII TTRTEHKCVL EAAR AMM-K-EGF EVTF LNVD DQGLIDLKE LEDAI	-----	234
 <i>Pf SufS</i>	NINTKVI SICH ASN VIGNIQ NIEKI IKKIKNVY PHIIII IDASQFAH IKYDI KMK KNK	-----	279
<i>Bs SufS</i>	TSNTKIVAVSHVSNV LGTVNPIKEMAKIAHDNG--AVIVVDGAQSTPHMKIDVQDLD---	-----	214
<i>Ec SufS</i>	DEKTRLLAITHVSNV LGTENPLAEMITLAHQHG--AKVLVDGAQAVMHH PVDVQALD---	-----	216
<i>At cNif1</i>	SPKTKLVAVHHVSNV LASSLPIEEIVVWAHDVG--AKVLVDACQSVPHMVV DVQKLN---	-----	265
<i>Pf IscS</i>	KDNTIMASF IF VNNEIGVIQDIENIGNL CKEKN--ILF HTDASQAA GKV P IDVQK MN---	-----	340
<i>Bs YrvO</i>	RDDTILVTV MYGNNEIGTVQPIE EIGELL KEHK--AYF HTDAVQAF GLLP IDV KNSH---	-----	192
<i>Ec IscS</i>	RDDTILVS IMHV NNIEIGVVQDIAAIGEMCRARG--I IYHV DATQSVGKLP IDLSQLK---	-----	196
<i>At mNif1</i>	RPDTGLVS IMAVN NEIGVVQPMEEIGMICKEHN--VPF HTDAAQAI GKV P IDV VKWN---	-----	244
<i>Sc Nfs1</i>	RPDTCLVS VMAVN NEIGVIQPIKEIGAICRK NK--IYF HTDAAQAYGK I HIDV NEMN	-----	289

<i>Pf Sufs</i>	SCP DILITSGHKFCASLGTGFIFINKELSSKYKFKPLIYGSNIITNVSKVSKFVTSLSE	339
<i>Bs Sufs</i>	--CDFFALSSHKMCGPTGVGVLYGKALLE--NMEPAEFGGEMIDFVGLYE-STWKELPW	269
<i>Ec Sufs</i>	--CDFYVFSGHKLGYGPTGIGILYVKEALLQ--EMPPWEGGGSMIATVSLSEGTTWTKAPW	272
<i>At cNif1</i>	--ADFLVASSHKMCGPTGIGFLYGKS DLLH--SMPPFLGGGEMISDVFLDH-STYAEPPS	320
<i>Pf IscS</i>	--IDLMSMSGHKLGYGPKGIGALYIKRKKP-NIRLNALIHGGGQ-----ER	382
<i>Bs YrvO</i>	--IDLLSVSGHKLNGPKGTFLYASK---DVKLSPLLFGGEQ-----ER	231
<i>Ec IscS</i>	--VDLMSFSGHKIYGPKGIGALYVRR-KP-RVRIEAQMHHGGH-----ER	237
<i>At mNif1</i>	--VALMSMSAHKIYGPKGVGALYVRR-RP-RIRLEPLMNGGGQ-----ER	285
<i>Sc Nfs1</i>	--IDLLSISSHKIYGPKGIGAIYVRR-RP-RVRLEPLLSGGGQ-----ER	330
<i>Pf Sufs</i>	LLETGTQNIPGILSMGISLEFFKKINWNVYQYEMYLYDLFIYYMNKYMKNHFV-QLPNL	398
<i>Bs Sufs</i>	KFEAGTPPIAGAIGLGAIDFLEEIGLDEISRHEHKLA-----AYALERFR-QLDGV	320
<i>Ec Sufs</i>	RFEAGTPNTGGIIGLGALEYVSALGLNNIAEYEQNLM-----HYALSQLE-SVPDL	323
<i>At cNif1</i>	RFEAGTPAIGEAIALGAAVDYLSGIGMPKIHEYVEIG-----KYLYEKLS-SLPDV	371
<i>Pf IscS</i>	GLRSGTLPTHLLIVGFGEAAKVCSLEMN-R---DEKKVRYFF---NYVKDYLTKHLDYI	433
<i>Bs YrvO</i>	KRAGTENVPGIVGLKEAIKLSSEERD-E---KNEKYQSFK---AIFADTLRDAGVAF	282
<i>Ec IscS</i>	GMRSGTLPVHQIVGMGEAYRIAKEEMA-T---EMERLRGLR---NRLWNGIKDI-EEV	287
<i>At mNif1</i>	GLRSGTGATQQIVGFGAACELAMKEME-Y---DEKWIKGLQ---ERLLNGVREKLDGV	336
<i>Sc Nfs1</i>	GLRSGTLAPPLVAGFGEAARLMKKEFD-N---DQAHIKRLS---DKLVKGLLSA-EHT	380
<i>Pf Sufs</i>	NLSYKKENINYKSHMQTHPPVHKYNDEQNFTNDHNITQSQTKSIHSQHDTFKIYTHDTR	458
<i>Bs Sufs</i>	TVYGPEE-----	327
<i>Ec Sufs</i>	TLYGPQN-----	330
<i>At cNif1</i>	RIYGPRP-----	378
<i>Pf IscS</i>	VFNCGQINR-----	442
<i>Bs YrvO</i>	EVNGDKEHS-----	291
<i>Ec IscS</i>	YLNGDLEHG-----	296
<i>At mNif1</i>	VVNGSMDSR-----	345
<i>Sc Nfs1</i>	TLNGSPDHR-----	389
<i>Pf Sufs</i>	KYGLKKIGILPLWSNTFSSFDLVTFLDF-KNICIRAGHHCASLLH--KYYLK-----	508
<i>Bs Sufs</i>	-----RAGLVTNFLDDVPHDVATVLDA-EGIAVRAGHHCAQPLM--KW-LDV-----	371
<i>Ec Sufs</i>	-----RLGVIAFNLGKHHAYDVGSLDN-YGIAVRTGHHCAMPLM--AY-YNV-----	374
<i>At cNif1</i>	SESVHARGALCSFNVEGLHPTDLATFLDQQHGVAIRSGHHCAQPLH--RY-LGV-----	428
<i>Pf IscS</i>	-----YYGNMNISFLFVEGESLLMSL--NEIALSSGSACTSSTLEPSYVLSI-GISED	493
<i>Bs YrvO</i>	-----LPHVNLNLFPGVSVEALLVNLDM-AGVAVSSGSACTAGSVLPShVLTAMFGEESD	345
<i>Ec IscS</i>	-----APNILNVSFNYVEGESLIMAL--KDLAVSSGSACTSASLEPSYVLRAL-GLNDE	347
<i>At mNif1</i>	-----YVGNLNLASFAYVEGESLLMGL--KEVAVSSGSACTSASLEPSYVLRAL-GVDED	396
<i>Sc Nfs1</i>	-----YPGCVNVSEAYVEGESLLMAL--RDIALSSGSACTSASLEPSYVHLHAL-GKDDA	440
<i>Pf Sufs</i>	--PDTSRISIYFYNTPQEIKYLAQQIASTSFMNEMKNEK-----	566
<i>Bs Sufs</i>	--TATARASFYLYNTEEEIDKLVEALQKTKEYFTNVF-----	429
<i>Ec Sufs</i>	--PAMCRASLAMYNTHEEVDRLVTLGLQRIHRLIG-----	432
<i>At cNif1</i>	--NASARASLYFYNTKDDVDAFIVALADTVSFFNSFK-----	486
<i>Pf IscS</i>	IAHTSIRIGFNRTFFFEVQQLCINLVKSVERLRSISPLYEMELEKKNPSNDDIPKFIWT	553
<i>Bs YrvO</i>	RLTSSIRISFGLGNTAEQVKTAAKHVADVVKRIT-----	405
<i>Ec IscS</i>	LAHSSIRFSLGRFTTEEEIDYTIELVRKSIGRLRDLSPLWEMYKQGV-----DLNSIEWA	402
<i>At mNif1</i>	MAHTSIRFGIGRFTTKEEDKAVELTVKQVEKLREMSPLYEMVKEGI-----DIKNIQWS	451
<i>Sc Nfs1</i>	LAHSSIRFGIGRFSTEEDYVVKA VASDRVKFLRELSPWEMVQEGI-----DLNSIKWS	495

<i>Pf Sufs</i>	--	546
<i>Bs Sufs</i>	--	406
<i>Ec Sufs</i>	--	406
<i>At cNif1</i>	--	463
<i>Pf IscS</i>	--	553
<i>Bs YrvO</i>	--	379
<i>Ec IscS</i>	HH	404
<i>At mNif1</i>	QH	453
<i>Sc Nfs1</i>	GH	497

Class II

Class I

B

Percentage (%) sequence identity among the cysteine desulfurases

	<i>Pf SufS</i>	<i>Bs SufS</i>	<i>Ec SufS</i>	<i>At cNif1</i>	<i>Pf IscS</i>	<i>Bs YrvO</i>	<i>Ec IscS</i>	<i>At mNif1</i>	<i>Sc Nfs1</i>
<i>Pf SufS</i>		45.57	41.53	40.98	28.41	33.89	34.37	29.62	29.73
<i>Bs SufS</i>	45.57		67.08	67.23	36.13	42.68	39.95	38.21	38.55
<i>Ec SufS</i>	41.53	67.08		63.11	35.66	38.86	36.95	36.56	35.75
<i>At cNif1</i>	40.98	67.23	63.11		30.24	38.82	37.62	36.65	34.31
<i>Pf IscS</i>	28.41	36.13	35.66	30.24		58.91	69.25	54.95	55.94
<i>Bs YrvO</i>	33.89	42.68	38.86	38.82	58.91		62.76	59.42	60.47
<i>Ec IscS</i>	34.37	39.95	36.95	37.62	69.25	62.76		76.30	73.51
<i>At mNif1</i>	29.62	38.21	36.56	36.65	54.95	59.42	76.30		66.87
<i>Sc Nfs1</i>	29.73	38.55	35.75	34.31	55.94	60.47	73.51	66.87	

Figure 7- figure supplement 1- source data 1. **(A)** Multiple sequence alignment used for **Figure 7- figure supplement 1**. **(B)** Table showing the percentage sequence identity among cysteine desulfurase orthologs based on the sequence alignment shown in panel (A).