

Supplementary Table 1. Summary of model building for the yeast exocyst complex

Subunit	Length	Built region	Domain	PDB code	Modeling
Sec3	1336	611-1332	CorEx motif (640-710)	-	De novo building
			CAT-A (740-881)	-	De novo building
			CAT-B (920-1035)	-	De novo building
			CAT-C (1040-1208)	-	De novo building
			CAT-D (1213-1332)	-	De novo building
Sec5	971	1-971	CorEx motif (155-231)	-	De novo building
			CAT-A (238-331)	-	De novo building
			CAT-B (350-619)	-	De novo building
			CAT-C (623-812)	-	De novo building
			CAT-D (813-936)	-	De novo building
Sec6	805	1-805	CorEx motif (18-85)	-	De novo building
			CAT-A (92-253)	-	De novo building
			CAT-B (297-396)	-	De novo building
			CAT-C (414-609)	2FJI	Rigid docking
			CAT-D (619-725)	2FJI	Rigid docking
Sec8	1065	23-1065	CorEx motif (91-177)	-	De novo building
			CAT-A (180-335)	-	De novo building
			CAT-B (338-570)	-	De novo building
			CAT-C (588-858)	-	De novo building
			CAT-D (874-968)	-	De novo building
Sec10	871	3-867	CorEx motif (58-172)	-	De novo building
			CAT-A (191-279)	5H11	Homology modeling
			CAT-B (317-428)	5H11	Homology modeling
			CAT-C (432-696)	5H11	Homology modeling
			CAT-D (697-825)	5H11	Homology modeling
Sec15	910	42-890	CorEx motif (78-186)	-	De novo building
			CAT-A (190-445)	-	De novo building
			CAT-B (453-667)	2A2F	Homology modeling
			CAT-C (668-830)	2A2F	Homology modeling
			CAT-D (838-890)	2A2F	Homology modeling
Exo70	623	1-623	CorEx motif (5-67)	-	De novo building
			CAT-A (74-190)	2B1E	Rigid docking
			CAT-B (194-340)	2B1E	Rigid docking
			CAT-C (341-513)	2B1E	Rigid docking
			CAT-D (514-623)	2B1E	Rigid docking
Exo84	753	170-753	CorEx motif (197-276)	-	De novo building
			PH domain (346-452)	1ZC4	Homology modeling
			CAT-A (527-622)	2D2S	Rigid docking
			CAT-B (625-753)	2D2S	Rigid docking

Supplementary Table 2. Plasmids and yeast strains used in this study

Name	Description
pGV373	<i>p415TEF-TOM20-mCherry</i>
pG1831	<i>pRS416-Cit1-GFP</i>
pG1872	<i>p415TEF-TOM20-mCherry-sec3(1-1336)</i>
pG1976	<i>p415TEF-TOM20-mCherry-sec3(1-620)</i>
pG1977	<i>p415TEF-TOM20-mCherry-sec3(1-740)</i>
pG1978	<i>p415TEF-TOM20-mCherry-sec3(601-1336)</i>
pG1979	<i>p415TEF-TOM20-mCherry-sec3(710-1336)</i>
pG215	<i>Prs304(TRP)-SEC3-GFP</i>
pG1980	<i>Prs304(TRP)-sec3(Δ621-710)-GFP</i>
GY1132	<i>MAT a trp1, leu2, his3, ura3, lys2</i>
GY1143	<i>MAT a trp1, leu2, his3, ura3, lys2, Sec5-GFP::URA3</i>
GY2624	<i>Mat α leu2-3, 112, ura3-52, trp1, his3Δ200, L-A+, GAL+, sec3::Kan</i>
GY3485	<i>MAT a his3Δ1 leu2Δ0 met15Δ0 ura3Δ0 SEC3-TAP::HIS3</i>
GY3486	<i>MAT a his3Δ1 leu2Δ0 met15Δ0 ura3Δ0 SEC5-TAP::HIS3</i>
GY3487	<i>MAT a his3Δ1 leu2Δ0 met15Δ0 ura3Δ0 SEC6-TAP::HIS3</i>
GY3489	<i>MAT a his3Δ1 leu2Δ0 met15Δ0 ura3Δ0 SEC15-TAP::HIS3</i>
GY3924	<i>Mat α ura3-52, leu2-3, 112, his3Δ200, trp1 Gal+, LA+, SEC6-Protein A-HIS3::SEC6</i>
GY3933	<i>MATa his3Δ1 leu2Δ0 met15Δ0 ura3Δ0 EXO70-TAP::HIS3, SEC8-GFP::LEU2</i>
GY3937	<i>MATa his3Δ1 leu2Δ0 met15Δ0 ura3Δ0 Exo70-TAP::HIS3, Exo84-GFP::LEU2</i>
GY3948	<i>Mat α leu2-3, 112, ura3-52, trp1::SEC3-GFP-TRP1, his3Δ200, L-A+, GAL+, sec3::Kan</i>
GY3949	<i>Mat α leu2-3, 112, ura3-52, trp1::sec3(Δ621-710)-GFP-TRP1, his3Δ200, L-A+, GAL+, sec3::Kan</i>

Supplementary Note 1. Evolutionary conversation of the CorEx motif in the exocyst subunits.

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CeEXOC1 182 LTIGERHIFAPM TPOQSL GANINSMDSSENSVQLLSSIDAALTOVESVEKEDRCDDILAFVRNSIELIEEKDSL SVVERKNKORLNEE VSPVNSLQAVTDS 288
DrEXOC1 197 FAIKDAQFOFIQO SREIHDL GANIQSVLASEQVMKMMHEHDNAISEADKFPETRLDTYEDLLGHVKETMERKIGKNAMIEIANNNNIKLMKE NKVISQLD-LPHS 302
huEXOC1 180 YATSNAAEAPAK SREIQVL GANIQSIMASEKQVNLMLKLLDALKKEVDQIETLKSSEYEMLOSVEKQMDQISESNHILHLSNTNNVKLSE EFLVNHMD-LAKG 285
ScSec3 636 STEDADSMIR DLRIAET YLFPNQLLSLQKIGPNIRPYEDKVNDECHRIIPTLSLFLMEMSNFSDNIENVESQDNGLQVESANKKLLWNT DELLKRTVS-LDEI 742

CeEXOC2 197 ONEAKRSEEMHKANLYSLINCVDTLAN HQALEKGENA-D-----HFAALKN SKLKDKSKAENVFAD LKRKDDADATRNALGV VRFKFIFFLSSKIEDSMKKG 301
DrEXOC2 190 ESOKEGQLSFLKSNAGSVIDQLDTMN RDKLQEDVKL-H-----GNETLNI ETSIENSISESQKIFTD LVRKEKADSTRSLVFA SRHKFLFCLPNSVDRRAKAG 294
huEXOC2 195 NKKSEGLAYVVKGLSTFFEAQDALSA HQKLEADGTEKV-----EGSMTQK ENVLNRSANTADTLFQE LGRKDKADSTRNALNV ORKFLFNLPLNIERNIKGG 300
ScSec5 141 QDOSIHLKQVLKGNFTKYVKIKKMLDQ YKFEDEKTNE-KNQCDSPKENGINVS NKKVDEVIRTTTFKLK MDNYQKILNYQATKF ELNKFYNLPKSLKRCITND 253

CeEXOC3 1 M-----DVDVEEAA EQ AAL QRPDQL PEL KRAD KKLAVFAMRTGVQGLEG RTA AHLQTASDD TA SQGVH RERLGGPPOLKEKLE RDANARHG 104
DrEXOC3 1 MD-----LQQLLEEQARQAA KD QNM QREGQL EQY HRIA KVASVEALRTKMGQGLDG RVG KQLETQMD RE RRRMD ERLLRGVPPEYDALEV REENTRHS 109
huEXOC3 1 MQCEDSTSPFTMKETDREAVATA QR AGM QRPDQL EQY RREA KVASVEARAAATQSLQD RTG SQLHNALND KD QQSLA SKDWRGSIINTESLKD KDAVVQHS 118
ScSec6 1 M-----SSDF QQ CDL KGLSL RDI EQLL EKSVYEQLNKESDRYYGE EES KLI NLSKMS TS KOQIN NKLGNINRFAINRYDI FRATKLYE 100

CeEXOC4 56 EKGKYGESGAL DR KNHQQDVEKCLVS RDVSSK SNCRERIHN NAHTVKSLELR DDKLLWHENAQQKSVCEIMAK EE REAFS IENLISKQO QQAADT TE 168
DrEXOC4 49 EAEPKRSDLR NE SRHQQLTQVLP SQVSSE TASRERIHA ENLGVCKRL QCRDELKMMWMDAVQHKVYLEMLEQ QE RKPQ VVGYTAKRQ LHASKA TD 161
huEXOC4 47 EEAYEKCDRD DE VQHYTELTAIRT QSITER TNSRNKIKQ ENLLSCKML HCKDELRLKWIEGIEHKHVLNLLDE EN KQVQ LEQCMASKH LSATDM VS 159
ScSec8 81 K---SQIGSH QD NEHSQVFNTVAS GKAVSS MQAQEQTLN NCLKEANEK TTDKGLQELNDNNLKYTKMIDVLVN EE LQIPE IENIRKEN HQVQL ER 190

CeEXOC5 2 -----FEEB GS QMLC QFQNIINLEKQMNEEKDYQKLRHLEKNCEAIDKMKQDHTMQAVSTK VHLG QLESDQP SRAHDAHQLMQHFD FLSD 100
huEXOC5 32 RGG-----PEAFDKRRLLEEVNH QELQMD RIQRAVEKLEQOQKEAKEFAKKVQELQKSNQVAFQHPQELDEHISVATK CHLG QLEGVNTP QRAVEAQLMKYFN ELDG 143
DrEXOC5 31 LQN-----SEDFHPVALHDTFIQT KD KILQ KQSSCERLESLESLQEKESHAKKIAKIQRHQTAIDVFGQ DEKINSVAGK MHLG QLENVNTP SRSVEAQLINPMS FLAA 143
ScSec10 55 KDQDAIREAIWKQLDPKRYIRTFEST KEKLNLN ETLNRQYFSEQVATQEVISHENVIKSKDLHTLLTLPDKLDRITNVQV SPFG KLETAIKK QNYIQSVELIRRYN EYGM 174

CeEXOC6 58 GDVQSFARLQQRISHYDK--N QKVCSEHYQ-SFVDAMQE MKLKEQCO-D KEETVAIDAEIQOISQRLCQKQEI RY KLMKNAKTAMDO AVC PVLENYAKLQEQMSNRK 172
DrEXOC6 27 NNTKQIGDQLEQRIRSHDK--E ERICNLYYD-GFIDSIQE LQVRTQAO-Q HNEVHSLDTSLRQISASLIQGGNDI RA QIESNLASATEA KSC PALECYMKTFOQARNFY 141
huEXOC6 42 NAHKFMKELDACIRNHDK--E EKMCNFHHQ-GFVDAITE LKVRTDAE-K KVQVTDNRRFODAGKEVIVHTEDI RC IQQRNITTVVEK QLC PVLEMYSKLQEQMSARV 156
ScSec15 75 NQLEDPVIDELENSEDNFQGLE QLLQBSQMNDKLETSID ANIQGMQVDT SSEISKFIQLRESANELIVKQMY NN KISKISEATIL TKV RILELSSKQCELTTEBK 193

CeEXOC7 22 MASDQ---NAAEAGIARK AQ EEW ONFOKNIAKSAO ROGIEQV EKFDORLSLEKNVLEPHISNGKLOKKNHNIQRL NTIDATLOFYGKTSIVENA NS 121
DrEXOC7 1 MN---NLDSSLAQHNK EK ATN ALLKORVDKYHD STQMSSTIFEFKRLGNLEQITLPIVYOETEQLQKRQONLEAT NCLESVLSHYDVSQVQCQL HQ 99
huEXOC7 1 MIPPOEASARRREIEDK QO EET SFIRDSLEKSDQ TKNMVSISSEFESRLMKLENSITPVHKQENLQRLQENVEKT SCIDHVISYHYHVASDTEKI RE 103
ScExo70 1 MP-----AE DI EAD LVLSQELQTSK TFEINKS KIAAATSNQSQSLFPELLARNVLTTLQRIEST NSVASVKDLANEASKYETI QK 90

CeEXOC-8 13 NFSPSD IRDRLEKVKIGDETRK RHLRSEMGALNHAQOET KNNVFR VQOFIDASK S-HLEREYO THS I QKQV ENLIMMTGD-----DKS 105
DrExo84A 8 NFSVEK TKDLTRRCVGG--GSD QQRKKEIEAYNETTAAT KOTCKK VMFQITAK SRNLESMYO SHI I QRNI ATMTDGKTS-----SHL 97
huEXOC8 21 GFEARL VKQLSQSQSDG---DRD QEHRQRIQALAEETAQN KRNVYQ YRQFIETAR S-YLESEMYO SHL T QKSS ESTPITLL-PAAAAAGAAAASGGEQVGGAGGRDHL 133
ScExo84 181 HFNKAD VHKIKGNAS---AIT DKFTSNITDLSIQVQEE KLNINK VNIIMTVNN N-VAMLEKR RAN N LNEV DQCTRIAEKRIQEQDQDQERQGNFNNEVSHSNSPA 293

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Sequences of full-length exocyst subunits from *Saccharomyces cerevisiae* (“Sc”), *C. elegans* (“Ce”), *Human* (“Hu”) and *Drosophila* (“Dr”) are aligned based on their secondary structures using PROMALS3D¹. The CorEx motifs are indicated by black lines under each alignment. Same residues in the alignment are colored in red and the conserved ones in orange. The gray background in each sequence shows predicted α -helices in the alignment.

Reference

1. Pei, J., Kim, B.H. & Grishin, N.V. PROMALS3D: a tool for multiple protein sequence and structure alignments. *Nucleic Acids Res.* **36**, 2295–2300 (2008).