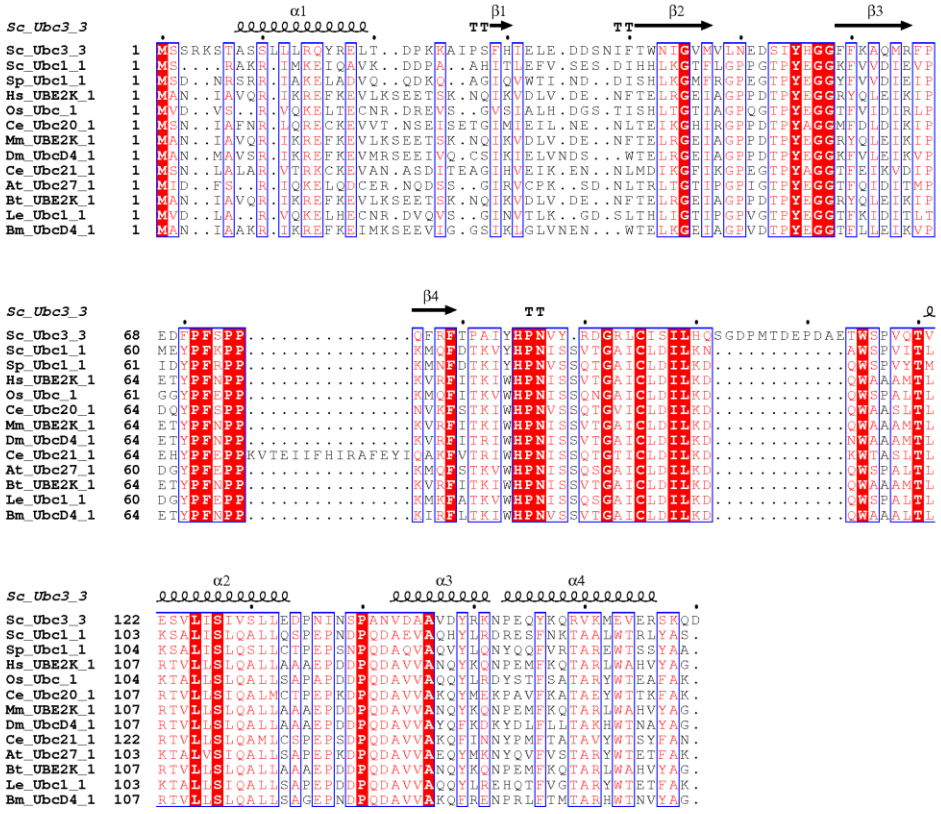
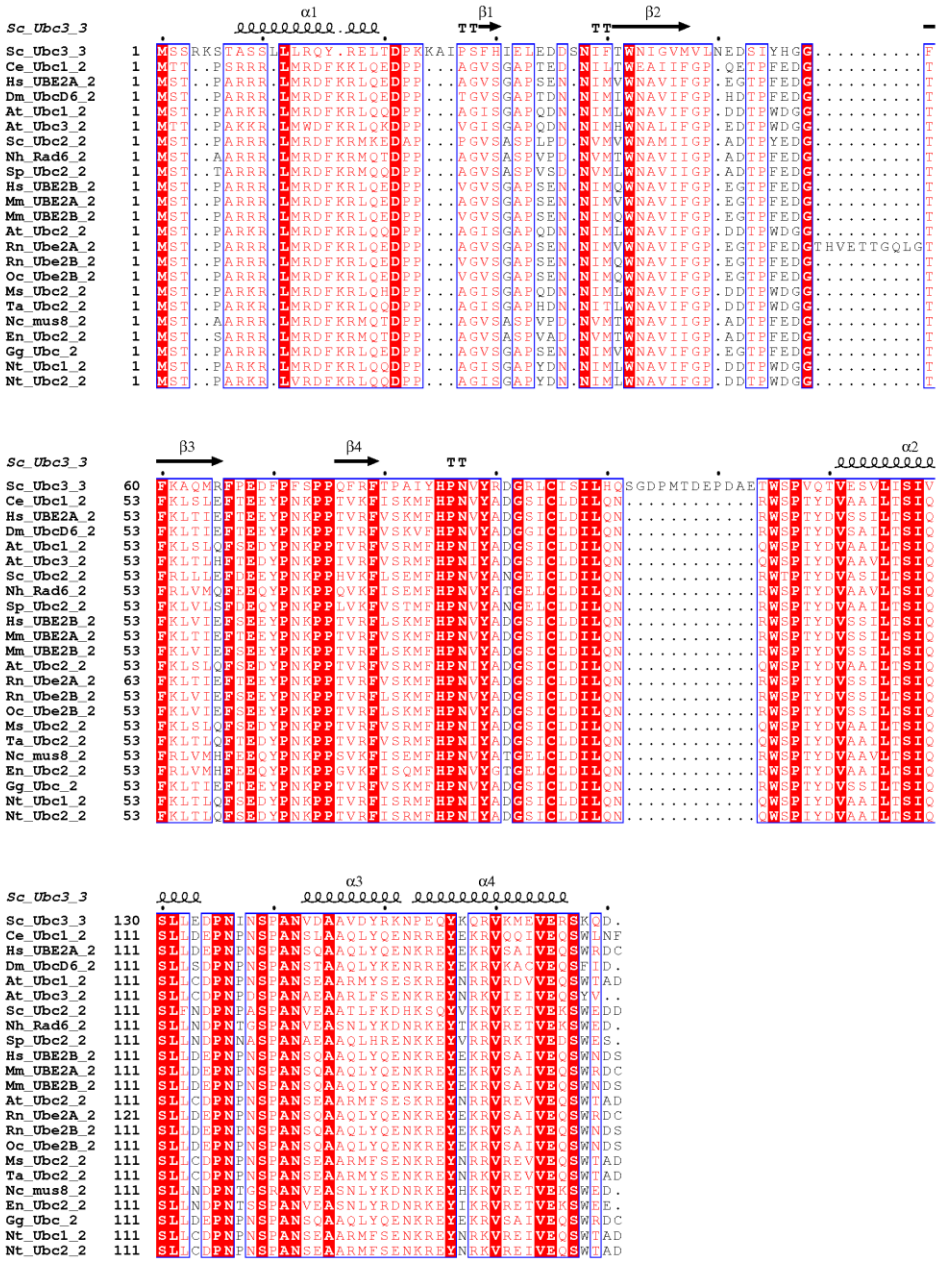
Figure 1. Intra-family multiple sequence alignment. In each alignment the families are indicated by '_x'.

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β1
β2
β3
β4
α2
α3
α4
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Ce_Ubc1_2 1 MTT..PSRRRLMRDFKKLQEDPP...AGVSGAPTED.NILTWEAIIFGP.QETPFEDG.....T
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At_Ubc1_2 1 MST..PARKRLMRDFKRLQQDPP...AGISGAPQDN.NIMLWNAVIFGP.DDTPWDGG.....T
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Sc_Ubc2_2 1 MST..PARRRLMRDFKRMKEDAP...PGVSGASPLPD.NVMVWNAMIIGP.ADTPYEDG.....T
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Sp_Ubc2_2 1 MST..TARRRLMRDFKRMQQDPP...AGVSGASPVSD.NVMLWNAVIIGP.ADTPFEDG.....T
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Hs_UBE2A_2 53 FKLTIEFTEEYPNKPPTVRFVSKMEHPNVYADGSICLDILQN.....RWSPTYDVSSILTSIQ
Dm_UbcD6_2 53 FKLTIEFTEEYPNKPPTVRFVSKVEHPNVYADGGICLDILQN.....RWSPTYDVSAILTSIQ
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Dm_UbcD6_2 111 SLLSDPNPNSPANSTAAQLYKENRREYEKRVKACVESQSFID.
At_Ubc1_2 111 SLLCDPNPNSPANSEAARMYSESKREYNRRVRDVVESQSWTAD
At_Ubc3_2 111 SLLCDPNPDSPANAEAARLFSENKREYNRKVIETVESQSYV..
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Oc_Ube2B_2 111 SLLDEPNPNSPANSQAAQLYQENKREYEKRVSAIVEQSWNDS
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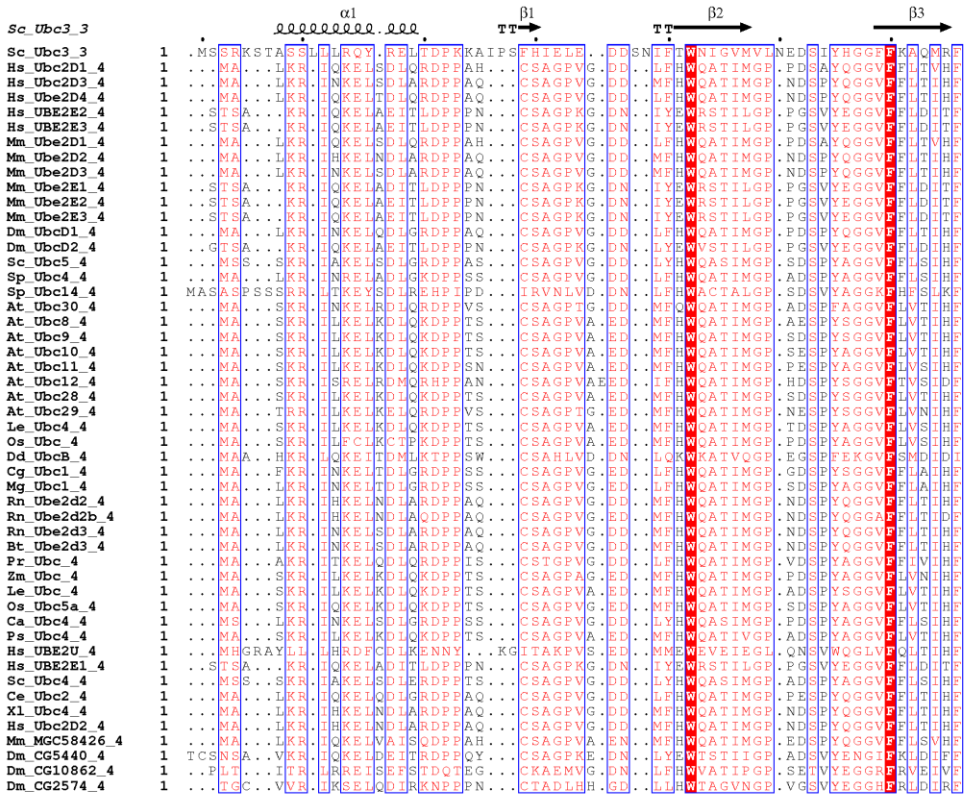
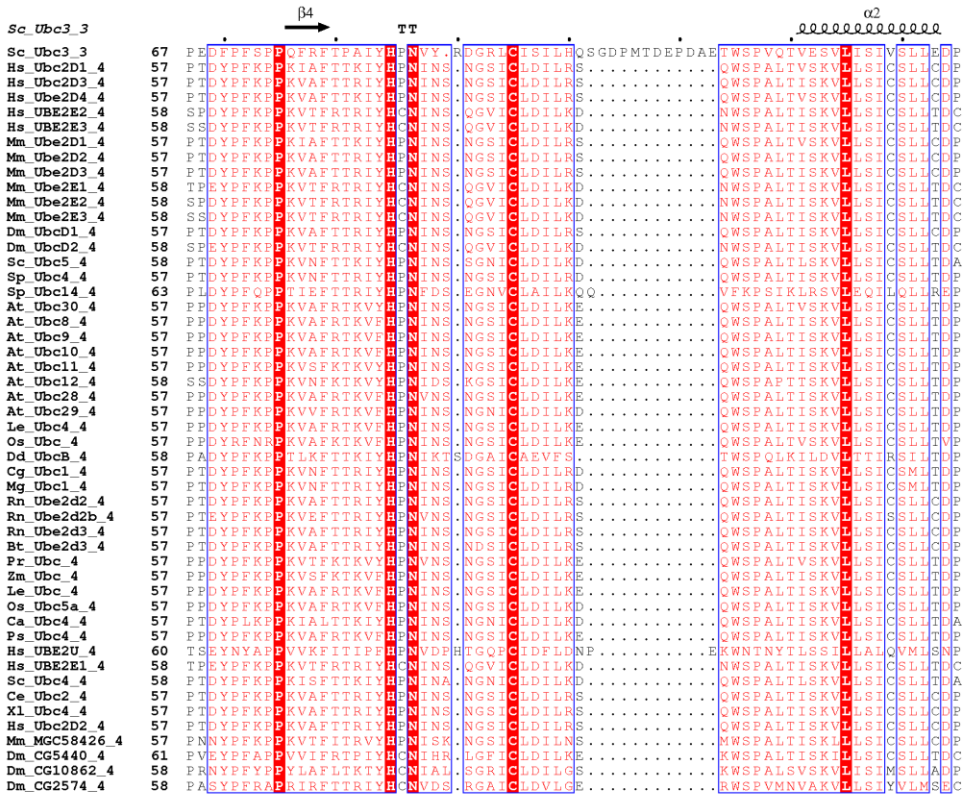

α1
β1
β2
β3
Sc_Ubc3_3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TT
α2
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Os_Ubc5a_4 57 PPDYPFKPPKVAFRTKVFHPNINS.NGSICLDILKD.....QWSPALTISKVLLSICSLLTDP
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Ps_Ubc4_4 57 PPDYPFKPPKVAFRTKVFHPNINS.NGSICLDILKE.....QWSPALTISKVLLSICSLLTDP
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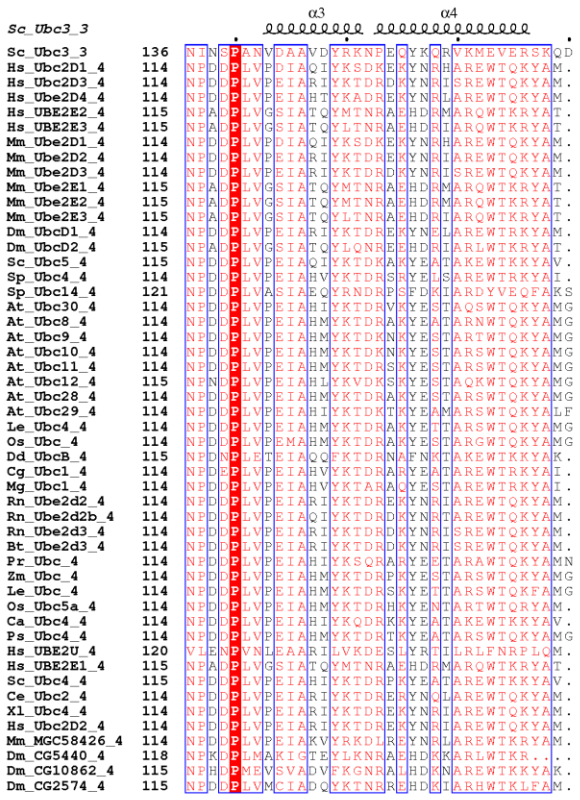
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α4
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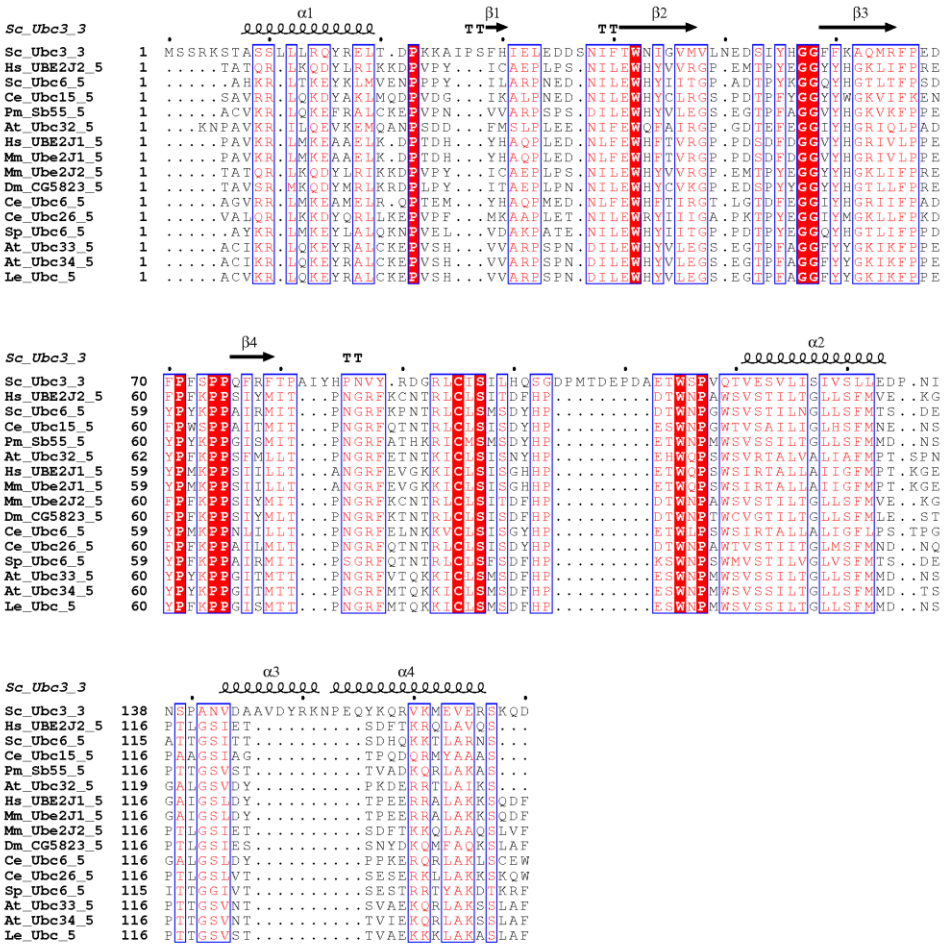
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β1
β2
β3
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Dm_CG5823_5 1 .....TAVSR.MKQDYMRLKRDPLPY...ITAEPLPN.NILEWHYCVKGP.EDSPYYGGYYHGTLLFPRE
Ce_Ubc6_5 1 .....AGVRR.LMKEAMELR.QPTEM...YHAQPMED.NLFEWHFTIRGT.LGTDFEGGIYHGRIIFPAD
Ce_Ubc26_5 1 .....VALQR.LKKDYQRLLKEPVPF...MKAAPLET.NILEWRYIIIGA.PKTPYEGGIYMGKLLFPKD
Sp_Ubc6_5 1 ......AYKR.LMKEYLALQKNPVEL...VDAKPATE.NILEWHYIITGP.PDTPYEGGQYHGTLIFPPD
At_Ubc33_5 1 .....ACIKR.LQKEYRALCKEPVSH...VVARPSPN.DILEWHYVLEGS.EGTPFAGGFYYGKIKFPPE
At_Ubc34_5 1 .....ACIKR.LQKEYRALCKEPVSH...VVARPSPN.DILEWHYVLEGS.EGTPFAGGFYYGKIKFPPE
Le_Ubc_5 1 .....ACVKR.LQKEYRALCKEPVSH...VVARPSPN.DILEWHYVLEGS.EGTPFAGGFYYGKIKFPPE
β4
α2
Sc_Ubc3_3 Sc_Ubc3_3 70 FPPSPPQFRFTPAIYHPNVY.RDGRICLSILHQSGDPMTDEPDAETWSPVQTVESVLISIVSLLEDP.NI
Hs_UBE2J2_5 60 FPFKPPSIYMIT...PNGRFKCNTRICLSITDFHP.....DTWNPAWSVSTILTGLLSFMVE..KG
Sc_Ubc6_5 59 YPYKPPAIRMIT...PNGRFKPNTRICLSMSDYHP.....DTWNPGWSVSTILNGLLSFMTS..DE
Ce_Ubc15_5 60 FPWSPPAITMIT...PNGRFQTNTRICLSISDYHP.....ESWNPGWTVSAILIGLHSFMNE..NS
Pm_Sb55_5 60 YPYKPPGISMIT...PNGRFATHKRICMSMSDYHP.....ETWNPMWSVSSILTGLLSFMMD..NS
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Dm_CG5823_5 60 FPFKPPSIYMLT...PNGRFKTNTRICLSISDFHP.....DTWNPTWCVGTILTGLLSFMLE..ST
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Sp_Ubc6_5 59 YPFKPPAIRMIT...PSGRFQTNTRICLSFSDFHP.....KSWNPSWMVSTILVGLVSFMTS..DE
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At_Ubc34_5 60 YPYKPPGITMTT...PNGRFMTQKKICLSMSDFHP.....ESWNPMWSVSSILTGLLSFMMD..TS
Le_Ubc_5 60 YPFKPPGISMTT...PNGRFMTQKKICMSMSDFHP.....ESWNPMWSVSSILTGLLSFMMD..NS
α3
α4
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Sc_Ubc6_5 115 ATTGSITT.....SDHQKKTLARNS...
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Pm_Sb55_5 116 PTTGSVST.....TVADKQRLAKAS...
At_Ubc32_5 119 GALGSVDY.....PKDERRTLAIKS...
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Mm_Ube2J1_5 116 GAIGSLDY.....TPEERRALAKKSQDF
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Dm_CG5823_5 116 PTLGSIES.....SNYDKQMFAQKSLAF
Ce_Ubc6_5 116 GALGSLDY.....PPKERQRLAKLSCEW
Ce_Ubc26_5 116 PTLGSLVT.....SESERKLLAKKSKQW
Sp_Ubc6_5 115 ITTGGIVT.....SESTRRTYAKDTKRF
At_Ubc33_5 116 PTTGSVNT.....SVAEKQRLAKSSLAF
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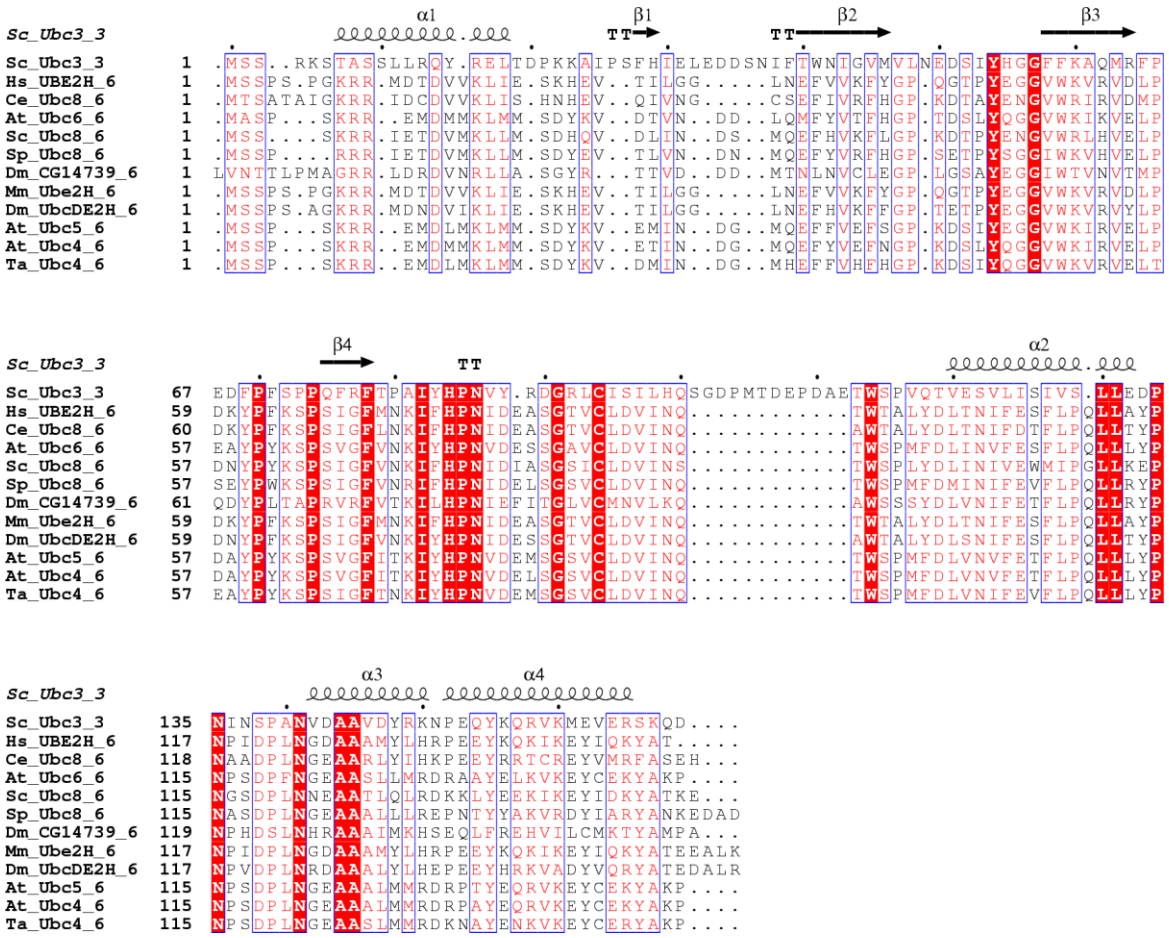
Le_Ubc_5 116 PTTGSVST.....TVAEKKKLAKASLAF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β1
β2
β3
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Ce_Ubc8_6 1 .MTSATAIGKRR.IDCDVVKLIS.HNHEV..QIVNG.....CSEFIVRFHGP.KDTAYENGVWRIRVDMP
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Sc_Ubc8_6 1 .MSS....SKRR.IETDVMKLLM.SDHQV..DLIN..DS..MQEFHVKFLGP.KDTPYENGVWRLHVELP
Sp_Ubc8_6 1 .MSSP....RRR.IETDVMKLLM.SDYEV..TLVN..DN..MQEFYVRFHGP.SETPYSGGIWKVHVELP
Dm_CG14739_6 1 LVNTTLPMAGRR.LDRDVNRLLA.SGYR...TTVD..DD..MTNLNVCLEGP.LGSAYEGGIWTVNVTMP
Mm_Ube2H_6 1 .MSSPS.PGKRR.MDTDVVKLIE.SKHEV..TILGG.....LNEFVVKFYGP.QGTPYEGGVWKVRVDLP
Dm_UbcDE2H_6 1 .MSSPS.AGKRR.MDNDVIKLIE.SKHEV..TILGG.....LNEFHVKFFGP.TETPYEGGVWKVRVYLP
At_Ubc5_6 1 .MSSP...SKRR..EMDLMKLMM.SDYKV..EMIN..DG..MQEFFVEFSGP.KDSIYEGGVWKIRVELP
At_Ubc4_6 1 .MSSP...SKRR..EMDMMKLMM.SDYKV..ETIN..DG..MQEFYVEFNGP.KDSIYQGGVWKIRVELP
Ta_Ubc4_6 1 .MSSP...SKRR..EMDLMKLMM.SDYKV..DMIN..DG..MHEFFVHFHGP.KDSIYQGGVWKVRVELT
Sc_Ubc3_3
β4
α2
Sc_Ubc3_3 67 EDFPFSPPQFRFTPAIYHPNVY.RDGRLCISILHQSGDPMTDEPDAETWSPVQTVESVLISIVS.LLEDP
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Ce_Ubc8_6 60 DKYPFKSPSIGFLNKIFHPNIDEASGTVCLDVINQ.....AWTALYDLTNIFDTFLPQLLTYP
At_Ubc6_6 57 EAYPYKSPSVGFVNKIYHPNVDESSGAVCLDVINQ.....TWSPMFDLINVFESFLPQLLLYP
Sc_Ubc8_6 57 DNYPYKSPSIGFVNKIFHPNIDIASGSICLDVINS.....TWSPLYDLINIVEWMIPGLLKEP
Sp_Ubc8_6 57 SEYPWKSPSIGFVNRIFHPNIDELSGSVCLDVINQ.....TWSPMFDMINIFEVFLPQLLRYP
Dm_CG14739_6 61 QDYPLTAPRVRFVTKILHPNIFFITGLVCMNVLKQ.....AWSSSYDLVNIFETFLPQLLRYP
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Dm_UbcDE2H_6 59 DNYPFKSPSIGFVNKIYHPNIDESSGTVCLDVINQ.....AWTALYDLSNIFESFLPQLLTYP
At_Ubc5_6 57 DAYPYKSPSVGFITKIYHPNVDEMSGSVCLDVINQ.....TWSPMFDLVNVFETFLPQLLLYP
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Ta_Ubc4_6 57 EAYPYKSPSIGFTNKIYHPNVDEMSGSVCLDVINQ.....TWSPMFDLVNIFEVFLPQLLLYP
Sc_Ubc3_3
α3
α4
Sc_Ubc3_3 135 NINSPANVDAAVDYRKNPEQYKQRVKMEVERSKQD....
Hs_UBE2H_6 117 NPIDPLNGDAAAMYLHRPEEYKQKIKEYIQKYAT.....
Ce_Ubc8_6 118 NAADPLNGEAARLYIHKPEEYRRTCREYVMRFASEH...
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Sc_Ubc8_6 115 NGSDPLNNEAATLQLRDKKLYEEKIKEYIDKYATKE...
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Dm_CG14739_6 119 NPHDSLNHRAAAIMKHSEQLFREHVILCMKTYAMPA...
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At_Ubc5_6 115 NPSDPLNGEAAALMMRDRPTYEQRVKEYCEKYAKP....
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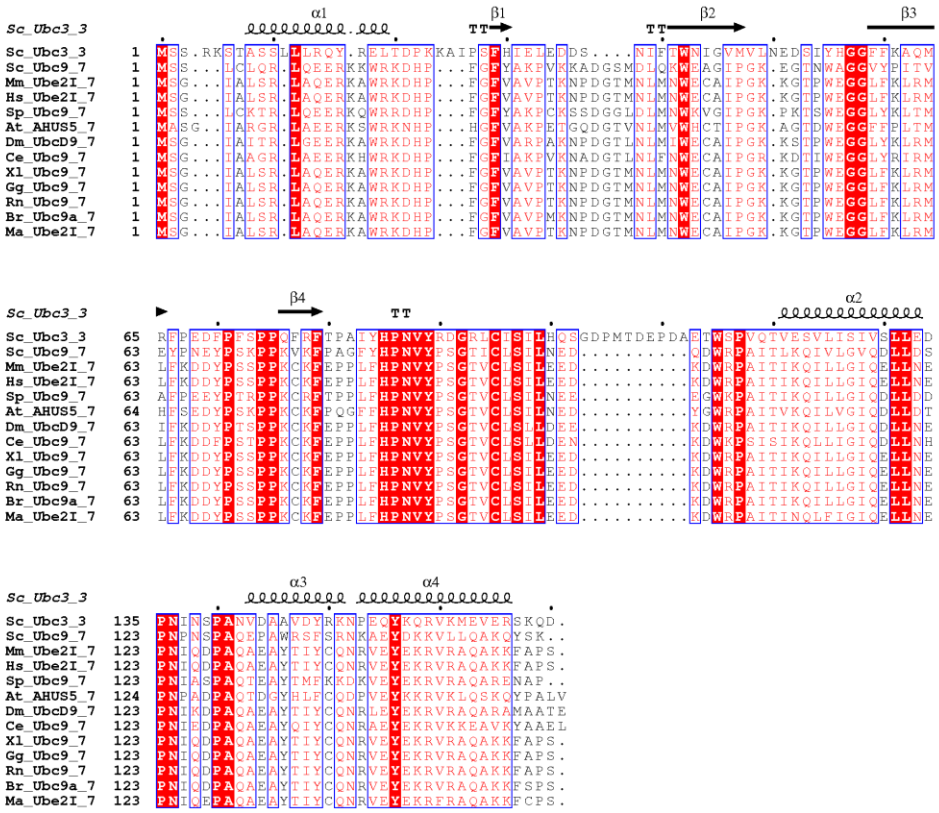
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β2
β3
β4
α2
α3
α4
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Hs_Ube2I_7 1 MSG...IALSR.LAQERKAWRKDHP...FGFVAVPTKNPDGTMNLMNWECAIPGK.KGTPWEGGLFKLRM
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Hs_Ube2I_7 123 PNIQDPAQAEAYTIYCQNRVEYEKRVRAQAKKFAPS.
Sp_Ubc9_7 123 PNIASPAQTEAYTMFKKDKVEYEKRVRAQARENAP..
At_AHUS5_7 124 PNPADPAQTDGYHLFCQDPVEYKKRVKLQSKQYPALV
Dm_UbcD9_7 123 PNIKDPAQAEAYTIYCQNRLEYEKRVRAQARAMAATE
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Rn_Ubc9_7 123 PNIQDPAQAEAYTIYCQNRVEYEKRVRAQAKKFAPS.
Br_Ubc9a_7 123 PNIQDPAQAEAYTIYCQNRVEYEKRVRAQAKKFSPS.
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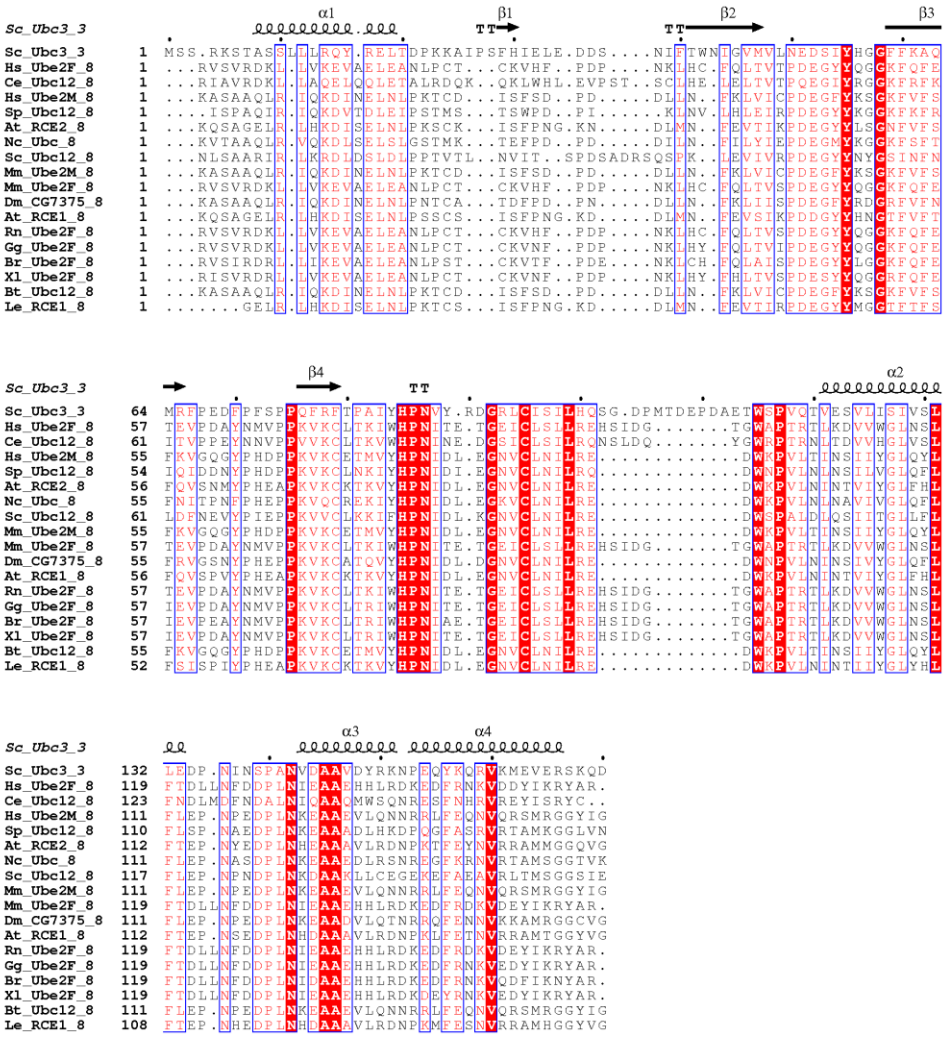
α1
β1
β2
β3
β4
α2
α3
α4
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At_RCE2_8 56 FQVSNMYPHEAPKVKCKTKVYHPNIDL.EGNVCLNILRE.............DWKPVLNINTVIYGLFHL
Nc_Ubc_8 55 FNITPNFFHEPPKVQCREKIYHPNIDL.EGKVCLNILRE.............DWKPVLNLNAVIVGLQFL
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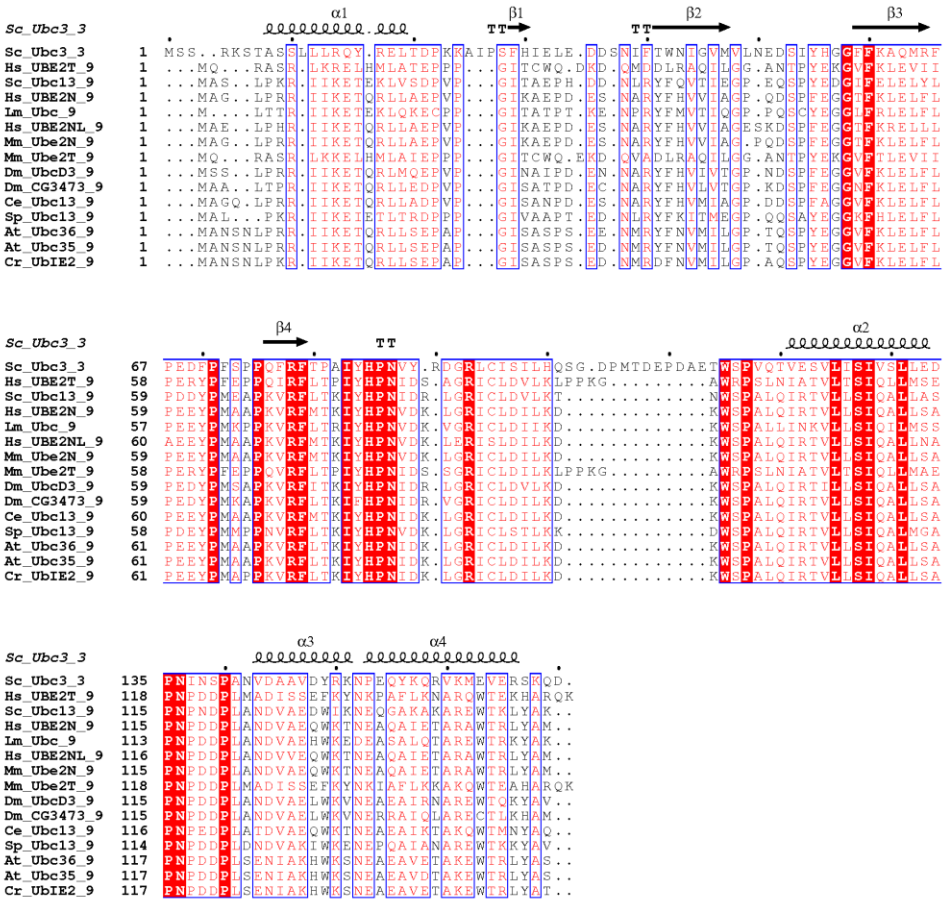
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β2
β3
β4
α2
α3
α4
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Hs_UBE2N_9 1 ...MAG..LPRR.IIKETQRLLAEPVPF...GIKAEPD.ES.NARYFHVVIAGP.QDSPFEGGTFKLELFL
Lm_Ubc_9 1 ...M....LTTR.IIKETEKLQKECPPF...GITATPT.KE.NPRYFMVTIQGP.PQSCYEGGLFRLELFL
Hs_UBE2NL_9 1 ...MAE..LPHR.IIKETQRLLAEPVPF...GIKAEPD.ES.NARYFHVVIAGESKDSPFEGGTFKRELLL
Mm_Ube2N_9 1 ...MAG..LPRR.IIKETQRLLAEPVPF...GIKAEPD.ES.NARYFHVVIAG.PQDSPFEGGTFKLELFL
Mm_Ube2T_9 1 ...MQ...RASR.LKKELHMLAIEPPPF...GITCWQ.EKD.QVADLRAQILGG.ANTPYEKGVFTLEVII
Dm_UbcD3_9 1 ...MSS..LPRR.IIKETQRLMQEPVPF...GINAIPD.EN.NARYFHVIVTGP.NDSPFEGGVFKLELFL
Dm_CG3473_9 1 ...MAA..LTPR.IIKETQRLLEDPVPF...GISATPD.EC.NARYFHVLVTGP.KDSPFEGGNFKLELFL
Ce_Ubc13_9 1 ...MAGQ.LPRR.IIKETQRLLADPVPF...GISANPD.ES.NARYFHVMIAGP.DDSPFAGGVFKLELFL
Sp_Ubc13_9 1 ...MAL...PKR.IIKEIETLTRDPPPF...GIVAAPT.ED.NLRYFKITMEGP.QQSAYEGGKFHLELFL
At_Ubc36_9 1 ...MANSNLPRR.IIKETQRLLSEPAPF...GISASPS.EE.NMRYFNVMILGP.TQSPYEGGVFKLELFL
At_Ubc35_9 1 ...MANSNLPRR.IIKETQRLLSEPAPF...GISASPS.ED.NMRYFNVMILGP.TQSPYEGGVFKLELFL
Cr_UbIE2_9 1 ...MANSNLPKR.IIKETQRLLSEPAPF...GISASPS.ED.NMRDFNVMILGP.AQSPYEGGVFKLELFL
Sc_Ubc3_3
Sc_Ubc3_3 67 PEDFPFSPPQFRFTPAIYHPNVY.RDGRLCISILHQSG.DPMTDEPDAETWSPVQTVESVLLSIVSLLED
Hs_UBE2T_9 58 PERYPFEPPQIRFLTPIYHPNID.SAGRICLDVLKLPPKG.....AWRPSLNIATVLTSIQLLMSE
Sc_Ubc13_9 59 PDDYPMEAPKVRFLTKIYHPNID.RLGRICLDVLKT.....NWSPALQIRTVLLSIQALLAS
Hs_UBE2N_9 59 PEEYPMAAPKVRFMTKIYHPNVD.KLGRICLDILKD.....KWSPALQIRTVLLSIQALLSA
Lm_Ubc_9 57 PEEYPMKPPKVRFLTRIYHPNVD.KVGRICLDIIKD.....KWSPALLINKVLLSIQILMSS
Hs_UBE2NL_9 60 AEEYPMAAPKVRFMTKIYHPNVD.KLGRISLDILKD.....KWSPALQIRTVLLSIQALLNA
Mm_Ube2N_9 59 PEEYPMAAPKVRFMTKIYHPNVD.KLGRICLDILKD.....KWSPALQIRTVLLSIQALLSA
Mm_Ube2T_9 58 PERYPFEPPQVRFLTPIYHPNID.SSGRICLDILKLPPKG.....AWRPSLNIATVLTSIQLLMAE
Dm_UbcD3_9 59 PEDYPMSAPKVRFITKIYHPNID.RLGRICLDVLKD.....KWSPALQIRTILLSIQALLSA
Dm_CG3473_9 59 PEDYPMKAPKVRFLTKIYHPNID.RVGRICLDILKD.....KWSPALQIRTVLLSIQALLSA
Ce_Ubc13_9 60 PEEYPMAAPKVRFMTKIYHPNID.KLGRICLDILKD.....KWSPALQIRTVLLSIQALLSA
Sp_Ubc13_9 58 PDEYPMMPPNVRFLTKIYHPNVD.KLGRICLSTLKK.....DWSPALQIRTVLLSIQALLMGA
At_Ubc36_9 61 PEEYPMAAPKVRFLTKIYHPNID.KLGRICLDILKD.....KWSPALQIRTVLLSIQALLSA
At_Ubc35_9 61 PEEYPMAAPKVRFLTKIYHPNID.KLGRICLDILKD.....KWSPALQIRTVLLSIQALLSA
Cr_UbIE2_9 61 PEEYPMAAPKVRFLTKIYHPNID.KLGRICLDILKD.....KWSPALQIRTVLLSIQALLSA
Sc_Ubc3_3
Sc_Ubc3_3 135 PNINSPANVDAAVDYRKNPEQYKQRVKMEVERSKQD.
Hs_UBE2T_9 118 PNPDDPLMADISSEFKYNKPAFLKNARQWTEKHARQK
Sc_Ubc13_9 115 PNPNDPLANDVAEDWIKNEQGAKAKARETWTKLYAK..
Hs_UBE2N_9 115 PNPDDPLANDVAEQWKTNEAQAIETARAWTRLYAM..
Lm_Ubc_9 113 PNPDDPLANDVAEHWKEDEASALQTARETWTRKYAK..
Hs_UBE2NL_9 116 PNPDDPLANDVVEQWKTNEAQAIETARAWTRLYAM..
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Dm_UbcD3_9 115 PNPDDPLANDVAELWKVNEAEAIRNARETQKYAV..
Dm_CG3473_9 115 PNPDDPLANDVAELWKVNERRAIQLARECTLKHAM..
Ce_Ubc13_9 116 PNPEDPLATDVAEQWKTNEAEAIKTAKQWTMNYAQ..
Sp_Ubc13_9 114 PNPDDPLDNDVAKIWKENEPQAIANAREWTKKYAV..
At_Ubc36_9 117 PNPDDPLSENIAKHWKSNEAEAVETAKEWTRLYAS..
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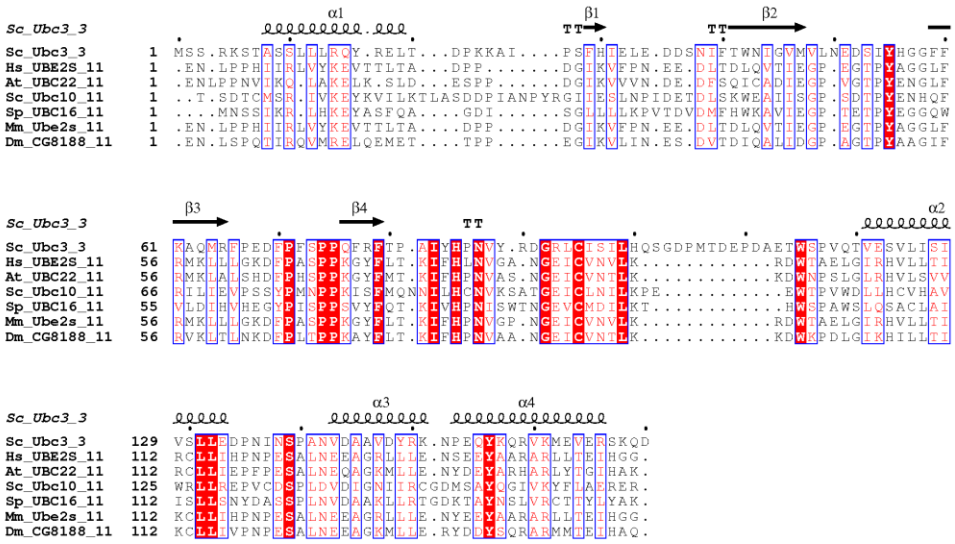
Cr_UbIE2_9 117 PNPDDPLSENIAKHWKSNEAEAVETAKEWTRLYAT..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At_UBC22_11 1 .ENLPPNVIKQ.LAKELK.SLD...ESPP......DGIKVVVN.DE.DFSQICADIEGP.VGTPYYENGLF
Sc_Ubc10_11 1 ..T.SDTCMSR.IVKEYKVILKTLASDDPIANPYRGIIESLNPIDETDLSKWEAIISGP.SDTPYYENHQF
Sp_UBC16_11 1 ....MNSSIKR.LHKEYASFQA....GDI.......SGLLLLKPVTDVDMFHWKAVIEGP.TETPYYEGGQW
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Dm_CG8188_11 1 .EN.LSPQTIRQVMRELQEMET....TPP......EGIKVLIN.ES.DVTDIQALIDGP.AGTPYYAAGIF
Sc_Ubc3_3
Sc_Ubc3_3 61 KAQMRFPEDFPFSPPQFRSTP.AIYHPNVY.RDGRLCISILHQSGDPMTDEPDAETWSPVQTVESVLISI
Hs_UBE2S_11 56 RMKLLLGKDFPASPPKGYFLT.KIFHLNVGA.NGEICVNVLK.....RDWTAELGIRHVLLTI
At_UBC22_11 56 RMKLALSHDFPHSPPKGYFMT.KIFHPNVAS.NGEICVNTLK.....KDWNPSLGLRHVLSVV
Sc_Ubc10_11 66 RILIEVPSSYPMNPPKISFMQNNILHCNVKSAIGEICLNILKPE.....EWTPVWDLLHCVHAV
Sp_UBC16_11 55 VLDIHVHEGYPISPPSVYFQT.KIVHPNISWTNGEVCMDILKT.....HWSPAWSLQSACLAI
Mm_Ube2s_11 56 RMKLLLGKDFPASPPKGYFLT.KIFHPNVGP.NGEICVNVLK.....RDWTAELGIRHVLLTI
Dm_CG8188_11 56 RVKLTLNKDFPLLPPKAYFLT.KIFHPNVAA.NGEICVNTLK.....KDWKPDLGIKHILLTI
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Sc_Ubc3_3 129 VSLLEDPNINSPANVDAAVDYRK.NPEIYKQRVKMEVERSKQD
Hs_UBE2S_11 112 RCLLIHPNPESALNEEAGRLLLE.NSEEYAARARLLTEIHGG.
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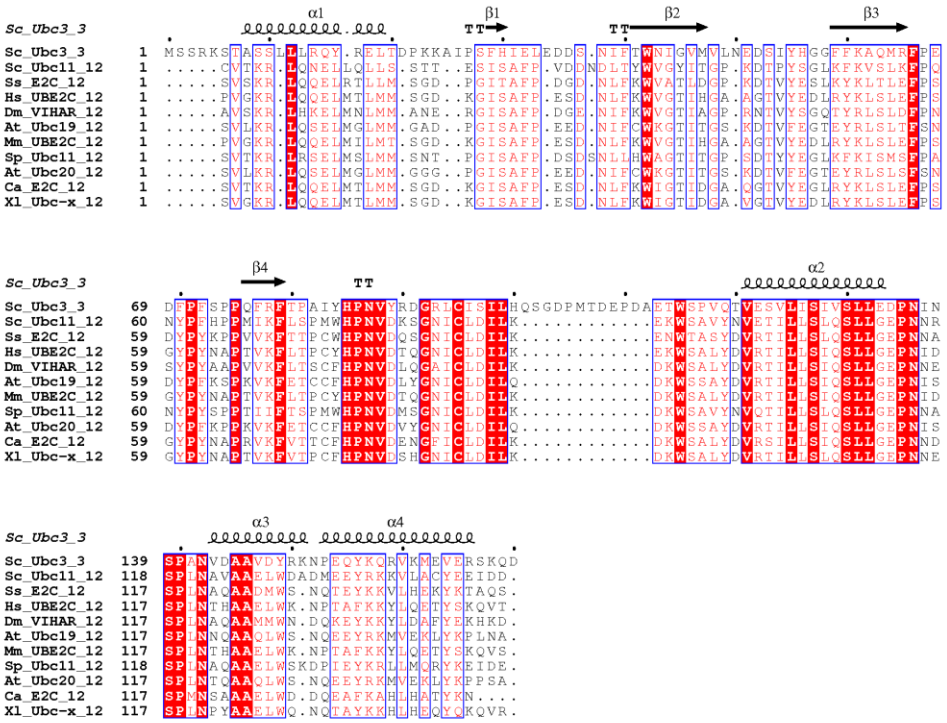
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β1
β2
β3
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Ss_E2C_12 1 .....SVSKR.LQQELRTLLM.SGD..PGITAFP.DGD.NLFKWVATLDGP.KDTVYESLKYKLTLEFPS
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Sp_Ubc11_12 1 .....SVTKR.LRSELMSLMM.SNT..PGISAFP.DSDSNLLHWAGTITGP.SDTYYEGLKFKISMSFPA
At_Ubc20_12 1 .....SVLKR.LQSELMGLMM.GGG..PGISAFP.EED.NIFCWKGTITGS.KDTVFEGTEYRLSLSFSN
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Xl_Ubc-x_12 1 .....SVGKR.LQQELMTLMM.SGD..KGISAFP.ESD.NLFKWIGTIDGA.VGTVYEDLRYKLSLEFPS
β4
α2
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Dm_VIHAR_12 59 SYPYAAPVVKFLTSCFHPNVDLQGAICLDILK.....DEWSALYDVRTILLSICSLLGEPNNE
At_Ubc19_12 59 DYPFKSPKVKFETCCFHPNVDLYGNICLDILQ.....DEWSSAYDVRTILLSICSLLGEPNIS
Mm_UBE2C_12 59 GYPYNAPTVKFLTPCYHPNVDTQGNICLDILK.....DEWSALYDVRTILLSICSLLGEPNID
Sp_Ubc11_12 60 NYPYSPPTIIFTSPMWHPNVDMSGNICLDILK.....DEWSAVYNVQTILLSLQSLLGEPNNA
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Ca_E2C_12 59 GYPYNAPRVKFVTTCFHPNVDENGFICLDILK.....DEWSALYDVRSILLSICSLLGEPNND
Xl_Ubc-x_12 59 GYPYNAPTVKFVTPCFHPNVDSHGNICLDILK.....DEWSALYDVRTILLSLQSLLGEPNNE
α3
α4
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Sc_Ubc11_12 118 SPPNAVAAELWDADMEEYRKKVLACYEEIDD.
Ss_E2C_12 117 SPPNAQAADMWS.NQTEYKKVLHEKYKTAQS.
Hs_UBE2C_12 117 SPPNTPAAELWK.NPTAFKKYLQETYSKQVT.
Dm_VIHAR_12 117 SPPNAQAAMMWN.DQKEYKKYLDAFYEKHKD.
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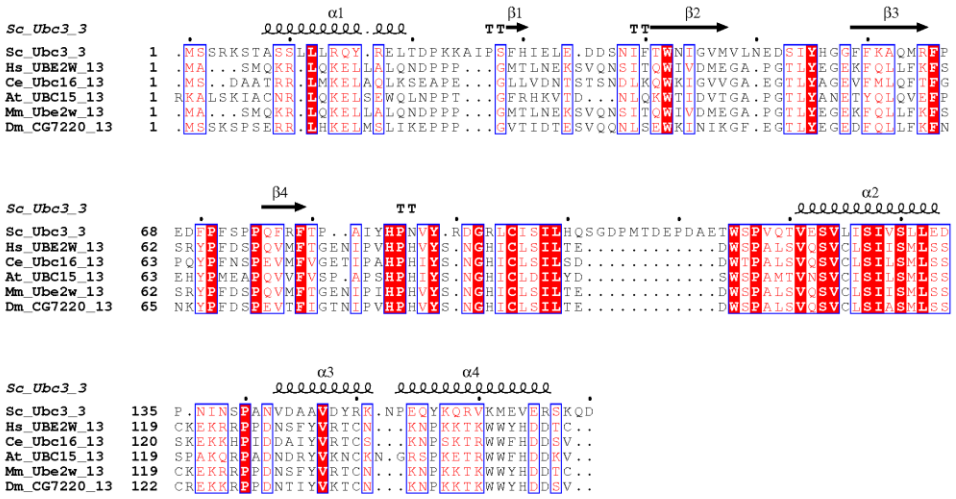
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β1
β2
β3
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β4
α2
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Hs_UBE2W_13 62 SRYPFDSPQVMFTGENIPVHPHYYS.NGHICLSILTE.....DWSPALSVQSVCISILSSLSS
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At_UBC15_13 63 EHYPMEAPQVVFVSP.APSHPHIYS.NGHICLDILYD.....SWSPAMTVNSVCISILSSLSS
Mm_Ube2w_13 62 SRYPFDSPQVMFTGENIPIHPHVYS.NGHICLSILTE.....DWSPALSVQSVCISILSSLSS
Dm_CG7220_13 65 NKYPFDSPEVTFLGTNIPVHPHVYS.NGHICLSILTE.....DWSPALSVQSVCLSIASMLSS
α3
α4
Sc_Ubc3_3 135 P.NINSPANVDAAVDYRK.NPEQYKQRVKMEVERSKQD
Hs_UBE2W_13 119 CKEKRRPPDNSFYVRTCN...KNPKKTKWWYHDDTC..
Ce_Ubc16_13 120 SKEKKHPIDDAIYVRTCS...KNPSKTRWWFHDDSV..
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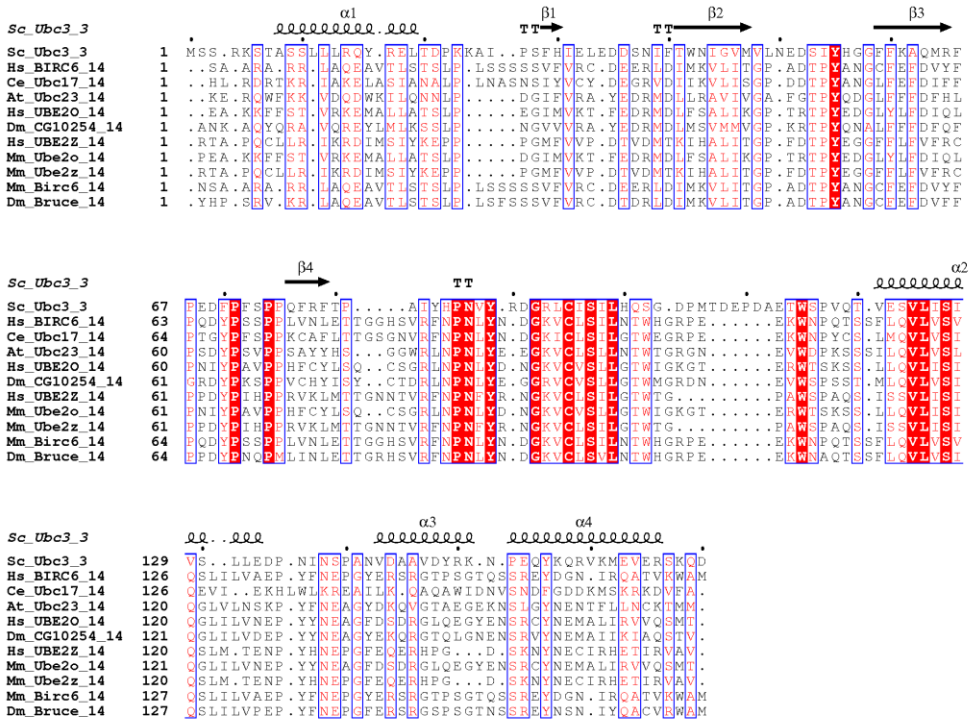
α1
β1
β2
β3
Sc_Ubc3_3
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Ce_Ubc17_14 1 ..HL.RDRTKR.IAKELASIANALPF.LNASNSIYVCY.DEGRVDIIKVLISGP.DDTPYANGLFEFDIFF
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Hs_UBE2O_14 1 ..EA.KKFFST.VRKEMALLATSLP.....EGIMVKT.FEDRMDLFSALIKGP.TRTPYEDGLYLFDIQL
Dm_CG10254_14 1 .ANK.AQYQRA.VQREYLMLKSSLP.....NGVVVRA.YEDRMDIMSVMMVGP.KRTPYQNALFFFDFQF
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Dm_Bruce_14 1 .YHP.SRV.KR.LAQEAVTLSTSLP.LSFSSSVFVRC.DIDRLDIMKVLITGP.ADTPYANGCFEFDVFF
β4
α2
Sc_Ubc3_3
Sc_Ubc3_3 67 TEDFPTSPPQFRFTP.....AIYRPNVYRDGRICISILHQSG.DPMTDEPDAETWSPVQT.VESVLISI
Hs_BIRC6_14 63 PQDYPSSPPLVNLETTGGHSVRFNPNLYNDGKVCLSILNTWHGRPE.....EKWNPQTSSFLQVLVSV
Ce_Ubc17_14 64 PTGYPFSPPKCAFLTTGSGNVRFNPNLYNDGKICLSILGTWEGRPE.....EKWNPYCS.LMQVLVSI
At_Ubc23_14 60 FSDYPSVPPSAYYHS...GGWRLNPNLYEEGKVCLSLLNTWTGRGN.....EVWDPKSSSILQVLVSL
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Mm_Ube2o_14 61 PNIYPAVPPHFCYLSQ..CSGRLNPNLYDNGKVCLSLLGTWIGKGT.....ERWTSKSS.LLQVLISI
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Dm_Bruce_14 64 PPDYPNQPMLINLETTGRHSVRFNPNLYNDGKVCLSVLNTWHGRPE.....EKWNAQTSSFLQVLVSI
α3
α4
Sc_Ubc3_3
Sc_Ubc3_3 129 VS..LLEDP.NINSPANVDAAVDYRK.N.PEQYKQRVKMEVERSKQD
Hs_BIRC6_14 126 QSLILVAEP.YFNEPGYERSRGTPSGTQSSREYDGN.IRQATVKWAM
Ce_Ubc17_14 126 QEVI..EKHLWLKREAILK.QAQAWIDNVSNDFGDDKMSKRKDVFA.
At_Ubc23_14 120 QGLVLNSKP.YFNEAGYDKQVGTAEGEKNSLGYNENTFLLNCKTMM.
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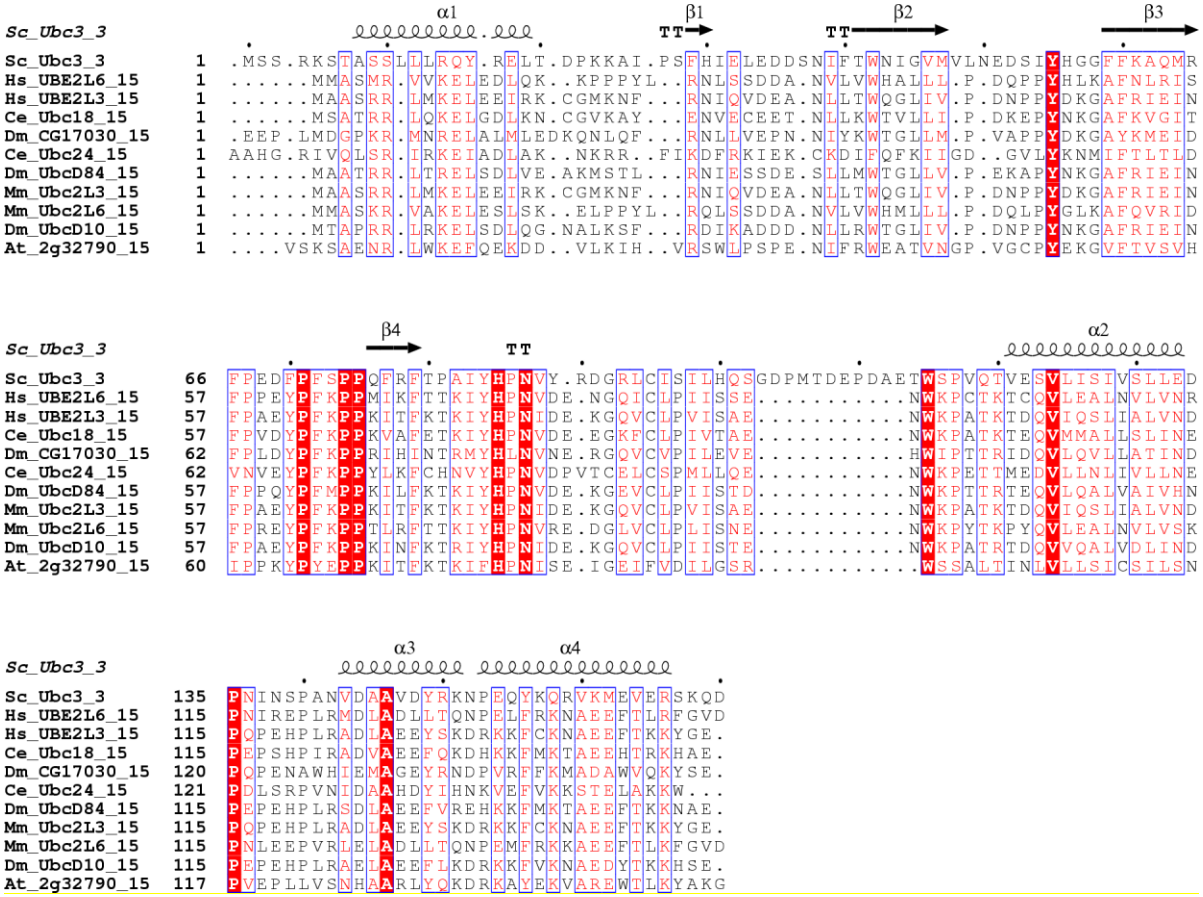
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β1
β2
β3
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β4
α2
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Hs_UBE2L6_15 57 FPPEYPFKPPMIKFTTKIYHPNVDE.NGQICLPIISSE..........NWKPCTKTCQVLEALNVLVNR
Hs_UBE2L3_15 57 FPAEYPFKPPKITFKTKIYHPNIDE.KGQVCLPVISAE..........NWKPATKTDQVIQSLIALVND
Ce_Ubc18_15 57 FPVDYPFKPPKVAFETKIYHPNVDE.EGKFCLPIVTAE..........NWKPATKTEQVMMALLSLINE
Dm_CG17030_15 62 FPLDYPFKPPRIHINTRMYHLNVNE.RGQVCVPILEVE..........HWIPTTRIDQVLQVLLATIND
Ce_Ubc24_15 62 VNVEYPFKPPYLKFCHNVYHPNVDPVTCELCSPMLLQE..........NWKPETTMEDVLLNLIVLLNE
Dm_UbcD84_15 57 FPPQYPFMPPKILFKTKIYHPNVDE.KGEVCLPIISTD..........NWKPTTRTEQVLQALVAIVHN
Mm_Ubc2L3_15 57 FPAEYPFKPPKITFKTKIYHPNIDE.KGQVCLPVISAE..........NWKPATKTDQVIQSLIALVND
Mm_Ubc2L6_15 57 FPREYPFKPPTLRFTTKIYHPNVRE.DGLVCLPLISNE..........NWKPYTKPYQVLEALNVLVSK
Dm_UbcD10_15 57 FPAEYPFKPPKINFKTRIYHPNIDE.KGQVCLPIISTE..........NWKPATRTDQVVQALVDLIND
At_2g32790_15 60 IPPKYPYFPPKITFKTKIFHPNISE.IGEIFVDILGSR..........WSSALTINIVLLSICSILSN
α3
α4
Sc_Ubc3_3
Sc_Ubc3_3 135 PNINSPANVDAAAVDYRKNPEQYKQRVKMEVERSKQD
Hs_UBE2L6_15 115 PNIREPLRMDLAADLLTQNPELFRKNAEEFTLRFGVD
Hs_UBE2L3_15 115 PQPEHPLRADLAEEYSKDRKKFCKNAEEFTKKYGE.
Ce_Ubc18_15 115 PEPSHPIRADVAEEFQKDHKKFMKTAEEHTRKHAE.
Dm_CG17030_15 120 PQPENAWHIEMAGEYRNDPVRFFKMADAWVQKYSE.
Ce_Ubc24_15 121 PDLSRPVNIDAAHDYIHNKVEFVKKSTELAKKW...
Dm_UbcD84_15 115 PEPEHPLRSDLAEEFVREHKKFMKTAEEFTKKNAE.
Mm_Ubc2L3_15 115 PQPEHPLRADLAEEYSKDRKKFCKNAEEFTKKYGE.
Mm_Ubc2L6_15 115 PNLEEPVRLELADLLTQNPEMFRKKAEEFTLKFGVD
Dm_UbcD10_15 115 PEPEHPLRAELAEEFLKDRKKFVKNAEDYTKKHSE.
At_2g32790_15 117 PVEPLLVSNHAARLYQKDRKAYEKVAREWTLKYAKG

Figure 2. Conformational landscape explored in Cdc34^{UBC} simulations. The free energy landscape is represented using projection of the Cdc34^{UBC} macro-trajectory along the principal components PC1 and PC2 of the essential subspace. The free energy is given in KJ/mol and indicated by the color bar. The label A-E indicates the region corresponding to the minimum free energy basins and the most populated structural clusters from cluster analysis.

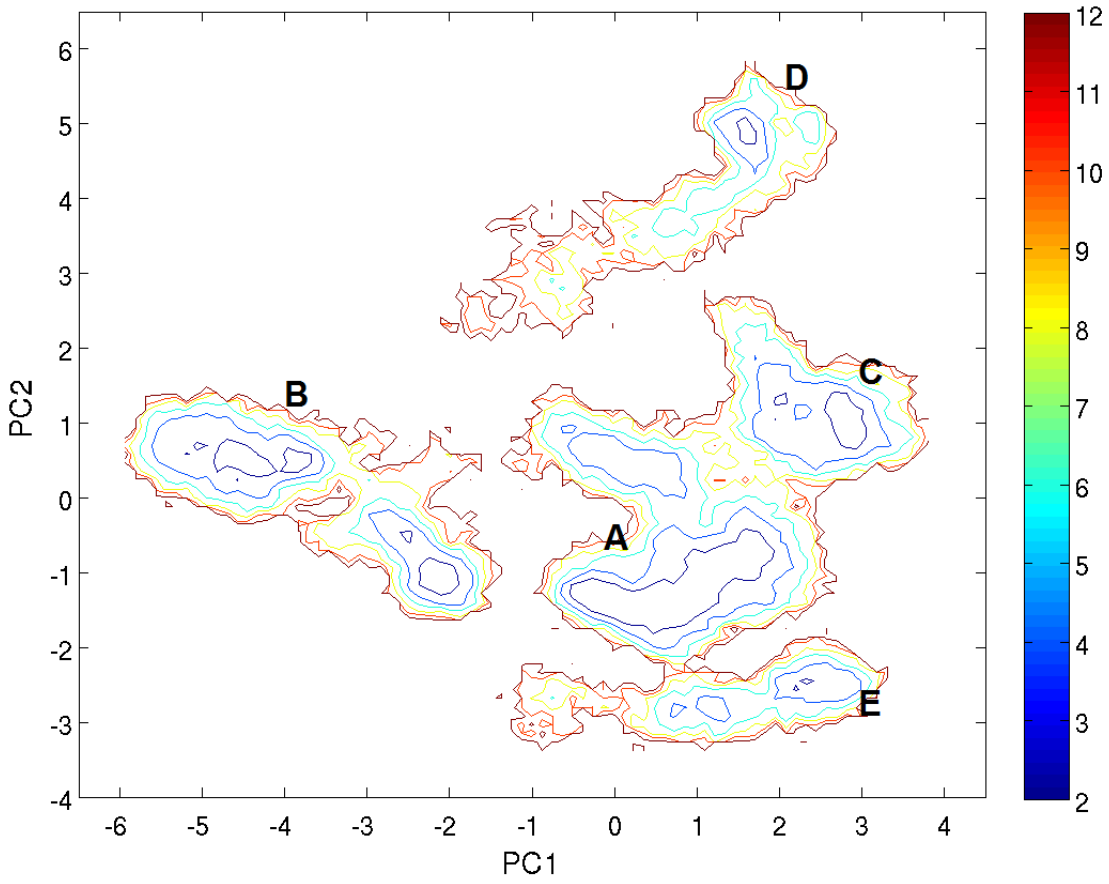


Figure 3. Flexibility profiles of non-phosphorylated Cdc34^{UBC}. Cα rmsf of ensemble trajectories of Cdc34^{UBC} and Cdc34^{UBC}–S130AS167A concatenated trajectories. The most persistent secondary structure during the simulations are represented schematically for each protein system. The rectangular box indicates the acidic loop.

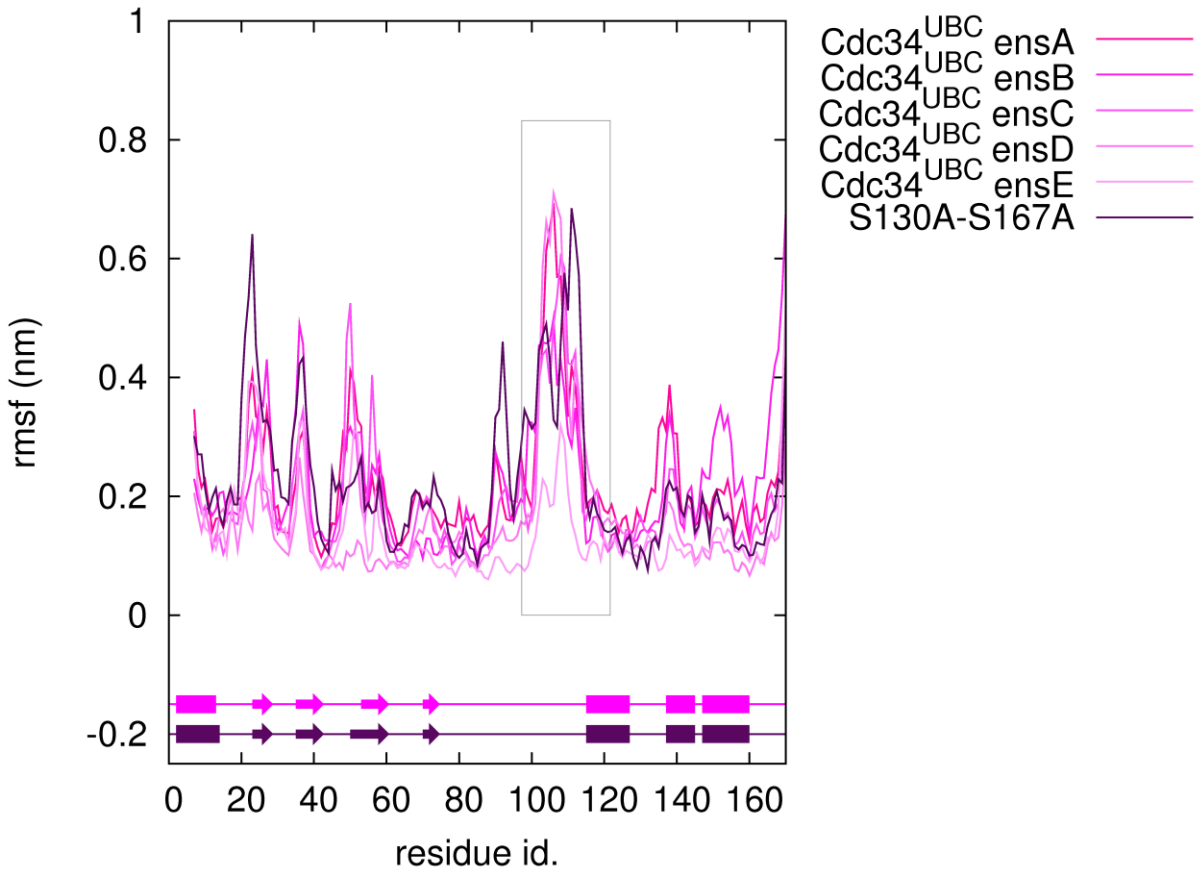


Figure 4. Flexibility profiles of phospho-Cdc34^{UBC}–pS130pS167 simulations. Cα rmsf of ensemble trajectories of Cdc34^{UBC}-pS130-pS167 and non-phosphorylated Cdc34^{UBC} are compared. The most persistent secondary structure during the simulations are represented schematically for each protein system. The rectangular box indicates the acidic loop.

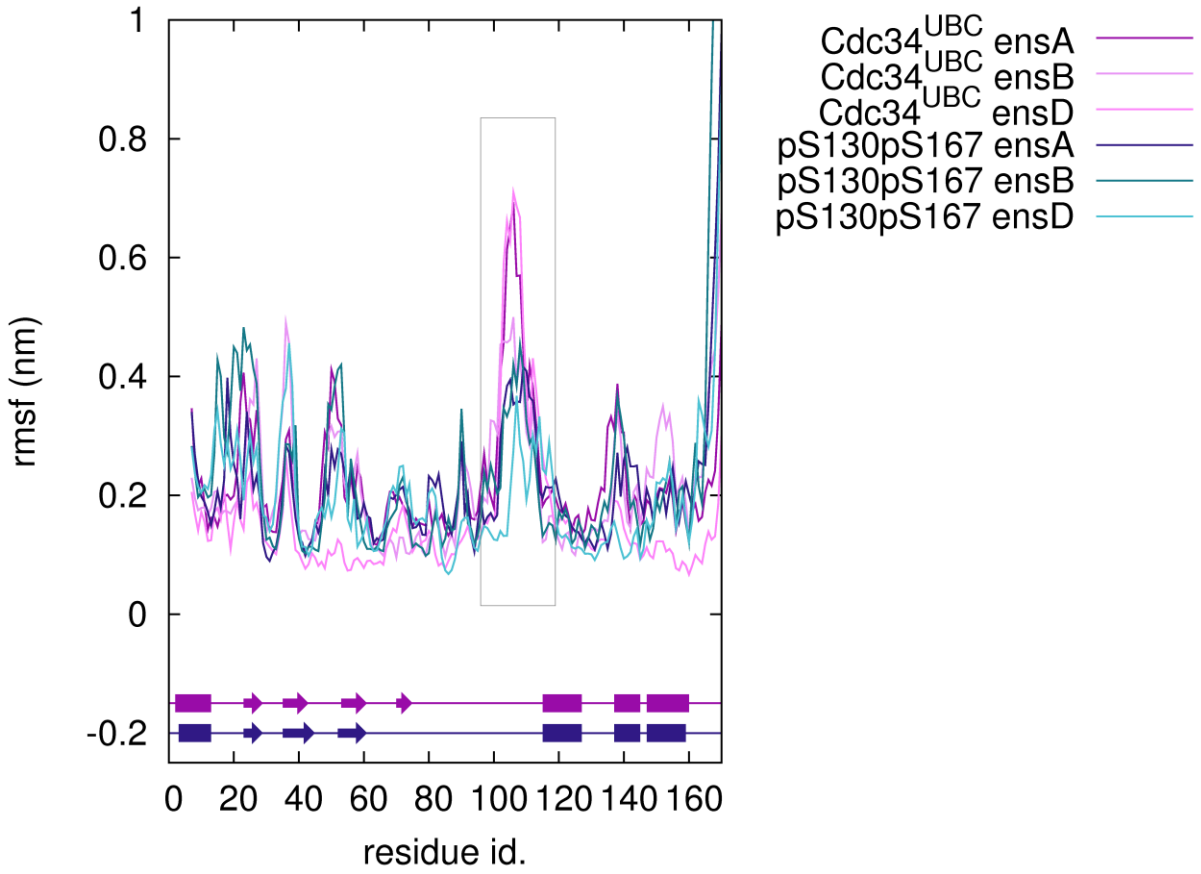


Figure 5. 3D structural superimposition of an average structure from phospho-Cdc34^{UBC}simulations, free NMR and X-ray structure of Ube2g2 and Ube2g2 in complex with the gp78 region of its E3 partner. The average structure of phospho Cdc34^{UBC} simulations is shown in blue, the NMR (PDB entry 2KLY) and X-ray (PDB entry 2CYX) structure of Ube2g2 are shown in dark and light green, respectively, the structure of Ube2g2 in complex with gp78 region of E3 partner (PDB entry 3H8K) is shown in orange. The catalytic cysteine is shown as yellow stick.

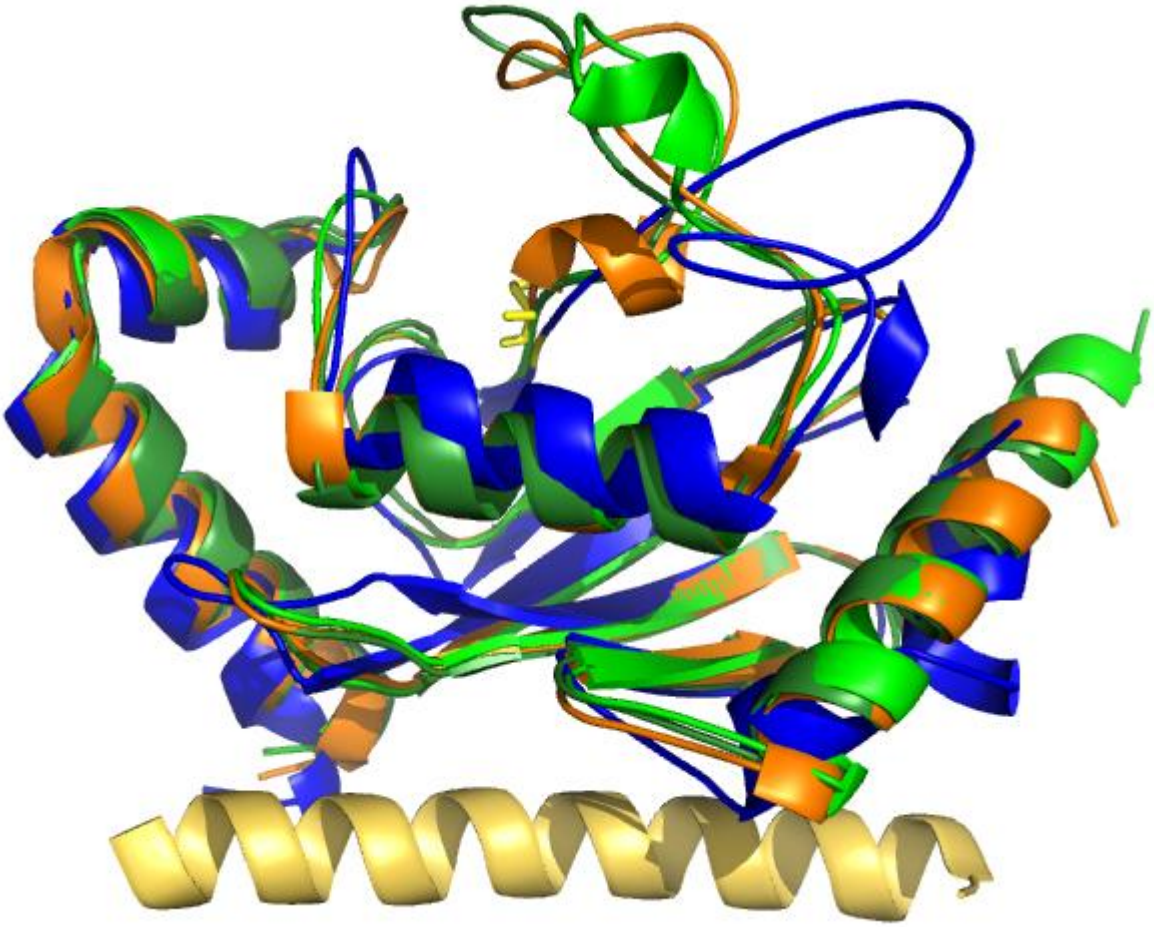


Figure 6. Projection of the simulations frames along the PC1 of Cdc34^{UBC}-S130D concatenated trajectory, indicated with different shades of grey. The rectangular box indicates the acidic loop and its aminoacidic composition.

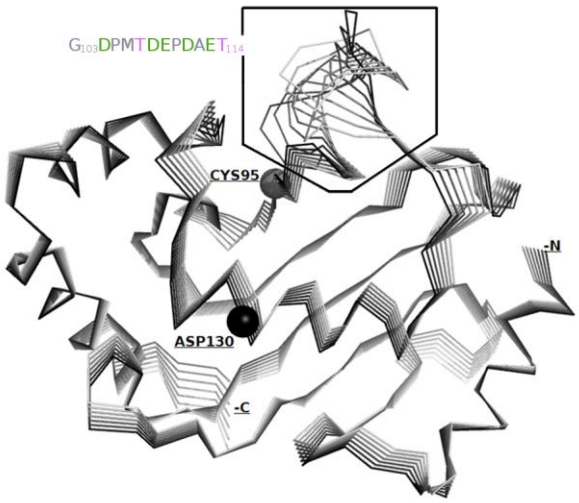


Figure 7. Root mean square fluctuation (rmsf) profiles of Cdc34^{UBC} and Cdc34-Δ12^{UBC} domains.

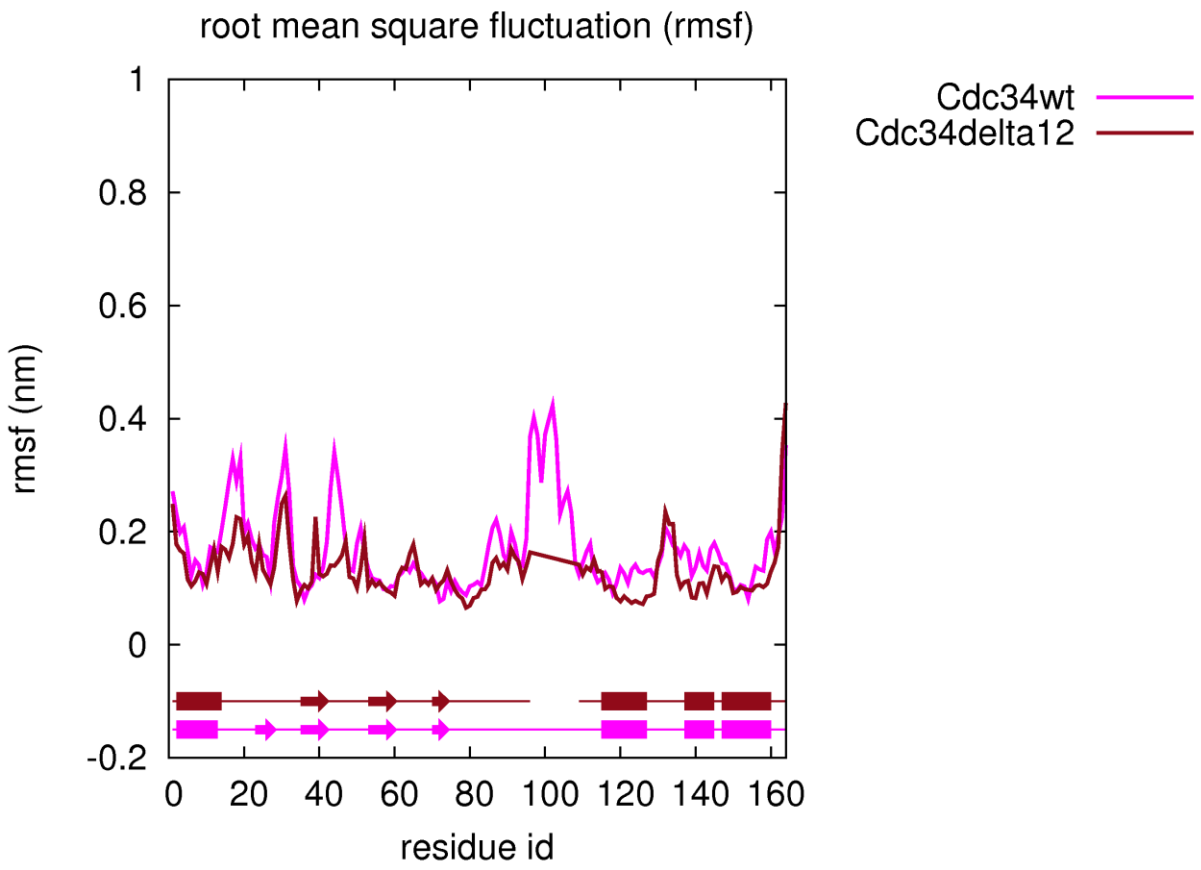
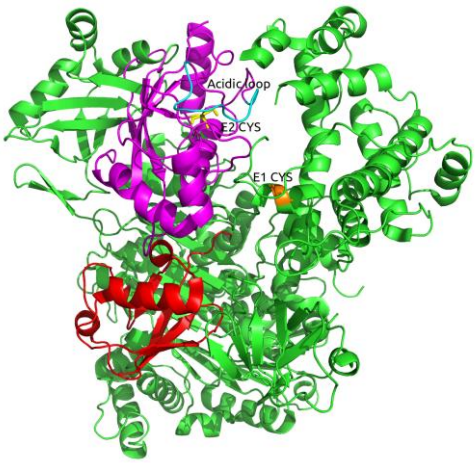
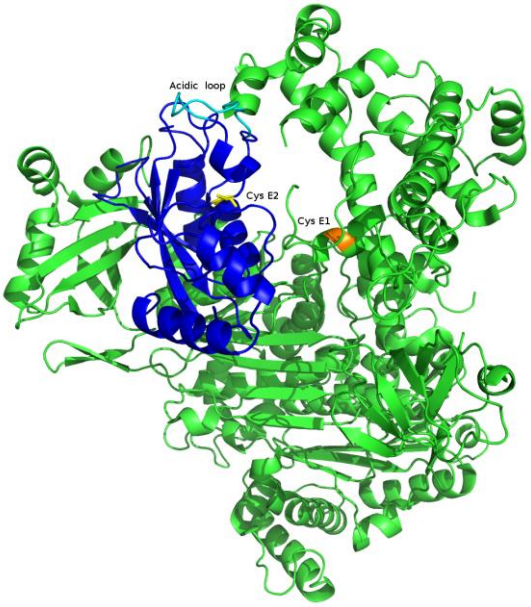


Figure 8. Model of Cdc34 both in closed (A) and open (B) conformations in complex with Uba1 E1 on the base of the known crystallographic structures of E2-E1 enzymes (PDB codes: 3CMM (Uba1) and 2PX9 (Ubc9 E2 in complex with SAE2 E1) and 2NVU (Ubc12 E2 in complex with Uba3 E1 and Nedd8).

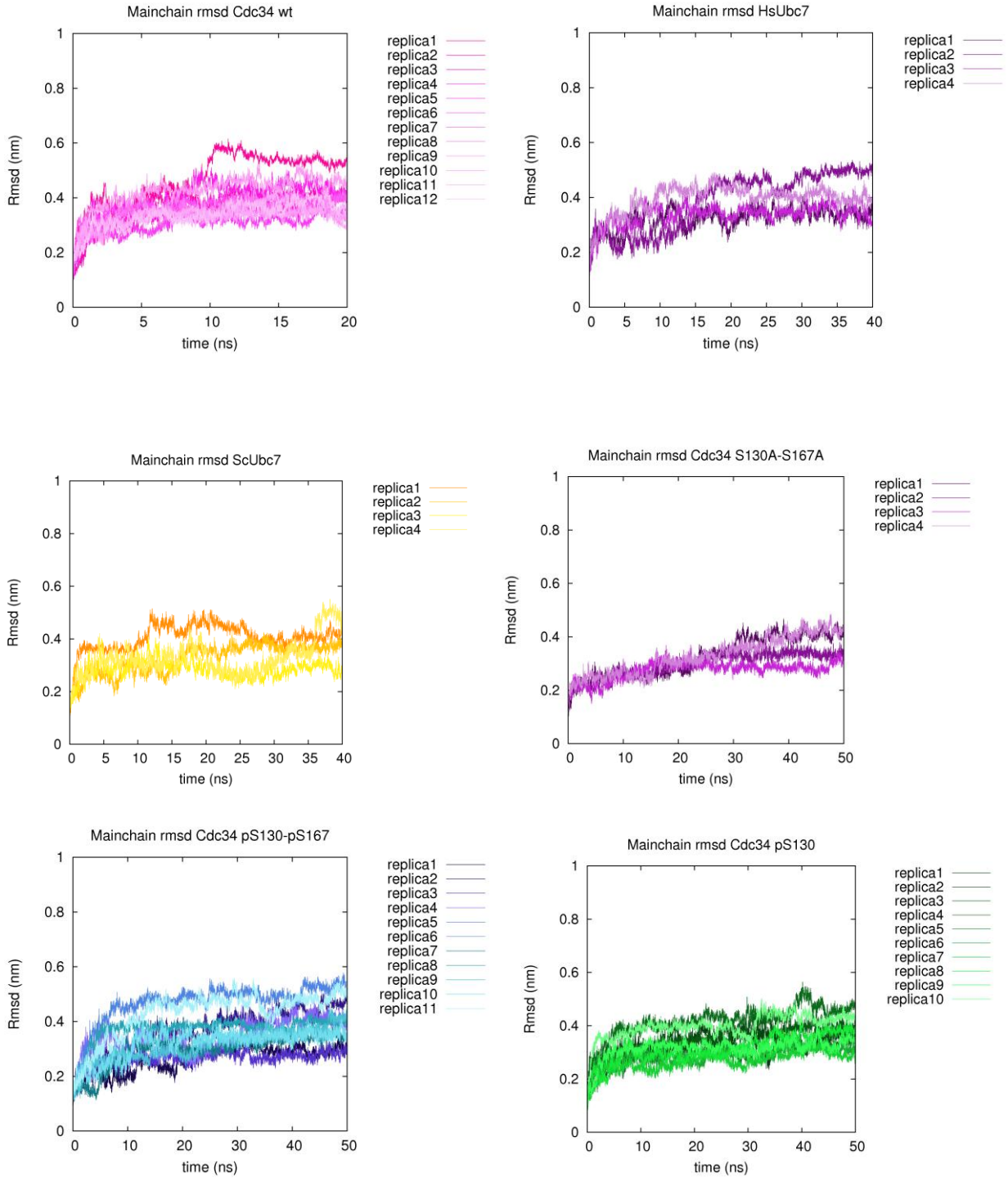


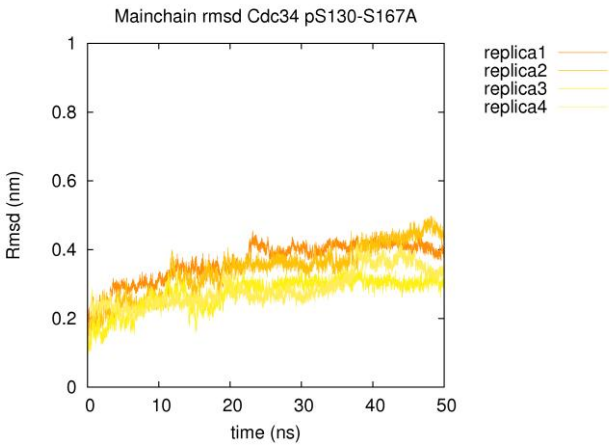
A

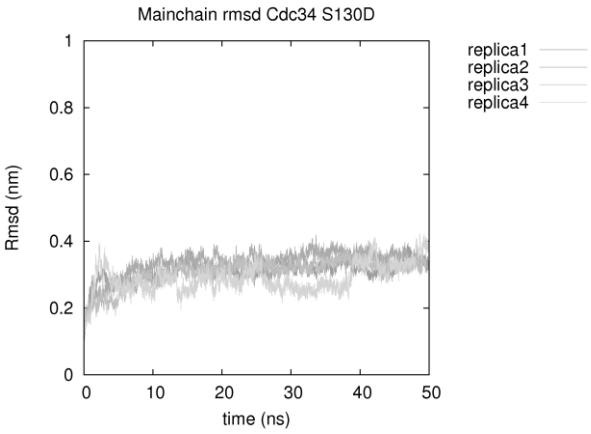


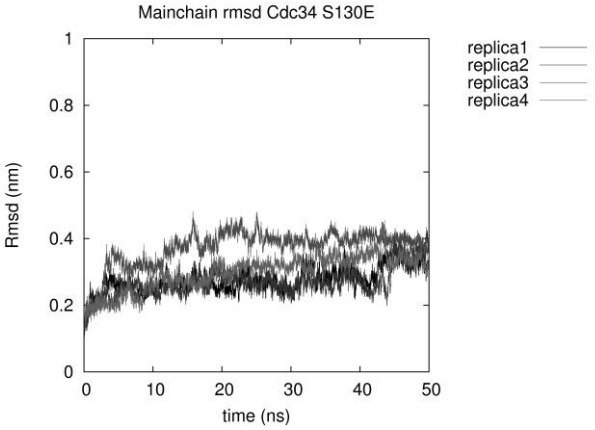
B

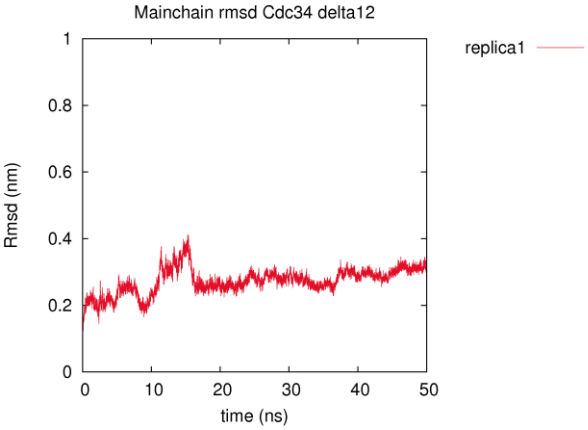
Figure 9. Mainchain root mean square deviation of single replicas of each simulated protein system.




Mainchain rmsd Cdc34 pS130-S167A
Rmsd (nm)
time (ns)
replica1
replica2
replica3
replica4


Mainchain rmsd Cdc34 S130D
Rmsd (nm)
time (ns)
replica1
replica2
replica3
replica4


Mainchain rmsd Cdc34 S130E
Rmsd (nm)
time (ns)
replica1
replica2
replica3
replica4


Mainchain rmsd Cdc34 delta12
Rmsd (nm)
time (ns)
replica1

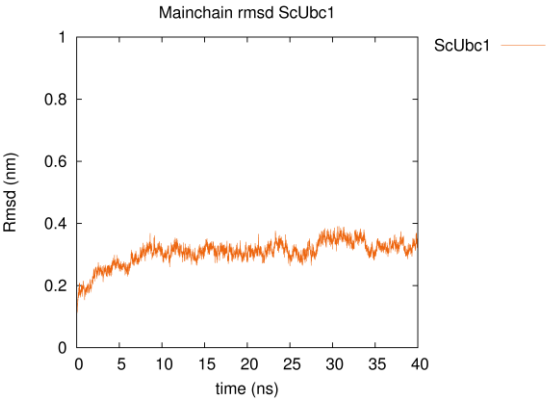

Mainchain rmsd ScUbc1
Rmsd (nm)
time (ns)
ScUbc1

Figure 10. Mainchain root mean square deviation of the structural elements of the common E2-fold (the mainchain atoms of the acidic loop are not included in the analysis) of the Cdc34 simulations.

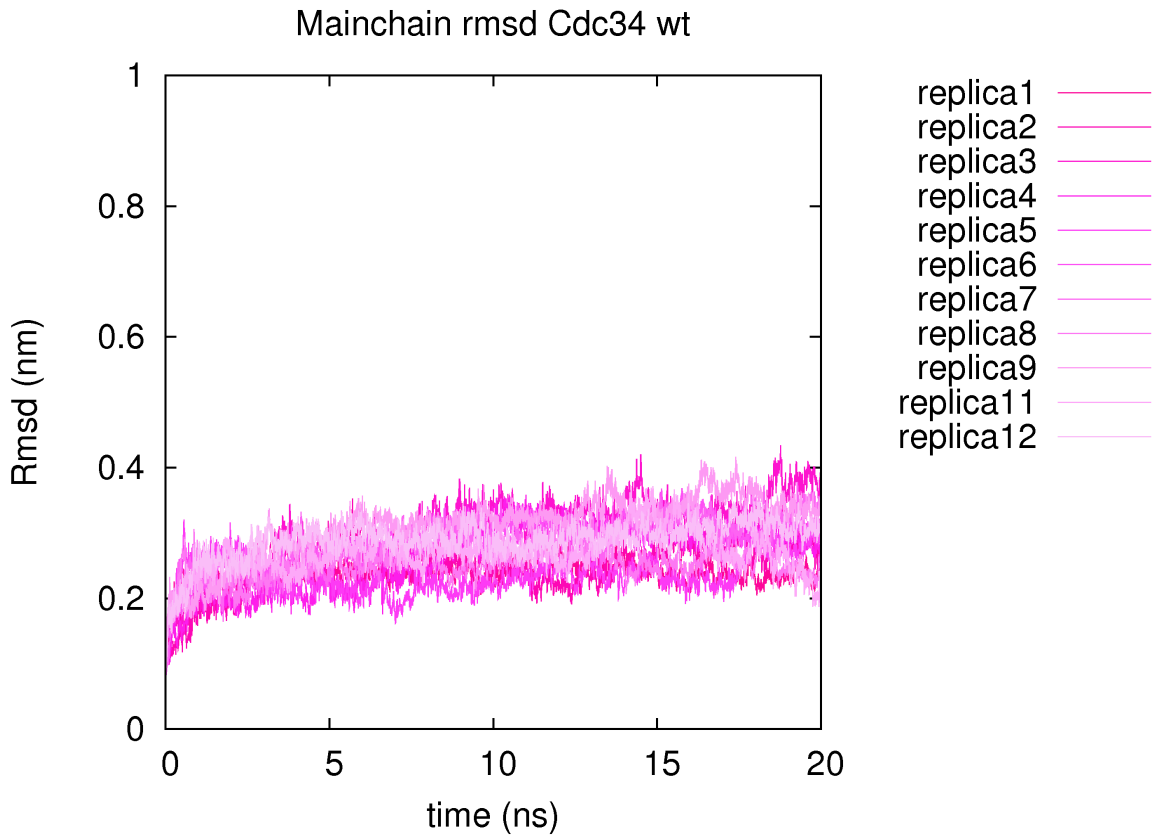
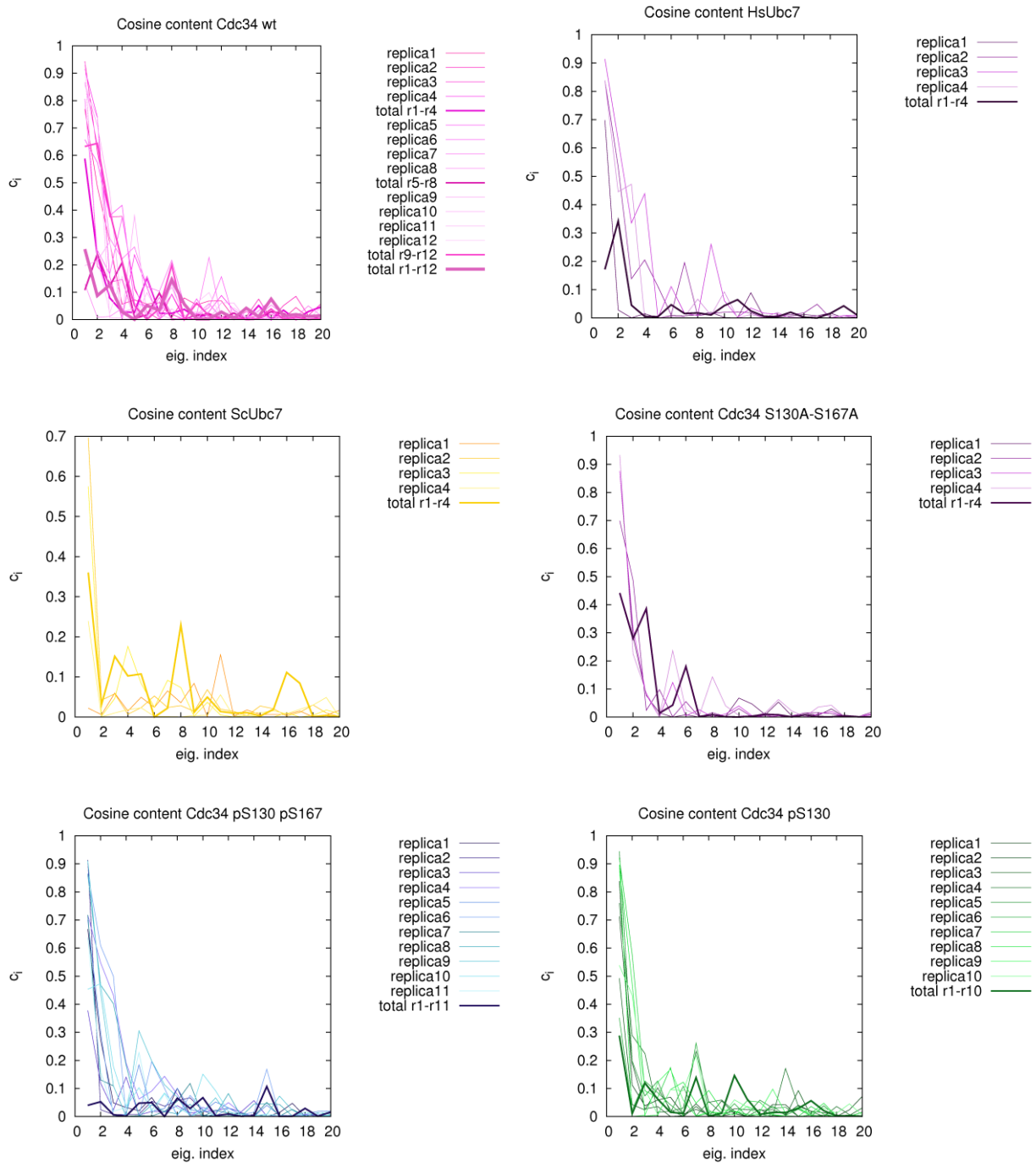
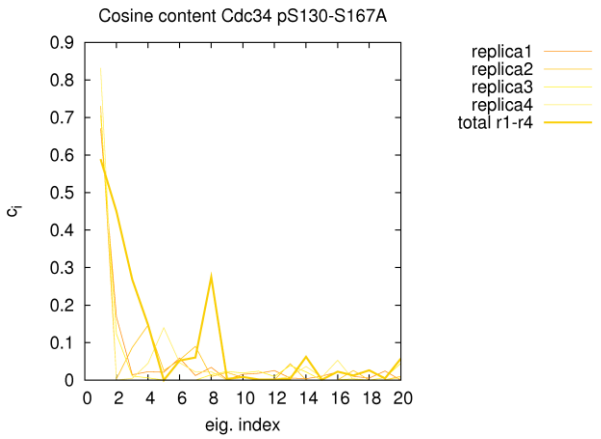
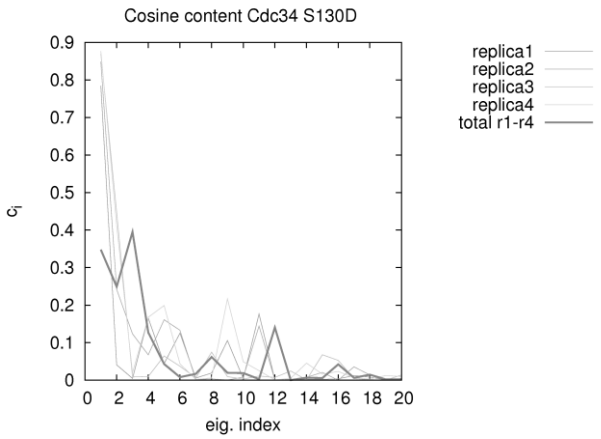
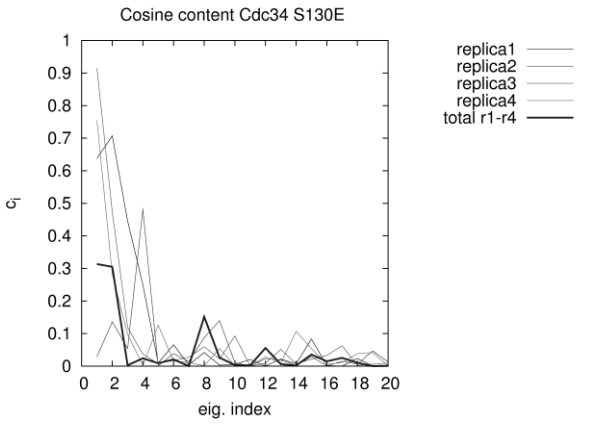


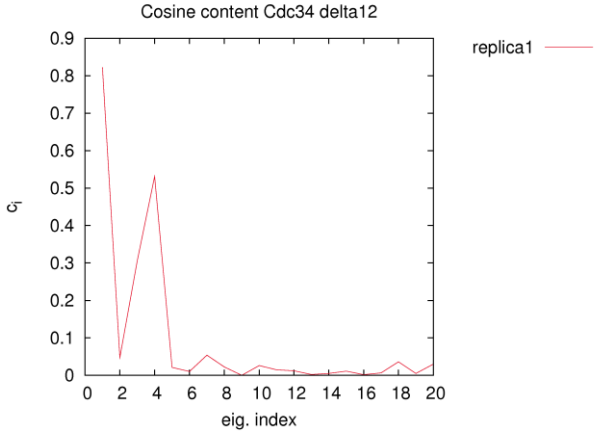
Figure 11. Cosine content along the first 20 principal components of single replicas and concatenated trajectories of different length for each simulated protein system.




Cosine content Cdc34 pS130-S167A
c_i
eig. index
replica1
replica2
replica3
replica4
total r1-r4


Cosine content Cdc34 S130D
c_i
eig. index
replica1
replica2
replica3
replica4
total r1-r4


Cosine content Cdc34 S130E
c_i
eig. index
replica1
replica2
replica3
replica4
total r1-r4


Cosine content Cdc34 delta12
c_i
eig. index
replica1