

Supplement to “Functional expression of adenosine A₃ receptor in yeast utilizing a chimera with the A_{2A}AR C-terminus”

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CLUSTAL O(1.2.4) multiple sequence alignment

SP P0DMS8 AA3R_HUMAN	MPNNSTALSLANVTYITMEIFIGLCAIVGNVLVICVVKLNPSLQTTTFYFIVSLALADIA	60
SP P29274 AA2AR_HUMAN	-----MPIMGSS <u>VYITVELAIAVLA</u> ILGNVLVCWAWLNSNLQNVTN <u>YFVVSLLAAADIA</u>	54
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SP P0DMS8 AA3R_HUMAN	VGVLVMPLAIVV SLGITIHFYSCLFMTCLLLIFTHASIMSLAIAVDRYLRVKLTVRYKR	120
SP P29274 AA2AR_HUMAN	VGVLAI PFAITI STGFCAACHGCLFIACFVLVLT <u>QSSIFSLLAIAI</u> DRYIAIRIPLRYNG	114
	****. :*: ***. :* : * : . :***: :*: :*: :*: :*****: * : : : :***:	
SP P0DMS8 AA3R_HUMAN	VTTHRR <u>IWLALGLCWLV</u> SFLVGLTPMF <u>GWNMKLTSEYHR</u> -----NVTFLSCQFVSVMRM	174
SP P29274 AA2AR_HUMAN	LVTGTR <u>AKGIIIAICWVL</u> SFAIGLTPMLG <u>GWNNCQPKEGKNHSQGC</u> GEOVACLFEDVVP <u>M</u>	174
	: * * : . :***: * :*****: * : : : . : * : * . : * : * : * :	
SP P0DMS8 AA3R_HUMAN	DYM VYFSFLTWIFIPLVVMCAIYLDIFYIIRNKLSNLNS --SKETGAFYGREFKTAKS	231
SP P29274 AA2AR_HUMAN	NYM VYFN <u>FACVLVPL</u> LL <u>MLG</u> VY <u>LRIFLAARRQLKQ</u> MESOPLPGERARSTLQKEVHAAKS	234
	:*****. * : : :***: * :** * * . :*. * : . : : : : * : . : : : : * : :***	
SP P0DMS8 AA3R_HUMAN	LFLVLFLFALS WLPLSI <u>INCIIYFN</u> GE---VP <u>QLVLYMGILLSHANSMMNPIVYAY</u> KIKK	288
SP P29274 AA2AR_HUMAN	LAI IV <u>GLFALCWLP</u> LH <u>IINCFTFFCPDCS</u> HAP <u>LWLMYLAIVL</u> SHTNSVNP <u>FIYAY</u> RIRE	294
	* : : : * : * : * : * : * : * : * : * : : : : : : : : : : : : : : : :	
SP P0DMS8 AA3R_HUMAN	FKETYLLILKACVV <u>CHPSDSL</u> -----DTSIEKNSE-----	318
SP P29274 AA2AR_HUMAN	FROTFRKIIRSHVL RQEPFKAAGTSARVLAAGHS <u>DGE</u> QVSRLNLGHPPGVWANGSAPH	354
	* :*: * : * : * : * : . : . : : : : : : : : :	
SP P0DMS8 AA3R_HUMAN	-----	
SP P29274 AA2AR_HUMAN	ERRPNGYALGLVSGGSQAESQNTGLP <u>DVE</u> LLSHELGVCPEPPGLDDPLAQDGAGVS	412

Figure S1 (A) Pairwise sequence alignment of A₂AR (P29274) and A₃R(P0DMS8) generated using the UNIPROT alignment tool. Transmembrane helices in A₂AR and putative transmembrane helices in A₃R are underlined, and the amino acids in bold. Identical amino acids denoted by *; conservative changes by period or colon. The palmitoylation site in A₃R (Cys 303) is double underlined, and colored red. The two D/E-X-D/E motifs in A₂AR (located at residues 330 and 382) are boxed and highlighted in yellow.

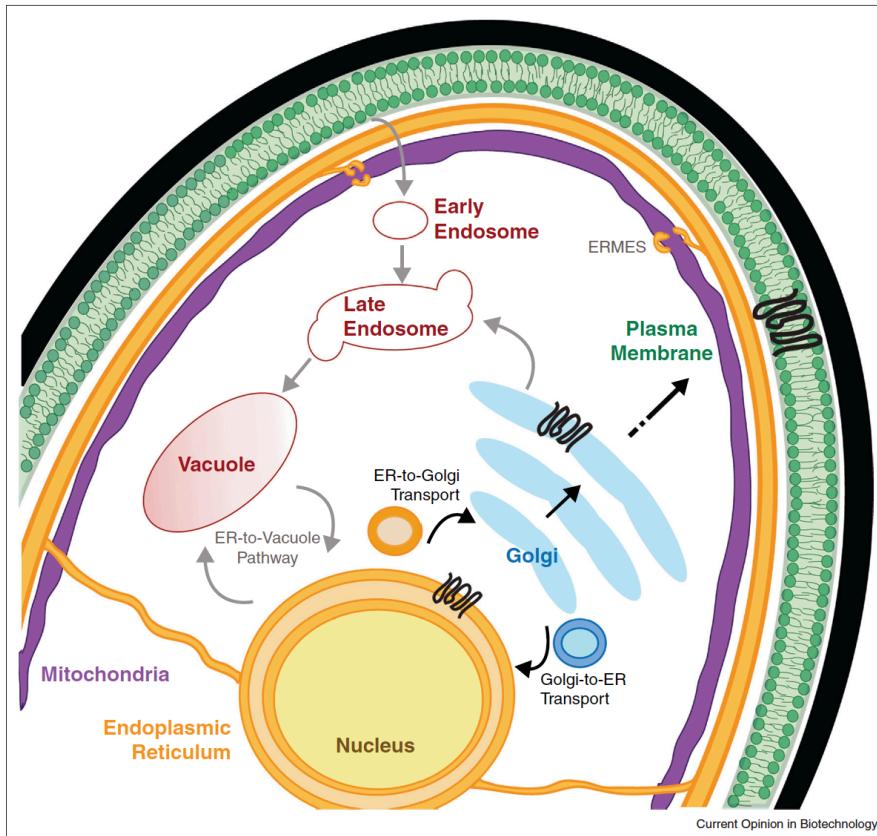


Figure S2 Protein trafficking in yeast. Endoplasmic reticulum (ER) to Golgi to plasma membrane trafficking is shown for membrane proteins. Reprinted with permission from Young and Robinson [1].

1. Young, C. L.; Robinson, A. S., Protein folding and secretion: mechanistic insights advancing recombinant protein production in *S. cerevisiae*. *Curr Opin Biotechnol* **2014**, 30, 168-77.