

Table S1 - Strains used in this work

strain name	genotype	Derivative of	reference
AM136	<i>MATa PRE2 (β5)-HA::Ylplac211</i>	JD47-13C	this work
AM200	<i>MATa ump1Δ::HIS3MX6 sse1Δ::KanMX6</i>	JD47-13C	this work
AM198-1D	<i>MATa pba3Δ::kan sse1Δ::Kan</i>	JD47-13C	this work
BY4741	<i>MATa his3-Δ1 leu2-Δ0, met15-Δ0 ura3-Δ0</i>	"WT"	Euroscarf
CM2	<i>MATa PRE6 (α4)-HA::Ylplac211</i>	JD47-13C	this work
CM3	<i>MATa PRE6 (α4)-HA::Ylplac211 ssa1-ts45 ssa2Δ::LEU2 ssa3Δ::TRP1 ssa4Δ::LYS2</i>	JB568	this work
CM5	<i>MATa PRE6 (α4)-HA::Ylplac211 sse1Δ::KanMX4</i>	BY4741	this work
CM13	<i>MATa PRE6 (α4)-HA::Ylplac211</i>	BY4741	this work
CM14	<i>MATa PRE6 (α4)-HA::Ylplac211</i>	JN54	this work
CM24	<i>MATα PRE6 (α4)-HA::Ylplac211 sse1/2Δp414TEFsse1-G205D</i>	W303	this work
CM25	<i>MATα PRE6 (α4)-HA::Ylplac211 sse1/2Δp414TEFSSE1WT</i>	W303	this work
CM29	<i>MATa P_{GAL1}PRE6-(α4)-HA::Ylplac211</i>	BY4741	this work
CM30	<i>MATa P_{GAL1}PRE6-(α4)-HA::Ylplac211 sse1Δ::kanMX4</i>	BY4741	this work
CM45	<i>MATα rpn4Δ::HIS3MX6</i>	ML163	this work
CM46	<i>MATa rpn4Δ::HIS3MX6 sse1Δ::KanMX4</i>	BY4741	this work
CM54	<i>MATa PUP1 (β2)-HA::Ylplac211</i>	BY4741	this work
CM55	<i>MATa PUP1 (β2)-HA::Ylplac211 sse1Δ::KanMX4</i>	BY4741	this work
CM61	<i>MATa PUP3 (β3)-HA::Ylplac211</i>	JD47-13C	this work
CM143	<i>MATa pre9Δ::KanMX4 sse1Δ::HIS3MX6</i>	Y04765	this work
CM90	<i>MATa PBA2-HA::Ylplac211</i>	JD47-13C	this work
CM128	<i>MATa PRE4 (β7)-HA::Ylplac211</i>	JD47-13C	this work
CM134	<i>MATa P_{GALS}SCL1::kanMX4 PRE6-2HA::Ylplac211</i>	JD47-13C	this work
CM152	<i>MATa PRE7 (β6)-HA::Ylplac211</i>	JD47-13C	this work
CM182	<i>MATa PRE8 (α2)-HA::Ylplac211 sse1Δ::TRP1</i>	PR93	this work
CM185	<i>MATa SCL1 (α1)-HA::Ylplac211 sse1Δ::TRP1</i>	PR93	this work
CM199	<i>MATa P_{GALS}-PRE6 (α4)::KanMX6 SCL1 (α1)-HA::Ylplac211</i>	JD47-13C	this work
CM252	<i>MATα SCL1 (α1)-HA::Ylplac211 ssa2Δ::LEU2 ssa3Δ::TRP1 ssa4Δ::LYS2</i>	JN516	this work
CM253	<i>MATa SCL1 (α1)-HA::Ylplac211 ssa1-ts45 ssa2Δ::LEU2 ssa3Δ::TRP1 ssa4Δ::LYS2</i>	JB568	this work
CM272	<i>MATα PRE6 (α4)-HA::Ylplac211 ssa1Δ::HIS3 ssa2Δ::LEU2</i>	MW123	this work
FP6	<i>MATa PRE3 (β1)-HA::Ylplac211</i>	JD47-13C	Pardelha, FP
FP8	<i>MATa PRE6 (α4)-HA::Ylplac211 pre9Δ::HIS3MX6</i>	JD305	Pardelha, FP
FP14	<i>MATa PRE6 (α4)-HA::Ylplac211 pre9Δ::HIS3MX6 sse1Δ::TRP1</i>	JD305	Pardelha, FP
FP16	<i>MATa PBA4-HA::Ylplac211</i>	JD47-13C	Pardelha, FP
JB568	<i>MATa ssa1-ts45 ssa2Δ::LEU2 ssa3Δ::TRP1 ssa4Δ::LYS2</i>	JN54	[41]
JN54	<i>MATa his3-11,3-15 leu2-3,2-112 lys2 trp1-Δ1 ura3-52</i>	"WT"	[41]
JN516	<i>MATα ssa2Δ::LEU2 ssa3Δ::TRP1 ssa4Δ::LYS2</i>	JN54	[41]
JD47-13C	<i>MATa his3-Δ200 leu2-3,112 lys2-801 trp1-Δ63 ura3-52</i>	"WT"	[21]
JD59	<i>MATa ump1Δ::HIS3MX6</i>	JD47-13C	[8]
JD127	<i>MATa PRE1 (β4)-HA::Ylplac211 UMP1-HA::Ylplac128</i>	JD47-13C	[8]
JD129	<i>MATa UMP1-HA::Ylplac128</i>	JD47-13C	[8]
JD139	<i>MATa PUP1 (β2)-HA::Ylplac211</i>	JD47-13C	[8]
JD183	<i>MATa PRE10 (α7)-HA::Ylplac211</i>	JD47-13C	this work
JD305	<i>MATa pre9Δ::HIS3MX6</i>	JD47-13C	this work
JM5	<i>MATa PRE8 (α2)-HA::Ylplac128</i>	JD47-13C	this work
JM6	<i>MATa PRE9 (α3)-HA::Ylplac211</i>	JD47-13C	this work
JM9	<i>MATa PUP2 (α5)-HA::Ylplac211</i>	JD47-13C	this work
JM10	<i>MATa PRE5 (α6)-HA::Ylplac211</i>	JD47-13C	this work
JM12	<i>MATa PRE6 (α4)-F6H::Ylplac204 PRE8 (α2)-HA::Ylplac128</i>	JD47-13C	this work
JM15	<i>MATa PRE6 (α4)-HA::Ylplac211 PRE5 (α6)-HA::LEU2</i>	JD47-13C	this work
JM17	<i>MATa SCL1 (α1)-HA::Ylplac211</i>	JD47-13C	this work
ML163	<i>MATα his3-Δ1 leu2-Δ0, lys-Δ0 ura3-Δ0 trp1-Δ0</i>	BY4742	London, M
MW123	<i>MATα leu2Δ trp ura3 his3 lys2 ssa1Δ::HIS3 ssa2Δ::LEU2</i>	JN54	[21]
PG24	<i>MATa PRE6 (α4)-HA::Ylplac211 ump1Δ::HIS3MX6</i>	JD47-13C	Gouveia, P
PG25	<i>MATa UMP1-HA::Ylplac128</i>	BY4741	Gouveia, P
PR93	<i>MATa sse1Δ::TRP1</i>	JD47-13C	this work
PR102	<i>MATa pba1Δ::HIS3MX6 sse1Δ::TRP1</i>	JD47-13C	this work
PR108	<i>MATa PRE6 (α4)-HA::Ylplac211 pba1Δ::HIS3MX6 sse1Δ::TRP1</i>	PR102	this work
PR110	<i>MATa PRE6 (α4)-HA::Ylplac211 pba3Δ::HIS3MX6 sse1Δ::TRP1</i>	JD47-13C	this work
PR118	<i>MATa pba1Δ::HIS3MX6</i>	JD47-13C	this work
PR122	<i>MATα PRE6 (α4)-HA::Ylplac211</i>	JN516	this work
PR125	<i>MATa PRE6 (α4)-HA::Ylplac211 sse1Δ::TRP1</i>	JD47-13C	this work
PR126	<i>MATa PRE9 (α3)-HA::Ylplac211 sse1Δ::TRP1</i>	PR93	this work
PR127	<i>MATa PUP2 (α5)-HA::Ylplac211 sse1Δ::TRP1</i>	JD47-13C	this work
PR129	<i>MATa pba1Δ::HIS3MX6 PRE6 (α4)-HA::Ylplac211</i>	JD47-13C	this work
PR132	<i>MATa PRE6 (α4)-HA::Ylplac211 pba3Δ::HIS3MX6</i>	JD47-13C	this work
PR135	<i>MATa PRE6 (α4)-HA::Ylplac211 sse1Δ::kanMX6 ump1Δ::HIS3MX6</i>	JD47-13C	this work
PR170	<i>MATa SCL1 (α1)-HA::Ylplac211</i>	JN54	this work
W303	<i>MATa ade2-1 trp can1-100 leu2-3,112 his3-11,-15 ura3</i>	"WT"	[40]
Y01576	<i>MATa pba3Δ::KanMX4</i>	BY4741	Euroscarf
Y02146	<i>MATa sse1Δ::KanMX4</i>	BY4741	Euroscarf
Y04765	<i>MATa pre9Δ::KanMX4</i>	BY4741	Euroscarf

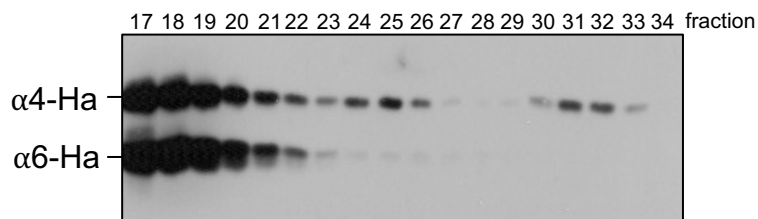


Figure S1. Gel filtration analysis of $\alpha 4$ and $\alpha 6$. A strain was generated expressing both $\alpha 4$ and $\alpha 6$ in an HA-tagged form. Gel filtration (Superdex200) and anti-HA western blot analysis was done as described in Figure 1.

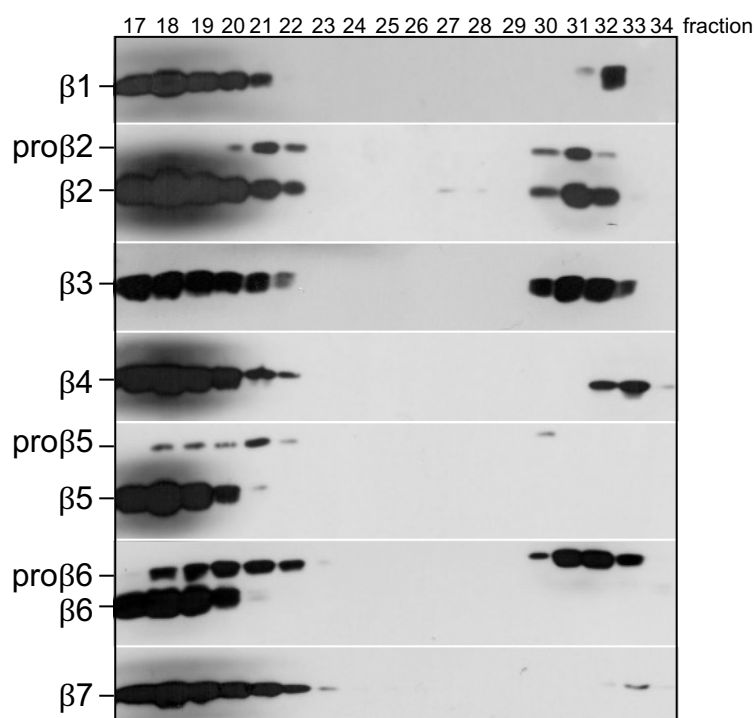


Figure S2. Gel filtration analysis of β subunits. Strains expressing HA-tagged versions of the seven distinct β subunits were used to analyze complexes formed by these subunits by Superdex200 gel filtration and anti-HA western blotting as described in Figure 1. The positions of the mature and unprocessed (pro) form of the β subunits are indicated on the left.



Figure S3. The subunit $\alpha 4$ synthesized in *E. coli* is largely insoluble. Total protein extract (T) from a bacteria strain overexpressing $\alpha 4$ -HA was analyzed by SDS-PAGE. (S) Soluble material. (P) Material found in the pellet.