# Table S1A Class I Mutants: Hygromycin B Sensitive Strains Suppressed by 100 mM KCl

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
|  | | | | YPAD+ |  | HB+ |
| Strain | ORF | Aliases | YPAD | 100 mM KCl | HB | 100 mM KCl |
| WT |  |  | **++++** | **++++** | **+++** | **++++** |
| Membrane Traffic Proteins (30) |  |  |  |  |  |  |
| *arl1Δ* | YBR164C | *DLP2* | ++++ | ++++ | ‐ | ++++ |
| *bro1Δ* | YPL084W | *VPS31* | ++++ | ++++ | ‐ | ++++ |
| *chs5Δ* | YLR330W | *CAL3* | ++++ | ++++ | ‐ | ++++ |
| *cog5Δ* | YNL051W | *COD4* | ++++ | ++++ | +/‐ | ++++ |
| *cog6Δ* | YNL041C | *COD2* | ++++ | ++++ | + | ++++ |
| *did4Δ* | YKL002W | *VPS2* | ++++ | ++++ | ‐ | ++++ |
| *gga1Δ* | YDR358W |  | ++++ | ++++ | + | ++++ |
| *gga2Δ* | YHR108W |  | ++++ | ++++ | ‐ | ++++ |
| *glo3Δ* | YER122C |  | +++ | +++ | ‐ | +++ |
| *gos1Δ* | YHL031C |  | ++++ | ++++ | +/‐ | +++ |
| *mon2Δ* | YNL297C | *YSL2* | ++++ | ++++ | +/‐ | +++ |
| *pep5D* | YMR231W | *VAM1, VPS11* | ++++ | ++++ | ‐ | ++++ |
| *rer1Δ* | YCL001W |  | ++++ | ++++ | ‐ | +++ |
| *rgp1Δ* | YDR137W |  | ++++ | ++++ | +/‐ | ++++ |
| *ric1Δ* | YLR039C |  | ++++ | ++++ | + | ++++ |
| *sec22Δ* | YLR268W | *SLY2, TSL26* | ++++ | ++++ | ‐ | +++ |
| *stp22D* | YCL008C | *VPS23* | ++++ | ++++ | ‐ | ++++ |
| *vam3Δ* | YOR106W | *PTH1* | ++++ | ++++ | ‐ | ++++ |
| *vam7Δ* | YGL212W | *VPS43* | ++++ | ++++ | ‐ | ++++ |
| *vps4Δ* | YPR173C | *DID6, GRD13* | ++++ | ++++ | ‐ | ++++ |
| *vps8Δ* | YAL002W | *FUN15, VPT8* | ++++ | ++++ | ‐ | ++++ |
| *vps9Δ* | YML097C | *VPL31, VPT9* | ++++ | ++++ | ‐ | ++++ |
| *vps20Δ* | YMR077C |  | ++++ | ++++ | ‐ | ++++ |
| *vps21Δ* | YOR089C | *YPT51* | +++ | ++++ | ‐ | ++++ |
| *vps24Δ* | YKL041W | *DID3* | ++++ | ++++ | ‐ | ++++ |
| *vps27Δ* | YNR006W | *GRD11, DID7* | ++++ | ++++ | ‐ | ++++ |
| *vps30Δ* | YPL120W | *APG6, VPT30* | ++++ | ++++ | ‐ | ++++ |
| *vps36Δ* | YLR417W | *GRD12, VAC3* | ++++ | ++++ | ‐ | ++++ |
| *vps41Δ* | YDR080W | *VAM2, VPL20* | ++++ | ++++ | ‐ | +++ |
| *ypt6Δ* | YLR262C |  | ++++ | ++++ | +/‐ | ++++ |
| Ion Transporters (2) |  |  |  |  |  |  |
| *gef1Δ* | YJR040W | *CLC* | ++++ | ++++ | ‐ | +++ |

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| *trk1Δ* | YJL129C | ++++ | ++++ | ‐ | ++++ |
| Protein Kinases (2) |  |  |  |  |  |
| *hal5Δ* | YJL165C | ++++ | ++++ | ‐ | ++++ |
| *sat4D* | YCR008W | *HAL4* ++++ | ++++ | ‐ | ++++ |
| Glycosylation (3) |  |  |  |  |  |
| *alg6Δ* | YOR002W | ++++ | ++++ | ‐ | ++++ |
| *hoc1Δ* | YJR075W | ++++ | ++++ | + | ++++ |
| *van1Δ* | YML115C | ++++ | ++++ | + | ++++ |
| Inositol kinases (3) |  |  |  |  |  |
| *arg82Δ* | YDR173C | *IPK2* ++++ | ++++ | ‐ | ++ |
| *fab1Δ* | YFR019W | *SVL7* +++ | +++ | + | +++ |
| *kcs1Δ* | YDR017C | ++++ | ++++ | ‐ | +++ |
| Metabolism (2) |  |  |  |  |  |
| *adh1Δ* | YOL086C | ++++ | ++++ | ‐ | ++++ |
| *ure2Δ* | YNL229C | ++++ | ++++ | ‐ | ++++ |
| Miscellaneous (16) |  |  |  |  |  |
| *arv1Δ* | YLR242C | ++++ | ++++ | ‐ | +++ |
| *bem1Δ* | YBR200W | ++++ | ++++ | ‐ | +++ |
| *cdc50Δ* | YCR094W | ++++ | ++++ | ‐ | +++ |
| *cyt1Δ* | YOR065W | *CTC1* +++ | +++ | +/‐ | +++ |
| *eft2Δ* | YDR385W | ++++ | ++++ | ‐ | +++ |
| *kap120Δ* | YPL125W | ++++ | ++++ | ‐ | ++++ |
| *lsb3D* | YFR024C | ++++ | ++++ | ‐ | ++++ |
| *nat3Δ* | YPR131C | ++++ | ++++ | ‐ | +++ |
| *nbp2Δ* | YDR162C | ++++ | ++++ | ‐ | ++++ |
| *ncs6D* | YGL211W | ++++ | ++++ | ‐ | ++++ |
| *ram1Δ* | YDL090C | ++++ | ++++ | + | ++++ |
| *reg1Δ* | YDR028C | ++++ | ++++ | +/‐ | +++ |
| *sap155Δ* | YFR040W | ++++ | ++++ | +/‐ | ++++ |
| *sse1Δ* | YPL106C | ++++ | ++++ | ‐ | +++ |
| *vph2Δ* | YKL119C | *CLS10, VMA12* ++++ | ++++ | ‐ | ++++ |
| Transcription / Replication (12) |  |  |  |  |  |
| *csi2Δ* | YOL007C | ++++ | ++++ | ‐ | +++ |
| *ctf4Δ* | YPR135W | +++ | +++ | ‐ | +++ |
| *eaf1Δ* | YDR359C | *VID21* ++++ | ++++ | ‐ | +++ |
| *irs4Δ* | YKR019C | ++++ | ++++ | + | ++++ |
| *mdm20Δ* | YOL076W | ++++ | ++++ | + | ++++ |

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| *rad6Δ* | YGL058W |  | ++++ | ++++ | ‐ | ++++ |
| *rtg1Δ* | YOL067C |  | +++ | +++ | +/‐ | ++++ |
| *scp160Δ* | YJL080C |  | ++++ | ++++ | + | ++++ |
| *sin3Δ* | YOL004W |  | ++++ | ++++ | + | ++++ |
| *sto1Δ* | YMR125W |  | ++++ | ++++ | ‐ | ++++ |
| *tho2Δ* | YNL139C | *RLR1* | +++ | +++ | ‐ | +++ |
| *tup1Δ* | YCR084C |  | +++ | +++ | ‐ | +++ |
| Ribosomal Proteins (4) |  |  |  |  |  |  |
| *rpl21aΔ* | YBR191W |  | ++++ | ++++ | +/‐ | ++++ |
| *rpl22aΔ* | YLR061W |  | ++++ | ++++ | ‐ | +++ |
| *rpl27aΔ* | YHR010W |  | ++++ | ++++ | ‐ | +++ |
| *rpp1bΔ* | YDL130W |  | ++++ | ++++ | +/‐ | ++++ |
| Unknown Function (3) |  |  |  |  |  |  |
| *fyv4Δ* | YHR059W |  | ++++ | ++++ | ‐ | +++ |
| *smi1Δ* | YGR229C |  | ++++ | ++++ | ‐ | ++++ |
|  | YDL133W |  | ++++ | ++++ | + | ++++ |

Strains from the deletion collection (Winzeler *et al.*, 1999) were screened for growth on medium containing 0.1 mg/ml hygromycin B compared to growth on medium without drug. The 156 strains listed in Tables S1A‐S1C were sensitive to hygromycin B (HB). The set of strains was further separated into three classes by the ability of KCl to suppress hygromycin B sensitive growth (0.075 – 0.1 mg/ml). The strains shown here were able to grow in the presence of hygromycin B if the medium was supplemented with 100 mM KCl. The membrane traffic mutants here were studied in more detail. See Table 3 and Figures 3 – 6 for details

# Table S1B Class II Mutants: Hygromycin B Sensitive Strains Suppressed by 500 mM KCl

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Strain | ORF | Aliases | YPAD | YPAD+500  mM KCl | HB | HB+500  mM KCl |
| WT |  |  | ++++ | ++++ | +++ | ++++ |
| Membrane Traffic Proteins  (12) |  |  |  |  |  |  |
| *arf1Δ* | YDL192W |  | ++++ | ++++ | ‐ | +++ |
| *mon1Δ* | YGL124C |  | ++++ | ++++ | ++ | ++++ |
| *pep7Δ* | YDR323C | *VPS19, VAC1* | ++++ | ++++ | ‐ | ++++ |
| *pep12Δ* | YOR036W | *VPS6, VPT13* | ++++ | ++++ | ‐ | +++ |
| *per1Δ* | YCR044C | *COS16* | ++++ | ++++ | ‐ | ++ |
| *swa2Δ* | YDR320C | *AUX1, BUD24* | ++++ | ++++ | ‐ | +++ |
| *sys1Δ* | YJL004C |  | ++++ | ++++ | ++ | ++++ |
| *vps1Δ* | YKR001C | *GRD1, VPT26* | ++++ | ++++ | ++ | +++ |
| *vps3Δ* | YDR495C | *PEP6, VPT17* | ++++ | ++++ | ‐ | +++ |
| *vps29Δ* | YHR012W | *PEP11* | ++++ | ++++ | ‐ | ++++ |
| *vps52Δ* | YDR484W | *SAC2* | ++++ | ++++ | ‐ | +++ |
| *vps75D* | YNL246W |  | ++++ | +++ | ++ | ++++ |
| Miscellaneous (7) |  |  |  |  |  |  |
| *grr1Δ* | YJR090C | *CAT80, COT2* | ++++ | ++++ | + | +++ |
| *hal3Δ* | YKR072C | *SIS2* | ++++ | ++++ | ++ | ++++ |
| *met22Δ* | YOL064C | *HAL2* | ++++ | ++++ | ++ | ++++ |
| *ncs2D* | YNL119W |  | ++++ | ++++ | ++ | ++++ |
| *slm4D* | YBR077C |  | ++++ | ++++ | ++ | ++++ |
| *snf3Δ* | YDL194W |  | ++++ | ++++ | ++ | ++++ |
| *ubx4D* | YMR067C |  | ++++ | ++++ | ++ | ++++ |
| Transcription and Replication  (2) |  |  |  |  |  |  |
| *ist3Δ* | YIR005W | *SNU17* | ++++ | ++++ | ++ | ++++ |
| *xrs2Δ* | YDR369C |  | +++ | +++ | + | +++ |
| Unknown (2) |  |  |  |  |  |  |
| *ilm1Δ* | YJR118C |  | ++++ | ++++ | ++ | ++++ |
|  | YDR161W |  | ++++ | ++++ | ++ | +++ |

As in the legend for Table S1A, strains were grown +/‐ 0.075 mg/ml hygromycin B but +/‐ 500 mM KCl. Strains that grew as well in the presence of hygromycin B and 500 mM KCl as they did without either addition were denoted Class II. Several strains grew modestly in the presence of hygromycin B if 100 mM KCl was added, but 500 mM was needed to achieve the same level of growth as in the absence of additions. Twenty‐three strains fell into Class II.

# Table S1C Class III Mutants: Hygromycin B Sensitive Strains Not Suppressed Well by 500 mM KCl

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Gene | ORF | Aliases | YPAD | YPAD+500 mM  KCl | HB | HB+500  mM KCl |
| WT |  |  | ++++ | ++++ | +++ | ++++ |
| Membrane Traffic Proteins (14) |  |  |  |  |  |  |
| *apl2Δ* | YKL135C |  | ++++ | ++++ | ‐ | + |
| *get1Δ* | YGL020C | *MDM39* | ++++ | ++++ | ‐ | ++ |
| *get2Δ* | YER083C | *RMD7* | ++++ | ++++ | ‐ | + |
| *nhx1Δ* | YDR456W | *VPS44* | ++++ | ++++ | ‐ | +/‐ |
| *pep3Δ* | YLR148W | *VPS18, VAM8* | ++++ | ++++ | ‐ | ‐ |
| *rvs161Δ* | YCR009C | *END6, FUS7* | ++++ | ++++ | ‐ | +/‐ |
| *rvs167Δ* | YDR388W |  | ++++ | ++++ | ‐ | ‐ |
| *snx3D* | YOR357C | *GRD19* | +++ | ++++ | ‐ | + |
| *tlg2Δ* | YOL018C |  | ++++ | ++++ | ‐ | +/‐ |
| *vps16Δ* | YPL045W | *VAM9, VPT16* | ++++ | ++++ | ‐ | ‐ |
| *vps33Δ* | YLR396C | *PEP14, VAM5* | ++++ | ++++ | ‐ | ‐ |
| *vps51Δ* | YKR020W | *WHI6, API3* | ++++ | ++++ | ‐ | ++ |
| *vps54Δ* | YDR027C | *LUV1* | +++ | +++ | ‐ | ‐ |
| *vps53Δ* | YJL029C |  | ++++ | ++++ | ‐ | ‐ |
| Phosphatases (3) |  |  |  |  |  |  |
| *ptc1Δ* | YDL006W | *KCS2, TPD1* | ++++ | ++++ | ‐ | ++ |
| *sac1Δ* | YKL212W | *RSD1* | ++++ | ++++ | ‐ | ‐ |
| *sit4Δ* | YDL047W | *LGN4* | +++ | +++ | + | ++ |
| Glycosylation (3) |  |  |  |  |  |  |
| *anp1Δ* | YEL036C | *MNN8, GEM3* | +++ | +++ | ‐ | ++ |
| *gup1Δ* | YGL084C |  | ++++ | ++++ | ‐ | ++ |
| *rot2Δ* | YBR229C | *GLS2* | ++++ | ++++ | ‐ | + |
| Lipid Metabolism (3) |  |  |  |  |  |  |
| *erg3Δ* | YLR056W | *SYR1, PSO6* | ++++ | ++++ | ‐ | + |
| *erg28Δ* | YER044C | *BUD18* | ++++ | ++++ | ‐ | ‐ |
| *plc1Δ* | YPL268W |  | +++ | +++ | ‐ | + |
| Miscellaneous (9) |  |  |  |  |  |  |
| *adk1Δ* | YDR226W | *AKY1* | +++ | +++ | ‐ | +/‐ |
| *bur2Δ* | YLR226W | *CST4* | +++ | +++ | ‐ | ‐ |
| *gas1Δ* | YMR307W | *GGP1, CWH52* | +++ | ++++ | ‐ | + |
| *gtr2Δ* | YGR163W |  | ++++ | ++++ | + | ++ |
| *nup133Δ* | YKR082W |  | +++ | +++ | +/‐ | ++ |

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| *pho80Δ* | YOL001W | *VAC5, TUP7* | ++++ | ++++ | ‐ | ++ |
| *pmp3Δ* | YDR276C |  | ++++ | ++++ | ‐ | ‐ |
| *shp1Δ* | YBL058W |  | ++++ | ++++ | ‐ | ++ |
| *slg1Δ* | YOR008C | *HCS77, WSC1* | ++++ | ++++ | ‐ | ++ |
| Transcription and Replication (22) |  |  |  |  |  |  |
| *bdf1Δ* | YLR399C |  | +++ | +++ | ‐ | + |
| *cdc40Δ* | YDR364C | *PRP17, SLT15* | +++ | ++++ | ‐ | ‐ |
| *ctk3Δ* | YML112W |  | +++ | +++ | ‐ | + |
| *dbp7Δ* | YKR024C |  | +++ | +++ | +/‐ | ++ |
| *dhh1Δ* | YDL160C |  | +++ | +++ | ‐ | ++ |
| *hap5Δ* | YOR358W |  | ++++ | ++++ | ‐ | + |
| *hmo1Δ* | YDR147W | *HSM2* | +++ | +++ | ‐ | ++ |
| *imp2Δ* | YIL154C |  | ++++ | ++++ | ‐ | + |
| *not5Δ* | YPR072W |  | +++ | +++ | ‐ | + |
| *pol32Δ* | YJR043C |  | ++++ | ++++ | ++ | +++ |
| *rad50Δ* | YNL250W |  | +++ | +++ | + | ++ |
| *ref2Δ* | YDR195W |  | ++ | ++ | ‐ | + |
| *rox3Δ* | YBL093C | *NUT3, SSN7* | ++++ | ++++ | ‐ | +/‐ |
| *rpb9Δ* | YGL070C | *SHI1, SSU73* | ++++ | ++++ | ‐ | + |
| *sfp1Δ* | YLR403W |  | +++ | +++ | ‐ | ‐ |
| *spt20Δ* | YOL148C | *ADA5* | +++ | +++ | ‐ | +/‐ |
| *srb2Δ* | YHR041C | *HRS2* | ++++ | ++++ | ‐ | ++ |
| *srb5Δ* | YGR104C |  | +++ | +++ | ‐ | + |
| *ssz1Δ* | YHR064C | *PDR13* | ++++ | ++++ | ‐ | ‐ |
| *taf14Δ* | YPL129W | *ANC1, SWP29* | ++++ | ++++ | ‐ | + |
| *tif4631Δ* | YGR162W |  | ++++ | ++++ | ‐ | ++ |
| *zuo1Δ* | YGR285C |  | ++++ | ++++ | ‐ | ‐ |
| Ribosomal Proteins (1) |  |  |  |  |  |  |
| *rpl31aΔ* | YDL075W |  | +/‐ | +/‐ | ‐ | ‐ |
| Unknown Function (2) |  |  |  |  |  |  |
|  | YDR532C  YOL015W |  | ++  ++++ | +++  ++++ | ‐  ‐ | +/‐  ++ |

As in the legend for Table S1B, strains were growth +/‐ 0.075 mg/ml hygromycin B and +/‐ 500 mM KCl. Strains unable to grow to the same extent in the presence of hygromycin B and 500 mM KCl as they do in the absence of the two additions were denoted as members of Class III. Fifty‐seven strains fell into this category.

# Table S2 Gene Ontology (GO) Terms

Tables S2A‐S2C are available for download at http://www.g3journal.org/lookup/suppl/doi:10.1534/g3.111.000166/‐/DC1. Tables S2A: Alphabetical Listing of Genes with Associated GO Terms

Tables S2B: Process, Function, and Component GO Terms Arranged by Significance for the Entire Set of 156 Genes Tables S2C: Process GO Terms Arranged by Significance for Genes in Each of the Three Classes

Note that each file has multiple tabs at bottom.

# Table S3 86Rb+ Uptake by Membrane Traffic Mutants of All Three Classes

Percent 86Rb+

|  |  |  |  |
| --- | --- | --- | --- |
| Class | Strain | ORF | Uptake |
|  | WT |  | 100% |
| **1** | ***arl1Δ*** | **YBR164C** | **68%** |
| **1** | ***bro1Δ*** | **YPL084W** | **8%** |
| **1** | ***chs5Δ*** | **YLR330W** | **33%** |
| **1** | ***cog5Δ*** | **YNL051W** | **67%** |
| 1 | *cog6Δ* | YNL041C | 116% |
| **1** | ***did4Δ*** | **YKL002W** | **62%** |
| 1 | *gga1Δ* | YDR358W | 81% |
| 1 | *gga2Δ* | YHR108W | 122% |
| **1** | ***glo3Δ*** | **YER122C** | **41%** |
| **1** | ***gos1Δ*** | **YHL031C** | **38%** |
| **1** | ***mon2Δ*** | **YNL297C** | **34%** |
| 1 | *pep5D* | YMR231W | 76% |
| 1 | *rer1Δ* | YCL001W | 151% |
| **1** | ***rgp1Δ*** | **YDR137W** | **56%** |
| **1** | ***ric1Δ*** | **YLR039C** | **47%** |
| **1** | ***sec22Δ*** | **YLR268W** | **49%** |
| **1** | ***stp22Δ*** | **YCL008C** | **13%** |
| **1** | ***vam3Δ*** | **YOR106W** | **75%** |
| **1** | ***vam7Δ*** | **YGL212W** | **39%** |
| **1** | ***vps4Δ*** | **YPR173C** | **55%** |
| **1** | ***vps8Δ*** | **YAL002W** | **58%** |
| **1** | ***vps9Δ*** | **YML097C** | **61%** |
| **1** | ***vps20Δ*** | **YMR077C** | **75%** |
| **1** | ***vps21Δ*** | **YOR089C** | **56%** |
| **1** | ***vps24Δ*** | **YKL041W** | **23%** |
| 1 | *vps27Δ* | YNR006W | 90% |
| **1** | ***vps30Δ*** | **YPL120W** | **47%** |
| **1** | ***vps36Δ*** | **YLR417W** | **29%** |
| **1** | ***vps41Δ*** | **YDR080W** | **52%** |
| **1** | ***ypt6Δ*** | **YLR262C** | **54%** |

|  |  |  |  |
| --- | --- | --- | --- |
| **2** | ***arf1Δ*** | **YDL192W** | **58%** |
| **2** | ***mon1Δ*** | **YGL124C** | **49%** |
| **2** | ***pep7Δ*** | **YDR323C** | **38%** |
| **2** | ***pep12Δ*** | **YOR036W** | **48%** |
| **2** | ***per1Δ*** | **YCR044C** | **52%** |
| **2** | ***swa2Δ*** | **YDR320C** | **0%** |
| **2** | ***sys1Δ*** | **YJL004C** | **66%** |
| **2** | ***vps1Δ*** | **YKR001C** | **24%** |
| **2** | ***vps3Δ*** | **YDR495C** | **33%** |
| **2** | ***vps29Δ*** | **YHR012W** | **47%** |
| 2 | *vps52Δ* | YDR484W | 109% |
| **2** | ***vps75Δ*** | **YNL246W** | **21%** |
| **3** | ***apl2Δ*** | **YKL135C** | **64%** |
| 3 | *get1D* | YGL020C | n.d. |
| 3 | *get2D* | YER083C | n.d. |
| **3** | ***nhx1Δ*** | **YDR456W** | **20%** |
| **3** | ***pep3Δ*** | **YLR148W** | **46%** |
| 3 | *rvs161D* | YCR009C | n.d. |
| 3 | *rvs167D* | YDR388W | n.d. |
| **3** | ***snx3Δ*** | **YOR357C** | **10%** |
| 3 | *tlg2Δ* | YOL018C | 276% |
| **3** | ***vps16Δ*** | **YPL045W** | **45%** |
| **3** | ***vps33Δ*** | **YLR396C** | **47%** |
| 3 | *vps51Δ* | YKR020W | 372% |
| 3 | *vps53Δ* | YJL029C | 142% |
| 3 | *vps54Δ* | YDR027C | 106% |

Cells were incubated with 86RbCl as described in the legend to Figure 2. Aliquots were removed at time 0 and at 30 min. Uptake was determined in triplicate. Each strain was tested at least twice on different days; the average of the independent determinations relative to wild type is shown. Results between experiments generally varied by less than 10%. The different mutants were sorted into 3 separate bins: strains exhibiting <75% of wild type uptake were conserved to be defective for uptake (in **bold**). Strains exhibiting >125% of wild type were considered to have excess uptake (underlined) and were examined for 86Rb+ efflux as described in the text. Strains exhibiting between >75% but <125% of wild type were considered to be indistinguishable from wild type.

# Table S4 Effect of K+ on CPY Secretion in Membrane Traffic Mutants

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Class | Gene | ORF | CPY | CPY on  KCl | CPY on Sorb |
| ‐ | WT | ‐ | ‐ | ‐ | ‐ |
| 1 | *arl1Δ* | YBR164C | +++ | +/‐ | +/‐ |
| 1 | *bro1Δ* | YPL084W | ++++ | ‐ | ‐ |
| 1 | *chs5D* | YLR330W |  | *non‐secretor* |  |
| 1 | *cog5Δ* | YNL051W | ++++ | + | ‐ |
| 1 | *cog6Δ* | YNL041C | ++++ | + | ‐ |
| **1** | ***did4Δ*** | **YKL002W** | **+++** | **‐** | **++** |
| 1 | *gef1D* | YJR040W | ++ | ‐ | ‐ |
| 1 | *gga1Δ* | YDR358W | +++ | ‐ | ‐ |
| 1 | *gga2Δ* | YHR108W | +++ | ‐ | ‐ |
| 1 | *glo3D* | YER122C | +/‐ | ‐ | ‐ |
| 1 | *gos1Δ* | YHL031C | ++++ | ++ | + |
| 1 | *mdm20D* | YOL076W | +/‐ | ‐ | ‐ |
| 1 | *mon2D* | YNL297C | +/‐ | +/‐ | +/‐ |
| 1 | *pep5Δ* | YMR231W | ++++ | ++++ | ++++ |
| 1 | *rer1D* | YCL001W |  | *non‐secretor* |  |
| 1 | *rgp1Δ* | YDR137W | ++++ | + | + |
| 1 | *ric1Δ* | YLR039C | ++++ | +++ | +++ |
| 1 | *sec22Δ* | YLR268W | +++ | + | + |
| 1 | *stp22Δ* | YCL008C | ++++ | ‐ | ‐ |
| 1 | *vam3D* | YOR106W | +/‐ | ‐ | ‐ |
| 1 | *vam7Δ* | YGL212W | ++++ | +++ | +++ |
| 1 | *van1D* | YML115C | ++ | ‐ | ‐ |
| 1 | *vph2Δ* | YKL119C | +++ | ‐ | ‐ |
| **1** | ***vps4Δ*** | **YPR173C** | **++++** | **+/‐** | **+++** |
| **1** | ***vps8Δ*** | **YAL002W** | **++++** | **+/‐** | **+++** |
| **1** | ***vps9Δ*** | **YML097C** | **++++** | **+/‐** | **+++** |
| 1 | *vps20Δ* | YMR077C | ++++ | +/‐ | +/‐ |
| 1 | *vps21Δ* | YOR089C | ++++ | + | + |
| **1** | ***vps24Δ*** | **YKL041W** | **++++** | **+/‐** | **+++** |
| **1** | ***vps27Δ*** | **YNR006W** | **++++** | **+/‐** | **+++** |
| 1 | *vps30Δ* | YPL120W | ++++ | ++++ | ++++ |
| 1 | *vps36Δ* | YLR417W | ++++ | + | + |
| 1 | *vps41Δ* | YDR080W | ++++ | ++ | ++ |
| 1 | *ypt6Δ* | YLR262C | ++++ | +++ | ++++ |

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| 2 | *arf1Δ* | YDL192W | ++++ | +++ | +++ |
| 2 | *mon1D* | YGL124C | +++ | +++ | ++ |
| **2** | ***pep7Δ*** | **YDR323C** | **++++** | **‐** | **++** |
| **2** | ***pep12Δ*** | **YOR036W** | **++++** | **‐** | **++** |
| 2 | *per1Δ* | YCR044C | +++ | ‐ | ‐ |
| 2 | *swa2Δ* | YDR320C | +++ | + | + |
| 2 | *sys1Δ* | YJL004C | ++++ | + | + |
| **2** | ***vps1Δ*** | **YKR001C** | **++++** | **+** | **+++** |
| **2** | ***vps3Δ*** | **YDR495C** | **++++** | **‐** | **+++** |
| 2 | *vps29Δ* | YHR012W | ++++ | ++++ | ++++ |
| 2 | *vps52Δ* | YDR484W | ++++ | +++ | ++++ |
| 2 | *vps75Δ* | YNL246W | ++++ | +/‐ | +/‐ |
| 3 | *nhx1Δ* | YDR456W | ++++ | ++++ | ++++ |
| 3 | *pep3Δ* | YLR148W | ++++ | +++ | +++ |
| 3 | *ptc1D* | YDL006W | ++ | ‐ | ‐ |
| 3 | *snx3D* | YOR357C | ++ | ‐ | ‐ |
| 3 | *tlg2Δ* | YOL018C | ++++ | ++++ | ++++ |
| 3 | *vps16Δ* | YPL045W | ++++ | ++++ | +++ |
| 3 | *vps33Δ* | YLR396C | ++++ | ++ | ++ |
| 3 | *vps51Δ* | YKR020W | ++++ | +++ | +++ |
| 3 | *vps53Δ* | YJL029C | ++++ | ++++ | ++++ |
| 3 | *vps54Δ* | YDR027C | ++++ | +++ | +++ |

The membrane traffic mutants in Classes I, II, and III were compared to the strains known to secrete CPY (Bonangelino *et al.*, 2002). We included in our analysis 5 strains obtained in our screen which secrete CPY but do not have GO terms that connote membrane traffic (*gef1D*, *mdm20D*, *van1D*, *vph2D* and *ptc1D;* see Tables S2A). All were grown overnight in rich medium, diluted to 1.0 OD600/ml, then subjected to serial 10‐fold dilutions. Cells were spotted onto rich medium without or with the additions of 0.5 M KCl or 1 M sorbitol using a replicator tool and grown overnight at 30oC. The next day, cells were overlaid with a nitrocellulose filter. After 15 ‐18 h, the filter was removed, washed free of cells, and prepared for Western analysis using a monoclonal anti‐CPY antibody (Roberts *et al.*, 1991). Strains are listed by class as in Tables S1A‐C and strains in which KCl specifically suppressed secretion are highlighted in bold. ‐ = no secretion, +/‐ = minimal secretion, + or ++ = moderate secretion, and +++ or ++++ = large amounts of secreted CPY.