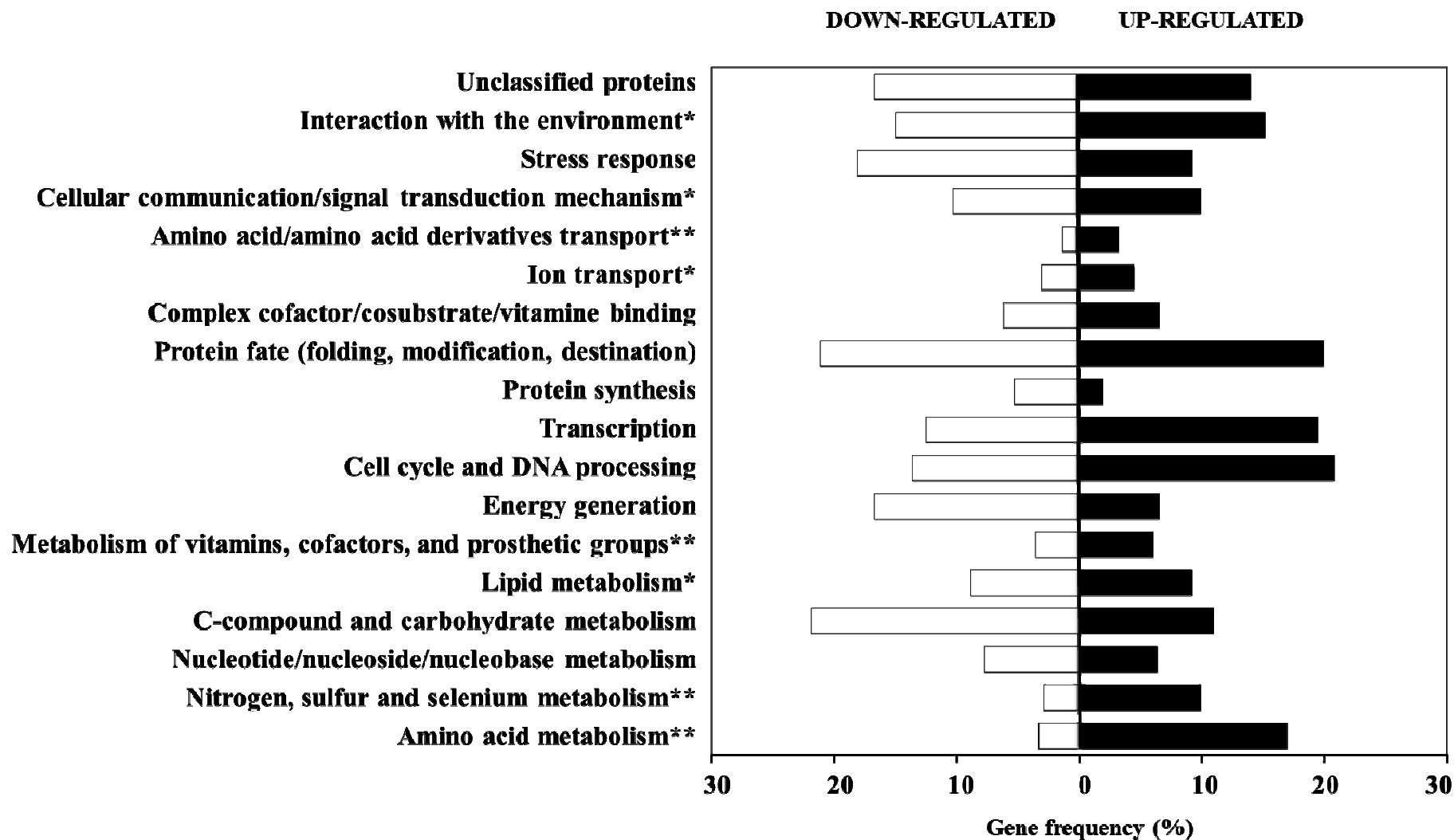
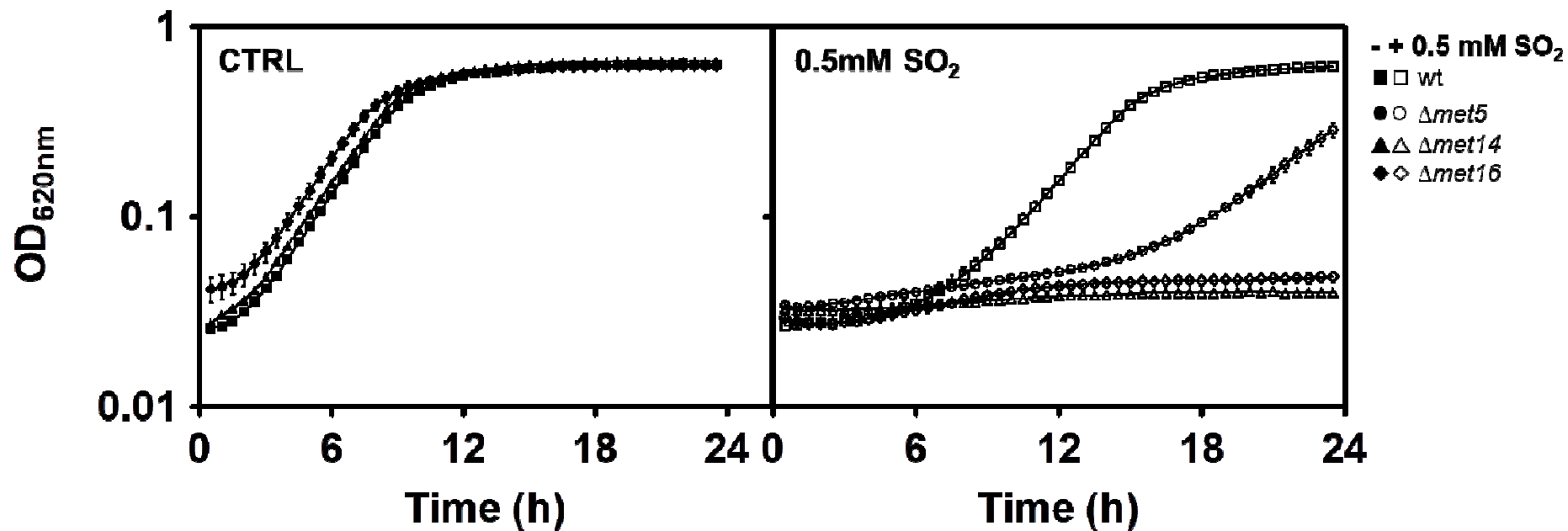


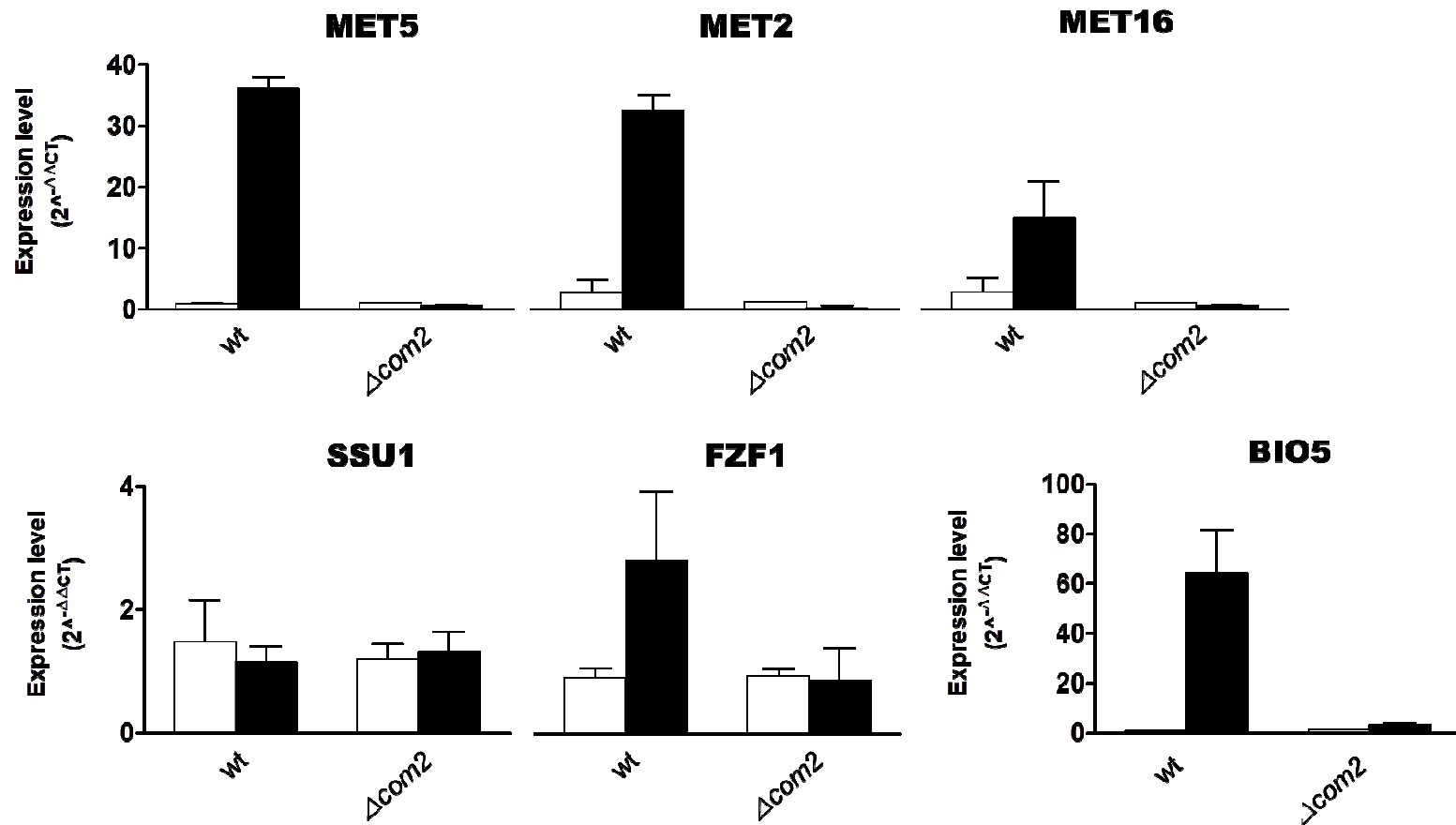
**Supplementary Fig. S1 - (A)** Growth curves of *S. cerevisiae* BY4741 (■, □) and of the deletion mutant  $\Delta com2$  (●, ○) in MMB medium (pH 3.5) (■, ●) or in the same basal medium supplemented with 0.5mM SO<sub>2</sub> (pH 3.5) (□, ○). The arrows indicate the time of cultivation at which cell sample were harvested to compare the transcriptomes. Growth was followed by measuring culture OD<sub>600nm</sub> and the concentration of viable cells was assessed as the number of colony-forming units/mL of cell culture (CFU mL<sup>-1</sup>). The growth curves shown are means of, at least, three independent growth curves that gave rise to the same growth pattern. **(B)** PCA analysis of global transcriptional response, showing variation in expression levels between the parental strain *S. cerevisiae* BY4741 (blue) and in the  $\Delta com2$  mutant (green), in cells untreated (filled marks) or treated with 0.5 mM SO<sub>2</sub> (open marks).



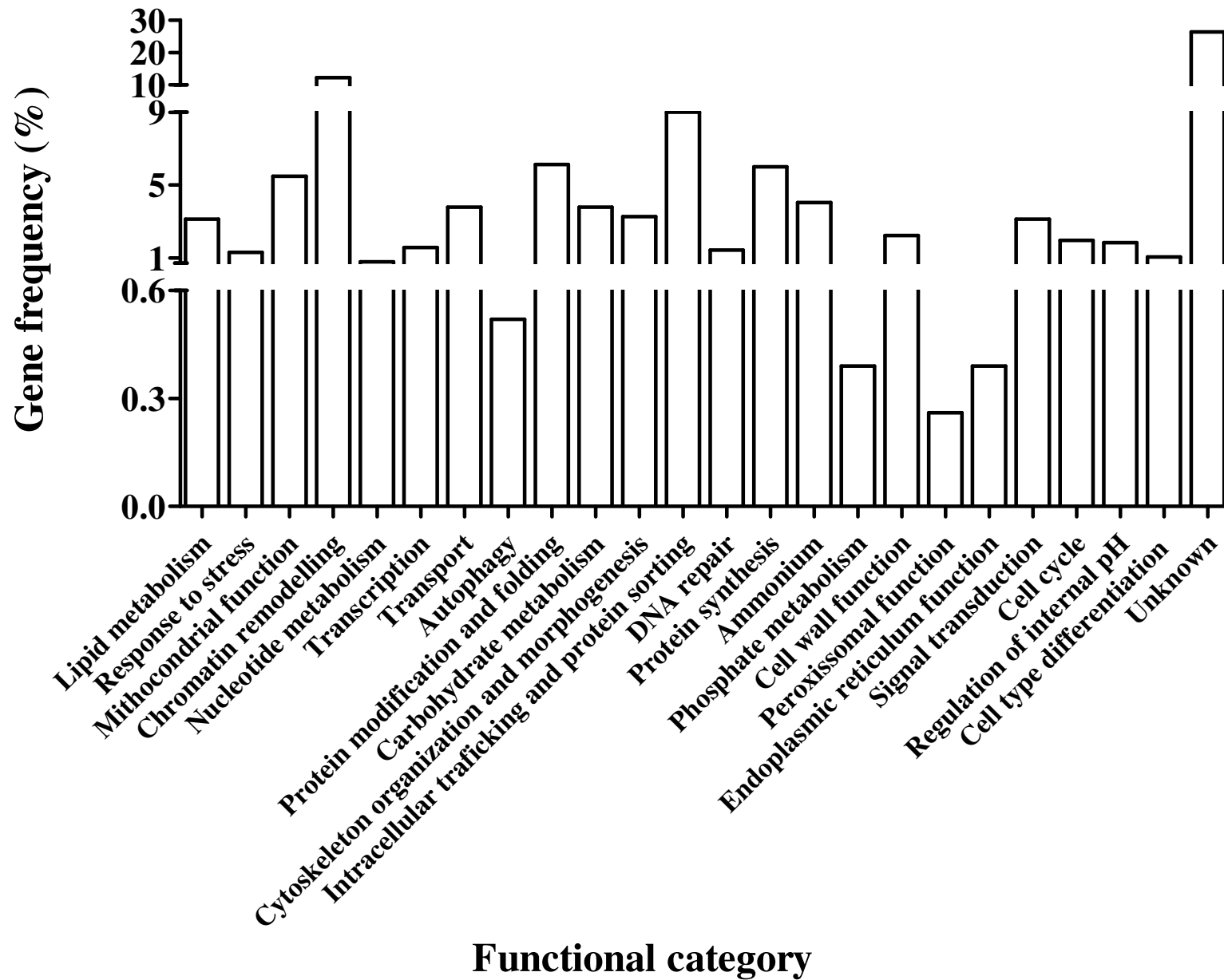
**Supplementary Fig. S2** – Functional clustering, of genes up- (black bars) or downregulated (white bars) in *S. cerevisiae* BY4741 cells cultivated for 60 min in the presence of 0.5 mM sulphur dioxide (at pH 3.5). The classification into the different classes was performed using MIPS functional catalogue and the enriched functional classes (those having an associated p-value below 0.01) are marked with \*.



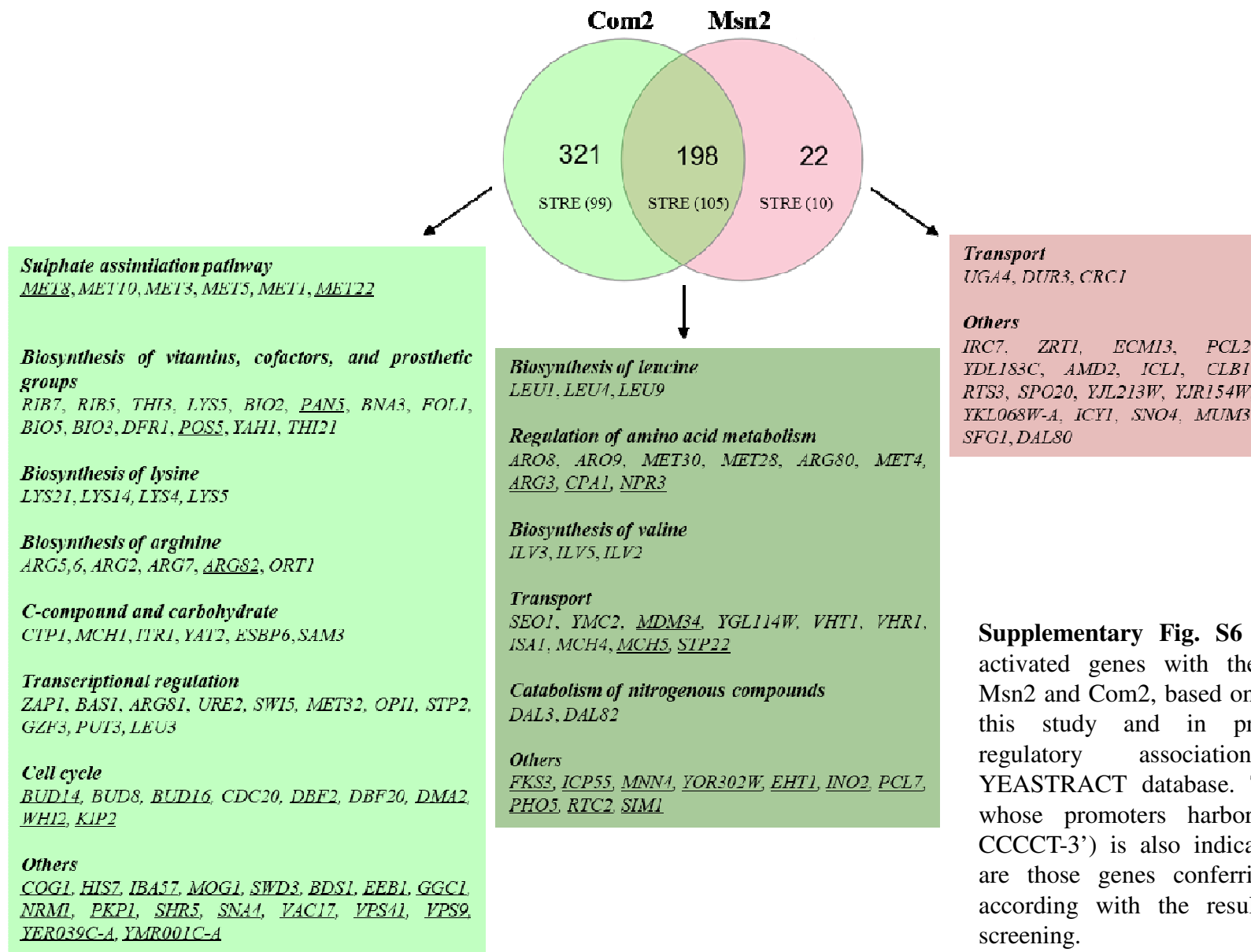
**Supplementary Fig. S3** - Comparison of the susceptibility towards SO<sub>2</sub> of the prototrophic *Saccharomyces cerevisiae* yfg $\Delta$ 0 strain and of the derived mutants  $\Delta met14$ ,  $\Delta met16$  and  $\Delta met5$ . Cells of the parental strain (wt) and of the indicated deletion mutants were grown until mid-exponential phase in liquid MMB medium (at pH 3.5) and then used to inoculate the same basal medium either supplemented (open symbols) or not supplemented with 0.5 mM SO<sub>2</sub> (filled symbols) (at pH 3.5). Cells were batch cultured at 30°C and growth was monitored based on OD<sub>620nm</sub>. The growth curves presented are representative of at least three independent growth experiments.



**Supplementary Fig. S4** - Comparison of the transcript levels, estimated by real-time RT-PCR, of MET5, MET2, MET16, BIO5, SSU1 and FZF1 genes produced in cells of the parental strain BY4741 or the  $\Delta com2$  mutant during cultivation for 60 min in MMB growth medium (at pH 3.5) unsupplemented (white bars) or supplemented with 0.5mM  $SO_2$  (black bars). The expression of each gene determined by real-time qRT-PCR was normalized to the expression of the house-keeping gene, *ACT1* (encoding the structural protein actin).

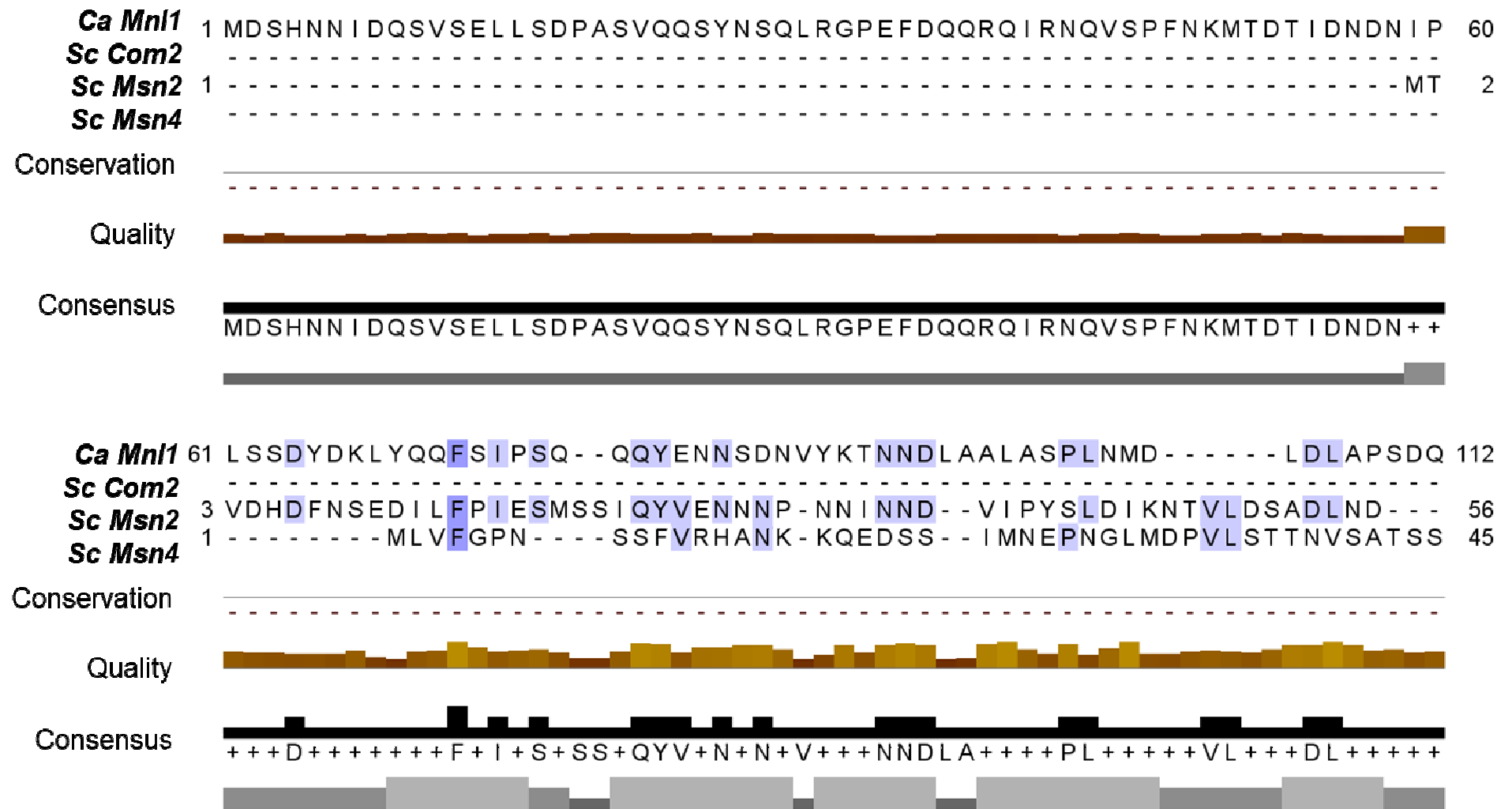


**Supplementary Fig. S5** – Functional categorization of genes whose deletion lead to increased sensitivity to 0.5, 1 or 1.5 mM of SO<sub>2</sub> (at pH 3.5). For each functional class the frequency of genes present in our dataset of SO<sub>2</sub>-resistance genes is shown.



**Supplementary Fig. S6** – Clustering of SO<sub>2</sub>-activated genes with the transcription factors Msn2 and Com2, based on the results obtained in this study and in previously documented regulatory associations described in YEASTRACT database. The number of genes whose promoters harbor a STRE-motif (5'-CCCCT-3') is also indicated. Underlined genes are those genes conferring tolerance to SO<sub>2</sub>, according with the results of the disruptome screening.

**Supplementary Fig. S7** - Amino acid sequence alignment of ScMsn2, ScMsn4, ScCom2 and *Candida albicans* Mnl1. The picture depicts the similarity registered at the level of the DNA-binding domain of these three regulators (highlighted as DBD in the end of the alignment) as well as the lack of similarity registered in domain HD1 comprising the nuclear export signal of Msn2. The two mapped regions are according with the motifs mapping described by Pfanzagl et al., 2018. For the alignment, the software “Multiple Sequence Alignment Clustal Omega” was applied. Refinements were conducted with the software “Jalview.” The blue color intensity correlates with the degree of conservation.

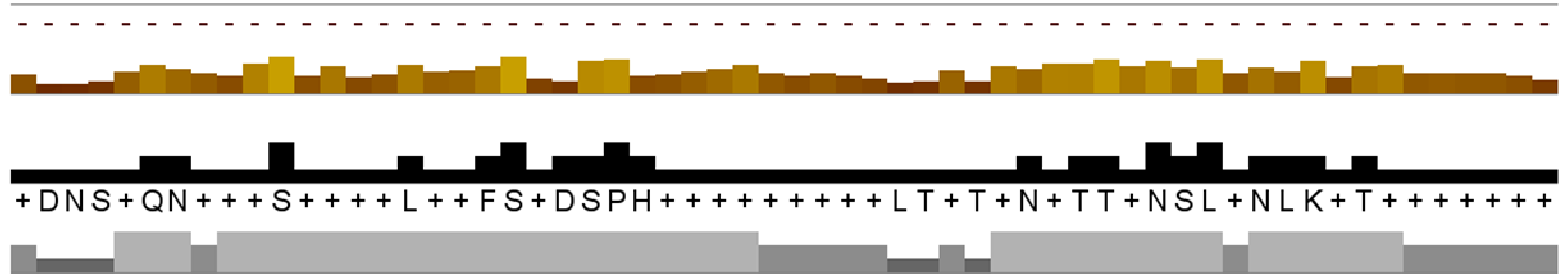


**Ca Mnl1** 113 Q - - QQP - QQ **S**QQQFL **N**DFSNL **L**PHLNNSQEQVQ - - - - **L**NGLT **S**N I **L**SNPQITVNMPGAY 164  
**Sc Com2** - - - - -  
**Sc Msn2** 57 - - - - I QNQET **S**LNLGL **P**PLS **F**DSPLPVTE - - - - - T - IPSTTD **N**SL - HLKADS - - - - - 97  
**Sc Msn4** 46 NDNSANNS I S **S**PEYTFGQF **S**MDS **P**HR **T**DATNTPI **L**TATTNTT **A**NS **L**MNLKDTASLATNW 105

Conservation

Quality

Consensus

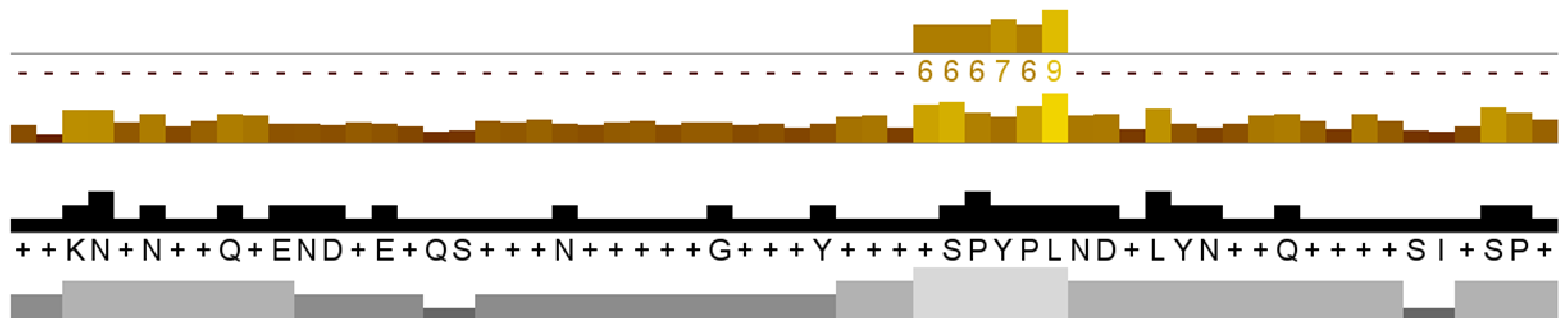


**Ca Mnl1** 165 H - Q **N**YNCDQME - - - - - - - - - - IDDSP **P**FT IES **D**LYFEQQTDNN **S**IVTSN 202  
**Sc Com2** 1 - - - - - - - - - - MSLYPL - - - - - 6  
**Sc Msn2** 98 - NKNRDART I **E**NDSEI - - KST **N**NASGSGANQY **T**TLTSPYPMND I **L**YNMNNPLQS - - P **S**PS 152  
**Sc Msn4** 106 KWKN **S**NNAQFVNDG **E**KQSSNANGKKN **G**GDKIYSSVAT **P**QALNDEL **K**NL **E**QLEKVV - - F **S**PM 163

Conservation

Quality

Consensus



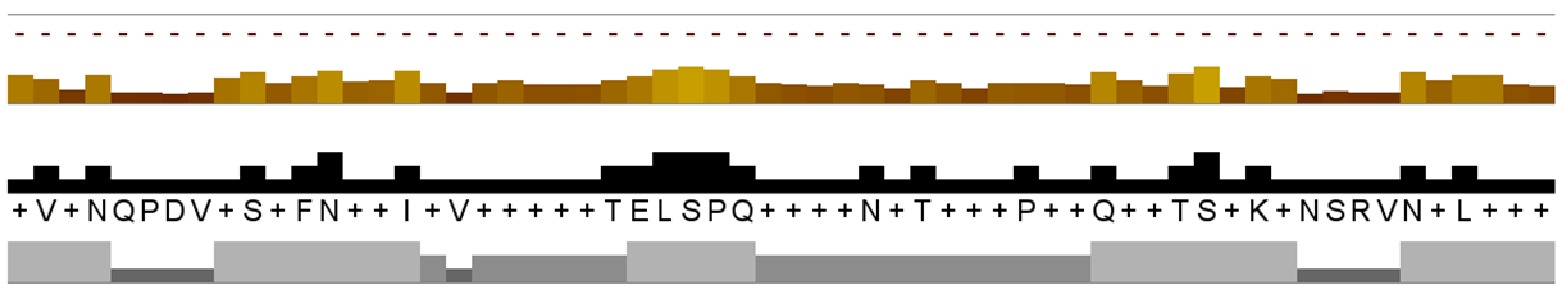


**Ca Mnl1** 203 RVHNQPDVESMFNNQLSVPSLQRTELSPPQHETNVT TGRPQESAFGSSFDNNSRVSSFANV 262  
**Sc Com2** -----  
**Sc Msn2** 153 SVPQ- - - NPTINPPIN-TASNETNLSPQTSNGNETL I SPRAQQHTS I KD- - - NRLSLP 203  
**Sc Msn4** 164 NPIN- - - DSHFNENI- - - - - ELSPH- - - - - QHATSPKT- - - - NLLEAE 194

Conservation

Quality

Consensus

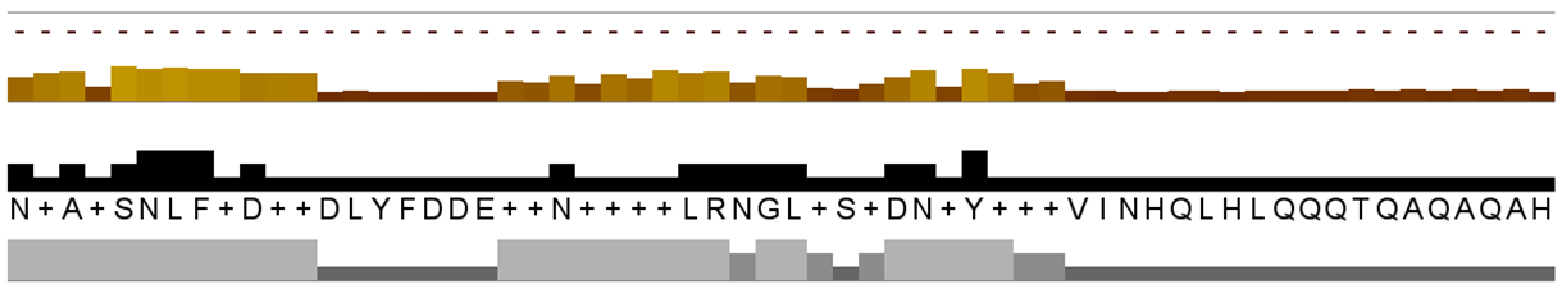


**Ca Mnl1** 263 NNAGTNL FVNGKDL YFDDESHHRTGRLRNGS I - - DSY Y AANVINHQLHLQQQTQAQAQAH 320  
**Sc Com2** -----  
**Sc Msn2** 204 NGANSNL F I DTN- - - - - PNNLNEKLRNQLNSDTNSY SNS - - - - - 237  
**Sc Msn4** 195 PS I YSNL F LDAR- - - - - LPNNANSTT - GL - - NDNDY N - - - - - 223

Conservation

Quality

Consensus

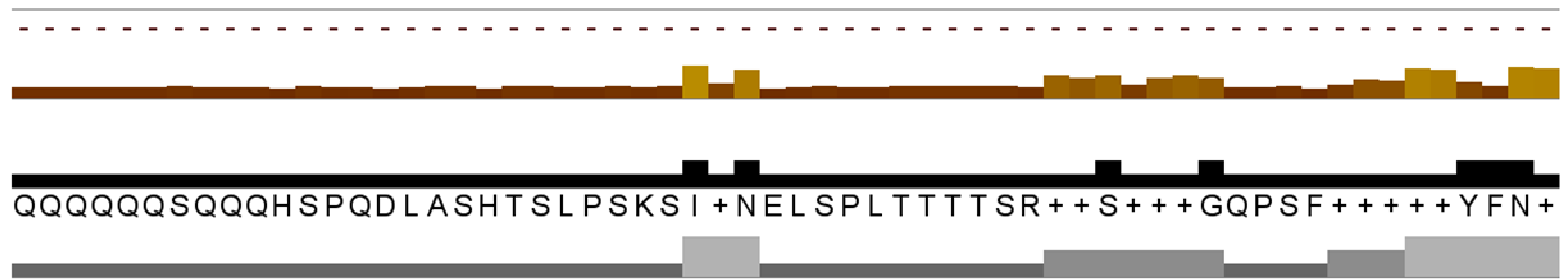


**Ca Mnl1** 321 QQQQQQSQQQHSPQDLASHTSLPSKSIYNELSPLTTTTSRASSIQSGQPSFFSAQQYFSR 380  
**Sc Com2** -----  
**Sc Msn2** 238 -----ISN-----SNSNSTG-----NLNSSYFNS 256  
**Sc Msn4** 224 -----LDD-----TNDNT 232

Conservation

Quality

Consensus



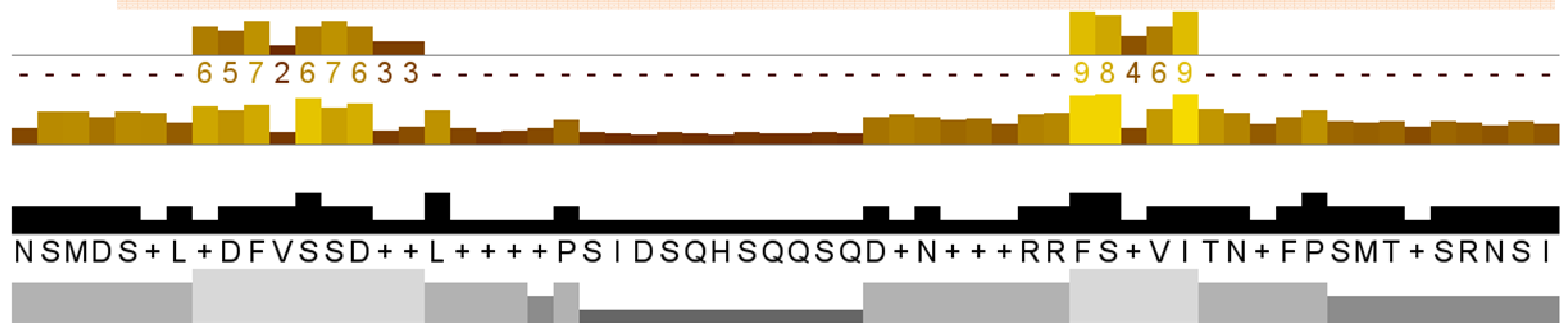
**HD1**

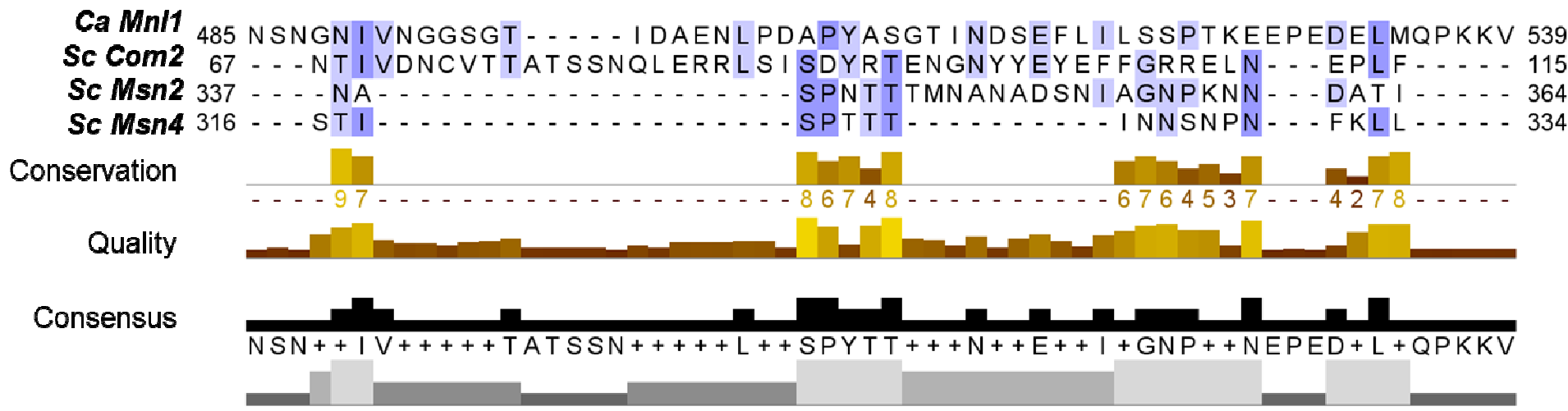
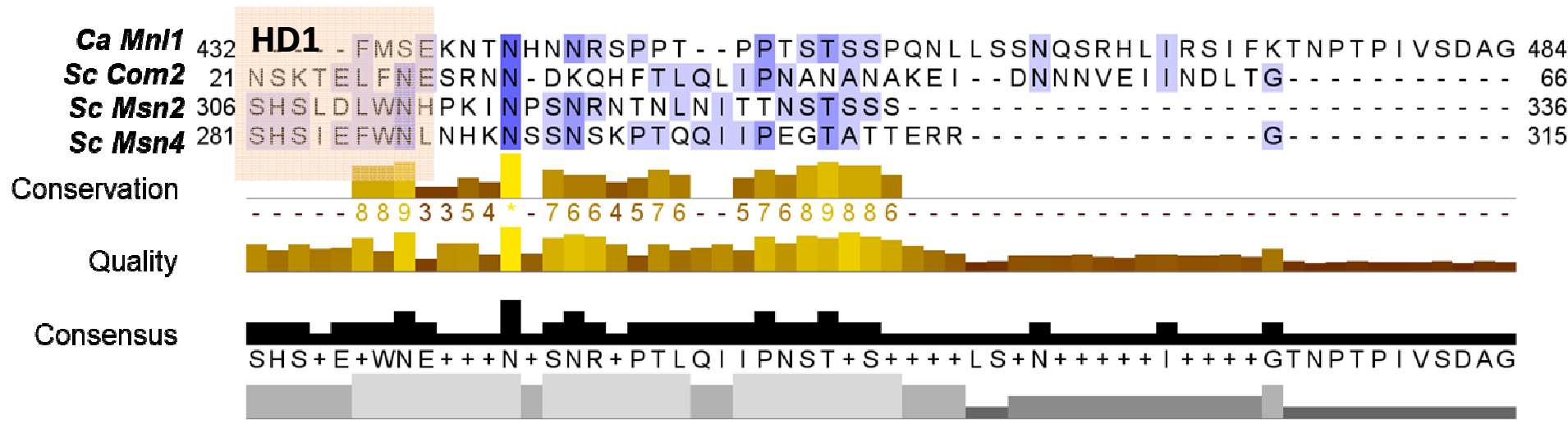
**Ca Mnl1** 381 NSMDQVPSSLHRPSFDLYNHRPSIDSQHSQQSQQRNARYTSFTSSISNILP----- 431  
**Sc Com2** 7 -----QRFESNDTV-----FSYTL----- 20  
**Sc Msn2** 257 LNIDSM LDDYVSSDLLLNDDDD-----DTNLSRRRFSDVITNQFP SMTNSRNSI 305  
**Sc Msn4** 233 NSMQSILEDVFSSEEALKFM-P-----DAGR DARRYSEVVTSSFP SMTDSRNSI 280

Conservation

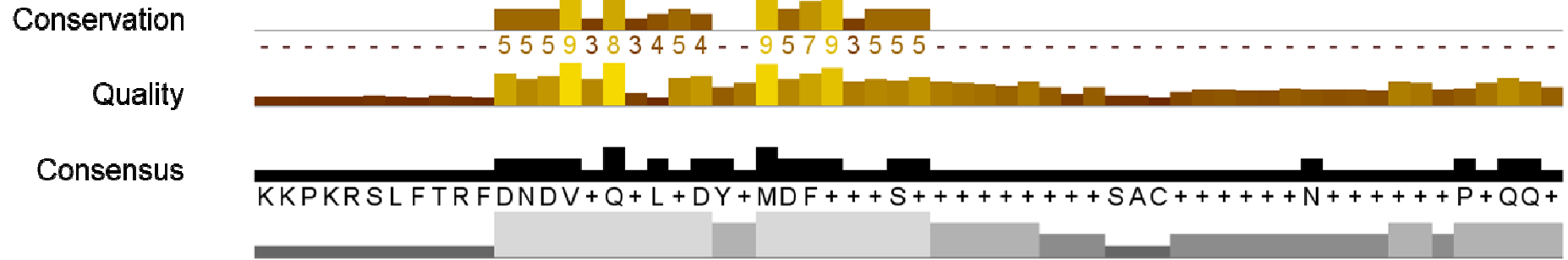
Quality

Consensus

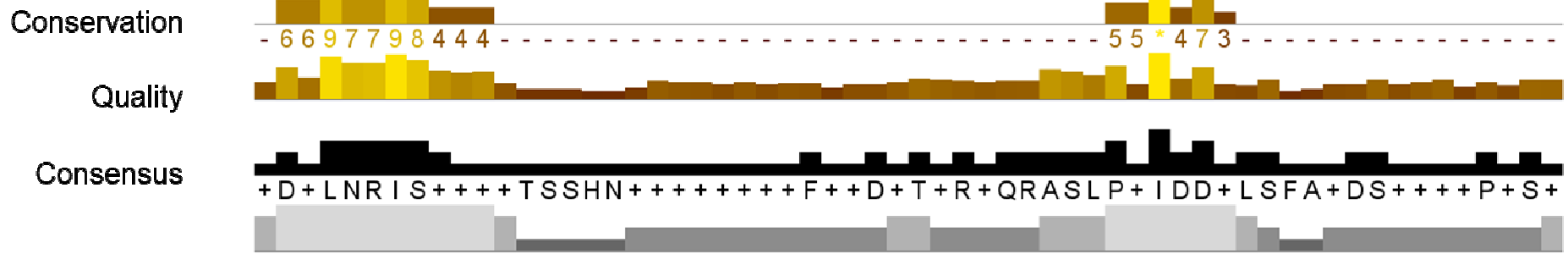




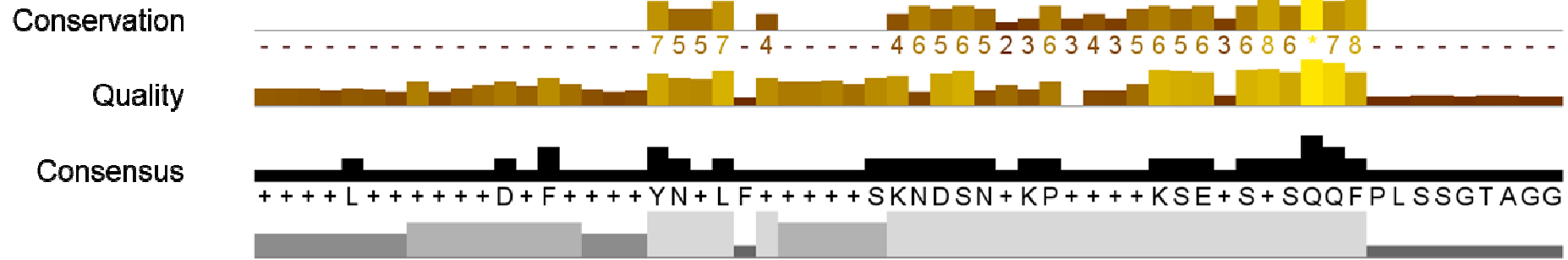
**Ca Mnl1** 540 KKPKRSLFTRFKTPVKQEQPDSEMAVSAPVD - - - - - ELKPDETV 578  
**Sc Com2** 116 - - - - - NNDIVENDDD - - IDLNNESDVLMVS - - - - - DDELEVNERFSF - LKQP 155  
**Sc Msn2** 365 - - - - - DNELTQILNEYNMNFNDNLGTSTSGKNKSACPSSFDANAMTKINPSQQL 413  
**Sc Msn4** 335 - - - - - DHDVSQALSGYSMDFSKDSGITKPKSIS - - - - - 362



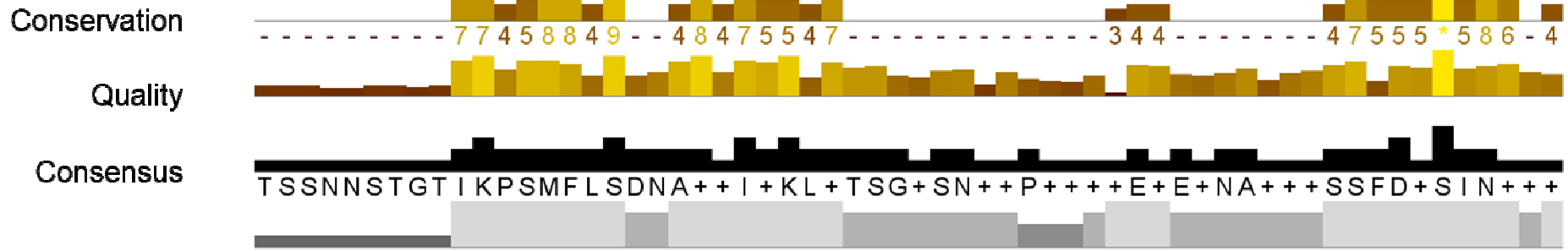
**Ca Mnl1** 579 IDMVGNISSHHS - - - - - SGTPSITTG - - - - - A 600  
**Sc Com2** 156 LDGLNRISSTNN - - - - - LKNLEIHEFIIDPT - - - - - ENIDDEL - - EDSFTTVPQSK 199  
**Sc Msn2** 414 QQQLNRVQHKQLTSSHNNSSSTMKSFNSDLYSRRQRASLPIIDDSLS - - - - - 460  
**Sc Msn4** 363 - SSLNRISSHSS - - - - - STTRQRASLPIHDIESFANDSVMANPLSD 403



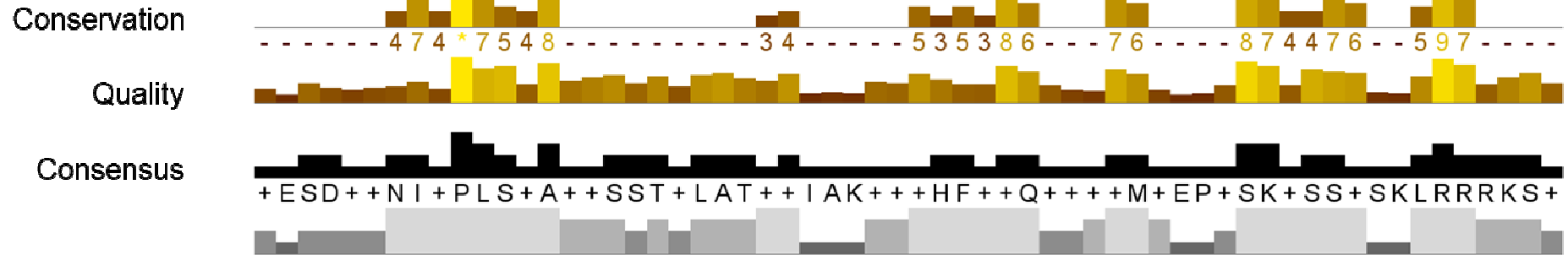
**Ca Mnl1** 601 GGNTLEHSISESSFQEPDYAALFENVGKRKNKSYRKPCKGKTKEEEQQQQQLPLSSGTAGG 660  
**Sc Com2** 200 - - - - - KKVRDYFK - - - LNIF - GSSSSSNNNSNSLGCPEIQTENSSSQKM - - - - - 239  
**Sc Msn2** 461 - - - - - - - - - - - YDLV - N - - - - KQDEDPKNDMLPNSNLSSSQQF - - - - - 487  
**Sc Msn4** 404 SASFLSEENEDDAFGALNYNSL - DATTMSAFDNNVDPFNILKSSPAQDQQF - - - - - 453



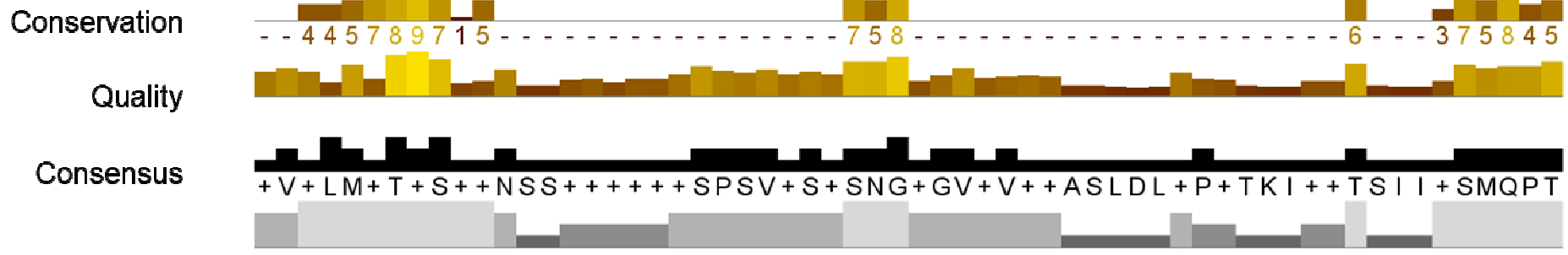
**Ca Mnl1** 661 TSSNNSTGTVEKTLFFN - - KTKIKKEPASERSLL - - - DNASAGNASASSSEASSIQNAI 715  
**Sc Com2** 240 - - - - - - - FKNRFFRSRKSTLIKSLP - - - - - LEQENEVLINSGFDVSSNEES 278  
**Sc Msn2** 488 - - - - - - - IKPSMILSDNASVIAKVATTGLSNDMPFLTEEGEQNANSTPNFDLSITQ - M 537  
**Sc Msn4** 454 - - - - - - - IKPSMMLSDNASAAAKLATSGVDNITPTPAFQR - - - - - RSYDISMNSSF 497



**Ca Mnl1** 716 G - SDDHNGP **PL**TS**A**PT**S**S**T** - LANAS - - - - - KRILSS - - - KL - - - I **SK**KK**S**T**S**KL**L**KEYTAA 762  
**Sc Com2** 279 DE**S**D**H**A**I** I**N****P**L**K**L**V**G**N**N - K**D**I**S**T**Q**S**I**A**K**T**T**N**P**F**K**S**G**S**D**F**K**M**I**E**P**V **S**K**F****S**N**D** - - **S**R**K** - - - - 331  
**Sc Msn2** 538 - - - - - **N**M**A****P**L**S**P**A**S**S**S**S**T**S**L**A**T**N**H - - - F**Y**H**H**F**P**Q**Q**G**H**T**M**N - - - **S**K**I**G**S**S - - **L**R**R**R**K**S - 582  
**Sc Msn4** 498 - - - - - **K**I**L****P**T**S**Q**A** - - - - - **H**H - - - A**A**Q**H**H**Q**Q**Q** - - **P**T**K**Q - - - A**T**V**S**P**N** - - **T**R**R**R**K**S**S** 532



**Ca Mnl1** 763 E L E D M C T V S S L - - - - - E T P V A T M I **S**K**G**I **E**V**E**V**D**L**A**S**L**D**L**P**P**D**T**K**I**F**P**T**S**I I N N K N R T 814  
**Sc Com2** 332 - - D L L A A I S E P S S P S P S A P S P S V Q S S S S S H G L V V R K - - - - - K - - - - - T - - - G S M Q K T 374  
**Sc Msn2** 583 A V P L M G T V P L T N - - Q Q N N I S S S S V N S T G N G A G V T K E R - - - - - R P S - - - Y R R - - - K S M T P S 629  
**Sc Msn4** 533 S V T L S P T I S H N N - - - - - N N G - K - - - V P V Q P R 554



### DBD

