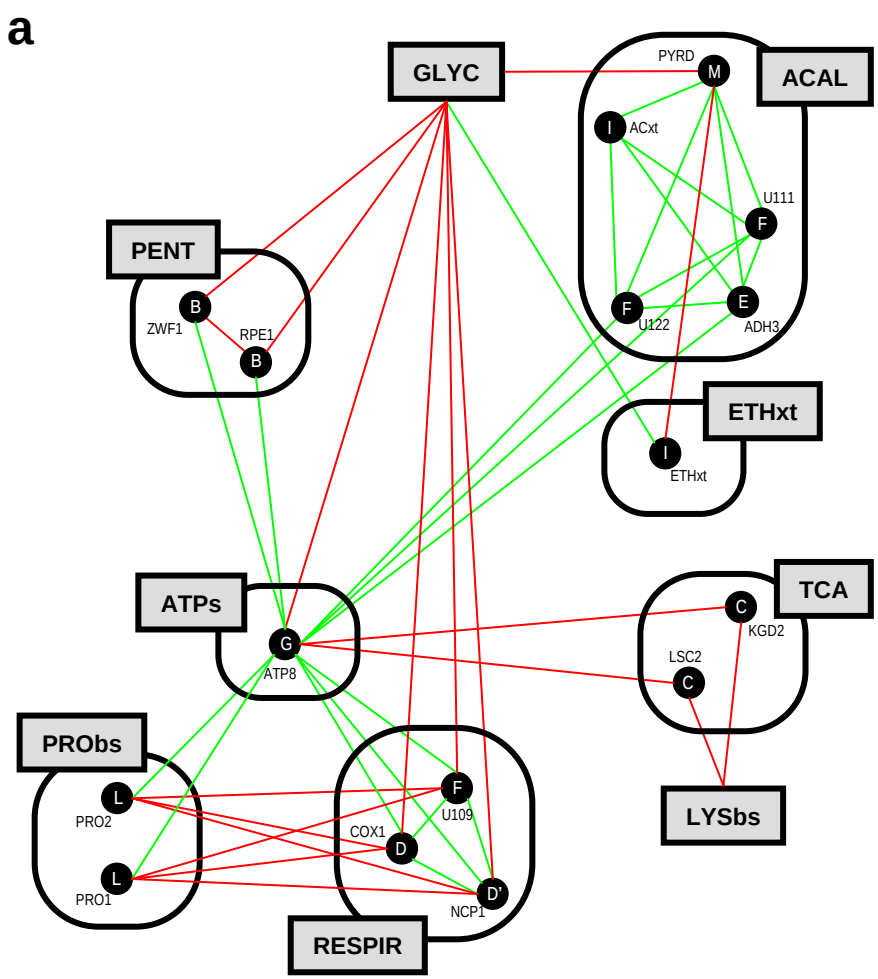
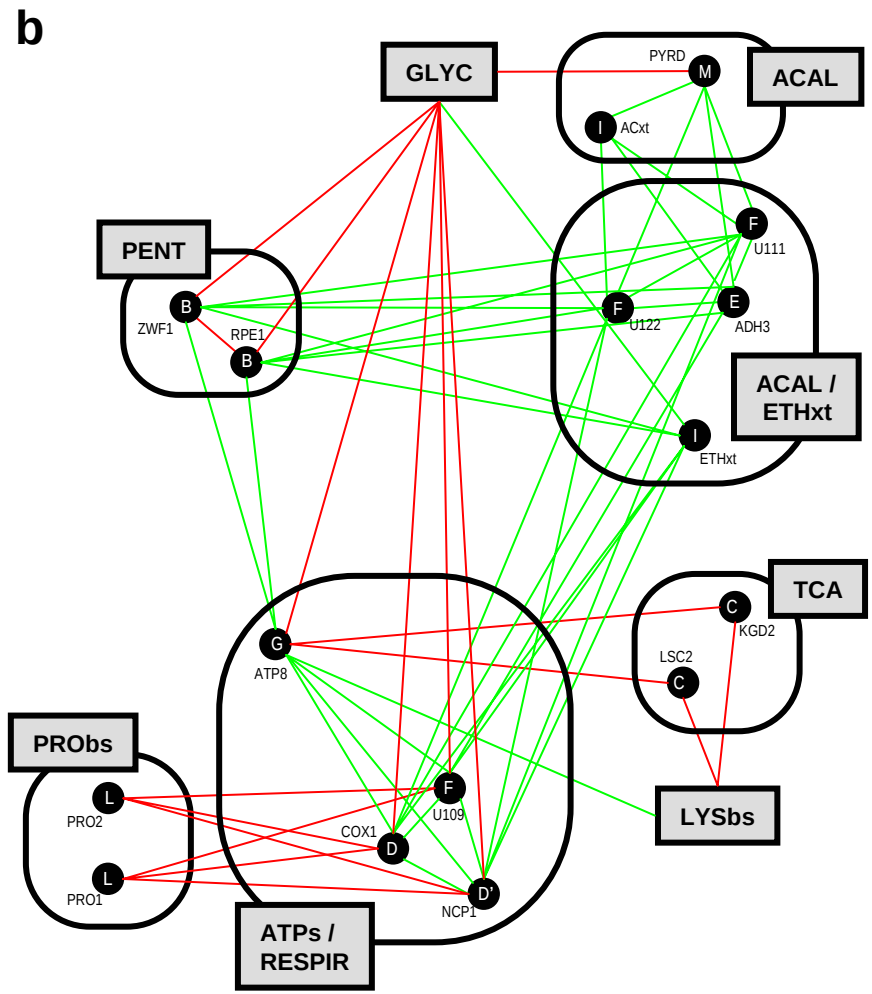
Supplementary Figure 5





Supplementary Fig. 5. Changes in monochromatically interacting epistasis modules following variation of oxygen uptake rate. A portion of the yeast FBA network obtained with Prism is shown **(a)** under nominal settings and **(b)** for a perturbation reducing the oxygen availability by half ("O-" perturbation, **Supplementary Table 1**). For the complete picture of the network see **Fig. 4a**. All aggravating (red) and buffering (green) links between the shown genes are indicated; the rest of the network remains unchanged when switching conditions. FBA predicts that the O_2 limitation results in a ten-fold reduction of the flux through the respiratory chain and a 50-fold decrease in the ATP synthetase flux. Consequently, energy generation is exclusively dependent on fermentative metabolism in the low oxygen network. As an epistatic interaction network is context-dependent, it is interesting to note that differences between these conditions occur exclusively in the energy-producing pathways shown here (fermentation and mitochondrial respiratory metabolism), as would be expected. Furthermore, changes in the gene-gene interaction network after O_2 limitation lead to a different grouping of genes into monochromatically interacting Prism modules. For example, the buffering interaction between ATP8 and proline biosynthesis genes becomes neutral in the O_2 limited network (b), allowing a monochromatic grouping of ATP8 with related respiratory chain genes, which have aggravating links with Proline biosynthesis (in the nominal conditions, such grouping of ATP8 and *RESPIR* would violate monochromaticity with *PRObs*). In this picture (b), the complete oxidative phosphorylation pathway is grouped in a single Prism module (*ATPs*/*RESPIR*). In the nominal conditions analysis (a), these modules are partially coupled through cofactor oxidation, proton pumping and ATP synthesis; however, as shown in **Supplementary Fig. 4b**, the respiratory chain has the additional role of oxidizing NADPH through *NCP1*. This activity contributes less to fitness when oxygen is further limited, where the respiratory chain has the exclusive role of providing mitochondrial ATP. This functional coupling is reflected in the clustering of *ATPs* and *RESPIR* genes. In a different example, the disappearance of the aggravating link between ETHxt and PYRD, together with the emergence of a new collection of buffering links connecting *ACAL*/*ETHxt* with *PENT* and *RESPIR*, leads to the division of the *ACAL* module. Interestingly, all the elements

required for fermentation and mitochondrial ethanol metabolism (ETHxt, ADH3, U111, and U122) are grouped in a single Prism module in the low oxygen case. Under the extreme O_2 limitation condition, pentose phosphate pathway, mitochondrial ethanol metabolism, and respiratory chain are all processes that have a low contribution to fitness and buffer one another because their unique role is the oxidation of mitochondrial NADH and cytosolic NADPH. Functional modules are named as in **Fig 4a** to reflect the main common metabolic processes of the genes involved: *ACAL* – acetaldehyde and ethanol metabolism, *ATPs* – ATP synthase, *ETHxt* – ethanol transport, *GLYC* – glycolysis, *LYSbs* – lysine biosynthesis, *PENT* – pentose phosphate pathway, *PRObs* – proline biosynthesis, *RESPIR* – respiratory chain, *TCA* – TCA cycle.

(For references, see **Supplementary References** online)