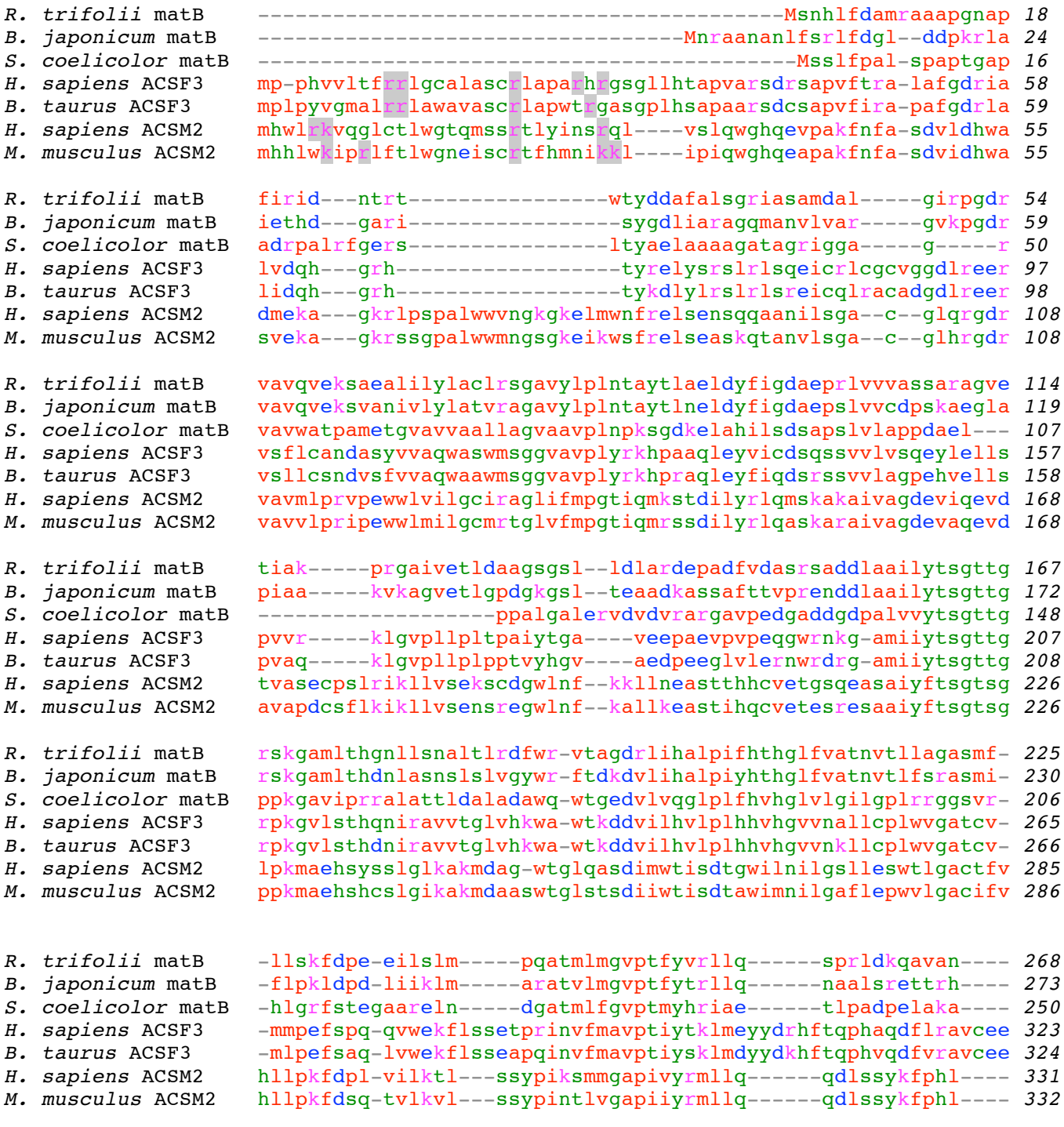
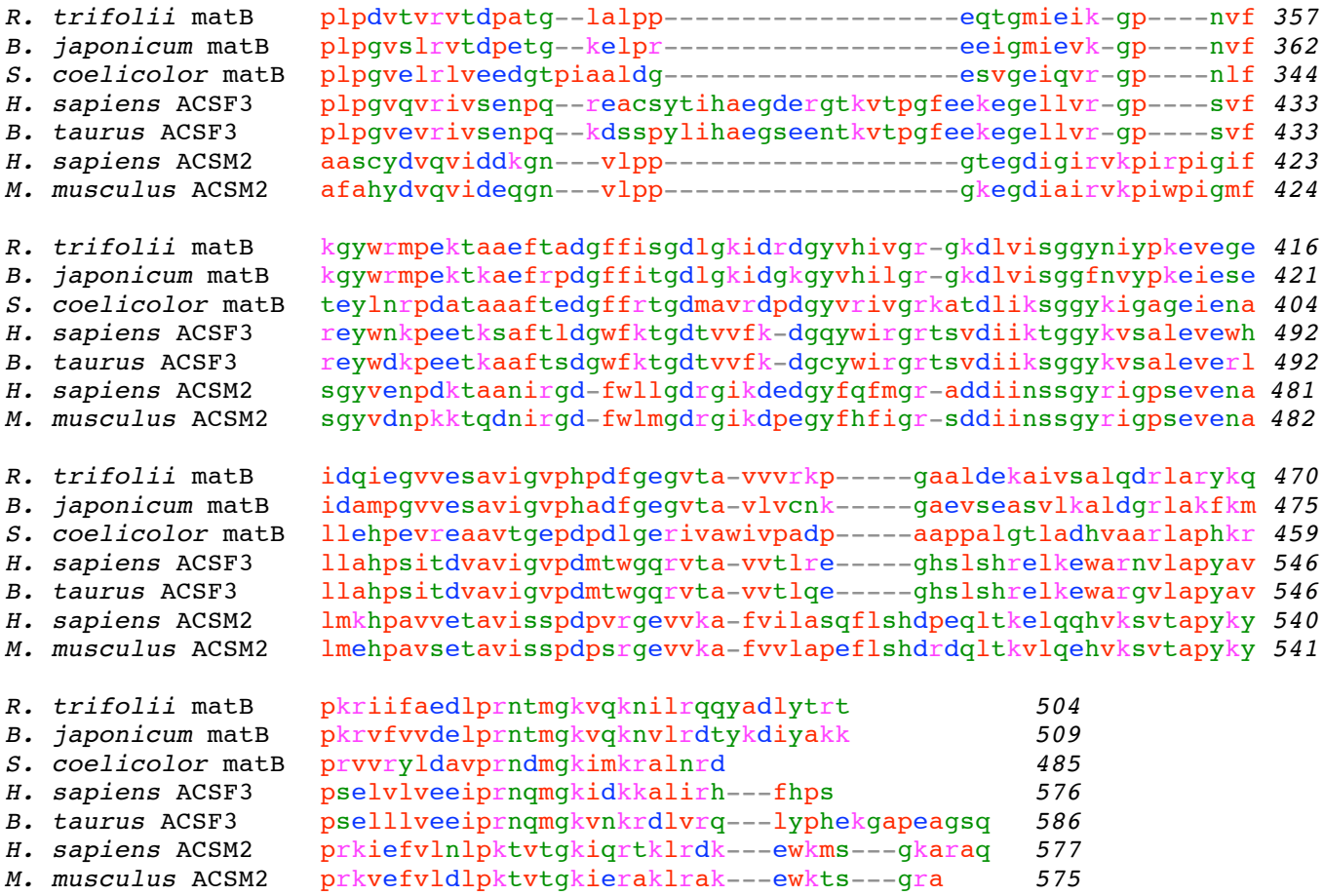
SUPPLEMENTARY MATERIAL FIG. 1





Supplementary FIG. 1. **Multiple sequence alignments of prokaryotic malonyl-CoA synthetases (matB) and mammalian acyl-CoA synthetases.** Alignments were performed using KALIGN at EMBL-EBI with the default parameters. Acidic residues are colored blue, basic residues magenta, hydrophobic residues red, others green. The mammalian proteins all have predominantly basic aminoterminal extensions typical of mitochondrial targeting sequences. Both ACSF3 and ACSM2 mammalian proteins exhibit approximately 60% sequence similarity with prokaryotic malonyl-CoA synthetases (matB). However, only ACSF3 proteins have the positionally conserved motif e**R**ygmte motif found in the prokaryotic matB proteins. Since this study was completed, a crystal structure was solved for the *S. coelicolor* matB that showed the Arg283, within this motif plays a key role in substrate recognition by forming a salt bridge with the 3-carboxylate of malonate (1). Significantly, a second residue involved in malonate binding, Ser261, is also universally conserved in matB and ACSF3 proteins (shaded yellow), but not in the ACSM2 proteins.

REFERENCE

1. Hughes, A. J. and Keatinge-Clay, A. (2011) *Chem Biol* **18,** 165-176