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Supplemental Information

Mutations in *SCO2* Are Associated with Autosomal-Dominant High-Grade Myopia

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Supplemental Inventory

Supplement 1. Summary of Exome Sequencing

Detailed summary table of four individuals who underwent exome sequencing. Last column (average) depicts average values across all four individuals.

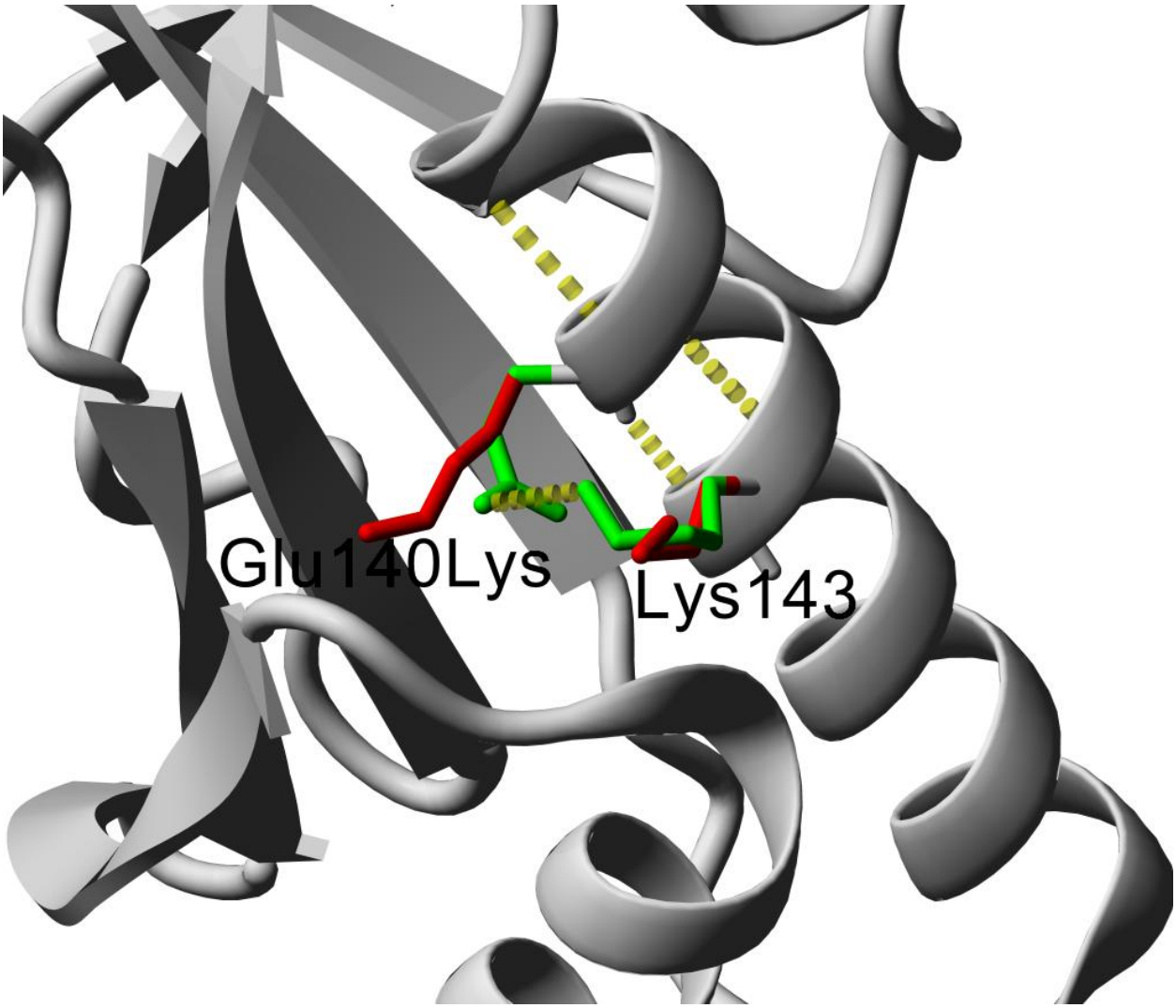
Exome Capture	**Affected**	**Affected**	**Unaffected**	**Unaffected**	**Average**
Individual	**III:2**	**IV:1**	**IV:2**	**III:1**	
All SNVs	252470	260113	222535	243168	244571.5
SNVs in dbSNP132	48865	49219	44678	45038	46950
Synonymous variant calls	1795	1807	1897	1849	1837
Nonsynonymous and canonical splice site calls	1864	1828	1979	1914	1896.25
Microindels	234	133	249	600	304

Coverage Analysis	**Affected**	**Affected**	**Unaffected**	**Unaffected**	**Average**
Total exonic targeted positions (bp)	37804019	37804019	37804019	37804019	37804019
In-target base pair calls (bp)	36216731	36292911	36310324	35878150	36174529
Off-target base pair calls (bp)	202846369	206534350	204202633	163279005	194215589
Total base pair calls in all reads (bp)	239063100	242827261	240512957	199157155	230390118
Proportion of target covered	95.80%	96.00%	96.00%	94.90%	95.6%
Total number of reads	23974834	24379907	21398893	23037982	23197904
Total number of reads overlapping target positions	16368831	16840346	14790170	16465811	16116289.5
Proportion of reads overlapping target	68.00%	69.00%	69.00%	71.00%	69.25%
Maximum depth	1760	2014	3258	4117	2787.25
Median depth	23	26	22	22	23.25
Coverage 1X	95.80%	96.00%	94.90%	96.00%	95.6%
Coverage 5X	88.00%	90.00%	84.00%	88.00%	87.5%
Coverage 10X	76.00%	80.00%	71.00%	76.00%	75.7%
Coverage 20X	54.00%	60.00%	51.00%	52.00%	54.2%

Micro-Insertion/Deletion	Affected	Affected	Unaffected	Unaffected	Average
Total microindels	3365	3424	2971	4312	3518
Seen by a too low proportion of reads	37	31	61	86	53.75
In reads with low average mapping quality	69	76	63	85	73.25
In reads with low average base quality in/around indel (<10)	37	66	75	71	62.25
In reads with high average mismatch count	788	772	610	937	776.75
In region with high mismatch rate (>0.2)	38	24	37	116	53.75
Outside exon/coding sequence/miRNA/UCRs	2001	2145	1722	2252	2030
In dbSNP132	166	185	161	172	171

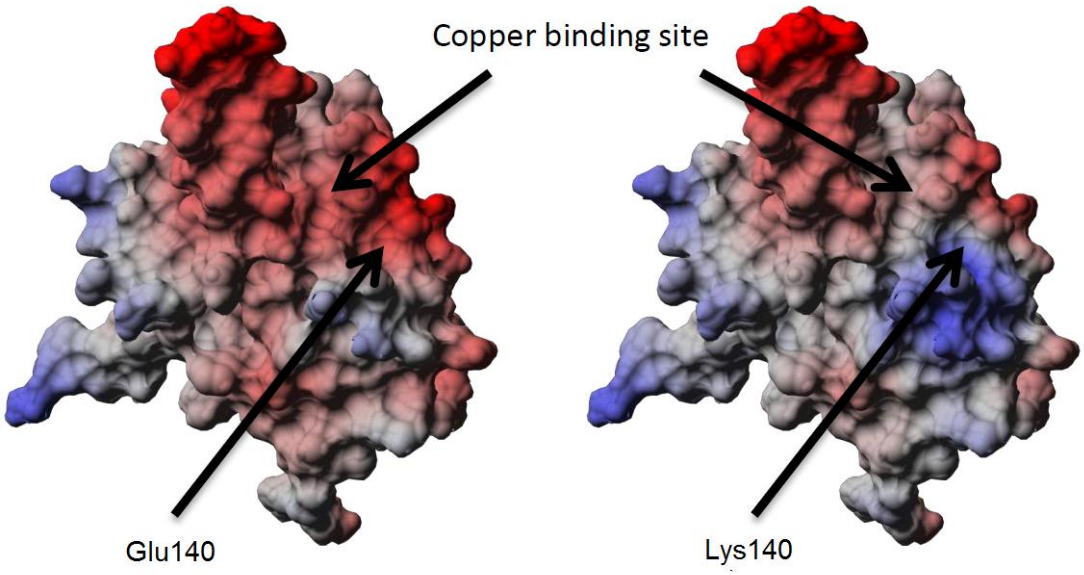
Supplement 2. Structural Modeling of p.Glu140Lys Mutation

Modeling structure of SCO2 where removal of a salt-bridge between Glu140 and Lys143 which can affect stabilization of the protein. Green represents wild-type, red represents mutant conformation**.** Yellow represents hydrogen bond along the helix backbone.



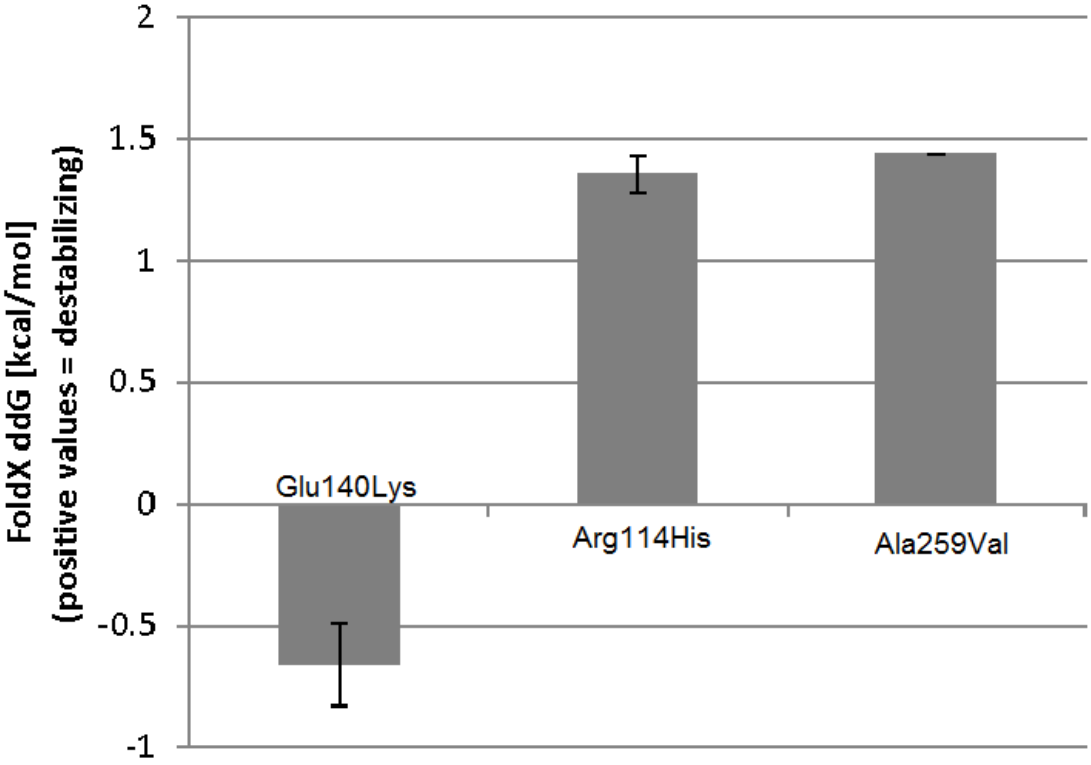
Supplement 3. Structure of SCO2 Depicting p.Gly140Lys Mutation Copper Binding Site

Electrostatic surface potential in the SCO2 structure for wildtype and mutant proteins was calculated with the Particle Mesh Ewald method implemented in YASARA. Blue indicates positive and red indicates negative charge potential. All protein structure Figures were created with YASARA. Figure depicts inversion of charge close to the copper ion which diminishes copper binding efficiency via negative electrostatic potential of the ion binding site.



Supplement 4. FoldX Energy Calculation of Mutations Identified in High Myopia Patients for SCO2

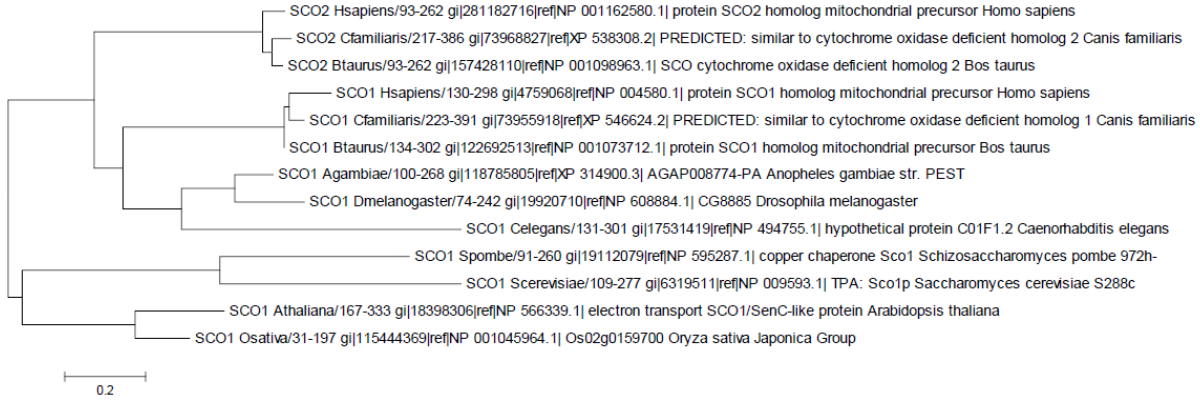
A quantitative estimation of the importance of the interactions contributing to the stability of proteins and protein complexes for mutations identified on SCO2. Positive values predicts destabilization of protein. For the structural stability analysis we first minimized chain A of PDB entry 2rli using the RepairPDB function of FoldX in YASARA. The free energy change for each missense mutation was then calculated with FoldX using the average of 5 repeated runs.

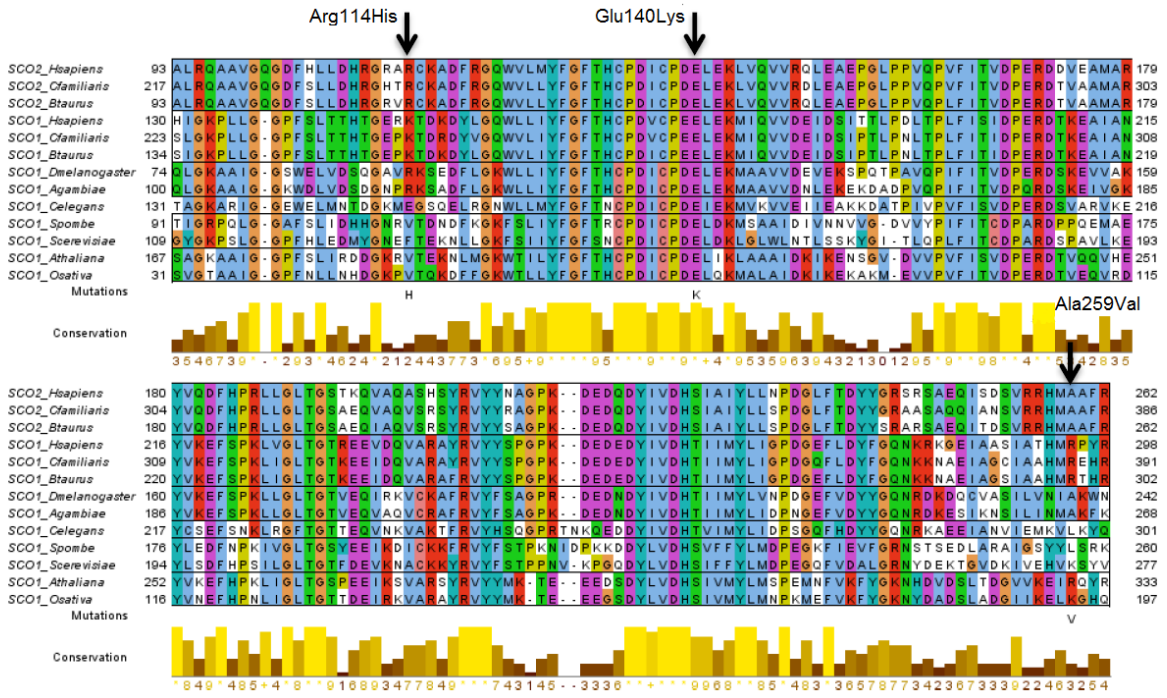


Supplement 5. SCO1 and SCO2 Homology, Conservation, and Location Of Mutations Identified within the Protein Sequence

A phylogenetic tree with MEGA5 using the Maximum Likelihood method based on the JTT matrix-based model. A discrete Gamma distribution was used to model evolutionary rate differences among sites (5 categories (+G, parameter = 1.9464)). The rate variation model allowed for some sites to be evolutionarily invariable ([+I], 17.7% sites). The tree is drawn to scale, with branch lengths measured in the number of substitutions per site.

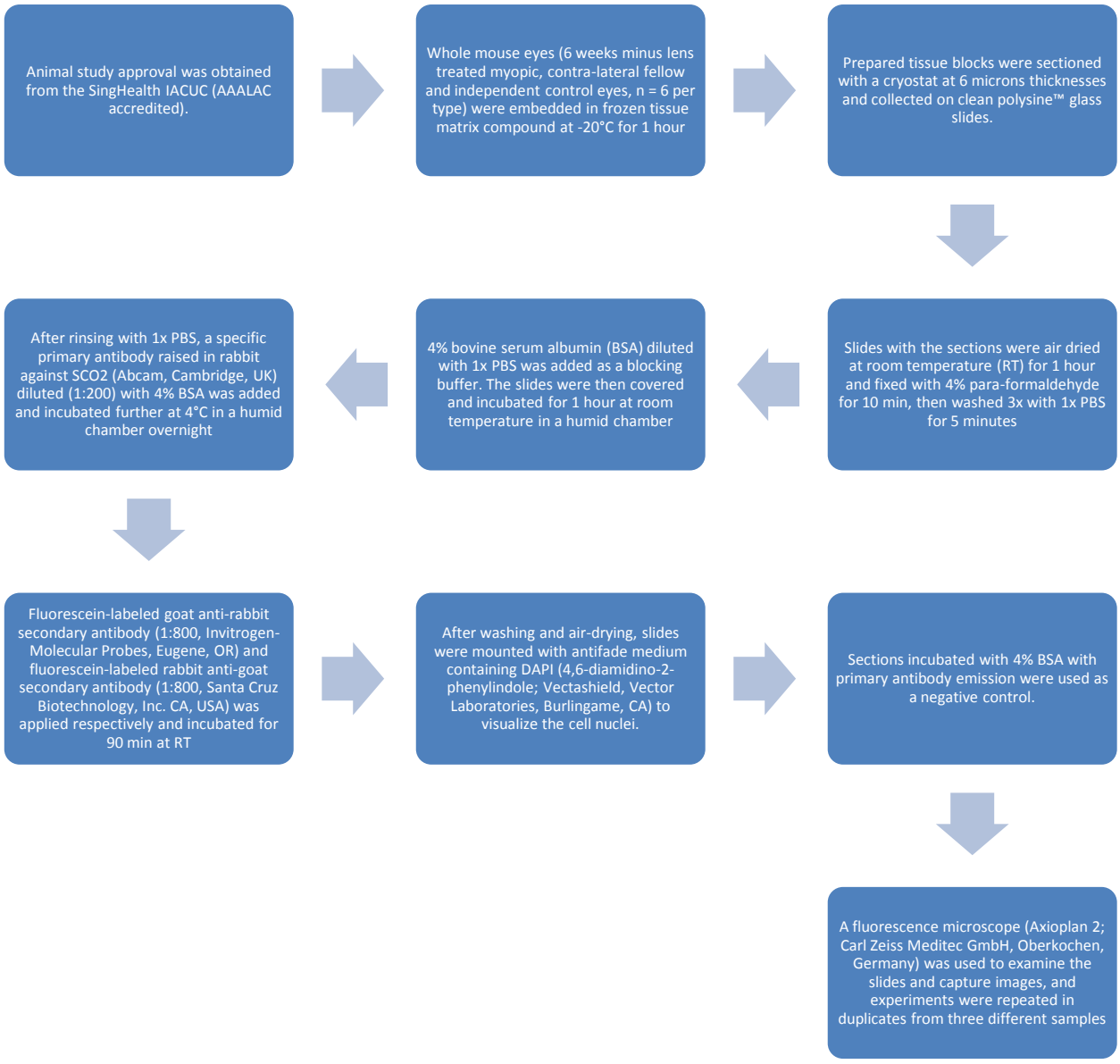
From the list of orthologues and paralogues identified with BLASTP against NCBI's nr database using our orthologue search protocol, we selected representative protein reference sequences from key model organisms and created a multiple alignment using MAFFT with L-INS-I parameter settings und curated and displayed the alignment in Jalview.





Supplement 6. Methods of Myopic Mouse Modeling

Detailed methodology of myopic mouse modeling for immunohistochemistry. Animals used in the experiement was IRB approved.



Supplement 7. cDNA Gene Expression

cDNA expression of *SCO2* and GAPDH on all available tissues. The *SCO2* primers used were F-gcttcctctcgtgcttggtc and R- CTTGACAAAAGCCAGGACCTC (177b bp). The *GAPDH* primers used were F-tcaccagggctgcttttaac and R- gacaagcttcccgttctcag (150bp).

Adult cDNA 1. Brain 2. Heart 3. Liver 4. Skeletal Muscle 5. Kidney 6. Lung 7. Placenta 8. Pancreas 9. Control cDNA --- **Fetal cDNA** 10. brain 11. Heart 12. Liver 13. Skeletal Muscle 14. Kidney 15. Lung 16. Thymus 17. Spleen 18. Control cDNA 19. Adult Retina 20. Adult RPE 21. Adult Choroid 22. Adult Sclera 23. 24 week Retina/RPE 24. 24 week Choroid 25. 24 week Sclera 26. NTC

Adult cDNA 27. Brain 28. Heart 29. Liver 30. Skeletal Muscle 31. Kidney 32. Lung 33. Placenta 34. Pancreas 35. Control cDNA --- **Fetal cDNA** 36. brain 37. Heart 38. Liver 39. Skeletal Muscle 40. Kidney 41. Lung 42. Thymus 43. Spleen 44. Control 45. Adult Retina 46. Adult RPE 47. Adult Choroid 48. Adult Sclera 49. 24 week Retina/RPE 50. 24 week Choroid 51. 24 week Sclera 52. NTC