

Supplementary Appendix

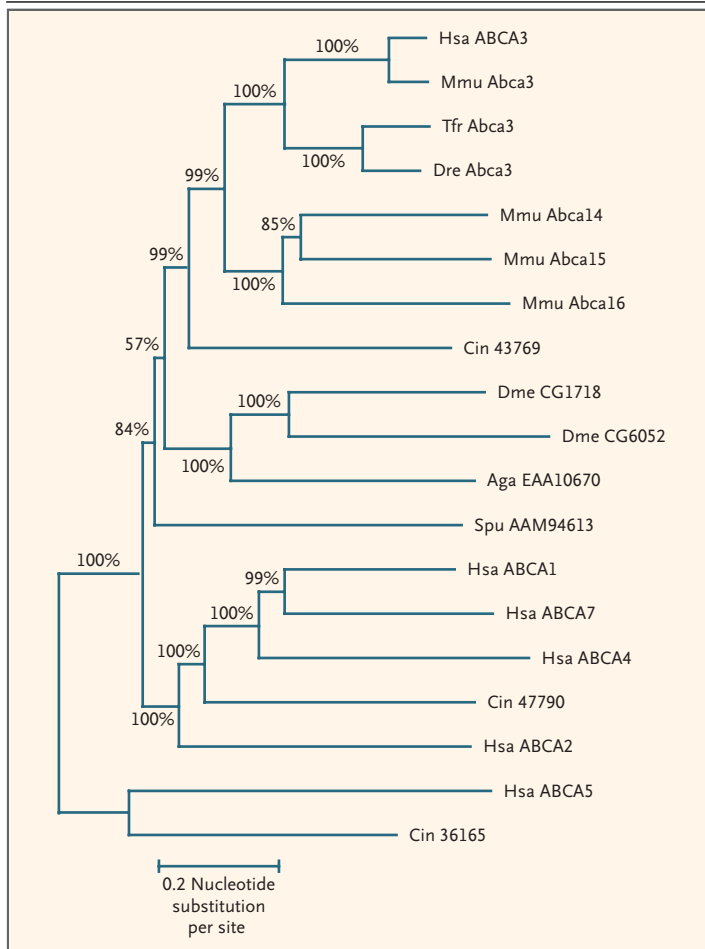
This appendix has been provided by the authors to give readers additional information about their work.

Supplement to: Shulenin S, Noguee LM, Annilo T, et al. *ABCA3* Gene Mutations in Newborns with Fatal Surfactant Deficiency. *N Engl J Med* 2004;350:1296-303.

Supplementary Appendix 1. Forward and Reverse Primers Used for Each Coding Exon.*

Exon No.	Sequence	Exon No.	Sequence
4	CACTCTGCGTGTTTCTATTGC	19	GGGGTGATGCTTTAGGAAC
4	GCATGCAGACAGCCCTTCC	19	GGCAGGAGAATCTCTTGAAC
5	GGAAGGGCTGTCTGCATGC	20	CAGTGCCTCTAACCATAGTC
5	CAGGTTGAAGTAGTGATGCG	20	TCTGCATGGGCTTACATGAG
6	AGTGACCTGAACCACGCAG	21	ACAGAACAGCACCTGCCG
6	GACCCAAGGAGTGACTGC	21	GGGATGCTGCTGAACCTC
7	TCCCACTCCACCCTGTTG	22	CATGCTCAGGTGGCAGGG
7	CGGCACTATTCACAACAGC	22	CTGCTGTCACAGGGAGTTG
8	CGAGAGCAGCTCCTCGAG	23	CTGCTGCCCTGCATGCCTTG
8	GGCAGAGCTTGCACTGAG	23	CCATGCATGGAGGGTAAAG
9	CCTGGCACATGCCATCATG	24	GTCTGAGGACCTCCAAATG
9	GACATTGACAGCTCCTCTC	24	CCACCACTCATGGCTAGC
10	GAGAGGAGCTGTCAATGTC	25	CGGATAAGGCCCTGTGC
10	CTTCCGATCACAGCCTCTG	25	CTCTGCAGTGACCACGTC
11	GTGTAGATGGCAAGTGCCAG	26	GAGGGGACATTGTTACAGG
11	GATGCTGCTGCCTTCAGTG	26	GGCCCAAGCAGAGACGTC
12	GTCAAATGTGCACACGTGTGC	27	GACCTCCACATCCACC
12	GCCACTCCCTGTGCACAG	27	GGACAGCCAGGTAGTCAG
13	TGGTTGTGCTTTGCTCGTC	28	AGGCTCAGACTGCTCTGC
13	CTGGAGATGTGGTCTGTGC	28	CTTGTCTCGCTCTCCAGAG
14	GCTTGGTTCCTTCTGAGACG	29	GCACCTCAAGGGTAGTCC
14	GCTGAGGTGCATCTCCTGC	29	GAGCTACCTGGGTACAC
15	GGTGCTGACGAGCAGGATC	30	GCCTCCACTGGGGACATG
15	GAGGTTCTGGTGAGAGGAAC	30	GGCTCTCCGATCAGGCTG
16	CAGCTACGTCAAGGAGAGG	31	CCTTCCTGTCTGCACAAGC
16	GCTTGAGTCCTCAAGGATG	31	GCACAGCAGATGGGAGAG
17	CATCCTTGGAGGACTCAAGC	32	CTCCAGTGCTGAGCACC
17	CCACCCAGAGGCAACAGAC	32	CAAACCAGCACGTATCAGG
18	GTGGGCATGAAGAGACCTG	33	CAGAGGACTCCCAGGTC
18	CAGATTCATCTGGGCTGATG	33	GAGAGAGGATGTAAGATGGG

* The first three exons are noncoding exons.



Supplementary Appendix 2. Phylogenetic Tree of ABCA3-Related Proteins.

The neighbor-joining tree shows the relation of the mammalian and fish ABCA3-like proteins and their position within the ABCA subfamily. The amino acid sequences of ABCA3 and other related transporters were aligned with the use of Clustal X, and phylogenetic analyses were carried out with the use of Mega2 software (<http://www.megasoftware.net/>). A maximal parsimony analysis yielded a tree with identical topology. The tree was rooted with human ABCA5 and a related sequence from ciona. A low frequency of nucleotide substitutions per site (as indicated by the length of the horizontal branch) suggests close genetic relatedness. Percentages indicate bootstrap values based on 1000 replicates. Aga denotes *Anopheles gambiae*, Cin *Ciona intestinalis*, Dme *Drosophila melanogaster*, Dre *Danio rerio*, Hsa *Homo sapiens*, Mmu *Mus musculus*, Spu *Strongylocentrotus purpuratus*, and Tfr *Takifugu rubripes*. Sequences from drosophila and anopheles are indicated by GenBank locus names. ABC genes from ciona were predicted with the use of a BLAST program (<http://genome.jgi-psf.org/ciona4/>). The scale bar indicates the number of substitutions per site with the use of the Poisson correction distance method.