

An Evolutionary Relationship Between Stearoyl-CoA Desaturase (SCD) Protein Sequences Involved in Fatty Acid Metabolism

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Abstract

Background: Stearoyl-CoA desaturase (SCD) is a key enzyme that converts saturated fatty acids (SFAs) to monounsaturated fatty acids (MUFAs) in fat biosynthesis. Despite being crucial for interpreting SCDs' roles across species, the evolutionary relationship of SCD proteins across species has yet to be elucidated. This study aims to present this evolutionary relationship based on amino acid sequences.

Methods: Using Multiple Sequence Alignment (MSA) and phylogenetic construction methods, a hypothetical evolutionary relationship was generated between the stearoyl-CoA desaturase (SCD) protein sequences between 18 different species.

Results: SCD protein sequences from *Homo sapiens*, *Pan troglodytes* (chimpanzee), and *Pongo abelii* (orangutan) have the lowest genetic distances of 0.006 of the 18 species studied. *Capra hircus* (goat) and *Ovis aries* (Sheep) had the next lowest genetic distance of 0.023. These farm animals are 99.987% identical at the amino acid level.

Conclusions: The SCD proteins are conserved in these 18 species, and their evolutionary relationships are similar.

Keywords: Multiple sequence alignment, Phylogenetic analysis, Stearoyl-CoA desaturase (SCD) proteins,

Introduction

Fatty acids are chief components of all living organisms, participating in various metabolic processes such as energy storage and as structural elements of biological membranes. They are the components of a wide variety of lipids including oils, waxes, phospholipids, and others.

Fatty acids occur in saturated and unsaturated forms, a fundamental feature of their physical properties.

Stearoyl-CoA desaturase (SCD) is an endoplasmic reticulum enzyme that catalyzes the biosynthesis of monounsaturated fatty acids from saturated fatty acids and is the critical gene responsible for the synthesis of triglycerides, phospholipids, and cholesterol esters (1, 2). Stearoyl-

CoA desaturase -1 (SCD-1) plays an important role in triacylglycerol (TG) accumulation and feeding-induced adiposity and hepatic steatosis (3-6).

Stearoyl-CoA desaturase is the rate-limiting enzyme that introduces the first cis-double bond at the delta-9 position of saturated fatty acids (SFAs) to thereby generate monounsaturated fatty acids (MUFAs) (7), which are major substrates for biosynthesis of polyunsaturated fatty acids (PUFAs) and complex lipids such as triglycerides, phospholipids, cholesterol esters, and wax esters for energy storage, as components of biological membranes, and as signaling molecules. The scd genes are universally present in living organisms. The

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Received: May 12, 2014; Accepted: Jun 17, 2014

number of *scd* genes varies from one to five, and are generally called *scd1*, *scd2*, *scd3*, *scd4*, and *scd5* in different organisms (7, 8). The evolutionary history revealed that the *scd* genes in vertebrates could be distinctly classified into *scd5* (8-10) and *scd1* types including homologs *scd2*, *scd3*, and *scd4* (8, 11). The divergence of *scd1* and *scd5* genes occurred early in vertebrate evolution due to the whole genome duplication (2R) (8). However, *scd* gene evolution has not been comprehensively studied. Our aim in this study was to evaluate the evolutionary relationships of SCD protein sequences from 18 different species (Table 1).

Materials and Methods

Obtaining protein sequences

All the functional protein sequences of the *scd* genes from the 18 organisms chosen were downloaded from the NCBI database (Table 1).

Multiple sequence alignment

These sequences were analyzed on ClustalW (<http://www.ebi.ac.uk/clustalw/>) for the multiple sequences alignment. Sequences were also analyzed using Geneious 7.1.2 (16), and a ClustalW algorithm was used to align multiple sequences in parallel (Fig. 1).

Construction of phylogenetic trees

The phylogenetic trees were first constructed using the neighbor joining method (14) from the MEGA5.2 package (12). Confidence on each node was assessed by 2000 bootstrap replications. (Fig. 2) (13). Also the maximum likelihood method (17) from a MEGA5.2 package (12) was used to construct a phylogenetic tree and 2000 replicates were used for bootstrap statistical test (13) (Fig. 3).

Pairwise distances

To measure genetic distances between sequences, a pairwise distances method from the MEGA5.2 package (12) was used.

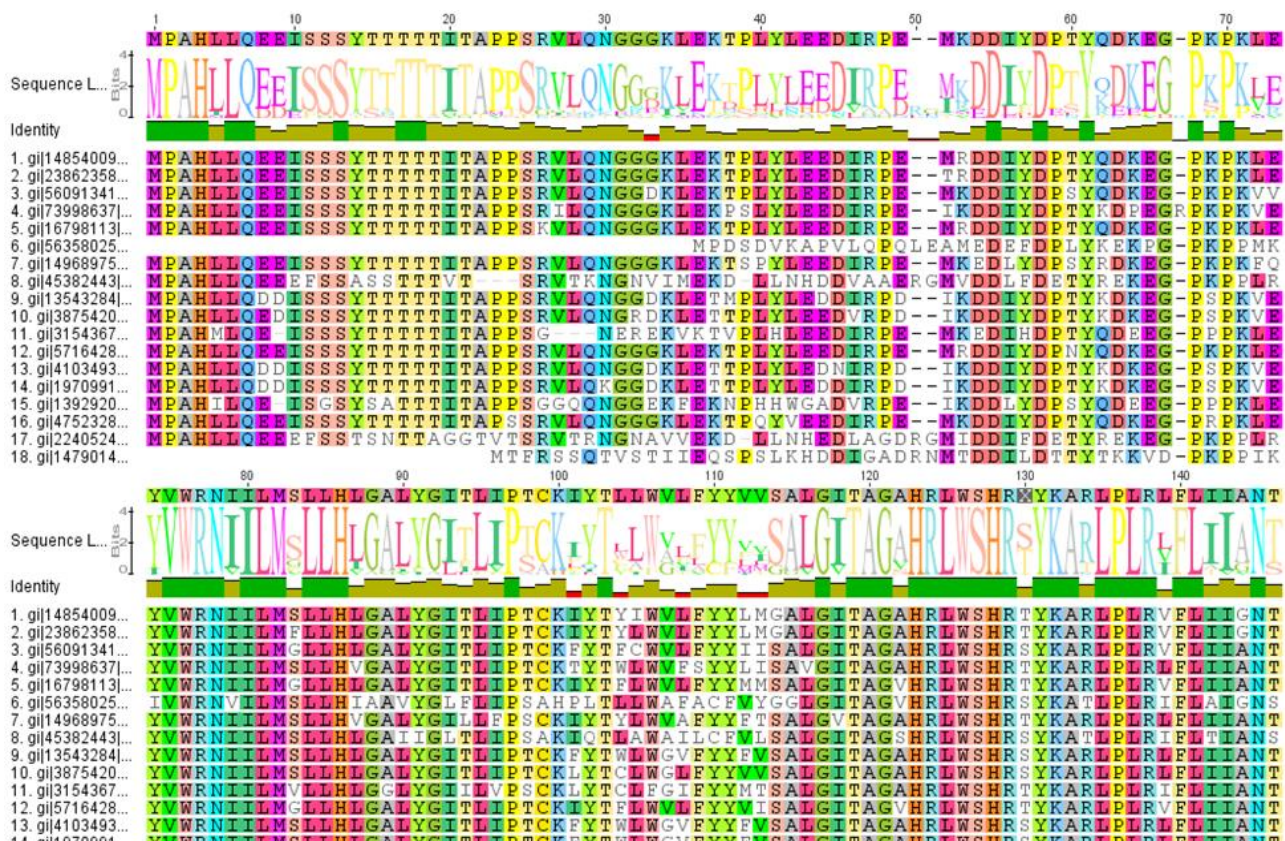


Fig. 1. Multiple sequence alignment result by Geneious 7.1.2 (16). Green boxes represent 100% identity between species. Green-brown regions represent 30-99% identity, and red regions represent 0-29% identity.

Evolutionary Relationship of Stearoyl-CoA Desaturase (SCD) Protein Sequences

Table 1. Descriptions of SCD protein sequences from the 18 species analyzed in this study.

| Seq. no. | Organism | Protein ID (NCBI Reference Sequence) | Locus | Definition | Version | Length |
|----------|---|--|--------------|---|--------------------------------|--------|
| 1 | <i>Bos Taurus</i> (Cattle) | NP_776384.3 | NP_776384 | acyl-CoA desaturase | NP_776384.3 GI:148540094 | 359 aa |
| 2 | <i>Bubalus bubalis</i> (Water buffalo) | CAZ16319.1 | CAZ16319 | stearoyl CoA desaturase | CAZ16319.1 GI:238623585 | 359 aa |
| 3 | <i>Camelus ferus</i> (Camel) | XP_006182984.1 | XP_006182984 | Predicted: acyl-CoA desaturase | XP_006182984.1 GI:560913419 | 359 aa |
| 4 | <i>Canis lupus familiaris</i> (Dog) | XP_543968.2 | XP_543968 | Predicted: acyl-CoA desaturase | XP_543968.2 GI:73998637 | 360 aa |
| 5 | <i>Capra aegagrus hircus</i> (Goat) | AAL29305.1 | AAL29305 | stearoyl coenzyme A desaturase | AAL29305.1 GI:16798113 | 359 aa |
| 6 | <i>Danio rerio</i> (Zebrafish) | NP_942110.2 | NP_942110 | stearoyl-CoA desaturase 5 | NP_942110.2 GI:563580257 | 326 aa |
| 7 | <i>Equus caballus</i> (Horse) | XP_001500414.1 | XP_001500414 | PREDICTED: acyl-CoA desaturase | XP_001500414.1 GI:149689754 | 359 aa |
| 8 | <i>Gallus gallus</i> (Chicken) | NP_990221.1 | NP_990221 | stearoyl-CoA desaturase 1 | NP_990221.1 GI:45382443 | 357 aa |
| 9 | <i>Homo sapiens</i> (Human) | AAH05807.1 | AAH05807 | SCD protein | AAH05807.1 GI:13543284 | 355 aa |
| 10 | <i>Macaca mulatta</i> (Rhesus monkey) | AFJ71651.1 | AFJ71651 | acyl-CoA desaturase | AFJ71651.1 GI:387542048 | 359 aa |
| 11 | <i>Mus musculus</i> (Mouse) | NP_033153.2 | NP_033153 | acyl-CoA desaturase 1 | NP_033153.2 GI:31543675 | 355 aa |
| 12 | <i>Ovis aries</i> (Sheep) | NP_001009254.1 | NP_001009254 | acyl-CoA desaturase | NP_001009254.1 GI:57164289 | 359 aa |
| 13 | <i>Pan troglodytes</i> (Chimpanzee) | JAA41265.1 | JAA41265 | stearoyl-CoA desaturase (delta-9-desaturase) | JAA41265.1 GI:410349323 | 359 aa |
| 14 | <i>Pongo abelii</i> (Sumatran orangutan) | NP_001125731.1 | NP_001125731 | acyl-CoA desaturase | NP_001125731.1 GI:197099102 | 359 aa |
| 15 | <i>Rattus norvegicus</i> (Norway rat) | NP_114029.1 | NP_114029 | acyl-CoA desaturase 2 | NP_114029.1 GI:13929208 | 358 aa |
| 16 | <i>Sus scrofa</i> (Pig) | NP_998946.1 | NP_998946 | acyl-CoA desaturase | NP_998946.1 GI:47523282 | 359 aa |
| 17 | <i>Taeniopygia guttata</i> (Zebra finch) | XP_002198152.1 | XP_002198152 | PREDICTED: acyl-CoA desaturase | XP_002198152.1 GI:224052475 | 360 aa |
| 18 | <i>Xenopus laevis</i> (African clawed frog) | NP_001087809.1 | NP_001087809 | stearoyl-CoA desaturase (delta-9-desaturase) | NP_001087809.1 GI:147901446 | 339 aa |

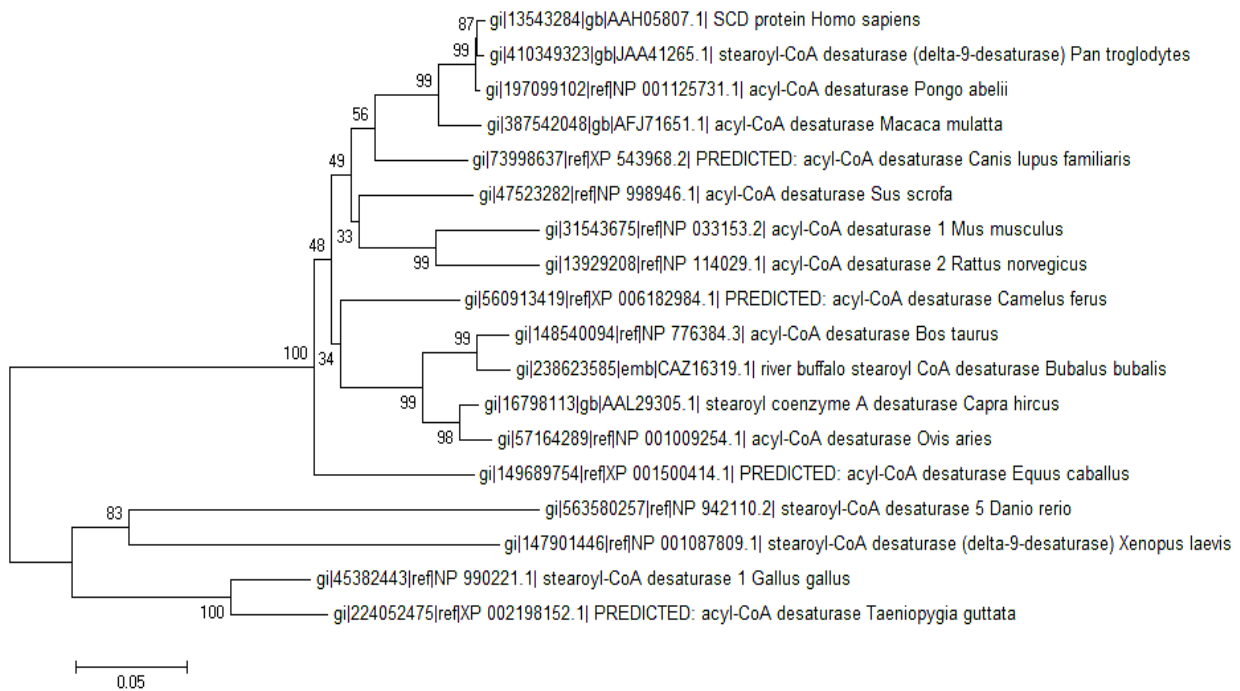


Fig. 2. Molecular phylogenetic analysis by the neighbor-joining method (14). The optimal phylogenetic tree with the sum of branch length = 1.28213934 is shown. The percentages of replicate trees in which the associated taxa clustered together in the bootstrap test (2000 replicates) are shown to the left of the branches (13). The tree is drawn to scale, with branch lengths in the same units as those of the evolutionary distances used to infer the phylogenetic tree. Evolutionary analyses were conducted in MEGA5.2 (12).

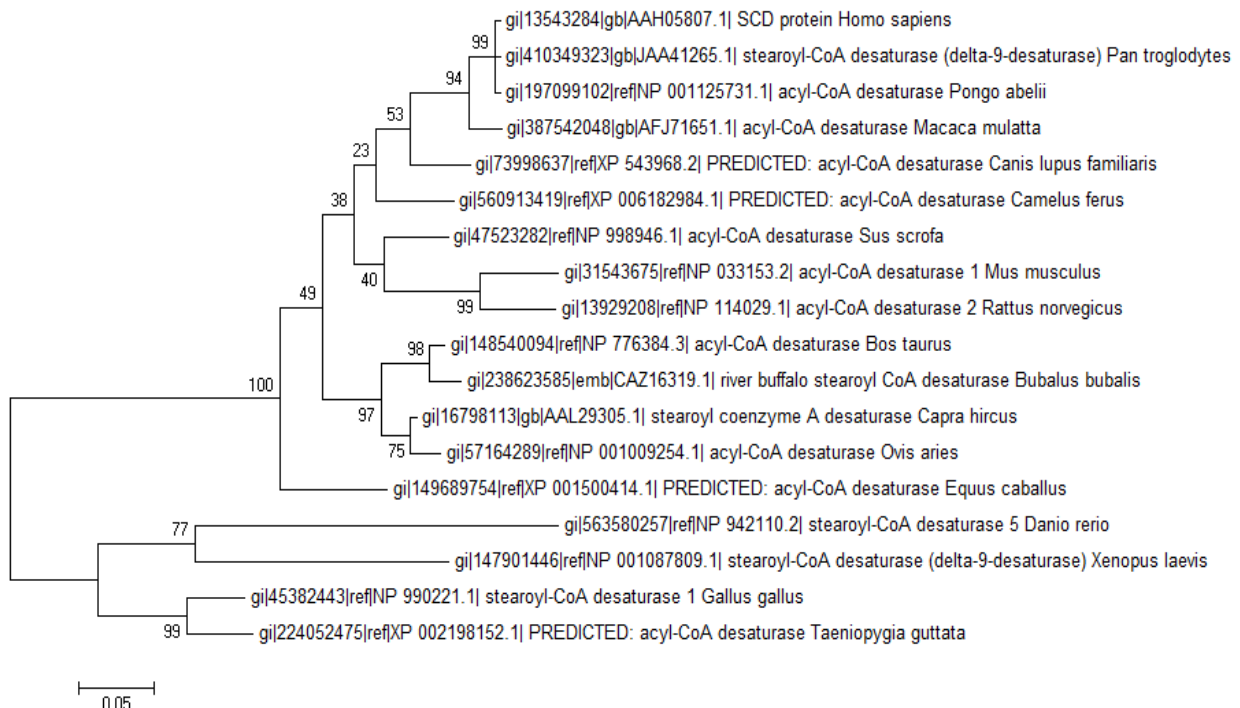


Fig. 3. Molecular phylogenetic analysis by the maximum likelihood method (17). The evolutionary history was inferred using the maximum likelihood method based on the JTT matrix-based model (17) with 2000 replicates of the bootstrap test.

Evolutionary Relationship of Stearoyl-CoA Desaturase (SCD) Protein Sequences

SCD proteins in human, chimpanzee, orangutan, rhesus monkey, camel, dog, pig, mouse, rat, cattle, buffalo, goat, sheep, horse, zebrafish, frog, chicken, and zebra finch (showed by green in Fig. 1).

Pairwise distances

The pairwise distances method of phylogenetic analysis relies on a measure of genetic distance between the sequences being classified. This analysis shows the divergence and percent identity of each sequence pair in the current alignment.

Sequence comparison between the SCD protein sequences (Fig. 4) indicated that the SCD Protein Sequences from *Homo sapiens*, *Pan troglodytes* (chimpanzee) and *Pon goabelii* (orangutan) have least genetic distance of 0.006 in these 18 species and are 99.994% identical at the amino acid level. Stearoyl-CoA desaturase protein sequences from *Capra hircus* (goat) and *Ovis aries* (sheep) have a genetic distance of 0.023 and are 99.987% identical at the amino acid level.

The maximum genetic distance of 0.473 occurred between *Danio rerio* and *Mus musculus* and *Danio rerio* and *Rattus norvegicus*. These proteins were 99.6% similar at the amino acid level (Fig.4).

Phylogenetic tree

This analysis shows evolutionary relationships predicted from the multiple sequence alignment. The

length of each pair of branches represents the distance between sequence pairs.

The phylogenetic trees (Figs 2 and 3) classified the species into three groups. Group 1 contains of four species (*Homo sapiens*, *Pan troglodytes*, *Pongo abelii*, and *Macaca mulatta*) with the lowest genetic distances, Group 2 contains 10 species (*Canis lupus*, *Camelus ferus*, *Sus scrofa*, *Mus musculus*, *Rattus norvegicus*, *Bos taurus*, *Bubalus bubalis*, *Capra hircus*, *Ovis aries*, and *Equus caballus*), and Group 3 contain four species (*Danio rerio*, *Xenopus laevis*, *Gallus gallus*, and *Taeniopygia guttata*).

The phylogenetic tree constructed by MEGA 5.2 shows that the evolutionary relationships between the SCD protein sequences from the 18 species analyzed are similar to their species evolutionary relationships.

Discussion

The SCD proteins are conserved in the 18 species analyzed in our study, and their evolutionary relationships are similar to the species evolutionary relationships.

Acknowledgments

The authors thank Mohsen Ghayoomian in department of biology from Ferdowsi University of Mashhad, Mashhad, Iran for his suggestions. The authors declare there is no conflict of interest.

| Seq. no. | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 | 15 | 16 | 17 | 18 |
|----------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|----|
| 1 | | | | | | | | | | | | | | | | | | |
| 2 | 0.029 | | | | | | | | | | | | | | | | | |
| 3 | 0.133 | 0.140 | | | | | | | | | | | | | | | | |
| 4 | 0.148 | 0.144 | 0.108 | | | | | | | | | | | | | | | |
| 5 | 0.059 | 0.059 | 0.111 | 0.144 | | | | | | | | | | | | | | |
| 6 | 0.448 | 0.433 | 0.463 | 0.443 | 0.458 | | | | | | | | | | | | | |
| 7 | 0.148 | 0.159 | 0.129 | 0.140 | 0.148 | 0.443 | | | | | | | | | | | | |
| 8 | 0.348 | 0.357 | 0.362 | 0.362 | 0.348 | 0.326 | 0.339 | | | | | | | | | | | |
| 9 | 0.151 | 0.151 | 0.115 | 0.090 | 0.137 | 0.458 | 0.159 | 0.348 | | | | | | | | | | |
| 10 | 0.151 | 0.155 | 0.111 | 0.090 | 0.137 | 0.453 | 0.155 | 0.348 | 0.039 | | | | | | | | | |
| 11 | 0.182 | 0.182 | 0.163 | 0.151 | 0.159 | 0.473 | 0.178 | 0.353 | 0.137 | 0.129 | | | | | | | | |
| 12 | 0.076 | 0.076 | 0.111 | 0.155 | 0.023 | 0.458 | 0.159 | 0.348 | 0.144 | 0.137 | 0.166 | | | | | | | |
| 13 | 0.151 | 0.151 | 0.115 | 0.090 | 0.137 | 0.453 | 0.159 | 0.348 | 0.006 | 0.039 | 0.137 | 0.144 | | | | | | |
| 14 | 0.151 | 0.151 | 0.115 | 0.090 | 0.137 | 0.448 | 0.151 | 0.339 | 0.006 | 0.039 | 0.137 | 0.144 | 0.006 | | | | | |
| 15 | 0.189 | 0.182 | 0.159 | 0.133 | 0.163 | 0.473 | 0.178 | 0.366 | 0.137 | 0.133 | 0.094 | 0.155 | 0.137 | 0.137 | | | | |
| 16 | 0.133 | 0.133 | 0.122 | 0.097 | 0.129 | 0.443 | 0.144 | 0.348 | 0.129 | 0.129 | 0.129 | 0.133 | 0.129 | 0.129 | 0.137 | | | |
| 17 | 0.353 | 0.362 | 0.362 | 0.366 | 0.344 | 0.335 | 0.362 | 0.080 | 0.348 | 0.348 | 0.385 | 0.344 | 0.348 | 0.339 | 0.394 | 0.357 | | |
| 18 | 0.453 | 0.453 | 0.418 | 0.418 | 0.438 | 0.353 | 0.433 | 0.291 | 0.433 | 0.443 | 0.438 | 0.453 | 0.433 | 0.428 | 0.458 | 0.409 | 0.304 | |

Fig. 4. Estimates of evolutionary divergence between SCD protein sequences. Pairwise distances methods of phylogenetic analyses were conducted using the Poisson correction model (15). The analysis involved 18 amino acid sequences. All positions containing gaps and missing data were eliminated. The overall average is 0.232. Evolutionary analyses were conducted in MEGA5.2 (12). This analysis shows the divergence of each sequence pair in the current alignment.

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