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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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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.

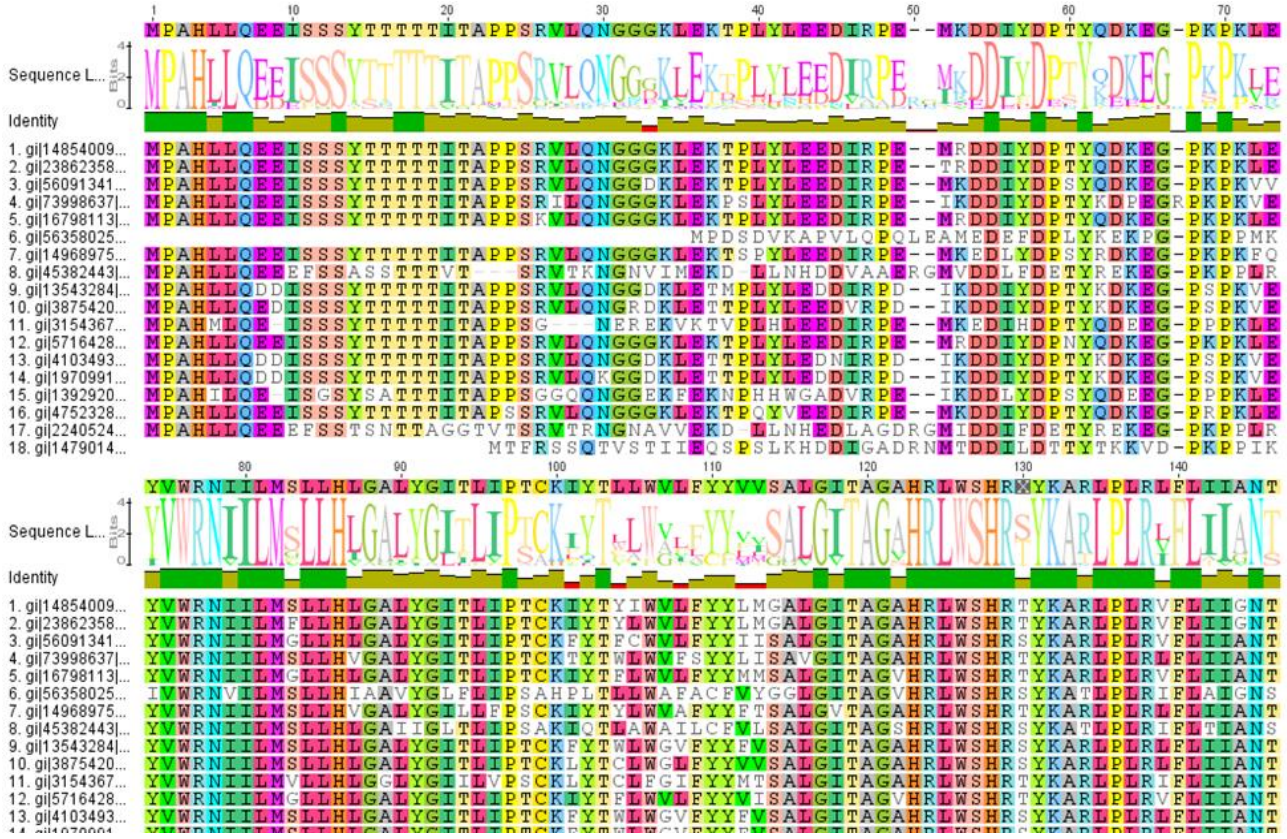

Fig. 1. Multiple sequence alignment result by Geneious 7.1.2 (16). Green boxes represent 100% identity between species. Green-brown regions represent 30-99% identity, and red regions represent 0-29% identity.

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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	*Bos Taurus* (Cattle)	NP_776384.3	NP_776384	acyl-CoA desaturase	NP_776384.3 GI:148540094	359 aa
2	*Bubalus bubalis* (Water buffalo)	CAZ16319.1	CAZ16319	stearoyl CoA desaturase	CAZ16319.1 GI:238623585	359 aa
3	*Camelus ferus* (Camel)	XP_006182984.1	XP_006182984	Predicted: acyl-CoA desaturase	XP_006182984.1 GI:560913419	359 aa
4	*Canis lupus familiaris* (Dog)	XP_543968.2	XP_543968	Predicted: acyl-CoA desaturase	XP_543968.2 GI:73998637	360 aa
5	*Capra aegagrus hircus* (Goat)	AAL29305.1	AAL29305	stearoyl coenzyme A desaturase	AAL29305.1 GI:16798113	359 aa
6	*Danio rerio* (Zebrafish)	NP_942110.2	NP_942110	stearoyl-CoA desaturase 5	NP_942110.2 GI:563580257	326 aa
7	*Equus caballus* (Horse)	XP_001500414.1	XP_001500414	PREDICTED: acyl-CoA desaturase	XP_001500414.1 GI:149689754	359 aa
8	*Gallus gallus* (Chicken)	NP_990221.1	NP_990221	stearoyl-CoA desaturase 1	NP_990221.1 GI:45382443	357 aa
9	*Homo sapiens* (Human)	AAH05807.1	AAH05807	SCD protein	AAH05807.1 GI:13543284	355 aa
10	*Macaca mulatta* (Rhesus monkey)	AFJ71651.1	AFJ71651	acyl-CoA desaturase	AFJ71651.1 GI:387542048	359 aa
11	*Mus musculus* (Mouse)	NP_033153.2	NP_033153	acyl-CoA desaturase 1	NP_033153.2 GI:31543675	355 aa
12	*Ovis aries* (Sheep)	NP_001009254.1	NP_001009254	acyl-CoA desaturase	NP_001009254.1 GI:57164289	359 aa
13	*Pan troglodytes* (Chimpanzee)	JAA41265.1	JAA41265	stearoyl-CoA desaturase (delta-9-desaturase)	JAA41265.1 GI:410349323	359 aa
14	*Pongo abelii* (Sumatran orangutan)	NP_001125731.1	NP_001125731	acyl-CoA desaturase	NP_001125731.1 GI:197099102	359 aa
15	*Rattus norvegicus* (Norway rat)	NP_114029.1	NP_114029	acyl-CoA desaturase 2	NP_114029.1 GI:13929208	358 aa
16	*Sus scrofa* (Pig)	NP_998946.1	NP_998946	acyl-CoA desaturase	NP_998946.1 GI:47523282	359 aa
17	*Taeniopygia guttata* (Zebra finch)	XP_002198152.1	XP_002198152	PREDICTED: acyl-CoA desaturase	XP_002198152.1 GI:224052475	360 aa
18	*Xenopus laevis* (African clawed frog)	NP_001087809.1	NP_001087809	stearoyl-CoA desaturase (delta-9-desaturase)	NP_001087809.1 GI:147901446	339 aa

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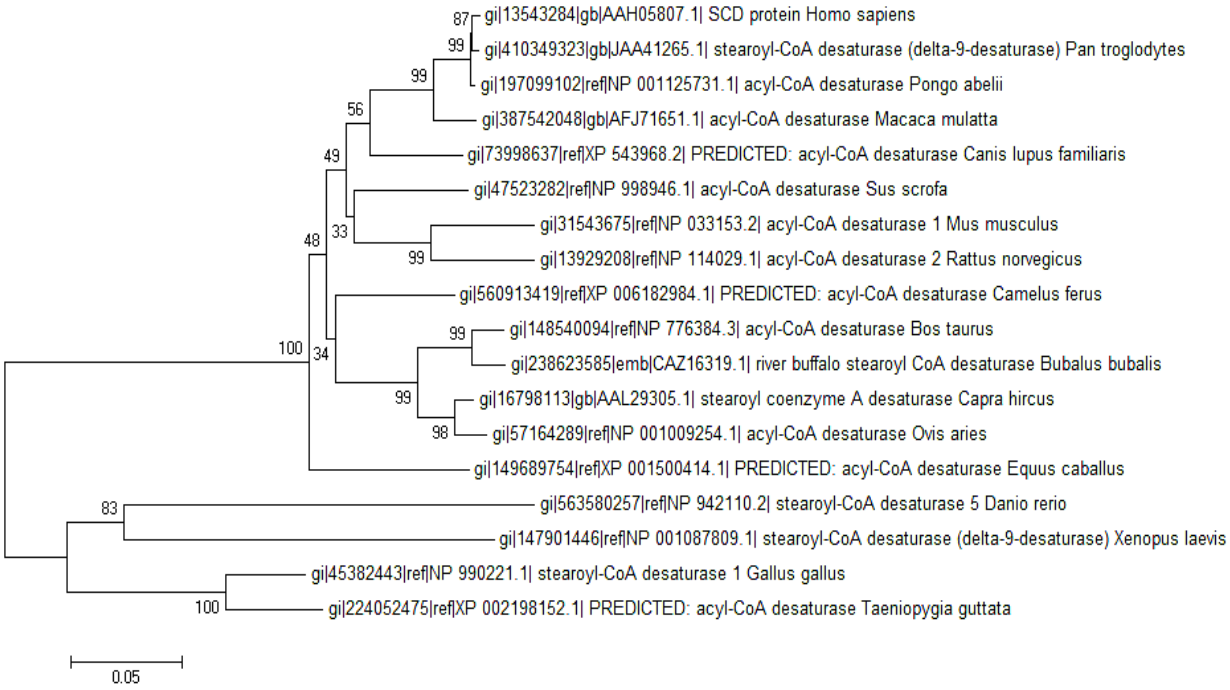

Fig. 2. Molecular phylogenetic analysis by the neighbor-joining method (14). The optimal phylogenetic tree with the sum of branch length = 1.28213934 is shown. The percentages of replicate trees in which the associated taxa clustered together in the bootstrap test (2000 replicates) are shown to the left 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).

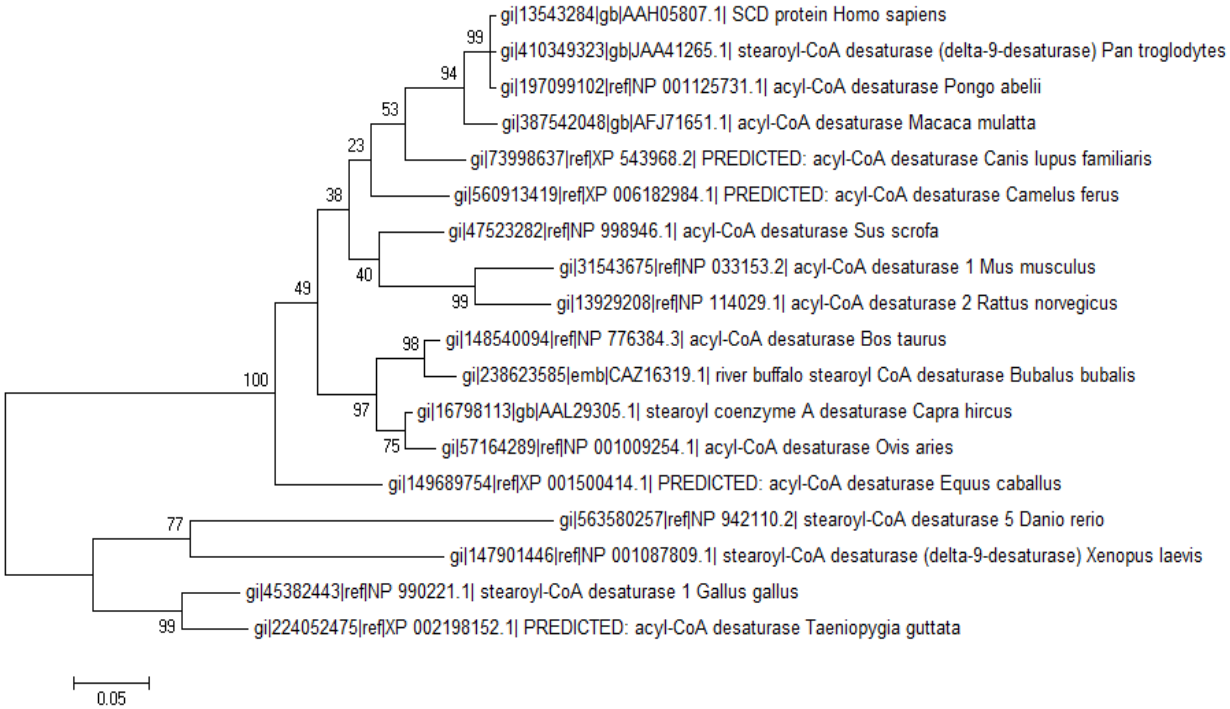

Fig. 3. Molecular phylogenetic analysis by the maximum likelihood method (17). The evolutionary history was inferred using the maximum likelihood method based on the JTT matrix-based model (17) with 2000 replicates of the bootstrap test.

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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 phylogenic 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 contains10 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 Taeniopy gia 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.

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Seq.nu.	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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