Multiple sequence alignment for SCD isoforms in humans and mice.

Key residues involved in substrate binding and iron binding are labeled as carets (^) and asterisks (*), respectively. The regions enclosed by dashed lines define three conserved His-box motifs. Sequence alignments were performed with Clustal Omega and edited in Jalview.

**Supplementary Figure 1**
X-ray fluorescence and X-ray absorption spectroscopy suggest that hSCD1 crystals contain zinc ions.
(a) The X-ray emission spectrum collected at 15000 eV shows two peaks at 8620 eV and 9549 eV corresponding to the Kα and Kβ X-ray emission lines of zinc; (b) The X-ray emission spectra collected at 15000 eV and 11000 eV (above the Zn K1s edge) show characteristic emission lines for zinc, while the spectrum collected at 9500 eV (below the Zn K1s edge) does not show these peaks. (c) X-ray absorption scan across the Zn K1s edge (9659 eV) shows the typical features of strong absorption.
Comparison of the dimetal center in hSCD1 with those in two soluble di-iron–containing enzymes.
(a) The di-metal center in hSCD1 is buried between TM2, TM4, CH2 and CH8. The zinc coordination residues (histidines) are shown as sticks. Zincs and the water molecule are illustrated as black and blue spheres, respectively. (b) The di-iron center in caster acyl-ACP desaturase (PDB code 1AFR) is surrounded by a four-helix bundle (α3, α4, α6 and α7). The iron coordination residues (glutamic acids and histidines) are shown as sticks. (c) The di-iron center in benzoyl-CoA epoxidase BoxB (PDB code 3PM5) is also surrounded by a four-helix bundle (αB, αC, αE and αF).
Supplementary Figure 4

Comparison of ligand interfaces in stearoyl-CoA–hSCD1 and benzoyl-CoA–epoxidase BoxB structures.

(a) The stearoyl-CoA-protein interaction in hSCD1; (b) the benzoyl-CoA-protein interaction in epoxidase BoxB
(PDB code 3PM5). Metals and substrates are shown as black spheres and yellow stick, respectively. Key residues involved in substrate interaction and metal coordination are shown as sticks. Hydrogen bonds and ionic interactions in the binding site are depicted as dashed lines.