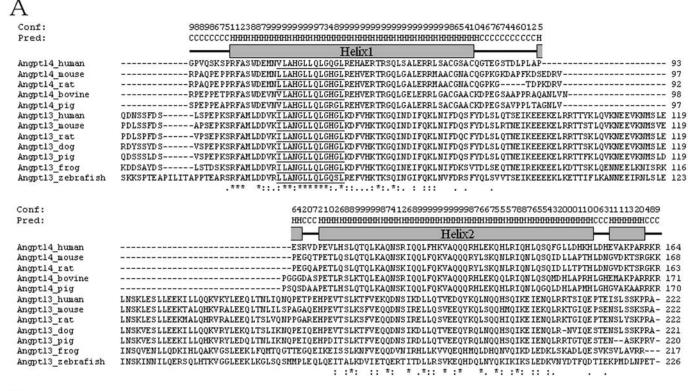
ONLINE SUPPLEMENTARY DATA (Yau at al)

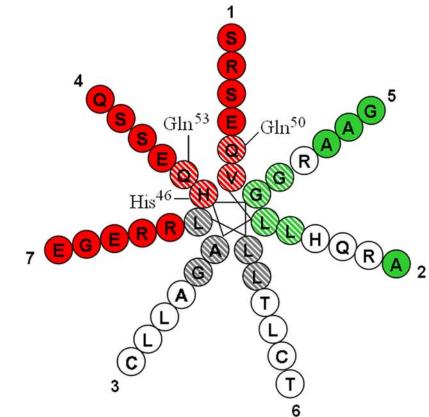
Primer	Oligonucleotide sequence
hA4-F-SalI	5'-TGAGGCGTCGACGGGACCCGTGCAGTCCAA-3'
hA4-R-XhoI	5'-TGAGGCCTCGAGCTATTCGCGCAGCCCCTG-3'
hA4-F-StyI	5'-TGAGGCGTCGACGGGACCCGTGCAGTCCAA-3'
H46A-F	5'-ATGAATGTCCTGGCGGCAGGACTCCTGCAGCTC-3'
H46A-R	5'-GAGCTGCAGGAGTCCTGCCGCCAGGACATTCAT-3'
Q50A-F	5'-GCGCACGGACTCCTGGCACTCGGCCAGGGGCTG-3'
Q50A-R	5'-CAGCCCCTGGCCGAGTGCCAGGAGTCCGTGCGC-3'
Q53A-F	5'-CTCCTGCAGCTCGGCGCAGGGCTGCGCGAACAC-3'
Q53A-R	5'-GTGTTCGCGCAGCCCTGCGCCGAGCTGCAGGAG -3'
465053A-F	5'-CTGGCGGCAGGACTCCTGGCACTCGGCGCAGGGCTG-3'
465053A-R	5'-CAGCCCTGCGCCGAGTGCCAGGAGTCCTGCCGCCAG-3'

Supplementary table-1: Primers used for sub-cloning human Angptl4-CCD and mutagenesis.

Supplementary Figure-1: Primary amino acid sequence analysis and secondary structural prediction for the NH₂-terminal CCD of Angptl4. A: multiple alignments of the amino acid sequences of CCD from all known Angptl3 and Angptl4 derived from different species. The alignments were generated by CLUSTAL W version 1.86 with default parameters. The amino acid sequences were aligned with their NH₂-terminal signal peptides omitted. The highly conserved 12-amino acid motif is underlined. <u>*</u> identical; <u>:</u> high similarity; <u>.</u> low similarity. The secondary structural prediction of hAngptl4-CCD was performed by using PSIPRED. Confidence of prediction (Conf) is shown on top (1-low; 9-high). Predicted structure (Pred) is annotated as "C" for free coil and "H" for α -helix. The diagrammatic annotation is shown above the sequence with strings as free coils and gray bars as α -helices. B. Helical-wheel plot showing the relative orientations of each repeat position in the Helix-1 at periodicity of "7/2" (i.e. 7 amino acid residues per 2 turns). The hydrophilic amino acid residues located on the solvent-exposed face are shown in green. The amino acid residues located on the solvent-exposed face are shown in green. The amino acid residues located on the solvent-exposed face.

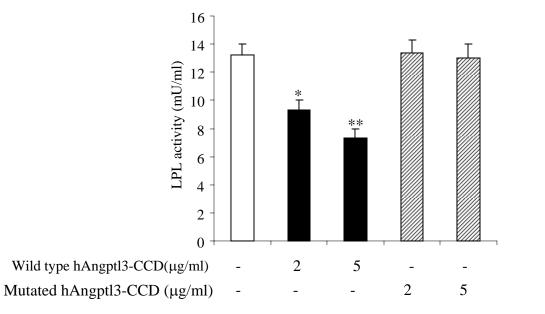
Supplementary Figure-1 (Yau et al)





В

Supplementary Figure-2 (Yau et al)



Supplementary Figure-2: The three polar amino acid residues within the coiled-coil domain are critical for the LPL inhibitory activity of Angptl3. Bovine LPL activities were measured as in Figure 1B, in the presence of different concentrations of wild type hAngptl3-CCD or mutated hAngptl3-CCD in which the three polar amino acid residues within the conserved motif were replaced by alanine. Open bar: control group; Black bar: wild type hAngptl3-CCD-treated groups; Hatched bar: mutated hAngptl3-CCD treated groups. *, p<0.05; **, p<0.01 *versus* control group or mutated hAngptl3-CCD treated groups (n=4-6).