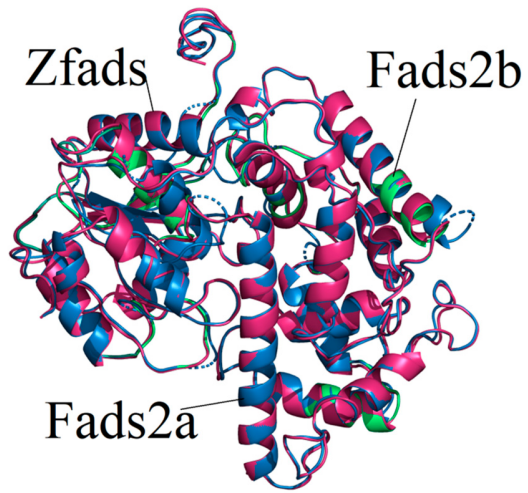


Figure S1. Multiple sequences alignments of protein sequence from common carp Fads2a and Fads2b.

Fads2a	MGGGGQQTDR	ITGTNARFST	YTWEEVQKHT	KSGDQWIVVE	RKVYNVSQWV
Fads2b	MGGGGQQTDR	LAGTNGRFGT	YTWEEVQKHT	KSGDQWIVVE	RKVYNVSQWV
		cytochrome b5-like heme/steroid binding domain			
Fads2a	KRHPGGLRII	GHYAGEDATE	AF ^T AFHPDLP	LVRKYMKPLL	IGELEASEPS
Fads2b	KRHPGGLRII	GHYAGEDATD	AF ^H AFHPNIQ	LVRKYMKPLL	IGELEASEPS
Fads2a	QDRQKN ^A ALV	EDFRALRERL	EAEGCFKTQP	LFF ^L LHLGHI	LLLE ^V IALML
Fads2b	QDRQKN ^G ALV	EDFRALRERL	EAEGCFKTQP	LFF ^I LHLGHI	LLLE ^A IALML
Fads2a	VWYFGTWIN	TAIV ^A VIMAT	AQSQAGWLQH	DFGHL ^S VCK ^S	SRW ^N HLVHKE
Fads2b	LWYFGTWIN	TAIV ^S VILAT	AQSQAGWLQH	DFGHL ^S VFK ^N	SRW ^D HLLHKE
		fatty acid desaturase domain			
Fads2a	VIGHLKGASA	GWWNHRHFQH	HAKPN ^V FKKD	PDVNMLN ^M FV	VG ^K VQPVEYG
Fads2b	VIGHLKGASA	GWWNHRHFQH	HAKPN ^I FKKD	PDVNMLN ^A FV	VG ^N VQPVEYG
Fads2a	VKK ^V KHLPYN	HQHKYFF ^F VG	PPLLIPV ^F FQ	FQI ^F HNMV ^S H	GLWVDL ^V WCI
Fads2b	VKK ^I KITLPYN	HQHKYFF ^F IG	PPLLIPV ^V FQ	FQI ^I QNM ^I TH	GLWVDL ^M WCI
Fads2a	SYVRYFLCY	TQFYGL ^F WAV	ILFN ^E VRFME	SHWFVWVTQM	SHIPM ^N IDYE
Fads2b	SYVRYFLCY	TQFYSV ^L WAV	LLFN ^I VRFME	SHWFVWVTQM	SHIPM ^D IDYE
Fads2a	KHQDWL ^S MQL	VATCNIEQSA	FNDWFSGHIN	FQIEHHLFPT	MPRHNYWRAA
Fads2b	KHQDWL ^N MQL	DATCNIEQSF	FNDWFSGHIN	FQIEHHLFPT	MPRHNYWRAA
Fads2a	PH ^V RALCDKY	GVKYQEKTLY	GAF ^A DIIRSL	EKS ^G EIWLDA	YLNK
Fads2b	PR ^V RALCDKY	GVKYQEKGLY	EAF ^V DIVRSL	EKS ^G EIWLDA	YLNK

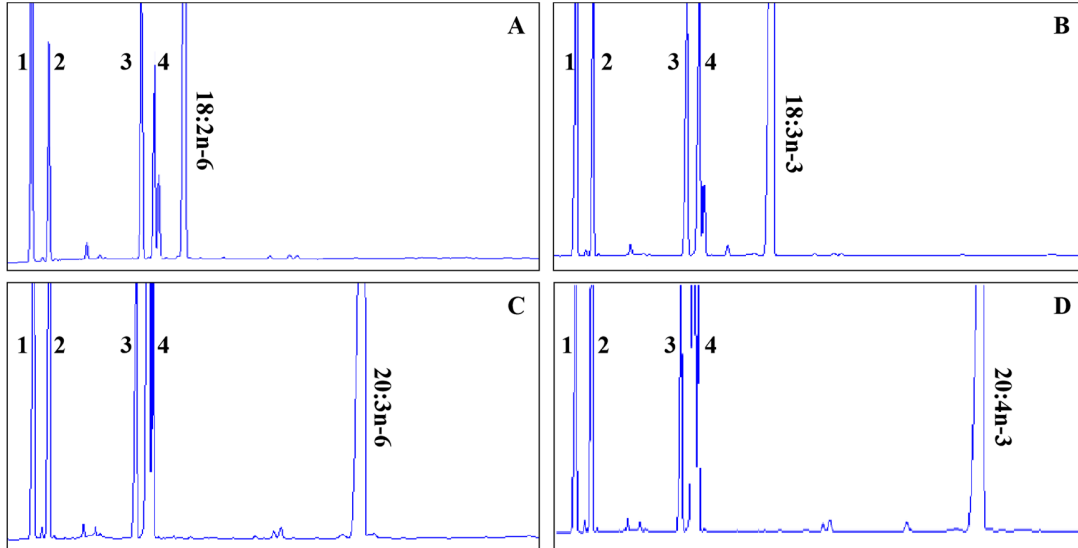
The red letters represent different amino acids between two proteins. The predicted domains were indicated by blue (cytochrome b5-like heme/steroid binding domain) and green (fatty acid desaturase domain) boxes.

Figure S2. The 3D structure of common carp Fads2a, Fads2b and zebra fish Fads2.



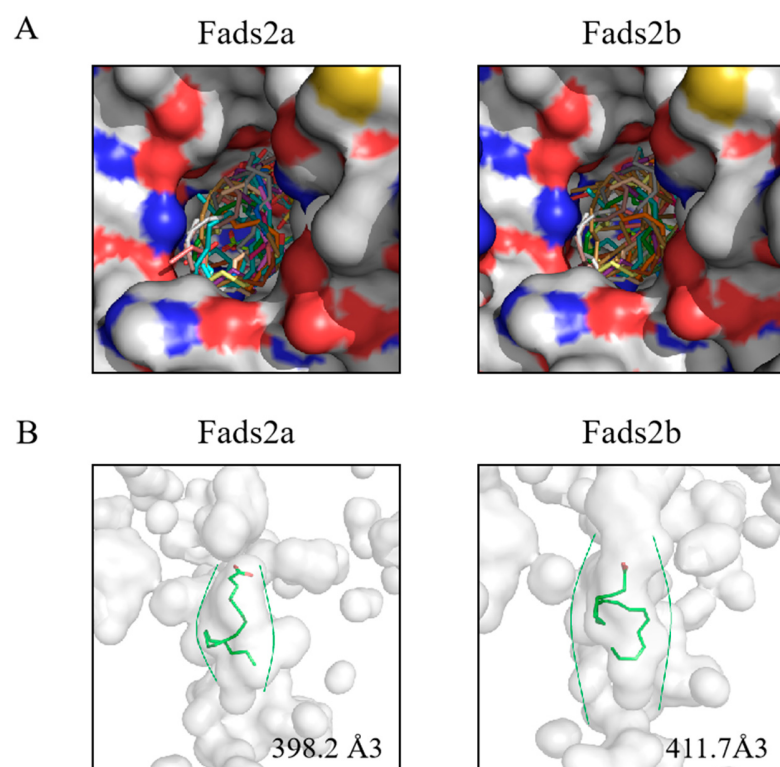
The 3D structure of common carp Fads2a (blue), Fads2b (green) and zebra fish Fads2 (Zfads, purple) protein of common carp predicted by homology modeling.

Figure S3. Characterization of desaturase activities of pYES2 without *fads2a* and *fads2b* cDNA inserts in transgenic yeast.



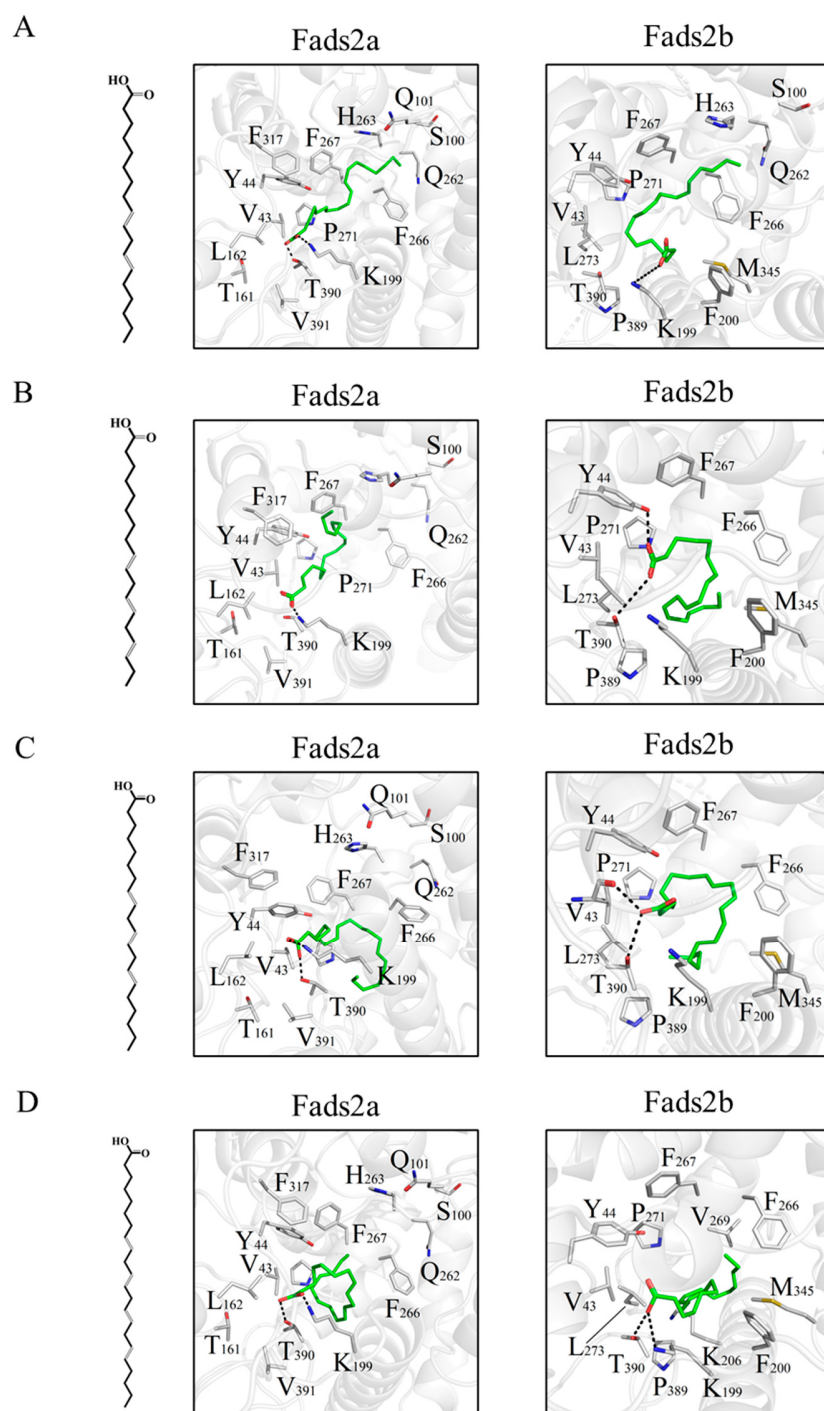
Characterization of desaturase activities of pYES2 without *fads2a* and *fads2b* cDNA inserts (negative control) in transgenic yeast (*S. cerevisiae*) towards grown in the presence of fatty acid substrates C18:2n-6 (A), C18:3n-3 (B), C20:3n-6 (C) and C20:4n-3 (D). Peaks 1 to 4 represent C16:0 (1), C16:1n-7 (2), C18:0 (3) and C18:1n-9 (4), respectively, which are the four main endogenous fatty acids of *S. cerevisiae*. The vertical axis displayed the flame-ionization detector response peak. The horizontal axis showed the peak retention time (min).

Figure S4. The modeling of common carp Fads2a and Fads2b.



(A) Results of molecular docking analysis between 10 fatty acid small molecules and Fads2a/Fads2b proteins in Common carp. (B) The pocket niche space of Fads2a and Fads2b proteins.

Figure S5. Docking pose of the $\Delta 6$ and $\Delta 5$ substrates small molecules in Fads2a and Fads2b protein.



Docking pose of the $\Delta 6$ and $\Delta 5$ substrates small molecules (green color) C18:2n-6 (A), C18:3n-3 (B), C20:3n-6 (C) and C20:4n-3 (D) in the backbone peptide structure of Fads2a/Fads2b protein (gray color).

Table S1. Genbank accessions of proteins used in the phylogenetic analysis.

Species	NCBI Reference Sequence
<i>Cyprinus carpio</i> Fads2a	XP_042570977.1
<i>Cyprinus carpio</i> Fads2b	XP_042609343.1
<i>Danio rerio</i>	NP_571720.2
<i>Homo sapiens</i>	NP_004256.1
<i>Mus musculus</i>	NP_062673.1
<i>Bos taurus</i>	NP_001076913.1
<i>Rattus norvegicus</i>	NP_112634.1
<i>Paralichthys olivaceus</i>	XP_019933638.1
<i>Lates calcarifer</i>	XP_018537439.1
<i>Salmo salar</i> Fads2.1	NP_001165251.1
<i>Salmo salar</i> Fads2.2	NP_001165752.1
<i>Carassius auratus</i> Fads2.1	XP_026094349.1
<i>Carassius auratus</i> Fads2.2	XP_026057919.1
<i>Sinocyclocheilus anshuiensis</i> Fads2.1	XP_016298063.1
<i>Sinocyclocheilus anshuiensis</i> Fads2.2	XP_016324433.1
<i>Larimichthys crocea</i>	NP_001290292.1
<i>Xiphophorus maculatus</i>	XP_005807011.1
<i>Poecilia formosa</i>	XP_007556956.1
<i>Oryzias latipes</i>	XP_004069638.1
<i>Oreochromis niloticus</i>	XP_005470692.1
<i>Trachinotus ovatus</i> Fads2a	QBA19079.1
<i>Trachinotus ovatus</i> Fads2b	QBA19080.1
<i>Lepisosteus oculatus</i>	AYG96562.1
<i>Siganus canaliculatus</i> Fads2.1	ADJ29913.1
<i>Siganus canaliculatus</i> Fads2.2	ABR12315.2
<i>Sparus aurata</i>	AAL17639.1

Table S2. Fatty acid composition of common carp embryos at different developmental stages.

Fatty acids (μg/mg)	0 hpf	24 hpf	48 hpf	72 hpf	96 hpf	120 hpf
C14:0	4.381±0.045	4.483±0.024	4.293±0.013	3.988±0.029	3.387±0.025	3.485±0.010
C15:0	1.531±0.029	1.956±0.008	2.046±0.009	2.328±0.014	2.579±0.012	2.487±0.010
C16:0	288.359±1.594	307.884±0.970	290.446±0.619	300.984±2.157	306.498±2.038	311.828±1.222
C18:0	165.870±7.783	106.741±0.318	89.112±0.235	105.497±0.540	117.402±0.449	128.205±0.164
C20:0	0.373±0.022	0.866±0.007	0.864±0.006	1.300±0.001	1.492±0.003	1.444±0.001
Total saturated	460.514±8.446	421.931±1.258	386.760±0.867	414.096±2.732	431.358±2.514	447.449±1.393
C16:1n-7	13.052±0.407	16.960±0.034	18.462±0.052	15.269±0.101	12.704±0.164	11.085±0.133
C18:1n-9	127.868±3.311	188.633±0.242	208.082±0.360	200.989±4.527	185.883±4.515	168.859±4.599
C20:1	11.747±0.348	9.880±0.030	10.910±0.036	10.185±0.046	9.687±0.027	8.354±0.020
Total monounsaturated	152.666±3.713	215.473±0.258	237.455±0.390	226.443±4.388	208.274±4.330	188.298±4.462
C18:2n-6	117.338±0.580	98.128±0.302	107.496±0.232	88.560±0.535	68.218±0.359	62.426±0.494
C18:3n-6	9.396±0.259	23.592±0.245	14.486±0.254	14.118±0.471	16.776±0.438	20.267±0.345
C20:2n-6	12.431±0.294	11.224±0.047	11.952±0.025	11.391±0.038	10.877±0.018	10.100±0.013
C20:3n-6	31.989±0.973	18.250±0.073	19.638±0.031	18.066±0.142	16.635±0.056	15.310±0.043
C20:4n-6	94.506±2.512	95.851±0.282	100.394±0.340	101.399±0.788	109.438±0.878	108.951±0.576
C22:4n-6	10.941±0.431	9.847±0.122	10.906±0.040	10.782±0.046	11.164±0.122	10.975±0.090
C22:5n-6	22.259±0.909	19.980±0.180	21.869±0.096	22.323±0.149	24.670±0.294	25.216±0.213
Total n-6 PUFA	298.860±3.657	276.872±0.613	286.741±0.640	266.640±1.070	257.778±1.939	253.245±1.582
C18:3n-3	5.702±0.111	3.740±0.001	4.131±0.009	2.997±0.024	2.008±0.013	1.984±0.025
C18:4n-3	0.630±0.019	1.304±0.344	0.644±0.007	0.980±0.381	0.802±0.334	0.561±0.131
C20:3n-3	1.281±0.034	1.867±0.263	1.500±0.327	1.821±0.158	1.755±0.144	1.875±0.087
C20:4n-3	0.591±0.037	0.758±0.209	0.789±0.236	1.019±0.245	1.086±0.531	0.323±0.010
C20:5n-3	4.050±0.056	3.415±0.019	3.534±0.014	3.335±0.028	3.158±0.026	3.249±0.041
C22:5n-3	4.506±0.187	5.110±0.198	5.035±1.129	4.950±0.382	4.385±0.335	5.024±0.869
C22:6n-3	71.200±1.467	69.579±0.308	73.341±0.594	77.643±0.669	89.435±0.965	98.058±0.745
Total n-3 PUFA	87.959±1.740	85.772±0.858	88.974±1.607	92.746±1.090	102.629±0.284	111.075±1.507
Total PUFA	386.819±5.295	362.644±1.470	375.714±1.169	359.385±1.957	360.408±2.202	364.320±3.085

Table S3. Primers for ORF sequence amplification of common carp *fads2a* and *fads2b*

Primers	Sequence (5'~3')	Product length
<i>fads2a</i> -ORF-F	AGTCAGAGTTTGATCAGTTATGGG	1335 bp
<i>fads2a</i> -ORF-R	GAGAGGTTTTATTTGTTGAGGTACG	
<i>fads2b</i> -ORF-F	GAGTTTGATCAGCGATGGGC	1335 bp
<i>fads2b</i> -ORF-R	GAGGTTTTATTTGTTGAGGTACGC	

Table S4. Primers for RACE amplification of common carp *fads2a* and *fads2b*

Primers	Sequence (5'~3')	Product length
5'RACE-F	AAGCAGTGGTATCAACGCAGAGT	820 bp
5'RACE- <i>fads2a</i> -R	TGAGCATGTTGACATCCGGG	
5'RACE-F	AAGCAGTGGTATCAACGCAGAGT	951 bp
5'RACE- <i>fads2b</i> -R	TGATGGTTGTAAGGCAGCGT	
3'RACE- <i>fads2a</i> -F	TCTTTGGGTGGACCTCGTGTGG	1069 bp
3'RACE Outer Primer	TACCGTCGTTCCACTAGTGATTT	
3'RACE- <i>fads2b</i> -F	GTGCTTTGGGCGGTGCTTCTG	952 bp
3'RACE Outer Primer	TACCGTCGTTCCACTAGTGATTT	
3'RACE- <i>fads2a</i> -F	TCTTTGGGTGGACCTCGTGTGG	1069 bp
3'RACE Inner Primer	CGCGGATCCTCCACTAGTGATTTCACTATAGG	
3'RACE- <i>fads2b</i> -F	GTGCTTTGGGCGGTGCTTCTG	952 bp
3'RACE Inner Primer	CGCGGATCCTCCACTAGTGATTTCACTATAGG	

Table S5. Primers for Yeast expression vector pYES2.0 inserts

Primers	Sequence (5'~3')	Restriction site
pYES2- <i>fads2a</i> -F	CCC <u>AAGCTT</u> AGTCAGAGTTTGATCAGTTATGGG	Hind III
pYES2- <i>fads2a</i> -R	CCG <u>CTCGAG</u> GAGAGGTTTTATTTGTTGAGGTACG	Xho I
pYES2- <i>fads2b</i> -F	CCC <u>AAGCTT</u> GAGTTTGATCAGCGATGGGC	Hind III
pYES2- <i>fads2b</i> -R	CCG <u>CTCGAG</u> GAGGTTTTATTTGTTGAGGTACGC	Xho I

Restriction sites are underlined in primers.

Table S6. Primers for time and spatial RT-qPCR of common carp

Primers	Sequence (5'~3')	Product length
<i>fads2a</i> -qPCR-F	CCAGTGGGTGAAGAGACACC	113 bp
<i>fads2a</i> -qPCR-R	ATTCCTCACCAGCGGAAGG	
<i>fads2b</i> -qPCR-F	GAAGAGACACCCCGGAGGAC	116 bp
<i>fads2b</i> -qPCR-R	GCGGCTTCATGTATTCCTCAC	
β -actin-F	TGCAAAGCCGGATTCGCTGG	100 bp
β -actin-R	AGTTGGTGACAATACCGTGC	

Table S7. Primer for WISH probe amplification

Primers	Sequence (5'~3')	Product length
WISH- <i>fads2a</i> -F	GCCTTTGCGGACATCATTAGG	365 bp
WISH- <i>fads2a</i> -R	AATCACACGACTTTCAGGCA	
WISH- <i>fads2b</i> -F	AGGGCTTGTATGAGGCCTTTG	345 bp
WISH- <i>fads2b</i> -R	TTCCAGGTCATTCATGCACGTT	

Table S8. Primers for Dual-luciferase reporter vector pGL3-Basic inserts

Primers	Sequence (5'~3')	Restriction site
<i>fads2a</i> -1-F (2.5K)	CTAG <u>CTAGC</u> GGGCAGGCTTCCAAGTCATA	Xho I
<i>fads2a</i> -2-F (2K)	CTAG <u>CTAGC</u> AGGTTTTTCACTGGAGGGCA	Xho I
<i>fads2a</i> -3-F (1.5K)	CTAG <u>CTAGC</u> ATGTGTCCCTGCTTTGACGG	Xho I
<i>fads2a</i> -4-F (1K)	CTAG <u>CTAGC</u> TGTGCCGATATCACGTGTCC	Xho I
<i>fads2a</i> -5-F (0.5K)	CTAG <u>CTAGC</u> GCAACGTTTTGGGCATTACAG	Xho I
<i>fads2a</i> -promoter-R	CCG <u>CTCGAG</u> CAGATCATTCGAGTGCCACC	Hind III
<i>fads2b</i> -1-F (2.5K)	CTAG <u>CTAGC</u> GCAGTGTGAGGACAGACAGAT	Xho I
<i>fads2b</i> -2-F (2K)	CTAG <u>CTAGC</u> AGTGAGCGAGTGCTGACATC	Xho I
<i>fads2b</i> -3-F (1.5K)	CTAG <u>CTAGC</u> CATGCAGGGCTGTGACCATT	Xho I
<i>fads2b</i> -4-F (1K)	CTAG <u>CTAGC</u> GTCATGAGCCAAGCTGATAAG	Xho I
<i>fads2b</i> -5-F (0.5K)	CTAG <u>CTAGC</u> GAAGGATGGTAAAATGCAACTG	Xho I
<i>fads2b</i> -promoter-R	CCG <u>CTCGAG</u> AGCGGAAGAGAGGCTCGC	Hind III

Restriction sites are underlined in primers.