Online Resource 2 for

Capability of DHA biosynthesis in a marine teleost, Pacific saury *Cololabis saira*: functional characterization of two paralogous Fads2 desaturases and Elovl5 elongase

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Online Resource 2 Thermocycling conditions of PCR conducted in this study

PCR	Forward primer	Reverse primer	Denature temperature (°C) and duration in seconds	Annealing temperature (°C) and duration in seconds	Extension temperature (°C) and duration in seconds	Number of cycles
fads2						
Partial CDS	fads2_partial_Fw	fads2_partial_Rv	94 (30)	58 (30)	72 (60)	35
fads2a						
5'RACE first	GeneRacer 5'	fads2a_5'RACE_R1	94 (30)	68 > 62* (30)	72 (60)	35
5'RACE nested	GeneRacer 5' nested	fads2a_5'RACE_Rn	94 (30)	68 > 62* (30)	72 (60)	35
3'RACE first	fads2a_3'RACE_F1	AdapterPrimer_R1	94 (30)	72 > 57* (30)	72 (60)	35
3'RACE nested	fads2a_3'RACE_Fn	AdapterPrimer_Rn	94 (30)	72 > 57* (30)	72 (60)	35
Complete CDS	fads2a_Fw	fads2a_Rv	98 (10)	55 (5)	72 (20)	35
Construction	fads2a_HindIII	fads2a_XbaI	98 (10)	55 (5)	72 (10)	35
Expression analysis	fads2a_RT_Fw	fads2a_RT_Rv				
fads2b						
5'RACE first	GeneRacer 5'	fads2b_5'RACE_R1	94 (30)	68 > 62* (30)	72 (60)	35
5'RACE nested	GeneRacer 5' nested	fads2b_5'RACE_Rn	94 (30)	68 > 62* (30)	72 (60)	35
3'RACE first	fads2b_3'RACE_F1	AdapterPrimer_R1	94 (30)	72 > 57* (30)	72 (60)	35
3'RACE nested	fads2b_3'RACE_Fn	AdapterPrimer_Rn	94 (30)	72 > 57* (30)	72 (60)	35
Complete CDS	fads2b_Fw	fads2b_Rv	98 (10)	55 (5)	72 (20)	35
Construction	fads2b_HindIII	fads2b_XbaI	98 (10)	55 (5)	72 (10)	35
Expression analysis	fads2b_RT_Fw	fads2b_RT_Rv	94 (30)	66 (30)	72 (30)	35

Online Resource 2 (continued)

PCR	Forward primer	Reverse primer	Denature temperature (°C) and duration in seconds	Annealing temperature (°C) and duration in seconds	Extension temperature (°C) and duration in seconds	Number of cycles
elovl5						
Partial CDS	elovl5_partial_Fw	elovl5_partial_Rv	94 (30)	50 (30)	72 (60)	35
5'RACE first	GeneRacer 5'	elovl5_5'RACE_R1	94 (30)	68 > 62* (30)	72 (60)	35
5'RACE nested	GeneRacer 5' nested	elovl5_5'RACE_Rn	94 (30)	68 > 62* (30)	72 (60)	35
3'RACE first	elovl5_3'RACE_F1	AdapterPrimer_R1	94 (30)	72 > 57* (30)	72 (60)	35
3'RACE nested	elovl5_3'RACE_Fn	AdapterPrimer_Rn	94 (30)	72 > 57* (30)	72 (60)	35
Complete CDS	elovl5_Fw	elovl5_Rv	98 (10)	50 (15)	72 (20)	35
Construction	elovl5_HindIII	elovl5_XbaI	98 (10)	55 (5)	72 (10)	35
Expression analysis	elovl5_RT_Fw	elovl5_RT_Rv	94 (30)	66 (30)	72 (30)	35
actb2						
Expression analysis	actb2_RT_Fw	actb2_RT_Rv	94 (30)	66 (30)	72 (30)	35

*Touchdown PCR was conducted. For decreasing annealing temperature 72 to 57°C and 68 to 62°C, it was decreased incrementally for the first 30 cycles by −0.5 and −0.2°C/cycle and then kept at 57 and 62°C for the remaining 5 cycles, respectively.