Comparative and Evolutionary Analyses Reveals the Conservation and Divergence of the Notch Pathway in the Lophotrochozoa

Xin He[a,b,c,f], Fucun Wu[a,b,e,f,*], Linlin Zhang[a,b,e,*], Li Li[a,d,e], Guofan Zhang[a,b,e]

[a] CAS and Shandong Province Key Laboratory of Experimental Marine Biology, Center for Ocean Mega-Science, Institute of Oceanology, Chinese Academy of Sciences, Qingdao, 266071, China
[b] Laboratory for Marine Biology and Biotechnology, Pilot National Laboratory for Marine Science and Technology, Qingdao 266237, China
[c] University of Chinese Academy of Sciences, Beijing 100039, China
[d] Laboratory for Marine Fisheries Science and Food Production Processes, Pilot National Laboratory for Marine Science and Technology, Qingdao 266237, China
[e] National and Local Joint Engineering Laboratory of Ecological Mariculture, Qingdao 266071, China
[f] The Innovation of Seed Design, Chinese Academy of Sciences, Wuhan 430072, China

[*] Corresponding author.
E-mail address: wufucun@qdio.ac.cn (FW), linlinzhang@qdio.ac.cn (LZ)

Table S1 Pfam Accession

Domain	**Notch**	**DSL**	**EGF**	**VWC**	**Presenilin**	**BTD**	**LAG1-DNAbind**	**HLH**
Pfam	PF00066	PF01414	PF00008	PF00093	PF01080.	PF09270.	PF09271	PF00010
Domain	**FGF**	**HH_signal**	**Insulin**	**MH2**	**Frizzled**	**Wnt**	**Activin_recp**	**TALPID3**
Pfam	PF00167	PF01085	PF00049	PF03166	PF01534	PF00110	PF01064	PF15324
Domain	**Homeobox**	**Forkhead**	**RAI16-like**	**STAT_bind**	**SH2**	**ERK-JNK_inhib**	**TGF_beta**	**Hairy_orange**
Pfam	PF00046	PF00250.	PF10257	PF02864	PF00017	PF15002	PF00019	PF07527

Table S2 The Sequence ID of the Major Genes in the Notch Pathway

	Notch	Delta	Jagged	Presenilin	Su(H)
Beroe ovata	UOYG01025213.1 UOYG01025214.1	-	-	UOYG01004404.1	UOYG01023648.1
Amphimedon queenslandica	ABZ79675.1 (NP_001266228.1)	XP_019852536.1 (*Aqu-Delta1*) XP_019858177.1 (*Aqu-Delta2*) NP_001292179.1 (*Aqu-Delta3*) NP_001292176.1 (*Aqu-Delta4*)	-	XP_019849527.1	XP_011410239.2
Nematostella vectensis	AEW42991.1 (*Nve-Notch*)	AEW42992.1 (*Nve-Delta*)	-	XP_001625236.1 (*Nve-Presenilin*)	XP_001633533.1 (*Nve-Su(H)*)
Hydra vulgaris	NP_001296638.1	XP_012561851.1	AEP83811.1	XP_012561939.1	XP_002162622.2
Hofstenia miamia	-	-	-	98014984|psn1 98020615|psn2	98031429|Su(H)
Schistosoma mansoni	XP_018647502.1 XP_018648078.1	XP_018655106.1	XP_018647863.1 XP_018649788.1 XP_018650604.1	ABS30416.1	XP_018650960.1
Echinococcus multilocularis	CDI98240.1	CDS35697.1 CDS37977.1	CDS39893.1	CDS40238.1	CDS42579.1
Lingula anatina	XP_013419105.1 (*Lan-Notch*)	XP_013393697.1 (*Lan-Delta*)	XP_013414285.1 (*Lan-Jagged*)	XP_013414065.1 (*Lan-Presenilin*)	XP_013403310.1 (*Lan-Su(H)*)
Platynereis dumerilii	CAJ38792.1	AKP06498.1	AKP06505.1	AKP06494.1	AKP06493.1
Capitella teleta	ELT93394.1	ELU04539.1	ELU03755.1	ELU08846.1	ELT98158.1
Helobdella robusta	XP_009013849.1, XP_009011066.1	XP_009015118.1	XP_009014186.1	XP_009026851.1 XP_009024256.1	XP_009017036.1 XP_009012889.1
Acanthopleura granulata	model.g31722.t1	asmbl_69547	model.g15382.t1	asmbl_61426	model.g1803.t1
Aplysia californica	XP_012943392.1	XP_012938459.1	XP_012939888.1	NP_001191418.1	XP_012944725.1
Elysia chlorotica	RUS91728.1	RUS85074.1	RUS89644	RUS69332.1	RUS90631.1
Biomphalaria	XP_013093464.1	XP_013094412.1	XP_013084238.1	XP_013065482.1	XP_013090963.1

glabrata					
Lottia gigantea	XP_009044486	XP_009061666.1 (*Lgi-Delta1*) XP_009046936.1 (*Lgi-Delta2*)	XP_009055674.1	XP_009047025.1	ESO97471.1
Haliotis discus hannai	T07122 (*Hdh-Notch*)	T19567 (*Hdh-Delta1*) T26181 (*Hdh-Delta2*)	T18809 (*Hdh-Jagged*)	T19349 (*Hdh-Presenilin*)	T13741 (*Hdh-Su(H)*)
Crassostrea gigas	CGI_10013186 (XP_011450291.1) (*Cgi-Notch*)	CGI_10009832 (*Cgi-Delta1*) CGI_10013656 (*Cgi-Delta2*)	CGI_10023897 (*Cgi-Jagged*)	CGI_10019579 (*Cgi-Presenilin*)	CGI_10018383 (*Cgi-Su(H)*)
Crassostrea virginica	XP_022322171.1	XP_022330248.1 XP_022320060.1	XP_022332849.1	XP_022332245.1	XP_022335897.1
Mizuhopecten yessoensis	XP_021352656.1	XP_021372652.1 XP_021356594.1	XP_021362559.1	XP_021349855.1	OWF47943.1
Chlamys farreri	CF64469.3	CF63471.32 CF493.17	CF17725.9	CF22817.10	CF64623.4
Pinctada fucata martensii	Pma_10025422 (*Pma-Notch*)	Pma_10018746 (*Pma-Delta1*) Pma_10023614 (*Pma-Delta2*)	Pma_10000893 (*Pma-Jagged*)	Pma_10014008 (*Pma-presenilin*) Pma_10009132 (*Presenilin-like*)	Pma_10008559 (*Pma-Su(H)*)
Octopus bimaculoides	XP_014776889.1	XP_014769856.1 XP_014771450.1	XP_014779348.1	XP_014785182.1	XP_014779920.1
Octopus vulgaris	XP_029649022.1	XP_029638366.1 XP_029644317.1	XP_029648604.1	XP_029642727.1	XP_029650905.1
Caenorhabditis elegans	NP_499007.1 NP_499014.1	NP_001024615.1	-	NP_508175.1, NP_492095.1	NP_001293739.1
Drosophila melanogaster	NP_001245510.1 (*Dme-Notch*)	NP_477264.1 (*Dme-Delta*)	NP_524527.3 (*Dme-Serrate*)	AAB53369.1 (*Dme-Presenilin*)	NP_476868.1 (*Dme-Su(H)*)
Apis mellifera	XP_006568051.2	XP_006566475.1	XP_006564793.1	XP_396620.5	XP_016767749.1
Strongylocentrotus purpuratus	XP_797451.4	NP_001027542.1	XP_011678556.1 XP_011678630.1 XP_011675859.1	XP_001178715.1	XP_011673426.1
Apostichopus japonicus	PIK60374.1 (*Aja-Notch*)	PIK54747.1 (*Aja-Delta*)	PIK49767.1 (*Aja-Jagged*)	PIK44116.1 (*Aja-Presenilin*)	PIK43190.1 (*Aja-Su(H)*)
Saccoglossus kowalevskii	XP_006818779.1	NP_001158418.1	XP_002740519.2	XP_002738746.1	XP_006811939.1
Styela clava	XP_039270515.1 (*Scl- Notch*)	XP_039248161.1 (*Scl-Delta*)	XP_039254307.1 (*Scl -Jagged*)	XP_039260250.1 (*Scl-Presenilin1*) XP_039254919.1 (*Scl-Presenilin2*)	XP_039265104.1 *(Scl-Su(H))*
Branchiostoma	XP_019643529.1	XP_019627312.1	XP_019633701.1	XP_019630833.1	XP_019626796.1

belcheri					XP_019647425.1
Danio rerio	NP_571516.1 (*Dre-Notch1a*) NP_571377.2 (*Dre-Notch1b*) NP_001108566.2 (*Dre-Notch2*) NP_571624.2 (*Dre-Notch3*)	NP_571029.2 (*Dre-dla*) NP_571033.1 (*Dre-dlb*) NP_571019.1 (*Dre-dlc*) NP_571030.2 (*Dre-dld*)	XP_005168261.1 (*Dre-jag1a*) NP_571938.2 (*Dre-jag1b*) XP_005160542.1 (*Dre-jag2b*)	NP_571099.1 (*Dre-psn1*) NP_571589.2 (*Dre-psn2*)	NP_942579.2 (*Dre-rbpja*) NP_001077020.1 (*Dre-rbpjb*)
Mus musculus	Notch1-NP_032740.3 Notch2-NP_035058.2 Notch3-NP_032742.1 Notch4-NP_035059.2	XP_011244569.1 NP_031892.2 NP_062327.2	NP_038850.1 XP_011242300.1	NP_001349200.1 NP_001122077.1	NP_001074397.1 NP_033062.1
Homo sapiens	Notch1-NP_060087.3 Notch2-NP_077719.2 Notch3-NP_000426.2 Notch4-NP_004548.3	NP_005609.3 NP_061947.1 NP_058637.1	NP_002217.3 NP_000205.1	NP_000012.1 NP_000438.2	NP_001268377.1 NP_001350506.1

Table S3 The *Hes*/*Hey*-related Genes among metazoans

	Hes	Hey	Helt	Cwo
Beroe ovata	-	-	-	-
Amphimedon queenslandica	-	AquHey-XP_003387453.1	-	-
Nematostella vectensis	NveHes2-AEW42994.1 or XP_001635328.1 NveHes4-AEW42997.1 NveHes3-XP_001635326.1	NveHey-AEW42993.1	-	-
Hydra vulgaris	HvuHes-XP_004208005.1	-	-	-
Hofstenia miamia	HmiHes-98048776	HmiHey-98033833	-	-
Schistosoma mansoni	SmaHes1-XP_018647563.1 SmaHes2-XP_018653208.1	SmaHey1-XP_018649397.1 SmaHey2-XP_018649396.1	-	-
Echinococcus	EmuHes-CDS38030.1	EmuHey-CDS426	-	-

multilocularis		97.1		
Lingula anatina	LanHES1-XP_013381587 LanHES2-XP_013381582 LanHES3-XP_013383390 LanHES4-XP_013383388 LanHES5-XP_013403236 LanHES6-XP_013412582 LanHES7-XP_013412583 LanHES8-XP_013412580 LanHES9-XP_013395724 LanHES10-XP_013395728 LanHES11-XP_013395690 LanHES12-XP_013390924	LanHey-XP_013414701	-	LanCwo1-XP_013419897 LanCwo2-XP_013380691
Platynereis dumerilii	PduHes1-AGS55435.1 PduHes2-AGS55436.1 PduHes3-AGS55437.1 PduHes4-AGS55438.1 PduHes5-AGS55439.1 PduHes6-AGS55440.1 PduHes7-AGS55441.1 PduHes8-AGS55442.1 PduHes9-AGS55443.1 PduHes10-AGS55444.1 PduHes11-AGS55445.1 PduHes12-AGS55446.1 PduHes13-AGS55447.1	PduHey1-AGS55448.1	-	PduCwo-AGS55449.1
Capitella teleta	CteHes1-ELU08377 CteHes2-ELU08376 CteHes3-ELU10814 CteHes4-ELU10815 CteHes5-ELT92193	CteHey1-ELU08981 CteHey2-ELU08980	-	CteCwo-ELU02556
Helobdella robusta	HroHES1-XP_009020819 HroHES2-XP_009022784 HroHES3-XP_009023789 HroHES4-XP_009023790 HroHES5-XP_009023792	HroHey-XP_009014197	-	-
Acanthopleura granulata	AgrHes1-model.g30019.t1 AgrHes2-model.g30018.t1 AgrHes3-model.g30014.t1 AgrHes4-model.g30046.t1 AgrHes5-model.g30020.t1 AgrHes6-model.g31256.t1 AgrHes7-model.g29508.t1	AgrHey-model.g19192.t1	AgrHelt-model.g4531.t1	-
Aplysia	AcaHes1-XP_005109821	AcaHey-XP_0129	--	AcaCwo-XP_012935276

californica	AcaHes2-XP_012945161 AcaHes3-XP_005104468	40775		
Elysia chlorotica	EchHes1-RUS81509.1 EchHes2-RUS85462.1 EchHes3-RUS85031.1 EchHes4-RUS87118.1 EchHes5-RUS83438.1 EchHes6-RUS72576.1	EchHey-RUS87300.1	-	EchCwo-RUS91494.1
Biomphalaria glabrata	BglHes1-XP_013095166 BglHes2-XP_013088757 BglHes3-XP_013075104 BglHes4-XP_013087343 BglHes5-XP_013088081	BglHey-XP_013067329	-	BglCwo-XP_013080510
Lottia gigantea	LgiHes1-XP_009064533 LgiHes2-XP_009058075 LgiHes3-XP_009057811 LgiHes4-XP_009058863 LgiHes5-XP_009049759 LgiHes6-XP_009053615 LgiHes7-XP_009053062 LgiHes8-XP_009058074 LgiHes9-XP_009064532 LgiHes10-XP_009064534 LgiHes11-XP_009064535	LgiHey-XP_009046688	LgiHelt-XP_009045357	LgiCwo-XP_009053558
Haliotis discus hannai	HdhHes1-T00679 HdhHes2-T00683	HdhHey-T01691	-	HdhCwo-T09094
Crassostrea gigas	CgiHes1-XP_011426791 CgiHes2-XP_011426827 CgiHes3-XP_011434420 CgiHes4-XP_011442866 CgiHes5-XP_011434419 CgiHes6-XP_011456332 CgiHes7-XP_011456334 CgiHes8-XP_011456335 CgiHes9-XP_011418427 CgiHes10-XP_011418430	CgiHey-XP_011446705	CgiHelt-XP_011454244	CgiCwo1-XP_011423628 CgiCwo2-XP_011423478
Crassostrea virginica	CviHes1-LOC111132480 CviHes2-LOC111133867 CviHes3-LOC111099184 CviHes4-LOC111135582 CviHes5-LOC111132221 CviHes6-LOC111132222 CviHes7-LOC111138134	CviHey-LOC111131773	CviHelt-LOC111120884	CviCwo1-XP_022302549.1 CviCwo2-XP_022301976.1
Mizuhopecten	MyeHes1-XP_021342518	MyeHey-XP_0213	MyeHelt-XP_021340917	MyeCwo-XP_021371008

yessoensis	MyeHes2-XP_021364693 MyeHes3-XP_021350063 MyeHes4-XP_021372905 MyeHes5-XP_021363795 MyeHes6-XP_021355435	56185		
Chlamys farreri	CfaHes1-CF20445.6 CfaHes2-CF57753.3 CfaHes3-CF59079.38 CfaHes4-CF64955.1 CfaHes5-CF59079.39	CfaHey-CF63447.3	CfaHelt-CF55315.5	CfaCwo-CF43523.5
Pinctada fucata martensii	PmaHes1-10022586 PmaHes2-10012161 PmaHes3-10022584	PmaHey1-10000662 PmaHey2-10019999	PmaHelt-10020900	PmaCwo1-10029931
Octopus vulgaris	OvuHes-XP_029657891.1	-	OvuHelt-XP_029638240.1	OvuCwo-XP_029635006.1
Octopus bimaculoides	ObiHes-XP_014772623	ObiHey-XP_014767679	ObiHelt-XP_014781134	ObiCwo-XP_014773539
Caenorhabditis elegans	CelHes-NP_500281.1	-	-	-
Drosophila melanogaster	DmeHes1-NP_524509 (*Dme-E(spl)m3*) DmeHes2-NP_524505 (*Dme-E(spl)mbeta*) DmeHes3-NP_536753 (*Dme-E(spl)m7*) DmeHes4-NP_524504 (*Dme-E(spl)mgamma*) DmeHes5-NP_525094 (*Dme-Hesr*), DmeHes6-HLH-NP_524503 (*Dme-E(spl)mdelta*) DmeHes7-NP_524511 (*Dme-E(spl)m5*) DmeHes8-NP_524513 (*Dme-E(spl)m8*) DmeHes9-NP_476923 (*Dme-dpn*), DmeHes10-NP_523599 (*Dme-Sidpn*) DmeHes11-NP_523977 (*Dme-hairy*)	DmeHey-NP_523657 (*Dme-Hey*)	-	DmeCwo-NP_524775 (*Dme-Cwo*)
Apis mellifera	AmeHes1-XP_393312.1 AmeHes2-XP_001120327.2 AmeHes3-XP_003249471.2 AmeHes4-XP_001120814.2 AmeHes5-XP_006559646.1	AmeHey-XP_026299131.1	-	AmeCwo-XP_026300267.1

Strongylocentrotus purpuratus	AmeHes6-XP_393948.3 SpuHes1-NP_001001768.2 SpuHes2-XP_787040.2 SpuHes3-XP_796692.2 SpuHes4-XP_030843279.1 SpuHes5-XP_030843275.1	SpuHey-XP_783676.2	-	-
Apostichopus japonicus	AjaHes1-PIK61026.1 AjaHes2-PIK35797.1 AjaHes3-PIK37883.1 AjaHes4-PIK58850.1	AjaHey-PIK40047.1	-	-
Saccoglossus kowalevskii	SkoHes1-NP_001158466.1	SkoHey1-NP_001158467.1 SkoHey2-XP_006811443	SkoHelt-NP_001161564.1	
Styela clava	SclHes1-XP_039251338.1 SclHes2-XP_039259018.1 SclHes3-XP_039250505.1	SclHey-XP_039261038.1	-	-
Branchiostoma belcheri	BbeHes1-XP_019637861.1 BbeHes2-XP_019637894.1 BbeHes3-XP_019637880.1 BbeHes4-XP_019628064.1 BbeHes5-XP_019628076.1 BbeHes6-XP_019628086.1 BbeHes7-XP_019628069.1 BbeHes8-XP_019633323.1 BbeHes9-XP_019627214.1 BbeHes10-XP_019633783.1	BbeHey1-XP_019641636.1 BbeHey2-XP_019647772.1	-	-
Danio rerio	DreHes1-NP_571155.1 (*Dre-her3*) DreHes2-NP_571154.2 (*Dre-her6*) DreHes3-NP_571948.1 (*Dre-her9*) DreHes4-NP_001116717.1 (*Dre-hes2.1*) DreHes5-NP_001038818.1 (*Dre-hes2.2*) DreHes6-NP_955918.3 (*Dre-her8a*) DreHes7-XP_003199574.1 (*Dre-her15.1*) DreHes8-XP_021324225.1 (*Dre-E(spl)m7*) DreHes9-NP_878295.1 (*Dre-her15.1*) DreHes10-NP_001159638.1 (*Dre-her8.2*)	DreHey1-NP_997726.1 (*Dre-hey1*) DreHey2-NP_571697.2 (*Dre-hey2*) DreHey3-NP_859425.1 (*Dre-heyl*)	DreHelt-NP_996948.1 (*Dre-helts*)	-

Mus musculus	MmuHes1-NP_032261.1 MmuHes2-NP_001288734.1 MmuHes3-NP_032263.2 MmuHes4-NP_001357684.1 MmuHes5-NP_062352.1 MmuHes6- NP_149030.2	MmuHey1-NP_034553.2 MmuHey2-NP_038932.1 MmuHey3-NP_038933.2	MmuHelt-NP_776150.2	-
Homo sapiens	HsaHes1-NP_001019769 HsaHes2-NP_001010926 HsaHes3-NP_061962 HsaHes4-NP_061115 HsaHes5-NP_005515 HsaHes6-NP_066993 HsaHes7-NP_003661 HsaHes8-NP_110389	HsaHey1-NP_036390 HsaHey-2NP_036391 HsaHey-3NP_055386	HsaHelt-NP_001025058	-

Table S4 The FPKM/RPKM and abbreviations of the developmental stages.
See excel file “supplementary Table S4.xlsx”.

Table S5 The Core Domains of Developmental Pathways.

	TGF-β[a]			Wnt[b]		Jak/STAT[c]		RPTK[d]		Notch[e]			Hedgehog[f]		RA[g]	Fox[h]	Hox[i]	ERK[j]
	Activin_recp	MH2	TGF_beta	Wnt	Frizzled	STAT_bind	SH2	Insulin	FGF	**EGF**	**DSL**	**bHLH**	HH_signal	TALPID3	RAI16-like	Forkhead	Homeobox	ERK-JNK_inhib
Bov	2	6	8	5	4	3	29	-	5	57	25	43	-	-	-	22	69	1
Aqu	2	4	8	5	5	2	87	-	2	225	12	23	1	-	2	19	33	1
Nve	3	5	7	19	7	1	25	2	14	643	24	51	2	-	2	27	94	1
Hvu	2	4	12	11	3	1	47	1	12	172	16	29	4	2	2	16	46	1
Hmi	1	6	6	6	8	1	60	-	3	33	1	39	1	1	-	17	90	2
Sma	1	9	2	8	9	-	33	-	-	43	6	35	1	-	2	16	75	1
Emu	-	7	3	7	9	-	44	-	-	42	8	31	2	-	2	16	81	-
Lan	4	10	11	13	5	2	65	2	3	1427	6	77	1	2	2	27	114	2
Cte	3	4	14	12	5	5	42	2	1	571	7	82	1	3	2	42	145	1
Hro	2	9	8	19	4	2	60	-	-	230	3	67	1	-	1	32	243	1
Agr	2	5	12	10	5	2	51	4	5	227	11	67	1	2	2	35	144	1
Ech	2	3	13	12	5	1	46	6	3	138	13	64	2	1	1	21	93	1
Aca	1	5	12	16	5	1	52	4	4	292	12	64	2	2	2	19	90	1
Bgl	1	7	14	18	5	2	55	2	3	280	16	73	1	2	3	18	86	1
Lgi	4	5	10	12	5	2	28	3	2	334	6	77	1	1	3	31	141	1
Hdh	3	4	9	14	6	1	58	3	4	447	19	77	6	1	2	25	145	-
Cgi	4	8	13	13	5	1	51	2	1	633	19	78	1	2	2	26	131	1
Cvi	4	13	18	13	5	3	78	4	2	821	24	101	3	2	2	33	144	1
Mye	4	6	13	12	5	3	63	2	2	593	27	69	6	2	2	25	134	1
Cfa	3	6	14	13	6	3	55	2	2	450	16	66	6	1	2	27	130	1
Pma	2	6	12	10	8	2	52	1	2	475	10	59	3	2	2	39	134	-
Ovu	1	5	10	12	7	1	50	1	2	425	16	51	2	-	2	16	82	1
Obi	1	6	11	13	5	1	42	1	4	267	7	48	3	1	2	19	81	1

Cel	3	20	5	5	8	3	43	1	2	88	12	54	-	-	1	24	131	1
Dme	5	6	7	8	6	1	31	6	3	105	2	55	1	-	1	18	108	-
Ame	10	5	8	6	2	1	35	2	4	139	2	71	1	-	2	16	99	1
Spu	2	8	18	16	8	1	60	5	5	2933	21	65	1	2	2	24	130	1
Aja	1	5	13	16	5	1	71	1	-	661	2	35	-	3	2	24	82	1
Sko	2	7	23	13	6	1	45	2	7	1161	6	63	1	2	3	41	142	1
Scl	3	4	14	17	8	3	55	-	6	452	3	46	2	2	2	35	115	2
Bbe	5	5	16	13	7	2	62	5	10	1499	2	86	3	2	2	27	141	1
Dre	13	15	14	26	15	8	147	14	32	417	10	149	4	1	8	69	232	1
Mmu	11	8	41	20	12	7	116	11	22	278	4	138	4	1	5	43	305	1
Hsa	10	7	21	40	12	8	128	15	26	336	4	126	4	1	4	48	290	1
Total	117	233	435	453	220	76	1966	104	193	**16894**	**372**	**2259**	72	41	74	937	4300	33

The line means none. The domains of Notch Pathway are underlined.

a Activin is a TGF receptor; MH2 is a domain of Smad; TGF_beta is a multifunctional peptide of TGF-β.

b Wnt is a family of Wnt signaling pathway. Frizzled is related to Wnt and Hedgehog.

c STAT proteins are a family of transcription factors of Jak/STAT, SH2 is a conserved domain included in both STAT and RPTK signaling pathway.

d Insulin, FGF and EGF are subfamilies of receptor protein-tyrosine kinase (RPTK) pathway.

e EGF is contained in the Notch ligand as well. DSL is the key domain of Notch ligands while bHLH is the key domain of Notch target genes.

f HH_signal is an amino-terminal signaling domain of Hedgehog pathway. TALPID3 is a family of eukaryotic proteins that are targets for Hedgehog signaling;

g RAI16-like is a retinoic acid-induced protein 16-like proteins of retinoic acid (RA) signaling pathway.

h Forkhead domain is a conserved DNA-binding domain of fox gene.

i Homebox is a domain of transcriptional regulators which encoded by Hox genes.

j ERK-JNK_inhib is an inhibitor of extracellular signal-regulated kinases (ERK).

Colors of background for the species represent different phylum. White is Ctenophora, light grey is Porifera, light blue is Cnidaria, dark blue is Xenacoelomorpha, blue is Lophotrochozoa, orange is Ecdysozoa, red is Ambulacrum (Echinodermata and Hemichordata) and green is Chordata (the same below). *Platynereis dumerilii* was discarded due to lack of proteome.

Abbreviation of species are showed in Materials & Methods.

Table S6 Domains of the Major Genes in the Notch Pathway

	Notch	DSL	EGF	VWC	Presenilin	BTD	LAG1-DN Abind	bHLH	Hairy orange
Bov	8	25	57	9	1	1	1	43	7
Aqu	5	12	225	4	1	1	1	23	1
Nve	3	24	643	10	1	1	1	51	5
Hvu	5	16	172	12	1	1	1	29	-
Hmi	-*	1*	33*	3*	1	1	1	39	1
Sma	2	6	43	-	1	1	1	35	1
Emu	4	8	42	1	1	1	1	31	-
Lan	13	6	1427	52	1	1	1	77	10
Cte	7	7	571	17	1	1	1	82	8
Hro	5	3	230	5	2	2	2	67	2
Agr	6	11	227	52	1	1	1	67	11
Ech	3	13	138	26	1	1	1	64	7
Aca	6	12	292	39	1	1	1	64	5
Bgl	8	16	280	16	1	1	1	73	10
Lgi	5	6	334	41	1	1	1	77	7
Hdh	5	19	447	30	1	2	2	77	11
Cgi	6	19	633	44	1	1	1	78	10
Cvi	7	24	821	74	1	1	1	101	9
Mye	8	27	593	42	1	1	1	69	7
Cfa	8	16	450	32	1	1	1	66	7
Pma	7	10	475	38	3	1	1	59	6
Ovu	8	16	425	38	1	1	1	51	1
Obi	5	7	267	39	1	1	1	48	4
Cel	6	12	88	12	2	1	1	54	-
Dme	4	2	105	9	1	1	1	55	11
Ame	4	2	139	34	1	1	1	71	9
Spu	6	21	2933	91	1	1	1	65	7
Aja	4	2	661	60	1	1	1	35	4
Sko	8	6	1161	45	1	1	1	63	5
Scl	5	3	452	47	2	2	2	46	3
Bbe	6	2	1499	90	1	2	2	86	11
Dre	16	10	417	68	2	3	3	149	15
Mmu	17	4	278	60	2	2	2	138	11
Hsa	18	4	336	63	2	2	2	126	11
Total	228	371	16861	1200	42	42	42	2259	217

Figure S1. DSL domain phylogeny across lophotrochozoans. (a) The phylogenetic tree was constructed using the DSL domains from 138 DSL family genes using maximum likelihood methods. Clades with different colours represent different domain arrangements shown in (b). Red indicates *Jagged* containing DSL, EGF, MNLL, and VWC. Orange indicates *Delta* containing DSL, EGF, and MNLL. Blue represents genes containing DSL and EGF domains. Purple represents genes containing DSL or DSL repeats but not EGF. Different DSL domains in the same gene are indicated by '-' after the gene ID. (b) Domain diagram of DSL family. DSL domain protein sequences are given in Supplementary Data 2.
See "Figure S1.pdf" file.

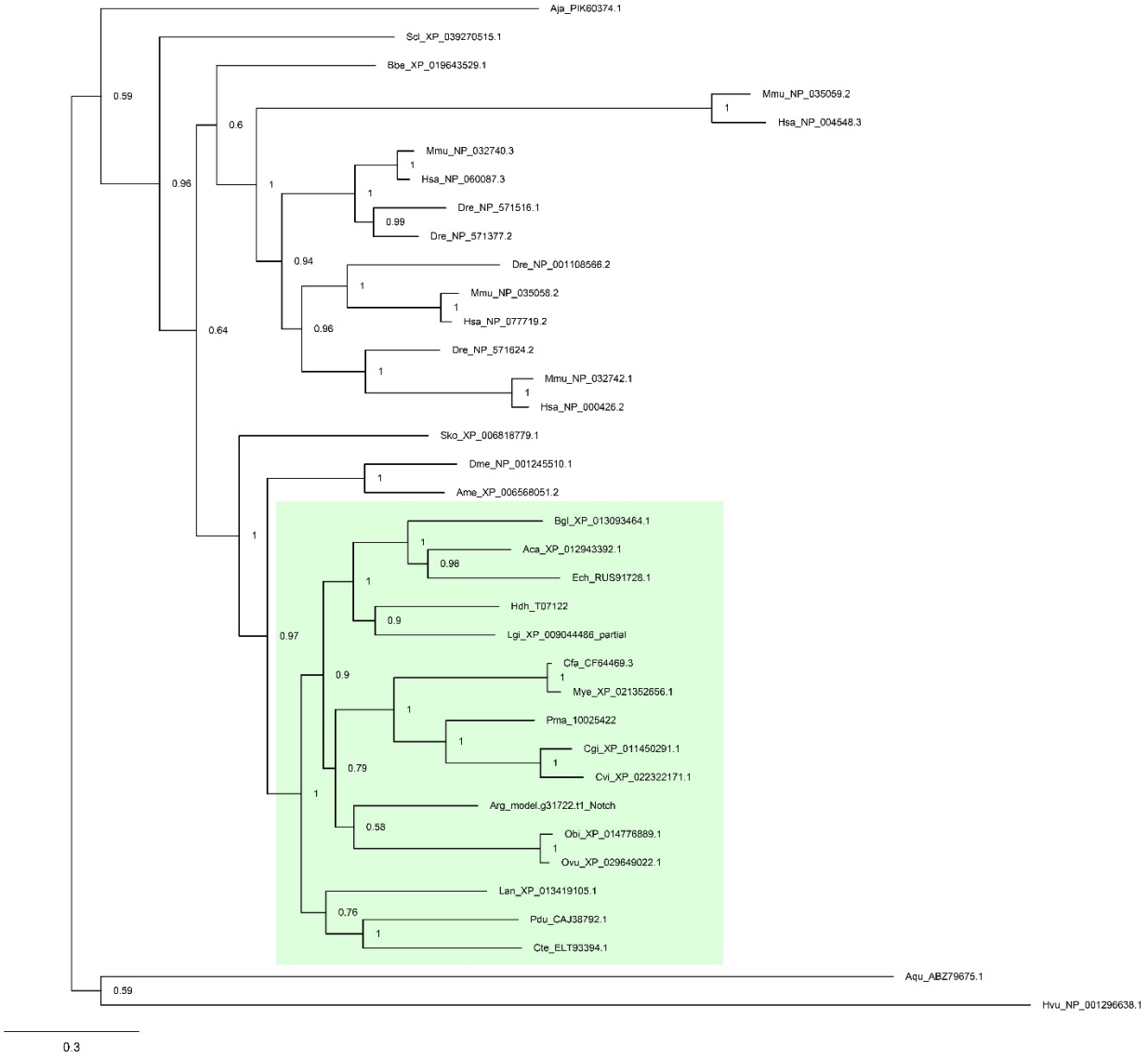

Figure S2. Phylogeny of *Notch* genes across metazoans. The phylogenetic tree was constructed using 36 *Notch* genes from 27 complete metazoan genomes using the maximum likelihood method. Fasttree support values are shown at the basal node. Lophotrochozoans are shown in green. The protein sequences of these genes are shown in Supplementary Data 4.

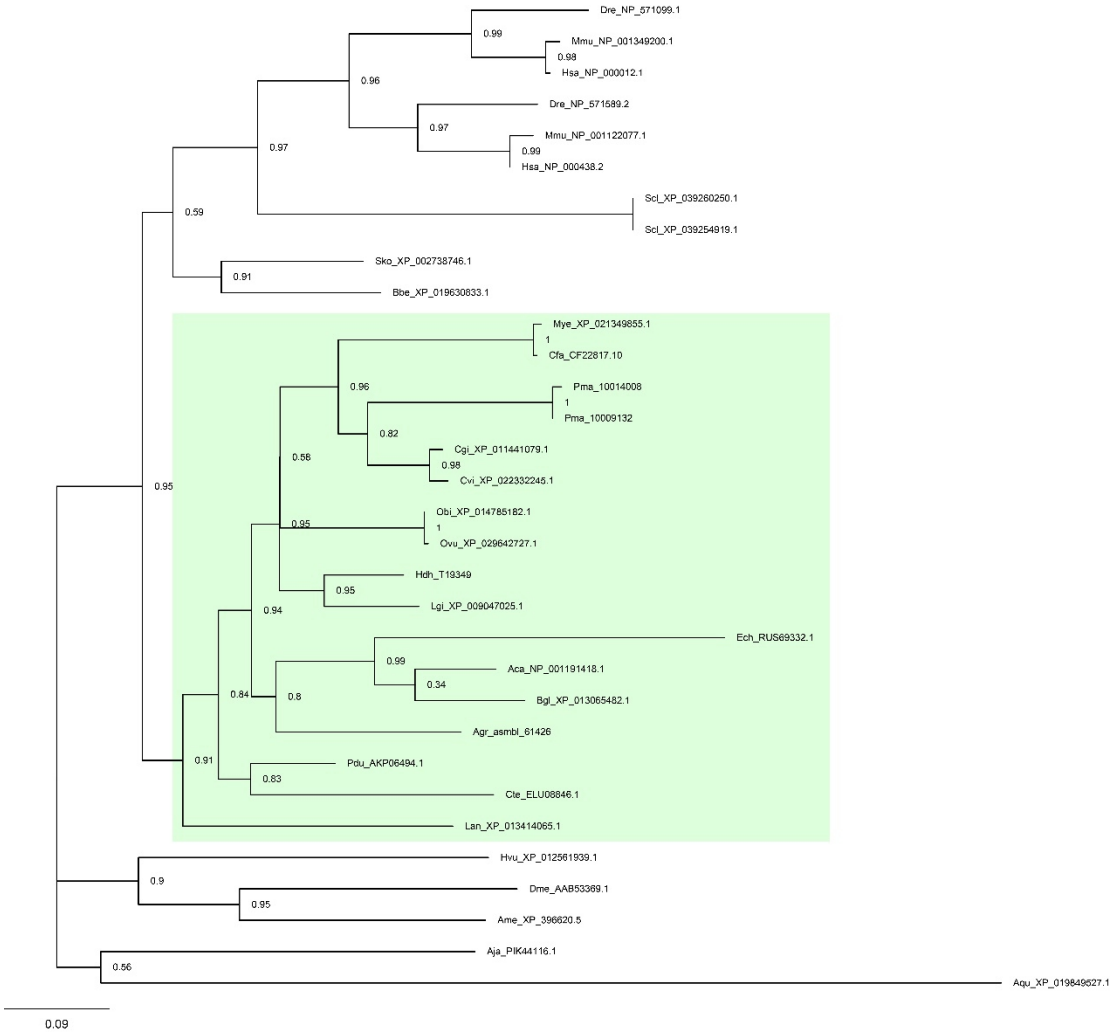

Figure S3. Phylogeny of *Presenilin* genes across metazoans. The phylogenetic tree was constructed using 32 *Presenilin* genes from 27 complete metazoan genomes using the maximum likelihood method. Fasttree support values are shown at the basal node. Lophotrochozoans are shown in green. The protein sequences of these genes are shown in Supplementary Data 5.

Figure S4. Phylogeny of *Su(H)* genes across metazoans. The phylogenetic tree was constructed using 31 *Su(H)* genes from 27 complete metazoan genomes using the maximum likelihood method. Fasttree support values are shown at the basal node. Lophotrochozoans are shown in green. The protein sequences of these genes are shown in Supplementary Data 6.

Figure S5. Transcriptomic data of the Notch pathway genes (*Notch*, *Delta*, *Jagged*, *Presenilin,* and *Su(H)*) in eleven species throughout development. The heatmaps show the expression trends. Red means high while white means low. Sequence IDs are shown in Supplementary Table S2 and S3. Developmental stage abbreviations are described in Supplementary Table S4.
See “Figure S5.pdf” file.