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B

me_Sox9a	PPTTPKTDC--SAKMDLKREGGLRPLPDGAPGRQLNIDFRDVDIGELSSDVISHIETFDV	293
Ntil_Sox9a	PPTTPKTDV--SSGKVDLKREVGLRSLPDGPGGRQLNIDFRDVDIGELSSDVISHIETFDV	295
plf_Sox9a	PPTTPKTDV--GSGKTDLKRAGLRLPDGNVRGQLNIDFRDVDIGELSSDVISHIETFDV	296
gup_Sox9a	PPATPKTDT--GSGKMDLKREAGLRLPDGNVRGQLNIDFRDVDIGELSSDVISHIETFDV	296
Am_Sox9a	PPATPKTDA--GSGKMDLKREAGLRLPEGNVGRQLNIDFRDVDIGELSSDVISHIETFDV	296
me_Sox9b	PPTTPKTDL--PSSKAIDLKREGR--PVQEG-TSRQLNIDFGTVDIGELSSDVISNIGSFDV	289
Ntil_Sox9b	PPTTPKTDL--PSSKAIDLKREGR--PMQEG-TSRQLNIDFGAVDIGELSTDVISNIGSFDV	289
Ghol_Sox9bX	PPTTPKTDL--SSSKADLKREGR--PIQEG-SSRQLNIDFGAVDIGELSCEVISNIGSFDV	289
Ghol_Sox9bY	PPTTPKTDL--SSSKADLKREGR--PIQEG-SSRQLNIDFGAVDIGELSCEVISNIGSFDV	289
gup_Sox9b	PPTTPKTDL--SSSKADLKREGR--PIQEG-SSRQLNIDFGAVDIGELSSDVISNIGSFDV	289
plf_Sox9b	PPTTPKTDL--SSSKADLKREGR--PIQEG-SSRQLNIDFGAVDIGELSSDVISNIGSFDV	289
Am_Sox9b	PPTTPKTDL--SSSKADLKREGR--PIQEG-SSRQLNIDFGAVDIGELSSDVISNIGSFDV	289
chick_Sox9	PPTTPKTDL--SSSKADLKREGR--PIQEG-SSRQLNIDFGAVDIGELSSDVISNIGSFDV	289
mouse_Sox9	PPTTPKTDAQQPGKQDQLKREGR--PLAEG-GRQPPHIDFRDVDIGELSSDVISNIETFDV	293
human_Sox9	PPTTPKTDV--QAGKVDLKREGR--PLAEG-GRQPP-IDFRDVDIGELSSDVISNIETFDV	291
	PPTTPKTDV--QPGKADLKREGR--PLPEG-GRQPP-IDFRDVDIGELSSDVISNIETFDV	291
	***:***** . * ***** : : * : *** *****:*****:*****	291
me_Sox9a	NEFDQYLPNGHPGAAPGSTAPVSGNYSIS-GAPPLSPQAGGGPAWMAKHSQQ---	349
Ntil_Sox9a	NEFDQYLPNGHPGST--NAAPVSYGSYSISSSGAPVSPQS--GGAWMAKSQNQQG--	348
plf_Sox9a	NEFDQYLPNGHPGPV--GATPVSYGSYSIS-GGAPLSPQ---PAWMSKSQNQQG--	346
gup_Sox9a	NEFDQYLPNGHPGPV--GATPVSYGSYSIS-GGAPLSPQ---PAWMSKSQNQQG--	346
Am_Sox9a	NEFDQYLPNGHPGPV--GATPVSYGSYSIS-GGAPLSPQ---PAWMSKSQNQQG--	346
me_Sox9b	DEFDQYLPPhSHAGMTGTA--QTYSNNYVINSSAVQTA-NVGAHAWMPKQ-----	338
Ntil_Sox9b	DEFDQYLPPhSHAGVNGAA--QAGYSSGYPINSSVGQPA-SVGAHAWMSKQQQQQQQ--	344
Ghol_Sox9bX	DEFDQYLPPhSHAGVAGPA--QAGYTGSYGISTSSVSQGA-GVGAHAWMSKQQ-Q----	340
Ghol_Sox9bY	DEFDQYLPPhSHAGVAGPA--QAGYTGSYGISTSSVSQGA-SVGAHAWMSKQQ-Q----	340
gup_Sox9b	DEFDQYLPPhSHAGVAGAA--QAGYTGSYGISTASVGQGA-GVGAHAWMSKQQ----	341
plf_Sox9b	DEFDQYLPPhSHAGVAGAA--QAGYTGSYGISTSSVGQGA-GVGAHAWMSKQQ----	339
Am_Sox9b	DEFDQYLPPhSHAGVVGAA--QAGYTGSYGISTASVGQGA-GVGAHAWMSKQQ-Q----	340
chick_Sox9	NEFDQYLPNGHPGVPATHQVTTYSGYGISSASS--P-AGAGHAWMAKQQQPQPQPPQ	350
mouse_Sox9	NEFDQYLPNGHPGVPATHQV-TYTGSYGISSTAPT--P-ATAGHVWMSKQQAPPQPPQ	347
human_Sox9	NEFDQYLPNGHPGVPATHQV-TYTGSYGISSTAAT--P-ASAGHVWMSKQQAPPQPPQ	347
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me_Sox9a	-----Q---HSLTPLGTSGGSEA--ALRRTQIKTEQL	376
Ntil_Sox9a	-----QOOOOHTLTTLGS SGASDAAQTQHRTQIKTEQL	381
plf_Sox9a	-----QQQITLTLLGGGGGSDAAQAQHRTQIKTEQL	379
gup_Sox9a	-----QQQITLTLLGGGG--GGSDAAQAQHRTQIKTEQL	378
Am_Sox9a	-----QQQITLTLLGGGG--GGSDAAQAQHRTQIKTEQL	378
me_Sox9b	-----QHSLATLGGGGDQSQQGQQRRTQIKTEQL	367
Ntil_Sox9b	-----QQQLSLLTLGGGEQSQQGQQRRTQIKTEQL	376
Ghol_Sox9bX	-----QOHTLTNLGGAGEEQQQQQQRRTQIKTEQL	370
Ghol_Sox9bY	-----QOHTLTTLGAAAEQGQQQQQRRTQIKTEQL	370
gup_Sox9b	-----OOHTLTTLGGAAGEOGOOQQQRRTQIKTEQL	371
plf_Sox9b	-----QOHTLTTLGGAAGEQGQQQQQRRTQIKTEQL	369
Am_Sox9b	-----QOHTLTTLGGAAGEQGQQQQQRRTQIKTEQL	370
chick_Sox9	A-----QPPAQHTLPALESEQ---GPAQQRPHIKTEQL	380
mouse_Sox9	QPPQAPQAPQAPPQQQAP-PQQPO-APQQQQQAHLTLLSSEP---G-QSQRTHIKTEQL	400
human_Sox9	QPPQAPPAPQAPPQPAAPPQQPAAPPQOPQOAHTLTLSSEP---G-QSQRTHIKTEQL	402
	-----:*****	402
me_Sox9a	SPSHYSEQQQGSP-----QNA-PYSPFNLQHYSPSSSYPPISRAQ--QYDYPDPQGGG	426
Ntil_Sox9a	SPSHYSEQQQGSP-----QHVSPYSPFNLQHYSPSS-SYPAISRAQQQQYDYPDHQGGG	433
plf_Sox9a	SPSHYSEQQQSSP-----QHV-PYSPFNIQHYSPPS-TYPAISRPQQ-YSEYSEHQGGG	429
gup_Sox9a	SPSHYSEQQQGSP-----QHV-PYSPFSMQLHYSPPS-TYPAISRPQQ-YSEYSEHQGGG	428
Am_Sox9a	SPSHYSEQQQGSP-----QHV-PYSPFSMQLHYSPPS-TYPAISRPQQ-YSEYSEHQGGG	428
me_Sox9b	SPSHYSEQQQSS-----PQHVSYGSFNLQHYSTS--SYPSITRA--QYDYSDHQNSA	414
Ntil_Sox9b	SPSHYSEQQQGS-----PQHVTYGSFNLQHYSTN--SYPSITRS--QYDYSDHQSGA	423
Ghol_Sox9bX	SPSHYSDQQGS-----PQHITYGSFNLQHYSPS--SYPSITRA--QYDYSEHEQSSA	417
Ghol_Sox9bY	SPSHYSDQQGS-----PQHITYGSFNLQHYSPS--SYPSITRA--QYDYSEHEQSSA	417
gup_Sox9b	SPSHYSDQQGS-----PQHITYGSFNLQHYSPS--SYPSITRA--QCDYSEHEQSSA	418
plf_Sox9b	SPSHYSDQQGS-----PQHITYGSFNLQHYSPS--SYPSITRA--QYDYSEHEQSSA	416
Am_Sox9b	SPSHYSDQQGS-----PQHITYGSFNLQHYSPS--SYPSITRA--QYDYSEHEQSSA	417
chick_Sox9	SPSHYSDQQGS-----PQHITYGSFNLQHYSPS--SYPSITRA--QYDYSEHEQSSA	433
mouse_Sox9	SPSHYSEQQQQHSPOQQQQQQQQQYGSFNLQHYGS--SYPPITRS--QYDYTEHQNS-	446
human_Sox9	SPSHYSEQQQQHSPO-----QISYSPFNLPHYSP--SYPPITRS--QYDYADHQNS-	446
	SPSHYSEQQQQHSPO-----QIAYSFPFNLPHYSP--SYPPITRS--QYDYTDHQNS-	448
	*****:*** * * *:*** * * *:*** * * *:*** * * *	448

me_Sox9a	F-----YSPAGAGQGSGLYSTFSYMSSPSQRPMYTPPIADNAGVPSIPQGSP-QHWEQAP	479
Ntil_Sox9a	TATASSYYSHAGAGQSSGLYSTFSYMSSPTQRPMYTPPIADNTGVPSIPQNSP-QHWDPAP	492
plf_Sox9a	ASYYSHAGAG--AGQGSGLYSTFSYMSPNQRPMYTPPIADNAGVPSIPQGSPQQHWEQAP	487
gup_Sox9a	ASYYSHAGAGAGQGSGLYSTFSYMSPNQRPMYTPPIADNAGVPSIPQGSPQQHWEQAP	488
Am_Sox9a	ASYYSHAGAGAGSGQGSGLYSTFSYMSPNQRPMYTPPIADNAGVPSIPQGSPQQHWEQAP	488
me_Sox9b	NSYYSHA----AGQGSNMYSTFSYMS-PSQRPMYTPPIADSTGVPSVPQTHSPQHWEQQP	468
Ntil_Sox9b	NSYYSHA----GGQGSSLYSTFSYMS-PNQRPMPYTPPIADTTGVPSVPQTHSPQHWE-QP	476
Ghol_Sox9bx	NSYYSHA----AGQGSSLYSTFSYMS-PSQRPMYTPPIADSTGVPSVPQTHSPQHWEQQP	471
Ghol_Sox9bY	NSYYSHA----AGQGSSLYSTFSYMS-PSQRPMYTPPIADSTGVPSVPQTHSPQYWEQQP	471
gup_Sox9b	NSYYSHA----AGQGSSLYSTFSYMS-PSQRPMYTPPIADSTGVPSVPQTHSPQHWEQQP	472
plf_Sox9b	NSYYSHA----AGQGSSLYSTFSYMS-PSQRPMYTPPIADSTGVPSVPQTHSPQHWEQQP	470
Am_Sox9b	NSYYSHA----AGQGSSLYSTFSYMS-PSQRPMYTPPIADSTGVPSVPQTHSPQHWEQQP	471
chick_Sox9	GSYYSHA----AGQSGGLYSTFTYMN-PTQRPMPYTPPIADTSGVPSIPQTHSPQHWE-QP	486
mouse_Sox9	GSYYSHA----AGQGSGLYSTFTYMN-PAQRPMYTPPIADTSGVPSIPQTHSPQHWE-QP	499
human_Sox9	SSYYSHA----AGQGTGLYSTFTYMN-PAQRPMYTPPIADTSGVPSIPQTHSPQHWE-QP	501
	***. .:*****:*. * *****. :****:*** *:***: * *	
me_Sox9a	VYTQLTRP 487	
Ntil_Sox9a	VYTQLTRP 500	
plf_Sox9a	VYTQLTRP 495	
gup_Sox9a	VYTQLTRP 496	
Am_Sox9a	VYTQLTRP 496	
me_Sox9b	IYTQLSRP 476	
Ntil_Sox9b	IYTQLSRP 484	
Ghol_Sox9bx	IYTQLSRP 479	
Ghol_Sox9bY	IYTQLSRP 479	
gup_Sox9b	VYTQLTRP 480	
plf_Sox9b	IYTQLSRP 478	
Am_Sox9b	IYTQLSRP 479	
chick_Sox9	VYTQLTRP 494	
mouse_Sox9	VYTQLTRP 507	
human_Sox9	VYTQLTRP 509	
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**Figure S5** Multiple species alignments of Gipc1 and Sox9 protein sequences.

(A) Gipc1 and (B) Sox9 protein alignment. The yellow shading in (B) highlights Sox9a and the blue one Sox9b sequences. The sequences were aligned with Clustal Omega. Am, Amazon molly; chick, chicken (*Gallus gallus*); Ghol, *Gambusia holbrooki*; gup, guppy; me, medaka; mol, Atlantic molly (*Poecilia mexicana*); Ntil, Nile tilapia; plf, platyfish. GenBank accession numbers: XP\_007572354.1 (Am Gipc1); XP\_007560319.1 (Am Sox9a); XP\_007556425.2 (Am Sox9b); XP\_025001585.1 (chick Gipc1); BAA25296.1 (chick Sox9); XP\_008436174.1 (gu Sox9a); NP\_001284373.1 (gu Sox9b); NP\_005707.1 (human (*Homo sapiens*) Gipc1); CAA86598.1 (human Sox9); XP\_023813268.1 (me Gipc1); AAX62152.1 (me Sox9a); AAX62151.1 (me Sox9b); XP\_014866717.1 (mol Gipc1); NP\_061241.1 (mouse Gipc1); NP\_035578.3 (mouse Sox9); XP\_003442153.1 (Ntil Gipc1); XP\_005448042.1 (Ntil Sox9a); XP\_003450167.1 (Ntil Sox9b); XP\_005801543.1 (plf Gipc1); XP\_005807407.1 (plf Sox9a); XP\_005801494.1 (plf Sox9b). Note that the NCBI databank refers to Sox9b as Sox9 in all of the used sequences except for medaka.