

**Supplementary Figure S1| Description of axial skeletons at the caudal level in the *Bdellostoma* species. The putative vertebral elements were described (red arrowheads)<sup>18</sup>.**

Eptatretus\_burgeri\_Twist FEDLQNRILANVREERQRTQSLNEAFSSLRKKIIPITLPSDKLSKIQTLKLAARYIDFLYQVLQNDDEMDNKMS - - - SCSYVAHERLSYAFSVWRME  
 Petromyzon\_marinus\_TwistA EPESQNRQRFVANVREERQRTQSLNDAFASLRKIIPTLP-DKLSKIQTLKLAARYIDFLYQVLQSDAAEGRA PV - PSCSYAATERLSYAFSVWRME  
 Scyliorhinus\_canicula\_Twist LEDIQTRVAVANVREERQRTQSLNDAFATLRKIIPTLPSDKLSKIQILKLATRYIDFLYQVLQNDDEMDSKV - - - TSCNYLSHERLSYAFSVWRME  
 Homo\_sapiens\_Twist1 YEELQTRVAVANVREERQRTQSLNEAFAALRKKIIPITLPSDKLSKIQTLKLAARYIDFLYQVLQSDDELDSKMA - - - SCSYVAHERLSYAFSVWRME  
 Mus\_musculus\_Twist1 YEELQTRVAVANVREERQRTQSLNEAFAALRKKIIPITLPSDKLSKIQTLKLAARYIDFLYQVLQSDDELDSKMA - - - SCSYVAHERLSYAFSVWRME  
 Monodelphis\_domestica\_Twist YEELQTRVAVANVREERQRTQSLNEAFAALRKKIIPITLPSDKLSKIQTLKLAARYIDFLYQVLQSDDELDSKMA - - - SCSYVAHERLSYAFSVWRME  
 Ornithorhynchus\_anatinus\_Twist YEELQTRVAVANVREERQRTQSLNEAFAALRKKIIPITLPSDKLSKIQTLKLAARYIDFLYQVLQSDDELDSKMA - - - SCSYVAHERLSYAFSVWRME  
 Gallus\_gallus\_Twist1 YEELQTRVAVANVREERQRTQSLNEAFAALRKKIIPITLPSDKLSKIQTLKLAARYIDFLYQVLQSDDELDSKMA - - - SCSYVAHERLSYAFSVWRME  
 Anolis\_carolinensis\_Twist1 YEELQTRVAVANVREERQRTQSLNEAFAALRKKIIPITLPSDKLSKIQTLKLAARYIDFLYQVLQSDDELDSKMA - - - SCSYVAHERLSYAFSVWRME  
 Xenopus\_tropicalis\_Twist1 FEELQSQRVAVANVREERQRTQSLNEAFAALRKKIIPITLPSDKLSKIQTLKLAARYIDFLCQVLQSDDELDSKMA - - - SCSYVAHERLSYAFSVWRME  
 Gasterosteus\_aculeatus\_Twist1b FDDLQTRVAVANVREERQRTQSLNEAFTSLRKKIIPITLPSDKLSKIQTLKLAARYIDFLCQVLESDDELARGT - - - SCSYVAHERLSYAFSVWRMG  
 Oryzias\_latipes\_Twist1b FDDLQTRVAVANVREERQRTQSLNEAFTSLRKKIIPITLPSDKLSKIQTLKLAARYIDFLYQVLQSDDEMDAKLA - - - SCNYLAHERLSYAFSVWRME  
 Danio\_erio\_Twist1a LEDLQTRVAVANVREERQRTQSLNEAFASLRKIIPTLPSDKLSKIQTLKLAARYIDFLCQVLQSDDELDSKMS - - - SCSYVAHERLSYAFSVWRME  
 Danio\_erio\_Twist1b FEELQTRVAVANVREERQRTQSLNEAFAALRKKIIPITLPSDKLSKIQTLKLAARYIDFLCQVLQSDDELDSKMA - - - SCSYVAHERLSYAFSVWRME  
 Mus\_musculus\_Twist2 FEELQSQRILANVREERQRTQSLNEAFAALRKKIIPITLPSDKLSKIQTLKLAARYIDFLYQVLQSDDEMDNKMT - - - SCSYVAHERLSYAFSVWRME  
 Xenopus\_tropicalis\_Twist2 FEDVHTQRILANVREERQRTQSLNDAFAELRKKIIPITLPSDKLSKIQTLKLAARYIDFLYQVLQSDDELHDKIA - - - SCNYLAHERLSYAFSVWRME  
 Gallus\_gallus\_Twist2 YEELQSQRILANVREERQRTQSLNEAFAALRKKIIPITLPSDKLSKIQTLKLAARYIDFLYQVLQSDDEMDSKMT - - - SCSYVAHERLSYAFSVWRME  
 Gasterosteus\_aculeatus\_Twist2 YEELQNRQCLANVREERQRTQSLNEAFSSLRKKIIPITLPSDKLSKIQTLKLAARYIDFLCQVLQSDDEMDNKMS - - - SCSYVAHERLSYAFSVWRME  
 Oryzias\_latipes\_Twist2 YEELQNRQVLANVREERQRTQSLNEAFASLRKIIPTLPSDKLSKIQTLKLAARYIDFLCQVLQSDDEMDSKMS - - - SCSYVAHERLSYAFSVWRME  
 Danio\_erio\_Twist2 FEDLHTQRVAVANVREERQRTQSLNDAFASLRKIIPTLPSDKLSKIQILKLAARYIDFLYQVLQSDDEMDAKLA - - - SCNYLAHERLSYAFSVWRME  
 Gallus\_gallus\_Twist3 FEDVHTQRVAVANVREERQRTQSLNDAFAELRKKIIPITLPSDKLSKIQTLKLAARYIDFLYQVLQSDDELHDKIT - - - SCNYLAHERLSYAFSVWRME  
 Gasterosteus\_aculeatus\_Twist3a FEDLHSQRVAVANVREERQRTQSLNDAFASLRKIIPTLPSDKLSKIQILKLAARYIDFLYQVLQSDDEMDAKLA - - - SCNYLAHERLSYAFSVWRME  
 Gasterosteus\_aculeatus\_Twist3b LEDPHGQRVAVANVREERQRTQSLNDAFASLRKIIPTLPSDKLSKIQTLKLAARYIDFLYQVLQNDDEMDTKLA - - - GCNYLAHERLSYAFSVWRME  
 Oryzias\_latipes\_Twist3a FEELHSQRVAVANVREERQRTQSLNDAFASLRKIIPTLPSDKLSKIQILKLAARYIDFLYQVLQSDDEMDAKLA - - - SCNYLAHERLSYAFSVWRME  
 Oryzias\_latipes\_Twist3b LEDPHAQRVAVANVREERQRTQSLNEAFASLRKIIPTLPSDKLSKIQTLKLAARYIDFLYQVLQSDQMDSKLA - - - GCNYLAHERLSYAFSVWRME  
 Saccoglossus\_kowalevskii\_Twist YEDLQNRVAVANVREERQRTQSLNEAFASLRKIIIPITLPSDKLSKIQTLKLAARYIDFLYQVLRSDVEVDTRLNMP TSCSYVAHERLSYAFSVWRME

**Supplementary Figure S2| Multiple alignment of conserved region of *Twist* genes.** Predicted amino acid sequences of *E. burgeri Twist* gene and various *Twist* proteins of one lamprey, eleven gnathostomes and one hemichordate species. Accession numbers are shown as follows; *Anolis\_carolinensis\_Twist1*, DAA06059; *Danio\_erio\_Twist1b*, NP\_001017820; *Danio\_erio\_Twist1a*, NP\_571059; *Danio\_erio\_Twist2*, NP\_001005956; *Eptatretus\_burgeri\_Twist*, AB594748; *Gallus\_gallus\_Twist1*, NP\_990070; *Gallus\_gallus\_Twist2*, NP\_990010; *Gallus\_gallus\_Twist3*, NP\_001096684; *Gasterosteus\_aculeatus\_Twist1b*, DAA06074; *Gasterosteus\_aculeatus\_Twist2*, DAA06075; *Gasterosteus\_aculeatus\_Twist3a*, DAA06076; *Gasterosteus\_aculeatus\_Twist3b*, DAA06077; *Homo\_sapiens\_Twist1*, NP\_000465; *Monodelphis\_domestica\_Twist*, XP\_001372508; *Mus\_musculus\_Twist1*, NP\_035788; *Mus\_musculus\_Twist2*, NP\_031881; *Ornithorhynchus\_anatinus\_Twist*, XP\_001514308; *Oryzias\_latipes\_Twist2*, DAA06067; *Oryzias\_latipes\_Twist1b*, DAA06066; *Oryzias\_latipes\_Twist3a*, DAA06068; *Oryzias\_latipes\_Twist3b*, DAA06069; *Petromyzon\_marinus\_TwistA*, ABY76996; *Scyliorhinus\_canicula\_Twist*, ABY76997; *Saccoglossus\_kowalevskii\_Twist*, NP\_001158484; *Xenopus\_tropicalis\_Twist1*, NP\_989415; *Xenopus\_tropicalis\_Twist2*, NP\_001096679.

Epatretus\_burgeri\_Pax1/9 GVFVNGRPLPNAIRIRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPTVVNHIREYKQDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGFLPQ  
 Lethenteron\_japonicum\_Pax1/9 GVFVNGRPLPNAIRLRIVEMAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPNVNHIRTYKQDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGNLSQ  
 Petromyzon\_marinus\_Pax1/9 GVFVNGRPLPNAIRLRIVEMARLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPNVNHIRTYKQDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGNLSQ  
 Petromyzon\_marinus\_Pax1/9b GVFVNGRPLPNPILRLRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPNVVKHIRDYKQDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGLSQ  
 Scyliorhinus\_canicula\_Pax1 GVFVNGRPLPNAIRLRIVESAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPTVVKYIREYKQDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGNLSH  
 Homo\_sapiens\_Pax1 GVFVNGRPLPNAIRLRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPNVVKHIRDYKQDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGSLAQ  
 Mus\_musculus\_Pax1 GVFVNGRPLPNAIRLRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPNVVKHIRDYKQDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGSLAQ  
 Canis\_familiaris\_Pax1 GVFVNGRPLPNAIRLRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPNVVKHIRDYKQDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGSLAQ  
 Bos\_taurus\_Pax1 GVFVNGRPLPNAIRLRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPNVVKHIRDYKQDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGSLAQ  
 Monodelphis\_domestica\_Pax1 GVFVNGRPLPNAIRLRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPNVVKHIRDYKQDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGNLSQ  
 Ornithorhynchus\_anatinus\_Pax1 GVFVNGRPLPNAIRLRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPTVVKHIRDYKQDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGNLSQ  
 Taeniopygia\_guttata\_Pax1 GVFVNGRPLPNAIRLRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPAVVKHIRDYKQDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGPLAP  
 Xenopus\_laevis\_Pax1 GVFVNGRPLPNAIRLRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPTVVKHIRDYKQDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGNLSQ  
 Oryzias\_latipes\_Pax1 GVFVNGRPLPNAIRLRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPNVVKHIRDYKQDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGNLSQ  
 Danio\_erio\_Pax1 GVFVNGRPLPNAIRLRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPNVVKHIRDYKQSDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGNLSQ  
 Homo\_sapiens\_Pax9 GVFVNGRPLPNAIRLRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPTVVKHIRDYKQDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGNLAQ  
 Mus\_musculus\_Pax9 GVFVNGRPLPNAIRLRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPTVVKHIRDYKQDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGNLAQ  
 Suncus\_murinus\_Pax9 GVFVNGRPLPNAIRLRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPTVVKHIRDYKQDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGNLTQ  
 Canis\_familiaris\_Pax9 GVFVNGRPLPNAIRLRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPTVVKHIRDYKQDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGNLAQ  
 Bos\_taurus\_Pax9 GVFVNGRPLPNAIRLRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPTVVKHIRDYKQDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGNLAQ  
 Monodelphis\_domestica\_Pax9 GVFVNGRPLPNAIRLRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPTVVKHIRDYKQDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGNLSQ  
 Ornithorhynchus\_anatinus\_Pax9 GVFVNGRPLPNAIRLRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPTVVKHIRDYKQDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGNLAQ  
 Gallus\_gallus\_Pax9 GVFVNGRPLPKAIRLRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPTVVKHIRDYKQDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGNLSQ  
 Taeniopygia\_guttata\_Pax9 GVFVNGRPLPNAIRLRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPTVVKHIRDYKQDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGNLSQ  
 Xenopus\_laevis\_Pax9 GVFVNGRPLPNAIRLRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPTVVKHIRDYKQDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGNLAQ  
 Oryctolagus\_cuniculus\_Pax9 GVFVNGRPLPNAIRLRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPNVVKHIRDYKQDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGSLAQ  
 Takifugu\_rubripes\_Pax9 GVFVNGRPLPNAIRLRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPTVVKHIRDYKQDPGIFAWERDRLLDAGGCDKFNLPVSSISRILRNKIGNLSQ  
 Oryzias\_latipes\_Pax9 GVFVNGRPLPNAIRLRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPTVVKHIRDYKQDPGIFAWERDRLLDAGVCDKFNLPVSSISRILRNKIGNLTP  
 Astyanax\_mexicanus\_Pax9 GVFVNGRPLPNAIRLRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPTVVKHIRDYKQDPGIFAWERDRLLDAGVCDKFNLPVSSISRILRNKIGNVQ  
 Danio\_erio\_Pax9b GVFVNGRPLPNAIRLRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPNVVKHIRDYKQDPGIFAWERDRLLDAGVCDKFNLPVSSISRILRNKIGNLSQ  
 Ciona\_intestinalis\_Pax1/9 GVFVNGRPLPNAIRLRIVELAQLGIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPNVVKHIRDYKQDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGNVLH  
 Branchiostoma\_floridae\_Pax1 GVFVNGRPLPNAIRLRIVELAHLVIRPCDISRQLRVSHGCVSKILARYNETGSLPGAIGGSKPRVTTPEVVNAIKDYKVRDPGIFAWERDRLLDAGVCDKYNVPSVSSISRILRNKIGNTTQ

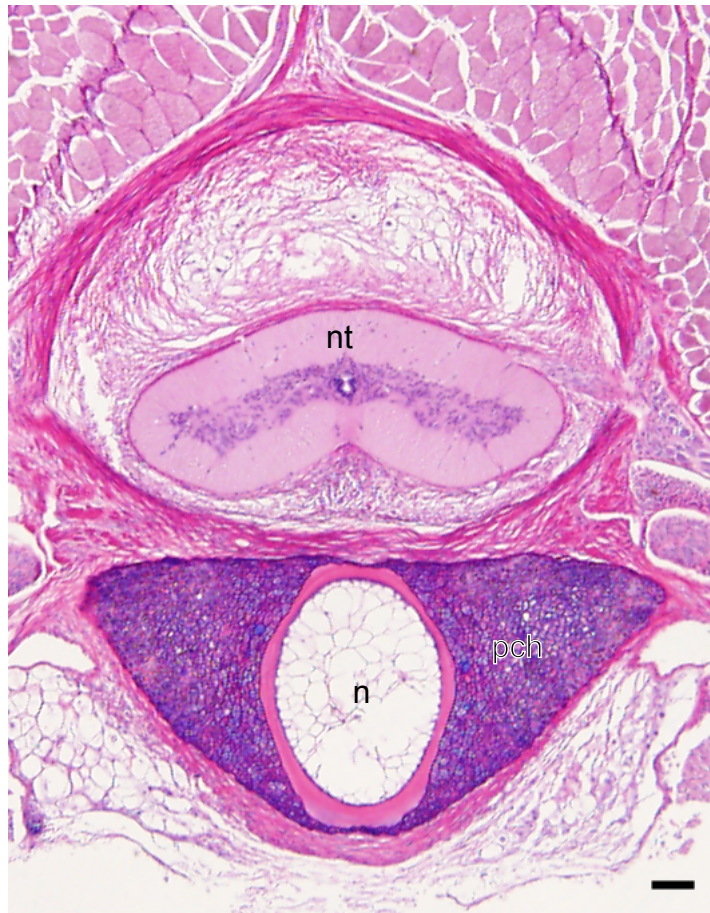
**Supplementary Figure S3| Multiple alignment of conserved region of Pax1/9 genes.** Predicted amino acid sequences of *E. burgeri Pax1/9* gene and various Pax1/9 proteins of three two lampreys, sixteen gnathostomes, and two non-vertebrate chordate species. Accession numbers are shown as follows; *Astyanax\_mexicanus\_Pax9*, ABG89862; *Bos taurus\_Pax9*, XP\_617873; *Bos taurus\_Pax1*, XP\_002692205; *Bos taurus\_Pax9*, NP\_001179298; *Branchiostoma floridae\_Pax1*, AAA81364; *Canis familiaris\_Pax1*, XP\_542866; *Canis familiaris\_Pax9*, XP\_547776; *Ciona\_intestinalis\_Pax1/9*, NP\_001027594; *Danio\_erio\_Pax1*, NP\_001074061; *Danio\_erio\_Pax9*, NP\_571373; *Danio\_erio\_Pax9b*, AAC60035; *Epatretus burgeri\_Pax1/9* AB594746; *Gallus gallus\_Pax9*, NP\_990243; *Homo sapiens\_Pax1*, AAH69134; *Homo sapiens\_Pax9*, NP\_006185; *Lethenteron japonicum\_Pax1/9*, BAB12396; *Monodelphis domestica\_Pax1*, XP\_001382098; *Monodelphis domestica\_Pax1/9*, XP\_001366875; *Mus musculus\_Pax1*, AAA39888; *Mus musculus\_Pax9*, NP\_035171; *Ornithorhynchus anatinus\_Pax1*, XP\_001506813; *Ornithorhynchus anatinus\_Pax9*, XP\_001511262; *Oryctolagus cuniculus\_Pax9*, XP\_002711030; *Oryctolagus cuniculus\_Pax9*, XP\_002718002; *Oryzias latipes\_Pax1*, NP\_001165520; *Oryzias latipes\_Pax9*, BAH86770; *Petromyzon marinus\_Pax1/9*, ABY76998; *Petromyzon marinus\_Pax1/9b*, ABY71250; *Scyliorhinus canicula\_Pax1*, ABY76999; *Suncus murinus\_Pax9*, BAF81894; *Taeniopygia guttata\_Pax1*, XP\_002197278; *Taeniopygia guttata\_Pax9*, XP\_002199853; *Takifugu rubripes\_Pax9*, AAG44703; *Xenopus laevis\_Pax1*, NP\_001090451; *Xenopus laevis\_Pax9*, NP\_001167485.



**Supplementary Figure S4| Expression pattern of *Twist* gene in the middle pharyngeal stage of *E. burgeri*.** *Twist* is strongly expressed in the mesenchymal cells located at the ventral side of the notochord. som, somite; nt neural tube; n, notochord. Bar = 100  $\mu$ m.

Eptatretus_burgeri_MyoD	HVRA PHGQHGP GPC LLWACKACKRKTSS TDRRKAA TMRERRRLRKVNEAFETLKRCTSSANPSQRLPKVEILRNAIRYIEGLQRLLR
Homo_sapiens_MyoD	HVRA PS GHHQA GRCLLWACKACKRKT TNADRRKAA TMRERRRLSKVNEAFETLKRCTSSNPNQRLPKVEILRNAIRYIEGLQALLR
Mus_musculus_MyoD	HVRA PS GHHQA GRCLLWACKACKRKT TNADRRKAA TMRERRRLSKVNEAFETLKRCTSSNPNQRLPKVEILRNAIRYIEGLQALLR
Bos_taurus_MyoD	HVRA PS GHHQA GRCLLWACKACKRKT TNADRRKAA TMRERRRLSKVNEAFETLKRCTSSNPNQRLPKVEILRNAIRYIEGLQALLR
Canis_familiaris_MyoD	HVRA PS GHHQA GRCLLWACKACKRKT TNADRRKAA TMRERRRLSKVNEAFETLKRCTSSNPNQRLPKVEILRNAIRYIEGLQALLR
Gallus_gallus_MyoD	HVRA PS GHHQA GRCLLWACKACKRKT TNADRRKAA TMRERRRLSKVNEAFETLKRCTSTNPNQRLPKVEILRNAIRYIESLQALLR
Xenopus_tropicalis_MyoD	HVRA PS GHHQA GRCLLWACKACKRKT TNADRRKAA TMRERRRLSKVNEAFETLKRCTSTNPNQRLPKVEILRNAIRYIESLQSLLR
Takifugu_rubripes_MyoD	H I R A P S G H H H A G R C L L W A C K A C K R K T T N A D R R K A A T L R E R R R L S K V N E A F E T L K R C T N T N P N Q R L P K V E I L R N A I S Y I E S L Q A L L R
Paralichthys_olivaceus_MyoD	HVRA PS GHHQA GRCLLWACKACKWK TTNADRRKAA TMRERRRLSKVNDAFETLKRCTSANPNQRLPKVEILRNAISYIESLQALLR
Oreochromis_niloticus_MyoD	HVRA PS GHHQA GRCLLWACKACKRKT TNADRRKAA TLRERRRLSKVNDAFETLKRCTTANPNQRLPKVEILRNAISYIESLQALLR
Micropterus_salmoides_MyoD	HVRA PS GHHQA GRCLLWACKACKRKT TNADRRKAA TLRERRRLSKVNDAFETLKRCTSANPNQRLPKVEILRNAISYIESLQALLR
Sparus_aurata_MyoD	HVRA PS GHHQA GRCLLWACKACKRKT TNADRRKAA TLRERRRLSKVNDAFETLKRCTSANPNQRLPKVEILRNAISYIESPQALLR
Ictalurus_punctatus_MyoD	H I R A P S G H H Q A G R C L L W A C K A C K R K T T N A D R R K A A T M R E R R R L S K V N D A F E T L K R C T S T N P N Q R L P K V E I L R N A I S Y I E S L Q A L L R
Ameiurus_catus_MyoD	H I R A P S G H H Q A G R C L L W A C K A C K R K T T N A D R R K A A T M R E R R R L S K V N D A F E T L K R C T S T N P N Q R L P K V E I L R N A I S Y I E S L Q A L L R
Oncorhynchus_mykiss_MyoD	H I R A P S G H H Q A G R C L L W A C K A C K R K T T N A D R R K A A T M R E R R R L S K V N D A F E T L K R C T S T N P N Q R L P K V D I L R N A I S Y I E S L Q G L L R
Danio_rerio_MyoD	HVRA PS GHHQA GRCLLWACKACKRKT TNADRRKAA TMRERRRLSKVNDAFETLKRCTSTNPNQRLPKVEILRNAISYIESLQALLR
Sternopygus_macrurus_MyoD	H I R A P S G H H Q A G R C L L W A C K A C K R K T T N A D R R K A A T M R E R R R L S K V N D A F E T L K R C T S T N P N Q R L P K V E I L R N A I S Y I E S L Q A L L R

**Supplementary Figure S5| Multiple alignment of conserved region of *MyoD* genes.** Predicted amino acid sequences of *E. burgeri MyoD* gene and various *MyoD* proteins of sixteen gnathostomes species. Accession numbers are shown as follows; *Ameiurus\_catus\_MyoD*, AAS67039; *Canis\_familiaris\_MyoD*, XP\_854756; *Danio\_rerio\_MyoD*, NP\_571337; *Eptatretus\_burgeri\_MyoD*, AB594747; *Gallus\_gallus\_MyoD*, P16075; *Homo\_sapiens\_MyoD*, NP\_002469; *Ictalurus\_punctatus\_MyoD*, AAS48083; *Micropterus\_salmoides\_MyoD*, ABY73734; *Mus\_musculus\_MyoD*, EDL22927; *Oncorhynchus\_mykiss\_MyoD1*, NP\_001118192; *Oreochromis\_niloticus\_MyoD*, ADA84033; *Paralichthys\_olivaceus\_MyoD*, ABA70719; *Sparus\_aurata\_MyoD*, AAL85337; *Sternopygus\_macrurus\_MyoD*, AAQ97204; *Taeniopygia\_guttata\_MyoD*, XP\_002198511; *Takifugu\_rubripes\_MyoD*, AAR39413; *Xenopus\_laevis\_MyoD*, AAA49900.



**Supplementary Figure S6| Transverse section at the level of the parachordal cartilage in the adult of *E. burgeri*.** The parachordal cartilage is stained by alcian blue. nt neural tube; n, notochord; pch, parachordal cartilages. Bar=100µm.

**Supplementary Table S1. Record of the forty-two obtained embryos from 2008 to 2009**

Developmental Stages	Designation	Date of after egg depositeon (dpd)
Neurula stage	Eb2009_01	108
	Eb2009_03	108
	Eb2009_06	109
	Eb2009_07	114
	Eb2009_27	135
	Eb2009_26	135
	Eb2009_30	139
	Eb2009_28	145
Early pharyngeal stage	Eb2009_05	109
	Eb2009_08	110
	Eb2009_09	114
	Eb2009_10	125
	Eb2009_12	124
	Eb2009_13	124
	Eb2009_14	121
	Eb2009_15	121
	Eb2009_23	130
	Eb2009_34	156
	Middle pharyngeal stage	Eb2009_02
Eb2009_04		100
Eb2009_11		125
Eb2009_16		126
Eb2009_17		126
Eb2009_18		127
Eb2009_19		127
Eb2009_20		127
Eb2009_21		130
Eb2009_22		130
Eb2009_24		138
Eb2009_25		135
Eb2009_29		142
Eb2009_31		145
Eb2009_32		145
Eb2008_E02		130
Eb2008_E03	130	
Late pharyngeal stage	Eb2008_B04	150
	Eb2009_33	153
	Eb2009_35	154
	Eb2009_36	172
	Eb2009_37	195
	Eb2009_38	195
	Eb2009_39	223
	Eb2009_40	242

**Supplementary Table S2. Primers for Degenerated RT-PCR**

Gene	Forward		Reverse		Description
	Primer name	Sequence	Primer name	Sequence	
<i>Pax1/9</i>	Pax19-GEVNQLG-18F	GGTGAAGTNAAYCARYTNGG	Pax19-GIFAWEI-110R	ATCTCCCANGCRAADATNCC	1st PCR
	Pax19-GGVFVNG-20F	GGAGGTGTNTTYGTNAAYGG	Pax19-DPGIFAW-107R	CCATGCGAADATNCCNGGRTC	Nested PCR
<i>Twist</i>	Tw-SLANVRE-69F	AGCCTCGCNAAYGTNMGNGA	Tw-WRMEGAW-148R	CCATGCACCYTCCATNCKCCA	1st PCR
	Tw-QSLNEAF-80F	CAGAGGCTNAAYGARGCNTT	Tw-YAFSVWR-143R	GCGCCANACNSWRAANGCRTA	Nested PCR
<i>MyoD</i>	MyoD-95F	ATGTGGGCNTGYAARGCNTG	MyoD-152R	CTCGATR TANCKDATNGCRT	1st PCR
	MyoD-107F	GTAAGACNACNACNATGGAY	MyoD-151R	ATGTANCKDATNGCRTT NCK	Nested PCR