

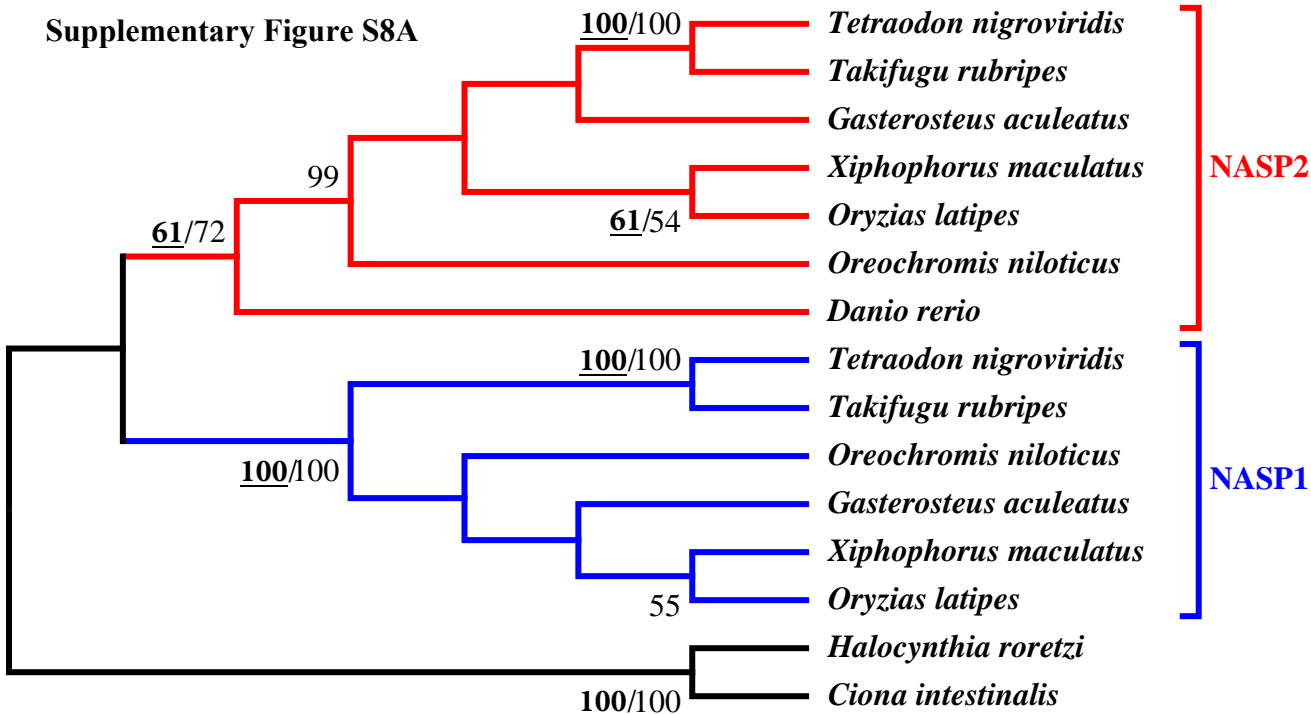
**Figure S8A:** Phylogenetic relationship among different fish specific NASP paralogs. Tree topology corresponds to the ML method under the JTT+G model as predicted by MEGA. Branch lengths do not reflect genetic distance. Alternatively, the same topology was also recovered by the neighbour joining method using p-distances. Confidence values for ML and NJ trees are based on 1000 bootstrap replicates and are indicated ( $\geq 50\%$ ) in light-face and boldface (underlined), respectively. The tree is rooted with *Halocynthia roretzi* and *Ciona intestinalis*.

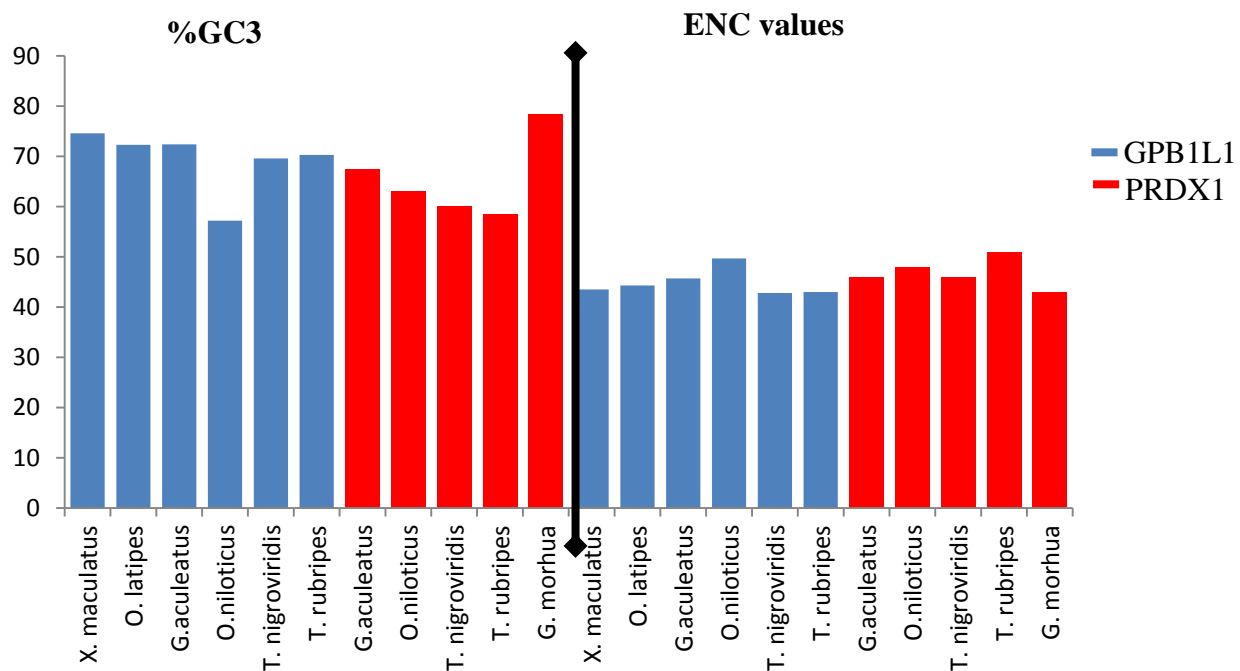
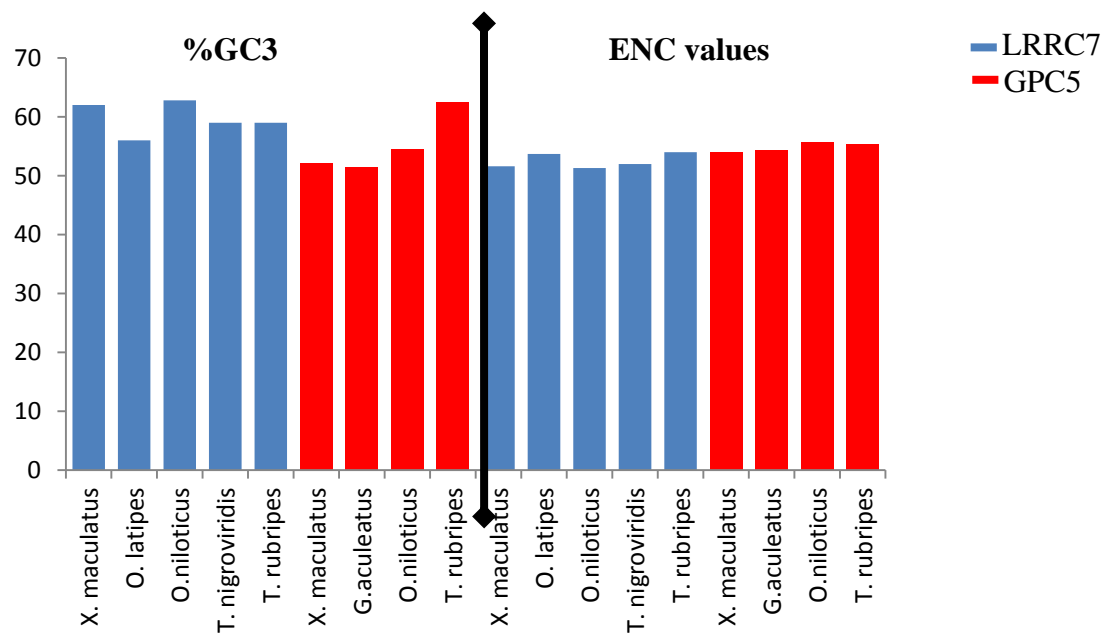
**Figure S8, B-C:** %GC3 content and ENC values are shown in chart diagrams for the fish-specific NASP1 and NASP2 neighbouring genes, respectively. Black line separates GC3 and ENC whereas blue and red colors represent different neighbouring genes as indicated in the chart legends.

**Figure S8, D-G:** Predicted fish specific NASP1/2 TPRs 1-4 sequence alignments are shown in figures B to E, respectively. Reconstructed ancestral sequence using the ML method is shown in black background whereas different amino acid substitutions in each TPR motif for two paralogous proteins are shown in blue.

**Figure S8, H-K:** Predicted NASP1/2 TPRs 1-4 sequence alignments are shown in figures H to K, respectively, for various lineages.

Supplementary Figure S8A



**B****C**

## D

<i>X.maculatus/1-34</i>	A	K	K	L	I	G	T	G	K	R	H	L	V	M	G	D	V	V	S	A	V	N	V	F	Q	E	A	C	S	M	L	A	A	K
<i>O.latipes/1-34</i>	A	K	K	L	I	G	T	G	K	R	H	L	V	M	G	D	V	V	S	A	V	N	V	F	Q	D	A	C	S	I	L	A	S	K
<i>G.aculeatus/1-34</i>	A	N	K	L	I	G	T	G	N	R	H	L	V	M	G	D	V	V	S	A	V	R	V	F	Q	D	A	C	S	M	L	A	A	R
<i>O.niloticus/1-34</i>	A	K	K	L	I	G	T	G	K	R	H	L	V	M	G	D	V	V	S	A	V	S	V	F	Q	E	A	C	G	M	L	A	E	K
<i>T.nigroviridis/1-34</i>	A	K	K	L	V	G	A	G	N	K	Y	L	V	L	G	D	V	V	S	A	V	G	V	F	Q	D	A	C	S	M	L	A	A	K
<i>T.rubripes/1-34</i>	A	K	K	L	V	G	A	G	N	K	Y	L	V	L	G	D	V	V	S	A	V	A	V	F	Q	D	A	C	S	M	L	A	A	K
<i>D.rerio/1-34</i>	A	K	K	L	I	G	T	G	S	R	H	L	V	M	G	D	V	V	S	A	V	S	V	F	Q	E	A	C	A	M	L	A	E	K
Ancestral/1-34	A	K	K	L	? G?	G	K	K	H	L	V	M	G	D	V?	S	A	V	N	A	F	Q	E	A	C	S?	L	A	E	K				
<i>X.maculatus/1-34</i>	A	N	K	L	V	G	T	G	K	K	F	L	V	T	G	K	V	V	E	A	V	S	A	L	Q	E	A	C	G	M	L	A	K	K
<i>O.latipes/1-34</i>	A	N	K	L	I	G	A	G	K	K	F	L	V	M	G	K	V	V	E	A	V	S	A	L	Q	E	A	C	G	M	L	A	K	Q
<i>G.aculeatus/1-34</i>	A	N	K	L	I	G	S	G	K	K	H	L	V	M	G	K	V	V	E	A	V	S	A	L	Q	E	A	C	G	M	L	A	Q	K
<i>O.niloticus/1-34</i>	A	N	K	L	I	G	T	G	K	K	H	L	V	M	G	K	V	V	E	A	V	S	T	L	Q	E	A	C	G	M	L	A	K	K
<i>T.nigroviridis/1-34</i>	A	N	K	L	V	G	A	G	K	K	H	L	V	M	G	K	V	V	E	A	V	N	S	L	Q	E	A	C	G	I	L	A	K	T
<i>T.rubripes/1-34</i>	A	N	K	L	I	G	A	G	K	K	H	L	V	M	G	K	V	V	E	A	V	N	S	L	Q	E	A	C	G	M	L	A	K	T

## E

<i>X.maculatus/1-34</i>	G	E	A	F	F	L	C	G	K	S	L	L	E	L	A	R	---	L	Q	L	A	W	E	M	L	E	V	A	K	V	I	F	K	R	K
<i>O.latipes/1-34</i>	G	E	A	V	F	L	Y	G	K	S	L	L	E	L	A	R	---	L	Q	L	A	W	E	M	L	E	V	A	K	L	I	Y	K	R	K
<i>G.aculeatus/1-34</i>	A	E	A	F	F	L	C	G	K	S	L	L	E	L	A	R	---	L	Q	L	A	W	E	M	L	E	V	A	K	V	I	Y	K	R	K
<i>O.niloticus/1-34</i>	G	E	A	F	F	F	C	G	K	S	L	L	E	L	A	R	---	L	Q	L	A	W	E	M	L	E	V	A	K	V	I	Y	K	R	K
<i>T.nigroviridis/1-34</i>	G	E	A	L	F	L	C	G	K	S	L	L	E	L	A	R	---	L	Q	L	A	W	E	M	L	E	V	A	K	V	I	Y	K	R	K
<i>T.rubripes/1-34</i>	G	E	A	L	F	L	C	G	K	S	L	L	E	L	A	R	---	L	Q	L	A	W	E	M	L	E	V	A	K	V	I	Y	K	R	K
<i>D.rerio/1-34</i>	G	E	V	F	F	F	C	G	K	A	L	L	E	L	A	R	---	L	R	L	A	W	E	M	L	E	V	A	K	V	I	Y	K	R	K
Ancestral/1-34	G	E	A	F	F	F	Y	G	K	S	L	L	E	L	A	R	---	L	Q	L	A	W	E	M	L	? L	A	K?	I?	L	R?				
<i>X.maculatus/1-34</i>	G	E	A	F	Y	W	C	G	K	A	L	L	D	L	A	R	---	L	Q	L	A	W	E	M	L	E	V	A	K	V	I	Y	R	R	K
<i>O.latipes/1-34</i>	G	E	A	F	L	W	C	G	K	A	L	L	D	L	A	R	---	L	Q	L	A	W	E	M	L	E	V	A	K	S	I	Y	K	R	K
<i>G.aculeatus/1-34</i>	G	E	A	F	F	W	C	G	K	A	L	L	D	L	A	R	---	L	Q	L	A	W	E	M	L	E	V	A	K	V	I	Y	K	R	K
<i>O.niloticus/1-34</i>	G	E	A	F	F	W	C	G	K	A	L	L	D	L	A	R	---	L	Q	L	A	W	E	M	L	E	V	A	K	V	I	Y	K	R	K
<i>T.nigroviridis/1-34</i>	G	E	A	F	F	W	C	G	K	A	L	L	D	L	A	R	---	L	Q	L	A	W	E	M	L	E	V	A	K	V	I	Y	K	R	K
<i>T.rubripes/1-34</i>	G	E	A	F	F	W	C	G	K	A	L	L	D	L	A	R	---	L	Q	L	A	W	E	M	L	E	V	A	K	V	I	Y	K	R	K

**F**

*X.maculatus/1-34* AQA YLKLGEVS AETGNYP QALEDFQECLI I QL KL  
*O.latipes/1-34* AQA H LKLGEVGAETGNYP QALEDFQECLAI QL QH  
*G.aculeatus/1-34* AQA YLKLGEVS AESGNYS QALEDFQECLAL QL KH  
*O.niloticus/1-34* AQA YLKLGEVS AESGNYP QALDDFQECLAL QL KH  
*T.nigroviridis/1-34* AQI YLKLGEVS AESGNYP QALEDFQECLCL QL KY  
*T.rubripes/1-34* AQI YLKLGEVS AESGNYP QALEDFQECLCL QL KH  
*D.rerio/1-34* AQI H LKLAEVGVESGNYS QALEDSNECLTL QL KH  
**Ancestral/1-34** AQA H LKLGEVS VESGNYP QA? EDFQ? CL? I QK? H  
*X.maculatus/1-34* AQT H LKLGEVS AESGNYP QALEDFQECLKVQL KH  
*O.latipes/1-34* AQA H LKLGEVS AESGNYP QALEDFQECLKL QVKH  
*G.aculeatus/1-34* AQA H S LKLGEVS S E SGNYP QALEDFQECLKL QVKH  
*O.niloticus/1-34* AQA H LKLGEVS AESGNYP QALEDFQECLKL QVKH  
*T.nigroviridis/1-33* AQA H LKLGEVS VESGNYP QALDDFQECLKL QAK-  
*T.rubripes/1-34* AQA H LKLGEVS VESGNYP QALDDFE ECLKL QL KH

**NASP-2****G**

*X.maculatus/1-34* AETHY HVATT L V F M D Q Y D Q A I K H Y N S S V K V I E T R  
*O.latipes/1-34* AETHY HVATT L C Y M D E Y R Q A I Q H Y N S S I E V I E N R  
*G.aculeatus/1-34* AETHY HVATT L C Y M D Q Y S Q A I Q H Y N S S I K V I E T R  
*O.niloticus/1-34* AETHY HVATT L C Y M D Q Y S Q A I Q H Y N S S I K V I E T R  
*T.nigroviridis/1-34* AETHY HVATT L C Y M D K Y S Q A I Q H Y N S S I E V I E K R  
*T.rubripes/1-34* AETHY HVATT L C Y M D K Y S Q A I Q H Y N S S I E V I E K R  
*D.rerio/1-34* TETHY Q L G T T Y S Y T T Q Y N Q A I E H F S N S I K V I E S R  
**Ancestral/1-34** AETHY Q L G L A Y S Y N ? Q Y D ? A I ? H F N ? S I ? V I E ? R  
*X.maculatus/1-34* AETHY Q L G V T H S L N L Q Y V P A I Q A L N N S I S V I K N R  
*O.latipes/1-34* AETHY Q L G L T Y S L N L Q Y S P A I E A L N N S I S V I K S R  
*G.aculeatus/1-34* AETHY Q L G L T Y S L D V Q Y G R A I E E L K S S I S V I K S R  
*O.niloticus/1-34* AETHY Q L G V T Y S L N T Q Y S E A I E S L K S S I S I I K N R  
*T.nigroviridis/1-34* AETHY Q L G L T H G L N L Q H G A A V A E L S R S I S V I K S R  
*T.rubripes/1-34* AETHY Q L G L T Y G L N L Q Y N Q A I A E L N R S I S V I K S R

**NASP-2**

