

Supplementary Table S1. Details of primers used in this study.

| Locus | Primer sequence (5' to 3') | Repeat | PCR product size (bp) | Chromosome | Gene | Genomic region | Gene function | Reference |
|-------------|---|---------------------|-----------------------|------------|---------------------------|-------------------------|--|---|
| BettaMS1.1 | F: CGTTCATTCCACTGGATCA R: AAGAGCTGGGGAAGTCCACT | (GT) _n | 378 | 1 | <i>bnc2</i> | intron | <ul style="list-style-type: none"> DNA-binding transcription factor activity developmental pigmentation pronephric nephron tubule development regulation of transcription, DNA-templated | Gaudet <i>et al.</i> , 2011; Patterson <i>et al.</i> , 2013; Kolvenbach <i>et al.</i> , 2019; Gaudet <i>et al.</i> , 2011 |
| BettaMS2.1 | F: GGGACATCAAACGAGAAGGA R: CTCCCAGCTCAAACCACACT | (CA) _n | 303,305 | 10 | LOC114864202 ¹ | intron | Unknow | - |
| BettaMS3.1 | F: TCGCTCTTCTTTATCGCTCA R: GGACGTAGCCATACCTA | (TC) _n | 394 | 4 | <i>dab1a</i> | between intron and exon | <ul style="list-style-type: none"> lateral motor column neuron migration ventral spinal cord development | Gaudet <i>et al.</i> , 2011 |
| BettaMS4.1 | F: GTGGACGAGAGTCTGACTGAAT R: GTGTAACGGCCCCAGGTTA | (TTAA) _n | 310 | 19 | <i>inpp4aa</i> | between intron and exon | <ul style="list-style-type: none"> phosphatidylinositol-3,4-bisphosphate 4-phosphatase activity | Gaudet <i>et al.</i> , 2011 |
| BettaMS5.1 | F: AGAATGGGCTGCAGTAGACG R: TTCAGCCTGTGTTTGCTCTG | (AC) _n | 345 | 3 | <i>gse1</i> | intron | <ul style="list-style-type: none"> protein binding | Rual <i>et al.</i> , 2015 |
| BettaMS6.1 | F: TTCAGCCTGTGTTTGCTCTG R: AGAATGGGCTGCAGTAGACG | (TG) _n | 345 | 3 | <i>gse1</i> | intron | <ul style="list-style-type: none"> protein binding | Rual <i>et al.</i> , 2015 |
| BettaMS7.1 | F: ACATAAACCACTGTGCAAC R: GATGAAACAGCTTTCCAGCA | (AC) _n | 367 | 4 | Non coding gene | - | - | - |
| BettaMS8.1 | F: AATTCTGCATTGGATGGG R: CATACTGTTAAGCCTTCGGT | (GT) _n | 304 | 4 | Non coding gene | - | - | - |
| BettaMS9.1 | F: GCTCGTCTTTAGGGGAGAGG R: ACATTCAGCCGCTAGTGAGG | (CA) _n | 345 | 5 | <i>si:dkey-49n23.1</i> | intron | <ul style="list-style-type: none"> chemorepellent activity semaphorin receptor binding axon guidance negative chemotaxis negative regulation of axon extension involved in axon guidance neural crest cell migration positive regulation of cell migration semaphorin-plexin signaling pathway | Gaudet <i>et al.</i> , 2011 |
| BettaMS10.1 | F: TCTGAGGAAGGAGGCGATTA R: GCGTGCACTGAAGCATAAAG | (CA) _n | 300 | 9 | <i>slc20a2</i> | between intron and exon | <ul style="list-style-type: none"> inorganic phosphate transmembrane transporter activity symporter activity phosphate ion transmembrane transport | Gaudet <i>et al.</i> , 2011 |

| | | | | | | | | |
|--------------|---|-----------------------|-----|----|---------------------------|-------------------------|--|--|
| BettaMS11.1 | F: AAGACGCTCCTTCATCCTGA R: CGTTCTCTCGTGTGCTGT | (CA) _n | 385 | 1 | LOC114854497 ¹ | intron | Unknow | - |
| BettaMS12.1# | F: GCCATCAGGCAGGAGATTAC R: GTTCCCTGCAGACAGAGGAG | (TCAC) _n | 383 | 1 | <i>smox</i> | between intron and exon | <ul style="list-style-type: none"> • oxidoreductase activity • polyamine oxidase activity • hemopoiesis | Gaudet <i>et al.</i> , 2011; Tijssen <i>et al.</i> , 2011 |
| BettaMS13.1 | F: GGGAGGGACGATCTCTTTTC R: CCCCTACTCTGCATGCTCTC | (CA) _n | 307 | 2 | LOC114870512 ¹ | between intron and exon | Unknow | - |
| BettaMS14.1 | F: GGGCTGCACCTTAAACTCAT R: GTCCACTGGGCTGATGTTCT | (TCCA) _n | 356 | 2 | LOC114850832 ¹ | between intron and exon | Unknow | - |
| BettaMS15.1 | F: GCTTTGAGTTGGGCTCAG R: TCCTCCGTGGACCTGACTAC | (CCAGCT) _n | 340 | 3 | Non coding gene | - | - | - |
| BettaMS16.1 | F: CACCTACAACCAGCAGCAGA R: CCACCCTGTTGGTAGGACAT | (AC) _n | 368 | 6 | LOC114856741 ¹ | between intron and exon | Unknow | - |
| BettaMS17.1 | F: GCGGGGTTCTGTCTCTAGTGT R: AGTAGTGCAGAGGGAGTGAG | (CA) _n | 451 | 14 | Non coding gene | - | - | - |
| BettaMS18.1 | F: CCCACATGATCTCACACAC R: CGTCCACATGAATGGAGAGA | (CA) _n | 377 | 11 | <i>myclb</i> | intron | <ul style="list-style-type: none"> • DNA binding • DNA-binding transcription factor activity • protein dimerization activity • developmental process | Gaudet <i>et al.</i> , 2011 |
| BettaMS19.1 | F: GGCTGACCACACAAGTATGC R: AGGCTGTGGTTGAAGTTGCT | (TAGA) _n | 378 | 5 | <i>magilb</i> | intron | <ul style="list-style-type: none"> • molecular adaptor activity • signal transduction | Gaudet <i>et al.</i> , 2011 |
| BettaMS20.1 | F: AGCGGCTGATTTCAAACATA R: GGGACTGGGAGATGAATGAA | (TCAC) _n | 326 | 21 | <i>znf292b</i> | between intron and exon | <ul style="list-style-type: none"> • DNA binding • DNA-binding transcription factor activity, RNA polymerase II-specific • regulation of transcription by RNA polymerase II | Gaudet <i>et al.</i> , 2011 |
| BettaMS1.2 | F: CGACGAGGCTGTGAAATACA R: AGCAGGTAAGTGGCAACC | (CA) _n | 170 | 10 | Non coding gene | - | - | - |

| | | | | | | | | |
|-----------------|--|-------------------|-----|----|------------------|-------------------------|---|---|
| BettaMS2.2 | F: ATTCTTTCTGCCGTAA R: AAAGAGGGCACTAAGCCA | (TG) _n | 200 | 20 | <i>meis2a</i> | intron | <ul style="list-style-type: none"> DNA binding animal organ morphogenesis brain development embryonic pattern specification embryonic viscerocranium morphogenesis eye development positive regulation of cell population proliferation positive regulation of transcription by RNA polymerase II regulation of transcription by RNA polymerase II | Gaudet <i>et al.</i> , 2011; Melvin <i>et al.</i> , 2013 |
| BettaMS3.2 # | F: CATCCGCGTTTCGATTGAA R: GTGGAAGGCCTCTCCTTGA | (TG) _n | 171 | 4 | Non coding gene | - | - | - |
| BettaMS4.2 | F: CCCACAAGTGAAAAAGGAT R: ATGCCCTAAGTGTGTCCAG | (CA) _n | 200 | 1 | <i>prss12</i> | intron | <ul style="list-style-type: none"> scavenger receptor activity serine-type endopeptidase activity zymogen activation | Gaudet <i>et al.</i> , 2011 |
| BettaMS5.2 | F: GGACAGATTCAGGGTTTAGG R: ACCAGGTACCAGCACAACA | (TG) _n | 195 | 3 | <i>pde3b</i> | between intron and exon | <ul style="list-style-type: none"> 3',5'-cyclic-AMP phosphodiesterase activity 3',5'-cyclic-nucleotide phosphodiesterase activity metal ion binding protein kinase B binding negative regulation of cAMP-mediated signaling signal transduction | Gaudet <i>et al.</i> , 2011 |
| BettaMS6.2 | F: CTTGGAGCCGAATGATATTG R: TACCTCATTCTCCCTCCT | (CA) _n | 246 | 5 | Non coding gene | - | - | - |
| BettaMS7.2 | F: GCCACCTGCTTAAGATACTT R: TAATCACTGCAGCCCCTCAT | (TG) _n | 290 | 4 | <i>ctnnd2a</i> | intron | <ul style="list-style-type: none"> cadherin binding cell-cell adhesion cell-cell junction assembly dendritic spine morphogenesis | Gaudet <i>et al.</i> , 2011 |
| BettaMS8.2 | F: ACCATAGTGTCAATCACCAG R: TACTGTGGCTTGTTGCCT | (CA) _n | 254 | 5 | <i>prickle2b</i> | intron | <ul style="list-style-type: none"> zinc ion binding cilium assembly Kupffer's vesicle development melanosome transport neural retina development | Mei <i>et al.</i> , 2014 |

| | | | | | | | | |
|--------------|---|---------------------|-----|----|-----------------|--------|---|---|
| BettaMS9.2 | F: GGCAAAATGATAGTGTGTGTC R: CTGACCAGCTGGGATAGAGG | (TG) _n | 248 | 17 | Non coding gene | - | - | - |
| BettaMS10.2# | F: CACAAACCCACAGACACAG R: GCATGGTGTGTGGAAGA | (AC) _n | 259 | 2 | Non coding gene | - | - | - |
| BettaMS11.2 | F: GAAGCCTTTATTCTGTAGACC R: CGTGTGCTGTTCTTTCGT | (CA) _n | 250 | 13 | <i>ulk2</i> | intron | <ul style="list-style-type: none"> ATP binding protein serine/threonine kinase activity autophagosome assembly autophagy of mitochondrion axon extension regulation of dendrite morphogenesis dendrite development | Gaudet <i>et al.</i> , 2011; Lee <i>et al.</i> , 2014; Taylor <i>et al.</i> , 2011; Colombo <i>et al.</i> , 2013 |
| BettaMS12.2 | F: AGGAAGACAGAGGACGACGA R: CCCCAAGGTCCTCCTGTTAT | (CA) _n | 251 | 10 | <i>efnb1</i> | intron | <ul style="list-style-type: none"> ephrin receptor binding axon guidance embryonic liver development ephrin receptor signaling pathway optic vesicle morphogenesis | Gaudet <i>et al.</i> , 2011; Cayuso <i>et al.</i> , 2016; Cavodeassi <i>et al.</i> , 2013 |
| BettaMS13.2 | F: ATTCACCCAGCAAAGGGAAG R: GGGCCACGAGATACTGTGTT | (CA) _n | 238 | 19 | Non coding gene | - | - | - |
| BettaMS14.2 | F: CCCGGTTTCTTGTCATTC R: CGCTGATGGAAATTGAGT | (TG) _n | 224 | 19 | Non coding gene | - | - | - |
| BettaMS15.2 | F: TGGGATTCTTCGGAGTTGTC R: TTCTTTCCAGCTTGCAATT | (TG) _n | 257 | 9 | <i>exoc4</i> | intron | <ul style="list-style-type: none"> chemical synaptic transmission exocytosis Golgi to plasma membrane transport protein targeting to membrane vesicle docking involved in exocytosis vesicle tethering involved in exocytosis | Gaudet <i>et al.</i> , 2011 |
| BettaMS16.2 | F: TGGTGCTGTGGTATTCCTGA R: ACTGTCAAATAGCTCATGC | (TATC) _n | 274 | 18 | Non coding gene | - | - | - |

| | | | | | | | | |
|--------------|--|--------------------|-----|----|---------------------------|-------------------------|---|--|
| BettaMS17.2 | F: TGAACAGAACGTACCTGTCCTC R: ATCAGTTGCAAGGCCAGTTT | (TG) _n | 226 | 15 | <i>cdh23</i> | intron | <ul style="list-style-type: none"> • cadherin binding • calcium ion binding • transmembrane transporter activity • auditory receptor cell development • hair cell differentiation • sensory perception of sound • protein localization to cilium • neuromast hair cell morphogenesis • locomotory behavior • inner ear receptor cell stereocilium organization • homophilic cell adhesion via plasma membrane adhesion molecules • equilibration • endocytosis • detection of mechanical stimulus involved in sensory perception of sound • detection of mechanical stimulus involved in sensory perception • cell projection organization • cell-cell adhesion via plasma membrane adhesion molecules | Gaudet <i>et al.</i> , 2011; Hailey <i>et al.</i> , 2012; Obholzer <i>et al.</i> , 2012; Sollner <i>et al.</i> , 2004; Chou <i>et al.</i> , 2017; Blanco-Sanchez <i>et al.</i> , 2014; Obholzer <i>et al.</i> , 2012; Maeda <i>et al.</i> , 2017 |
| BettaMS18.2# | F: CGGCCTGACAGAAAACAGA R: GCGGGTTTTGTGTAGTTGG | (TG) _n | 231 | 12 | LOC114866883 ¹ | intron | Unknow | - |
| BettaMS19.2 | F: TGGTGCATTGGTGCATTAT R: CGGAAGAACTGGAGGAAGTG | (ATT) _n | 242 | 18 | Non coding gene | - | - | - |
| BettaMS20.2 | F: ACAGAAGAGTGGATGTGTGT R: CGACTCCTTCACTCCTCAG | (GT) _n | 317 | 17 | LOC114844365 ¹ | between intron and exon | Unknow | - |

¹Uncharacterized gene.

References#

1. Gaudet, P.; Livstone, M.S.; Lewis, S.E.; Thomas, P.D. Phylogenetic-based propagation of functional annotations within the Gene Ontology consortium. *Brief. Bioinform.* **2011**, *12*, 449–462. doi:10.1093/bib/bbr042.
2. Patterson, L.B.; Parichy, D.M. Interactions with iridophores and the tissue environment required for patterning melanophores and xanthophores during zebrafish adult pigment stripe formation. *PLoS Genet.* **2013**, *9*, e1003561. doi:10.1371/journal.pgen.1003561.
3. Kolvenbach, C.M.; Dworschak, G.C.; Frese, S.; Japp, A.S.; Schuster, P.; Wenzlitschke, N.; Yilmaz, Ö.; Lopes, F.M.; Pryalukhin, A.; Schierbaum, L.; et al. Rare variants in *BNC2* are implicated in autosomal-dominant congenital lower urinary-tract obstruction. *Am. J. Hum. Genet.* **2019**, *104*, 994–1006. doi:10.1016/j.ajhg.2019.03.023.
4. Rual, J.F.; Venkatesan, K.; Hao, T.; Hirozane-Kishikawa, T.; Dricot, A.; Li, N.; Berriz, G.F.; Gibbons, F.D.; Dreze, M.; Ayivi-Guedehoussou, N.; et al. Towards a proteome-scale map of the human protein-protein interaction network. *Nature* **2005**, *437*, 1173–1178. doi:10.1038/nature04209.
5. Tijssen, M.R.; Cvejic, A.; Joshi, A.; Hannah, R.L.; Ferreira, R.; Forrai, A.; Bellissimo, D.C.; Oram, S.H.; Smethurst, P.A.; Wilson, N.K.; et al. Genome-wide analysis of simultaneous GATA1/2, RUNX1, FLI1, and SCL binding in megakaryocytes identifies hematopoietic regulators. *Dev. Cell* **2011**, *20*, 597–609. doi:10.1016/j.devcel.2011.04.008.
6. Melvin, V.S.; Feng, W.; Hernandez-Lagunas, L.; Artinger, K.B.; Williams, T. A morpholino-based screen to identify novel genes involved in craniofacial morphogenesis. *Dev. Dyn.* **2013**, *242*, 817–831. doi:10.1002/dvdy.23969.
7. Mei, X.; Westfall, T.A.; Zhang, Q.; Sheffield, V.C.; Bassuk, A.G.; Slusarski, D.C. Functional characterization of Prickle2 and BBS7 identify overlapping phenotypes yet distinct mechanisms. *Dev. Biol.* **2014**, *392*, 245–255. doi:10.1016/j.ydbio.2014.05.020.
8. Lee, S.; Page-McCaw, P.; Gamse, J.T. Kctd12 and Ulk2 partner to regulate dendritogenesis and behavior in the habenular nuclei. *PLoS One* **2014**, *9*, e110280. doi:10.1371/journal.pone.0110280.
9. Taylor, R.W.; Qi, J.Y.; Talaga, A.K.; Ma, T.P.; Pan, L.; Bartholomew, C.R.; Klionsky, D.J.; Moens, C.B.; Gamse, J.T. Asymmetric inhibition of Ulk2 causes left-right differences in habenular neuropil formation. *J. Neurosci.* **2011**, *31*, 9869–9878. doi:10.1523/JNEUROSCI.0435-11.2011.
10. Colombo, A.; Palma, K.; Armijo, L.; Mione, M.; Signore, I.A.; Morales, C.; Guerrero, N.; Meynard, M.M.; Pérez, R.; Suazo, J.; et al. Daam1a mediates asymmetric habenular morphogenesis by regulating dendritic and axonal outgrowth. *Dev.* **2013**, *140*, 3997–4007. doi:10.1242/dev.091934.
11. Cayuso, J.; Dzementsei, A.; Fischer, J.C.; Karemore, G.; Caviglia, S.; Bartholdson, J.; Wright, G.J.; Ober, E.A. EphrinB1/EphB3b coordinate bidirectional epithelial-mesenchymal interactions controlling liver morphogenesis and laterality. *Dev. Cell.* **2016**, *39*, 316–328. doi:10.1016/j.devcel.2016.10.009.
12. Cavodeassi, F.; Ivanovitch, K.; Wilson, S.W. Eph/Ephrin signalling maintains eye field segregation from adjacent neural plate territories during forebrain morphogenesis. *Dev.* **2013**, *140*, 4193–4202. doi:10.1242/dev.097048.

13. Hailey, D.W.; Roberts, B.; Owens, K.N.; Stewart, A.K.; Linbo, T.; Pujol, R.; Alper, S.L.; Rubel, E.W.; Raible, D.W. Loss of Slc4a1b chloride/bicarbonate exchanger function protects mechanosensory hair cells from aminoglycoside damage in the zebrafish mutant *persephone*. *PLoS Genet.* **2012**, *8*, e1002971. doi:10.1371/journal.pgen.1002971.
14. Obholzer, N.; Swinburne, I.A.; Schwab, E.; Nechiporuk, A.V.; Nicolson, T.; Megason, S.G. Rapid positional cloning of zebrafish mutations by linkage and homozygosity mapping using whole-genome sequencing. *Dev.* **2012**, *139*, 4280–4290. doi:10.1242/dev.083931.
15. Söllner, C.; Rauch, G.J.; Siemens, J.; Geisler, R.; Schuster, S.C.; Müller, U.; Nicolson, T.; Tübingen Screen, C. Mutations in *cadherin 23* affect tip links in zebrafish sensory hair cells. *Nature* **2004**, *428*, 955–959, doi:10.1038/nature02484.
16. Chou, S.W.; Chen, Z.; Zhu, S.; Davis, R.W.; Hu, J.; Liu, L.; Fernando, C.A.; Kindig, K.; Brown, W.C.; Stepanyan, R.; et al. A molecular basis for water motion detection by the mechanosensory lateral line of zebrafish. *Nat. Commun.* **2017**, *8*, 2234. doi:10.1038/s41467-017-01604-2.
17. Blanco-Sánchez, B.; Clément, A.; Fierro, J., Jr.; Washbourne, P.; Westerfield, M. Complexes of Usher proteins preassemble at the endoplasmic reticulum and are required for trafficking and ER homeostasis. *Dis. Model. Mech.* **2014**, *7*, 547–559. doi:10.1242/dmm.014068.
18. Maeda, R.; Pacentine, I.V.; Erickson, T.; Nicolson, T. Functional analysis of the transmembrane and cytoplasmic domains of PCDH15A in zebrafish hair cells. *J. Neurosci.* **2017**, *37*, 3231–3245. doi:10.1523/JNEUROSCI.2216-16.2017.

Supplementary Table S2. Information on fighting fish genomes.

| Name | Organism | Sex | Instrument | Strategy | Source | Layout | Run |
|------|------------------------|--------|------------------|------------------|---------|--------|-------------|
| B2 | <i>Betta splendens</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR4790873 |
| C1 | <i>Betta splendens</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR4790878 |
| D1 | <i>Betta splendens</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR4790879 |
| D2 | <i>Betta splendens</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR4790875 |
| H1 | <i>Betta splendens</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR4790883 |
| Orn4 | <i>Betta splendens</i> | Male | Illumina HiSeq X | WGS ¹ | GENOMIC | PAIRED | SRR18231397 |
| Orn5 | <i>Betta splendens</i> | Male | Illumina HiSeq X | WGS ¹ | GENOMIC | PAIRED | SRR18231396 |
| Orn6 | <i>Betta splendens</i> | Male | Illumina HiSeq X | WGS ¹ | GENOMIC | PAIRED | SRR18231395 |
| Orn7 | <i>Betta splendens</i> | Male | Illumina HiSeq X | WGS ¹ | GENOMIC | PAIRED | SRR18231394 |
| Orn8 | <i>Betta splendens</i> | Male | Illumina HiSeq X | WGS ¹ | GENOMIC | PAIRED | SRR18231393 |
| Orn9 | <i>Betta splendens</i> | Male | Illumina HiSeq X | WGS ¹ | GENOMIC | PAIRED | SRR18231392 |
| BBE1 | <i>Betta bellica</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR3904015 |
| BBE2 | <i>Betta bellica</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR3904011 |
| BBR1 | <i>Betta brownorum</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR3904028 |
| BBR2 | <i>Betta brownorum</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR3904022 |
| BBU1 | <i>Betta burdigala</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR3904017 |
| BBU2 | <i>Betta burdigala</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR3904012 |
| BIM1 | <i>Betta imbellis</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR4766250 |

| | | | | | | | |
|-------|----------------------------|--------|-------------|------------------|---------|--------|------------|
| BIM2 | <i>Betta imbellis</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR3332318 |
| BLI1 | <i>Betta livida</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR3904016 |
| BLI2 | <i>Betta livida</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR3904013 |
| BMA1 | <i>Betta mahachaiensis</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR4766262 |
| BMA2 | <i>Betta mahachaiensis</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR3904031 |
| BSM1 | <i>Betta smaragdina</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR3904029 |
| BSM2 | <i>Betta smaragdina</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR3904047 |
| BTU1 | <i>Betta tussyae</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR3904033 |
| BTU2 | <i>Betta tussyae</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR3904027 |
| BCH1 | <i>Betta channoides</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR3904023 |
| BCO1 | <i>Betta compuncta</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR3904018 |
| BHI1 | <i>Betta hipposideros</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR3904020 |
| BID1 | <i>Betta ideii</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR3904021 |
| BMAC1 | <i>Betta macrostoma</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR4766265 |
| BMAC2 | <i>Betta macrostoma</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR4766273 |
| BMAN1 | <i>Betta mandor</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR3904026 |
| BPU1 | <i>Betta pulchra</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR3904019 |
| BPU2 | <i>Betta pulchra</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR3904014 |
| BRU1 | <i>Betta rubra</i> | Unknow | HiSeq X Ten | WGS ¹ | GENOMIC | PAIRED | ERR3904024 |

¹Whole Genome Sequencing

Supplementary Table S7. Descriptive statistics at microsatellite markers level based on 21 Siamese fighting fish (*Betta splendens*, Regan, 1910)

| Markers | N_a | H_o | H_e | PIC | Source |
|-------------|-------|-------|-------|-------|-------------------------------------|
| BettaMS10.1 | 22 | 0.95 | 0.94 | 0.94 | This study |
| BettaMS14.1 | 13 | 0.24 | 0.88 | 0.87 | This study |
| BettaMS2.2 | 10 | 0.48 | 0.84 | 0.82 | This study |
| BettaMS14.2 | 1 | 0.00 | 0.00 | 0.00 | This study |
| BettaMS4 | 15 | 0.86 | 0.91 | 0.90 | Chailertrit <i>et al.</i> , 2014 |
| BettaMS5 | 14 | 0.52 | 0.91 | 0.90 | Chailertrit <i>et al.</i> , 2014 |
| BettaMS8 | 4 | 0.29 | 0.33 | 0.31 | Chailertrit <i>et al.</i> , 2014 |
| BettaMS15 | 5 | 0.57 | 0.71 | 0.66 | Chailertrit <i>et al.</i> , 2014 |
| BettaMS17 | 15 | 0.75 | 0.87 | 0.86 | Chailertrit <i>et al.</i> , 2014 |
| BettaMS23 | 12 | 0.95 | 0.87 | 0.85 | Chailertrit <i>et al.</i> , 2014 |
| BettaMS25 | 11 | 0.24 | 0.78 | 0.76 | Chailertrit <i>et al.</i> , 2014 |
| BettaMS40 | 13 | 0.38 | 0.87 | 0.86 | Chailertrit <i>et al.</i> , 2014 |

Number of alleles (N_a); allelic richness (H_o); expected heterozygosity (H_e); polymorphic information content (PIC).

References

1. Regan, C.T. The Asiatic fishes of the family Anabantidae. In *Proceedings of the Zoological Society of London*, 1909; pp. 767–787.
2. Chailertrit, V.; Swatdipong, A.; Peyachoknagul, S.; Salaenoi, J.; Srikulnath, K. Isolation and characterization of novel microsatellite markers from Siamese fighting fish (*Betta splendens*, Osphronemidae, Anabantoidei) and their transferability to related species, *B. smaragdina* and *B. imbellis*. *Genet. Mol. Res.* 2014, 13, 7157–7162. doi:10.4238/2014.September.5.1.

