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1 **Genome-wide association study and genomic prediction of tolerance to acute**  
2 **hypoxia in rainbow trout**

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22

23 **Abstract**

24

25 Hypoxia is one of the major threats to the aquaculture sector resulting in substantial

26 economic losses to the fish farmers. Thus, tolerance to hypoxia is of high economic interest to

27 be genetically improved by breeding programs. Rainbow trout (*Oncorhynchus mykiss*) is one

28 of the most cultured salmonid species worldwide, with well-developed breeding programs.

29 Still, studies of genetic potential to improve hypoxia tolerance in this species are rare. In the

30 present study, 1,320 individuals of rainbow trout were used for a genome-wide association

31 study of acute hypoxia tolerance based on imputed high-density genotypes to explore the

32 genetic architecture and related candidate genes affecting hypoxia response. Three significant

33 (Omy31\_1, Omy31\_2, Omy20) and two putative (Omy15, Omy28) quantitative trait loci (QTLs)

34 were detected, but each of them only explained between 0.2% and 0.8% of the genetic

35 variance of acute hypoxia tolerance. However, heritability was estimated at a moderate value

36 of 0.24 – 0.28, that suggests a solid potential to improve hypoxia tolerance in the studied

37 rainbow trout population by genetic selection. Moreover, it was shown that genomic

38 prediction for hypoxia tolerance would lead to a relative increase of ~ 11% for genomic

39 selection (GS) accuracy compared to the pedigree-based selection, considering a reference

40 population of 1000 individuals. Finally, sixteen genes (*ids, fmr1, arx, lonrf3, commd5, map4k4,*

41 *smu1, b4galt1, re1, abca1, noa1, igfbp7, noxo1, bcl2a, trim25, mylk3*) were proposed as

42 potential functional candidates involved in hypoxia tolerance. Taking all proposed candidate

43 genes within two main QTLs on Omy31 (12 out of 16 genes), we may hypothesize that the

44 complex response to acute hypoxia in rainbow trout, i.e., the interplay between behavioural,

45 morphological, and physiological responses, is primarily encoded by two supergenes.

46 However, further functional validation of their effects may help to specify the biological

47 mechanisms triggering a response to acute hypoxia in rainbow trout.

48 **Key words:** GWAS, Heritability, Hypoxia resistance, Hypoxia tolerance, *Oncorhynchus mykiss*

49

## 50 **1. Introduction**

51 A lack of oxygen referred to as hypoxia is one of the greatest challenges that most life  
52 can face. In terrestrial ecosystems, conditions of low oxygen are rare. In contrast, low  
53 concentration of dissolved oxygen (DO), is much more common in aquatic ecosystems (Diaz,  
54 2001; Townhill et al., 2017). The DO is a primary limiting factor in aquaculture because fish  
55 have aerobic metabolism requiring DO at efficient levels (Abdel-Tawwab et al., 2019). Hypoxia  
56 is also a very complex condition caused by several interrelated factors that might be divided  
57 into three main groups: 1) substandard rearing conditions due to the technical failure of water  
58 aeration or oxygenation or too high stocking densities regarding post-prandial oxygen  
59 consumption (Abdel-Tawwab et al., 2019); 2) eutrophication associated with increased  
60 anthropogenic nutrient loading of lakes, rivers and coastal waters leading to blooms of algae  
61 and phytoplankton, the dissimilation or death of which subsequently leads massive depletion  
62 of dissolved oxygen (Friedrich et al., 2014); 3) global warming as the main aspect of climate  
63 change leading to increased microbial respiration rates and reduced oxygen solubility with  
64 increasing water temperatures (McBryan et al., 2013; Sae-Lim et al., 2017; Reid et al., 2019).

65 Hypoxia is one of the major threats for aquaculture sector (Farrell and Richards, 2009;  
66 Abdel-Tawwab et al., 2019) affecting seriously health and welfare of fish including growth,  
67 reproduction, behaviour, immunity, and other energy-demanding activities (Gallage et al.,  
68 2016; Abdel-Tawwab et al., 2019; Reid et al., 2019). Consequently, hypoxia often leads to  
69 severe losses to fish farmers. Thus, tolerance to hypoxia is of high economic interest to be  
70 genetically improved by breeding programs (Sae-Lim et al., 2017).

71 Of the total European aquaculture production, 80 – 83% originated from at least 37  
72 different breeding programs that are primarily focused on production traits like growth or  
73 processing yields (Janssen et al., 2017). Moreover, improving of disease resistance to several  
74 pathogens in both freshwater and marine fishes is under strong interest (Yáñez et al., 2014;  
75 Gjedrem and Rye, 2016; Houston et al., 2020). However, improving resilience to  
76 environmental variation like e.g., tolerance to temperature, hypoxia or salinity is starting to  
77 be an interesting challenge for aquaculture breeding programs (Allal and Nguyen, 2022).

78 Through studies using mammalian model organisms, three hypotheses for oxygen-  
79 sensing and downstream response have been proposed: a “membrane hypothesis,” a  
80 “mitochondrial/metabolic hypothesis,” and a more recent “gas transmitter hypothesis”.  
81 These hypotheses are in close parallel to fish physiological response to hypoxia conditions  
82 (Gattuso et al., 2018).

83 Many molecular and physiological responses to hypoxia are regulated by transcription  
84 factors named hypoxia-inducible factors (HIFs), heterodimeric proteins with an oxygen-  
85 sensitive  $\alpha$ -subunit, HIF- $\alpha$  and an oxygen-insensitive  $\beta$ -subunit, HIF- $\beta$  (Nikinmaa and Rees,  
86 2005; Pelster and Egg, 2018; Dzhililova and Makarova, 2020). HIF- $\alpha$  is a key regulator of many  
87 hypoxia-related proteins and signalling pathways that facilitate adaptation of organisms to  
88 hypoxic environments e.g.: insulin-like growth factor binding protein (*igfbp*, a growth  
89 inhibitory protein), vascular endothelial growth factor (VEGF), mitogen-activated protein  
90 kinases (MAPK), reactive oxygen species (ROS), reactive nitrogen species (RNS), oxidative  
91 phosphorylation (OXPHOS), NADPH oxidase or nitric oxide synthase (NOS, a RNS regulating  
92 enzyme) (Fu et al., 2000; Zhu et al., 2013; Fago and Jensen, 2015; Sahoo et al., 2016; Gattuso  
93 et al., 2018; Thomas et al., 2019; Dzhililova and Makarova, 2020).

94           The acute response of fish to hypoxia mainly combines physiological changes in order  
95 to maintain normal oxygen supply to tissues (hyperventilation, bradycardia, increased blood  
96 O<sub>2</sub>-carrying capacity and redistribution of blood flow) with metabolic changes to provide  
97 adequate amounts of ATP (adenosine triphosphate) with less oxygen through stimulation of  
98 anaerobic glycolysis and gluconeogenesis pathways (Farrell and Richards, 2009; Richards,  
99 2011; Genz et al., 2013; Gattuso et al., 2018). Due to the use of carbohydrates to produce ATP  
100 in the absence of oxygen, glycogen and glucose levels immediately rise and further resulting  
101 in lactate accumulation and circulation (Omlin and Weber, 2010; Li et al., 2018; Abdel-Tawwab  
102 et al., 2019; Léger et al., 2021). Under chronic hypoxia, lipolysis replaces glycolysis, and lipids  
103 are the main energy source of fish (Li et al., 2018). Besides, both acute and chronic hypoxia  
104 stress promotes cell apoptosis and oxidative stress leading to DNA damage (Poon et al., 2007;  
105 Mahfouz et al., 2015; Mustafa et al., 2015; Birnie-Gauvin et al., 2017; Chowdhury and Saikia,  
106 2020), destroys the innate immune system, and makes fish more susceptible to pathogens  
107 (Giomi et al., 2016; Esteve et al., 2017; Sae-Lim et al., 2017; Abdel-Tawwab et al., 2019).  
108 Furthermore, fishes can show various behavioural responses such as rising to the surface to  
109 breathe the uppermost layer of water in contact with air, increasing activity to escape the  
110 hypoxic area or decreasing activity to reduce oxygen demand (Domenici et al., 2007; Domenici  
111 et al., 2013; Bowyer et al., 2014).

112           Rainbow trout (*Oncorhynchus mykiss*) is one of the most cultured salmonid species  
113 worldwide (FAO, 2020), with well-developed breeding programs resulting in significant  
114 selection responses in growth, slaughter yields and disease resistance (Gjedrem and Rye,  
115 2016; Chavanne et al., 2016; Boudry et al., 2021). In France, four commercial breeding  
116 programs are carried out. Besides, French rainbow trout production is mostly developed with  
117 all-female diploid or triploid stocks (Piferrer et al., 2009) reared in a land-based facility in a

118 concrete race-way with oxygen enrichment of the water by liquid oxygen injected in the water  
119 or mechanical aeration. Precise continuous monitoring of oxygen level is done with electronic  
120 probes and adapted software according to oxygen concentration fluctuations due to feeding  
121 delivery and consumption. However, an increase in trout robustness is demanded to adapt  
122 fishes to the rapid daily changes of oxygen content associated in summer with the increased  
123 temperature variation of the river's waters. Both acute and long-term exposure to low  
124 dissolved oxygen levels that are less than 4 mg/L (while optimum values are in the range from  
125 8 to 10 mg/L) can lead to massive mortality and growth depression caused by reduced growth  
126 hormone-I (GH-I) and insulin like growth factor I-II (IGF I-II) levels in rainbow trout (Hou et al.,  
127 2020; Aksakal and Ekinci, 2021; Royer et al., 2021). Although hypoxia is a common threat in  
128 rainbow trout farming, studies of genetic potential to hypoxia tolerance are still rare (Scott et  
129 al., 2015; Borey et al., 2018; Strowbridge et al., 2021; Lagarde et al., Submitted). Thus,  
130 understanding the genetic architecture of hypoxia tolerance could help to improve the  
131 resilience of rainbow trout stocks by genetic selection.

132         To select for genetically environmental-tolerant individuals, enough genetic variability  
133 must exist within the species. Although hypoxia tolerance is an interesting trait for a trout  
134 breeding program, heritability estimates for this trait are still missing in salmonids. As far as  
135 we know, there were only two estimates in the literature to date showing high heritability for  
136 acute hypoxia tolerance (0.50 and 0.61, respectively) in common carp (*Cyprinus carpio*) (Nagy  
137 et al., 1980) and large yellow croaker (*Larimichthys crocea*) (Ding et al., 2022).

138         Numerous genome-wide association studies (GWAS) have been run in the last decade  
139 to identify genomic regions (Quantitative trait loci – QTLs) and functional candidate genes  
140 associated with traits of interest in different aquaculture species (Robledo et al., 2017;  
141 Houston et al., 2020). However, only a few GWAS for hypoxia tolerance have been conducted

142 so far, and on limited number of fish species other than salmonids: Nile tilapia (Li et al., 2017;  
143 Yu et al., 2021) (*Oreochromis niloticus*), two catfish species (Wang et al., 2017; Zhong et al.,  
144 2017) (*Ictalurus punctatus* and *Ictalurus furcatus*), golden pompano (San et al., 2021)  
145 (*Trachinotus ovatus*) and large yellow croaker (Ding et al., 2022). These studies revealed  
146 multiple QTLs and potential candidate genes that might be responsible for response to  
147 hypoxia.

148 The aims of this study were i) to estimate the variance components of hypoxia  
149 tolerance; ii) to predict the genomic selection (GS) accuracy compared to pedigree-based  
150 BLUP selection for hypoxia tolerance in rainbow trout; iii) to perform GWAS to investigate the  
151 genetic architecture of hypoxia tolerance and to propose potential candidate genes for the  
152 main QTLs. To our knowledge, this is the first GWAS evaluating the genetic architecture and  
153 accuracy of genomic prediction of acute hypoxia tolerance in rainbow trout.

154

## 155 **2. Material and Methods**

### 156 **2.1 Ethics Statement**

157 This study used fin clips collected by the breeding company “Milin Nevez” (Bretagne  
158 Truite Group, Plouigneau, France) as part of their commercial breeding programs. All  
159 challenges were carried out in accordance with the European guidelines (Directive 2010–63-  
160 EU) and the corresponding French legislation. Animal experiment procedures were approved  
161 by the ethics committee on animal experimentation COMETH ANSES/ENVA/UPC No. 16 and  
162 were authorized by the French Ministry of Higher Education, Research, and Innovation under  
163 number APAFIS#24441-2020022417122193 v3, project N°20-032.

164

### 165 **2.2 Establishment and rearing of challenged fish**



166 The experimental stock was established (6.11. 2019) from 190 dams and 98 sex-  
167 reversed neomales of a commercial selected all-female line of Milin Nevez breeding company  
168 using ten independent full-factorial mating designs. The eggs were split by the dam (meaning  
169 190 separated batches, one for each dam), but only the 90 best spawnings were conserved  
170 for mixing at the larval stage. So, in the end, the experimental stock came from 90 dams and  
171 98 neomales. From each of the parents used in the mating, a fin clip was collected and stored  
172 in 98% ethanol for later DNA extraction and genotyping.

173 The progeny stock was then reared under common commercial conditions until 9  
174 months. 1,508 individuals were PIT tagged by Bretagne Truite and transferred to the SYSAAF-  
175 ANSES Fortior Genetics platform (ANSES, Plouzané, France) and acclimatized for four days  
176 before acute hypoxia challenge. Out of 1,508 fish, 1,351 individuals were challenged to  
177 hypoxia and fin-clipped for DNA and finally only 1,320 DNA samples were sent for genotyping.

178

### 179 **2.3 Hypoxia challenge**

180 The challenge to hypoxia was sub-divided into seven batches (one per day starting  
181 from 17.8. 2020), and in each batch, a random sample of fish was challenged (Table 1.) At the  
182 beginning of each trial, the initial oxygen level was recorded. The gradual decline of oxygen  
183 was conducted by bubbling nitrogen and monitored every five minutes until the end of the  
184 trial of a given day (Supplementary Fig. S1). Water temperature was constantly monitored  
185 during the trial and ranged from 15.2°C to 17.3°C. Hypoxia tolerance phenotype was defined  
186 as the time to loss of equilibrium (TLE) in minutes with a value set to 0 for the first fish losing  
187 its equilibrium. When fish lost their equilibrium, they were removed from the tank, identified  
188 (PIT-tag reading), weighed and euthanized in a concentrated bath of Eugenol (180mg/L). The  
189 corresponding TLE and oxygenation levels ( $\text{mg/l}^{-1}$ ) were recorded for each individual. The

190 challenge ended when the last fish lost its equilibrium and was removed from the tank. The  
191 parents were not challenged and were only used as a reference population for the imputation  
192 of their offspring genotypes.

## 193 **2.4 Genotyping and imputation**

194 Fin samples from hypoxia-challenged fish (1,320 ind.) and their parents (188 ind.) were  
195 sent to the INRAE genotyping platform Gentyane (Clermont-Ferrand, France) for DNA  
196 extraction and genotyping. The challenged fish were genotyped for 57,501 SNPs using the  
197 medium-density (MD) Rainbow Trout Axiom<sup>®</sup> 57K SNP array from Thermo Fisher (Palti et al.,  
198 2015). Parents were genotyped for 664,531 SNPs using a newly constructed high-density (HD)  
199 Rainbow Trout Axiom<sup>®</sup> 665K SNP array (Bernard et al., 2022).

200 The first round of quality control was done by Thermo Fisher software Axiom Analysis  
201 Suite™ with threshold values of 97% for SNP call rate and 95% for sample call rate for both  
202 MD and HD SNPs. All parents (288) passed the preliminary quality control, but twenty-three  
203 challenged fish samples did not pass quality control, and thus 1,297 progeny samples  
204 remained for the following step of quality control. Then, SNPs with probe polymorphism and  
205 multiple locations on the Arlee genome reference assembly (accession number:  
206 USDA\_Omyka\_1.1.; Gao et al., 2021) were discarded as described in Bernard et al. (2022).

207 Finally, PLINK v1.9 software (Chang et al., 2015) was used for keeping only SNPs with  
208 deviation from Hardy-Weinberg equilibrium with a p-value > 0.0001 and a minor allele  
209 frequency (MAF > 0.05). On the HD genotypes, 418,925 SNPs remained for the parents, while  
210 29,091 SNPs remained on the MD genotypes of the challenged progeny. Parentage  
211 assignment was done using 1,000 randomly sampled markers with the R package APIS (Griot  
212 et al., 2020) with a positive assignment error rate set to 2%.

213 The imputation of the MD genotypes into HD genotypes for the 1,297 challenged  
214 offspring was run using FIMPUTE3 software (Sargolzaei et al., 2014) utilizing quality-filtered  
215 genotypes and pedigree information from 188 parents. The correctness of imputation was  
216 checked by mendelian testing and by observation of changes in original MD genotypes that  
217 varied from 1 (0.004%) to 106 SNPs (0.37%) per individual. The final dataset contained 1,297  
218 phenotyped and genotyped (418,925 imputed SNPs) progeny, which were subjected to  
219 genomic analysis.

220

## 221 **2.5 Estimation of breeding values and variance components**

222 The following statistical model was derived to describe the hypoxia response:

$$223 \quad TLE_{ijk} = \mu + trial_i + dam_j + anim_k + e_{ijkl}$$

224 where  $TLE_{ijk}$  is the TLE performance for animal  $k$ , produced by dam  $j$  and tested day  $i$ ,  $\mu$  the  
225 overall mean for hypoxia tolerance trait in minutes,  $trial$  is the fixed effect of challenge trial ( $i$   
226 = 1 – 7);  $dam$  is the random maternal effect ( $j$  = 1 – 90);  $anim$  is the random genetic effect of  
227 the animal ( $k$  = 1 – 1,297) and  $e_{ijkl}$  is the random residual. In total, 49,402 animals were related  
228 through the pedigree relationship matrix (A) for BLUP evaluation, tracing back eight  
229 generations of ancestors of the 1,320 phenotyped animals.

230 For genomic-based evaluations, the pedigree matrix A was replaced by a genomic relationship  
231 matrix G (VanRaden, 2008), considering either MD (29,091 SNPs) or HD (418,925 SNPs)  
232 genotypes.

233 Variance components were estimated using the restricted maximum likelihood  
234 method and AIREML algorithm in BLUPF90 software (Misztal et al., 2002).

235 The selection accuracies of a pedigree-based selection approach (BLUP) and a genomic  
236 selection approach (GBLUP) were assessed from estimated breeding values (EBV and GEBV,

237 respectively) that were derived through consideration of either the pedigree relationship  
238 matrix (A) or the genomic matrix (G) using the software package BLUPf90 (Misztal et al., 2014)

239 To evaluate the accuracy of (G)EBVs, 100 replicates of Monte Carlo ‘leave-one-group-  
240 out’ cross-validation tests were run considering the same procedure as described in  
241 D’Ambrosio et al. (2020). For each replicate, 297 fish were randomly chosen for the validation  
242 set, and 1,000 fish were chosen for the training set. The phenotypes recorded in the validation  
243 population were then hidden and breeding values were estimated using BLUP and GBLUP  
244 models. The accuracy ( $r$ ) for each replicate was computed as:

$$245 \quad r = \text{cor}((G)EBV, y^*)/h$$

246 where  $\text{cor}((G)EBV, y)$  is the correlation between the (G)EBV and the phenotypes  $y^*$  adjusted  
247 for batch and maternal effects of the 297 individuals belonging to the validation population,  
248 and  $h$  is the square root of the pedigree-based heritability. Moreover, the degree of bias in  
249 BLUP and GBLUP estimations was evaluated by the regression coefficient of the adjusted  
250 phenotypes on the (G)EBVs. In the absence of selection bias, this coefficient is expected to be  
251 equal to 1; in the case of EBV over-dispersion (inflation), the coefficient is below 1, and in the  
252 case of EBV under-dispersion the value is above 1. The selection accuracies and the inflation  
253 coefficients of different tested models (BLUP, GBLUP – MD and GBLUP – HD) were presented  
254 as average values over all 100 replicates.

255

## 256 **2.6 Bayesian-based GWAS**

257 A Bayesian variable selection model with a Bayes  $C\pi$  approach (Habier et al., 2011) was  
258 used to perform GWAS to locate QTL regions and estimate the proportions of genetic variance  
259 explained by the identified QTLs. In this model, only a certain proportion of SNPs ( $\pi$ ) are  
260 assumed to have a non-zero effect on the phenotype. The marker effects are estimated

261 through the Markov Chain Monte Carlo (MCMC) algorithm that considers a mixture of  
 262 markers, of which proportion  $\pi$  has effects that follow a normal distribution  $N(0, \sigma_a^2)$  and  
 263 proportion  $1 - \pi$  have zero effect. The following general model was used:

$$264 \quad TLE_{ijl} = \mu + trial_i + dam_j + \sum_{k=1}^n \delta_{lm} a_k g_{kl} + \varepsilon_{ijlm}$$

265 where  $TLE_{ijl}$  is the time to loss of equilibrium of individual  $l$ ,  $\mu$  is the overall mean for hypoxia  
 266 tolerance trait in minutes,  $trial$  is the fixed effect of challenge trial ( $i = 1 - 7$ ),  $dam$  is the random  
 267 maternal effect ( $j = 1 - 90$ ),  $n$  is the total number of SNPs used in the analysis,  $a_k$  is the additive  
 268 effect of the reference allele for the  $k^{\text{th}}$  SNP, with genotype  $g_{kl}$  (coded as 0, 1, or 2) for  
 269 individual  $l$ , and  $\varepsilon_{ijlm}$  is the residual term for the  $l$ th individual in the  $m^{\text{th}}$  cycle of the MCMC  
 270 algorithm.

271 At each cycle  $m$ , the decision to include SNP  $k$  in the model depended on the indicator  
 272 variable  $\delta_{lm}$ . This indicator variable was sampled from a binomial distribution with a  
 273 probability  $\pi$  that  $\delta_{lm}$  was equal to 1 (i.e., the SNP has a non-zero effect  $a_k$ ) and a probability  
 274  $(1 - \pi)$  that  $\delta_{lm}$  was equal to 0 (i.e., the SNP  $k$  is thus not accounted for in the model fit at cycle  
 275  $m$ ). The proportion  $(1 - \pi)$  was sampled from a beta distribution,  $B(\alpha, \beta)$ , in which parameter  
 276  $\alpha$  was set as the total number of markers ( $n = 418,925$  – HD;  $n = 29,091$  – MD) and  $\beta$  was set  
 277 at 400, corresponding to a model retaining about 400 SNPs with non-zero effects at each cycle.

278 The BESSiE software (version 1.0) was used to compute this model (Boerner and Tier,  
 279 2016). A total of 410,000 cycles of Gibbs sampling were performed with a burn-in of 10,000  
 280 iterations using HD imputed genotypes. One Gibbs sample was saved every 40 iterations for  
 281 further analysis. For MD analysis, a total of 210,000 cycles of Gibbs sampling were performed  
 282 with a burn-in of 5,000 iterations. One Gibbs sample was saved every 20 iterations for further  
 283 analysis. To check the convergence, the MCMC algorithm was initiated three times with three

284 different seeds for the random number generator. In addition to checking the consistency of  
285 peak SNPs identified across seeds, convergence was assessed by visual inspection of plots of  
286 the posterior density of genetic, maternal and residual variances and by high correlations ( $r$   
287  $>0.99$ ) between the GEBVs estimated from the different seeds of the MCMC algorithm.

288 The evidence for association between the phenotype and each SNP was assessed by  
289 the Bayes Factor (BF):

$$290 \quad \text{BF} = \frac{P_i/(1 - P_i)}{\pi/(1 - \pi)}$$

291 where  $P_i$  is the probability that the  $i^{\text{th}}$  SNP has a non-zero effect in the model.

292 The existence of QTL was evaluated considering the values (hereafter named logBF) of  
293 twice the natural logarithm of BF for a peak SNP above the threshold value of 6 as proposed  
294 by Michenet et al. (2016). All SNPs close to the peak SNP and having a logBF greater than 2.5  
295 were considered in the QTL region. The algorithm started at the peak SNP for each  
296 chromosome, followed by a sliding window of 150 kb on both sides of the peak SNP. A sliding  
297 window was applied until no SNP had a logBF greater than 2.5 in the current window, defining  
298 the borders of our QTL region, i.e., the credibility interval for the location of the causal  
299 mutation. In addition, we considered the putative existence of a QTL if  $5 \leq \log\text{BF} < 6$  for a peak  
300 SNP and the corresponding QTL region explained at least 0.1% of the total genetic variance.

301 The genetic variation explained by a QTL region was calculated by the explained  
302 variance of each SNP in QTL region as:

$$303 \quad \sigma_{QTL}^2 = \sum_i 2p_i(1 - p_i)a_i^2$$

304 With  $p_i$  the MAF of the SNP $_i$  and  $a_i$  the effect of the SNP located in the QTL region.

305 All annotated genes within each QTL region are listed in Supplementary Table S1 based  
306 on the NCBI annotation released for *Oncorhynchus mykiss* Arlee genome reference assembly

307 USDA\_OmykA\_1.1. (GCA\_013265735.3) (Gao et al., 2021). The potential candidate genes that  
308 were meaningfully associated with acute hypoxia were subjected to further discussion.

## 309 **2.7 Comparison of TLE performances across different genotypes**

310 Homozygote standardized TLE difference was derived as the performance difference  
311 between the two homozygous genotypes AA = genotype coded as 0, and BB = genotype coded  
312 as 2) expressed in % of genetic standard deviation ( $\sigma_a$ ):  $(BB - AA) / \sigma_a$ . Standardized TLE  
313 dominance effect was calculated as the difference between the performance of the  
314 heterozygote genotype (AB = genotype coded as 1) and the average performance of the two  
315 homozygous genotypes expressed in % of genetic standard deviation:  $[(AB - (AA + BB)/2)] / \sigma_a$ .

316

## 317 **3. Results**

### 318 **3.1 Hypoxia challenge**

319 The mean weight of 1,320 challenged fish was  $50.8 \pm 11.8$  g, and the average time to  
320 loss of equilibrium (TLE) was  $98.9 \pm 71.8$  min. (Table 1). The cumulative time to loss of  
321 equilibrium within each trial is shown in Figure 1. The initial level of dissolved oxygen (DO)  
322 started from 8.7 to 9.2 mg/L<sup>-1</sup> and was gradually reduced to 2 – 3 mg/L<sup>-1</sup> two hours after the  
323 beginning of the challenge (Table 1, Supplementary Fig. S1). Then, after 3 hours of challenge,  
324 the DO was kept constant (1 – 2 mg/L<sup>-1</sup>) until the end of a given trial. The average level (across  
325 all trials) of DO when the first fish lost its equilibrium was 3.5 mg/L<sup>-1</sup>, 50% of fish at 1.7 mg/L<sup>-1</sup>  
326 and the last fish at 1.3 mg/L<sup>-1</sup> (Table 1).

327

### 328 **3.2 Parentage assignment**

329 Of 1,320 genotyped fish, 1,314 (99.5%) were uniquely assigned to a single parental pair  
330 using pedigrees. The number of progeny per sire varied from 3 to 27, with an average of 13,

331 and from 1 to 32 per dam, with an average of 14. Assigned fish (single parental pair) belonged  
332 to 646 full-sib families.

### 333 **3.3 Variance components**

334 The estimates of variance components for TLE using different models and density of  
335 genotypes are shown in Table 2. The highest heritability estimate was based on pedigree  
336 (0.28). Genomic heritability estimates (0.22 – 0.24) were slightly lower than the pedigree  
337 estimate. Maternal effects explained a modest but significant part of phenotypic variance  
338 across all studied models (0.09 – 0.10).

339

### 340 **3.4 Efficiency of genomic selection**

341 The criteria for assessing the efficiency of the selection (pedigree or genomic) are  
342 summarized in Table 3 (accuracy ( $r$ ) and bias ( $b$ )).

343 Inflation coefficients were statistically not indistinguishable from 1, while slightly lower  
344 for GBLUP than for BLUP EBVs, so (G)EBVs were not biased. On average, GBLUP accuracy was  
345 similar for MD and HD genotypes (0.65) and was 11% higher than BLUP accuracy (0.59).  
346 However, of the 100 replicates of the validation and training populations, a gain in accuracy  
347 between GBLUP and BLUP was observed for only 82 replicates (Figure 2).

348

### 349 **3.5 QTLs for acute hypoxia tolerance**

350 In summary, GWAS detected three significant and two putative QTLs using HD imputed  
351 genotypes (Figure 3) with the statistical characteristics described in Table 4. However, none  
352 of them explained over 1% of genetic variation. Hence, the genetic architecture of hypoxia  
353 tolerance seems to be highly polygenic.



354 The main QTL was named Omy31\_1. It explained 0.8% of the genetic variance and  
355 spanned the region from 19.93 to 21.56 Mb on the chromosome Omy31. This was the only  
356 significant QTL that could be identified using MD genotypes (results not presented). All genes  
357 annotated within the QTL Omy31\_1 are graphically visualized in Figure 4. The peak SNP for  
358 this QTL was located at 20.876 Mb in the intergenic region between *aff2* (AF4/FMR2 family  
359 member 2) and *ids* (iduronate 2-sulfatase isoform X1 – X2) genes. The Bayesian analysis  
360 identified the same peak SNP position of the QTL using two different seeds.

361 A second QTL region was found on the same chromosome (Omy31\_2), spanning from  
362 33.43 to 33.97 Mb and explaining only 0.3% of the genetic variance. The peak SNP for this QTL  
363 was located at 33.812 Mb, in the *smu1* gene (WD40 repeat-containing protein SMU1).

364 The third significant QTL explained only 0.2% of the genetic variance and spanned the  
365 region from 22.62 to 22.99 Mb on Omy20. Its SNP was located on 22.880 Mb in the gene *noxo1*  
366 (NADPH oxidase organizer 1).

367 Two other putative QTLs were also identified for hypoxia tolerance. The first one was  
368 on Omy15, spanned the region from 17.58 to 18.32 Mb, and explained 0.2% of the genetic  
369 variance. The peak SNP for this QTL was located at 17.668 Mb in the *bcl2a* (apoptosis regulator  
370 Bcl-2) gene. The second putative QTL was located on Omy28 between 14.92 and 15.09 Mb  
371 and also explained 0.2% of the genetic variance. The peak SNP for this QTL was located close  
372 to the *mylk3* (myosin light chain kinase 3) gene.

373

### 374 **3.6 Comparison of TLE performances across different genotypes**

375 Homozygote standardized TLE difference and standardized TLE dominance effect were  
376 calculated for the peak SNPs of all significant and putative QTLs (Table 4). Statistical

377 differences ( $p < 0.05$ ) in the TLE performance for genotypes at all identified QTLs are listed in  
378 Supplementary Table S2.

379 The performance difference between the two homozygous genotypes ranged from  
380 56.5% to 118.5% of genetic standard deviation in favour of the homozygotes BB (coded 2 in  
381 the GWAS). The TLE performance comparisons for genotypes at QTLs on Omy31\_1 and Omy28  
382 are illustrated in Figure 5. For Affx-569844518 (peak SNP of QTL Omy31\_1), the TLE  
383 performance of the homozygote (GG) was better ( $128.6 \pm 91.4$  min.) in comparison to the  
384 homozygote (AA) ( $89.6 \pm 65.7$  min.) and heterozygote ( $112.3 \pm 77.3$  min.) (Figure 5A). For Affx-  
385 1237683396 (peak SNP of QTL Omy28), the TLE performance of the homozygote (CC) was  
386 higher ( $120.9 \pm 81.3$  min.) compared to the homozygote (AA) ( $93.5 \pm 69.5$  min.) and  
387 heterozygote ( $91.2 \pm 65.2$  min.) (Figure 5B). A complete dominance effect of the unfavourable  
388 allele (A) was observed at Affx-1237683396 (QTL on Omy28) as the performances of the  
389 heterozygous individuals were similar to those of the (AA) homozygotes (Supplementary Table  
390 S2). A full additive QTL with codominance effects of the two alleles was observed on Omy20  
391 (the dominance effect was estimated close to zero, 1.7%).

392

#### 393 **4. Discussion**

394 Besides major diseases such as flavobacteriosis, IPN or SHV (Yáñez et al., 2014; Fraslin  
395 et al., 2020b), hypoxia is one of the most critical threats to rainbow trout aquaculture  
396 production (Van Raaij et al., 1996; Hou et al., 2020). Yet, a limited number of studies dealing  
397 with the genetic basis of hypoxia tolerance have been conducted on salmonid species (Anttila  
398 et al., 2013; Scott et al., 2015; Borey et al., 2018; Lagarde et al., Submitted), and genetic  
399 determinism of this trait is still unknown. Hence, we aimed to estimate the genetic

400 architecture of acute hypoxia tolerance by estimating this heritability of this trait and its  
401 potential for genomic selection and by proposing potential candidate genes for the main QTLs.

402 A new high-density (HD) 665K SNP array (Bernard et al., 2022) was used for the first  
403 time for GWAS on a rainbow trout population. This HD chip was used for the imputation step  
404 using HD genotypes of parents to get imputed HD genotypes of challenged offspring.  
405 Genotype imputation of challenged fish from MD (medium-density) to HD showed a high  
406 potential for identifying new QTLs and refining positions of QTL identified on MD genotypes.

407

#### 408 **4.1 Estimation of the variance components of hypoxia tolerance: potential for selective** 409 **breeding**

410 We found a maternal effect explaining a significant part of the phenotypic variance of  
411 hypoxia tolerance (Table 2.), mostly typical for growth at early stages in salmonids (Blanc,  
412 2002; Fishback et al., 2002; Haffray et al., 2012). One explanation might be the  
413 transgenerational epigenetics effect of dams (Wolf and Wade, 2009). Accordingly, Ho and  
414 Burggren (2012) demonstrated that parental hypoxic exposure in adult zebrafish (*Danio rerio*)  
415 has profound epigenetic effects on the morphological and physiological phenotype of their  
416 offspring. However, as the females and their previous generations were reared in the same  
417 conditions, additional investigations are needed to identify the rearing factors that may have  
418 indirectly induced this potential epigenetic effect. Another hypothesis could be based on  
419 different hypoxia-tolerant mitochondrial genomes as was reported in mitochondria  
420 differences of cerebral cortex cells of tolerant- and susceptible-to-hypoxia rats (Dudchenko et  
421 al., 1993; Luk'yanova et al., 1995). However, in fish, no data are to date available concerning  
422 the involvement of mitochondria in O<sub>2</sub> sensing, and thus this hypothesis could be of interest  
423 for further research investigation (Gattuso et al., 2018).

424           Regardless of the evaluation model applied, we estimated moderate heritability of TLE,  
425 with similar values estimated through MD or HD genomic relationship matrices, as previously  
426 observed in rainbow trout by Fraslin et al. (2020a). To our knowledge, there were only two  
427 estimates in the literature to date showing high heritability for hypoxia tolerance (0.50 and  
428 0.61, respectively) in common carp (Nagy et al., 1980) and large yellow croaker (Ding et al.,  
429 2022). Furthermore, previous studies dealing with acute hypoxia challenges showed  
430 significant inter-family differences in Atlantic salmon (Anttila et al., 2013) or catfish (Wang et  
431 al., 2017; Zhong et al., 2017), as well as between-isogenic lines differences (Borey et al., 2018;  
432 Lagarde et al., Submitted) and between-strains differences (Scott et al., 2015; Strowbridge et  
433 al., 2021) in rainbow trout. So, there is substantial evidence for standing additive genetic  
434 variation in hypoxia tolerance in fishes, which might be exploited by genetic selection.  
435 However, genetic correlations between hypoxia tolerance and other production traits (e.g.,  
436 harvest weight, slaughter yields, muscle fat etc.) need to be further assessed before  
437 integrating hypoxia tolerance in a future breeding program of rainbow trout.

438

#### 439 **4.2 Genomic prediction for hypoxia tolerance: increased accuracy compared to pedigree-** 440 **based selection**

441           Genomic selection (GS) is currently implemented in several aquaculture breeding  
442 programs (Boudry et al., 2021) and has been shown to improve selection accuracy, thus  
443 leading to higher genetic gains (Robledo et al., 2017). Besides, in fish species, GS is especially  
444 interesting for traits that cannot be measured directly on selection candidates (R2D2  
445 Consortium et al., 2021), for example, disease resistance and meat quality, but also for traits  
446 associated with environmental changes like salinity, thermal or hypoxia tolerance (Gjedrem  
447 and Rye, 2016; Robledo et al., 2017; Houston et al., 2020).

448 Our results showed that genomic prediction for TLE would lead to a relative increase  
449 of ~ 11% for GS accuracy compared to the pedigree-based selection, considering a reference  
450 population of 1000 individuals. However, only a few studies investigated selection accuracy  
451 for environmental stress-related traits such as heat tolerance in Pacific abalone (*Haliotis discus*  
452 *hannai*) (Liu et al., 2022) and salinity tolerance in eastern oyster (*Crassostrea virginica*)  
453 (McCarty et al., 2022) or growth-related traits under chronic thermal stress in rainbow trout  
454 (Yoshida and Yáñez, 2022). Still, both genomic prediction accuracies and relative increase in  
455 accuracy in comparison to PBLUP were in a similar range as in this study. Genomic selection  
456 may thus significantly improve tolerance to hypoxia and other environmental stress-related  
457 traits, supported by the advantage of phenotyping only the reference population, but the  
458 relative expected gains need to be balanced with the operational costs of genotyping  
459 (Sonesson and Meuwissen, 2009).

460 Even though genotyping costs are gradually decrease yearly, dense SNP arrays are still  
461 expensive. Genotype imputation to generate high-density markers is thus a cost-effective  
462 method. In general, the accuracy of genomic prediction is highly associated with the genotype  
463 density used, which means that increasing marker densities should generate higher accuracies  
464 (Tsai et al., 2016; Correa et al., 2017; Yoshida et al., 2019). However, we observed no increase  
465 in prediction accuracy using HD-imputed genotypes compared to MD genotypes. It illustrated  
466 that high-density (imputed) markers might not be necessary for genomic prediction as  
467 previously reported in pigs (Song et al., 2019), cattle (Gunia et al., 2014; Van Binsbergen et al.,  
468 2015), and salmonids (Yoshida et al., 2018a; Yoshida et al., 2018b). Accordingly, recent  
469 genomic studies are widely focused on optimising low-density SNP panels to get strong  
470 predictive accuracy still outperforming BLUP (Vallejo et al., 2017; Vallejo et al., 2018; Kriaridou  
471 et al., 2020; Griot et al., 2021).

472

473 **4.3 Genetic architecture of hypoxia tolerance: a highly polygenic trait and identification of**  
474 **potential candidate genes**

475 As far as we know, this is the first study that identified significant QTLs associated with  
476 tolerance to hypoxia in rainbow trout. We found three significant QTLs (one on Omy20 and  
477 two on Omy31) and two putative QTLs (on Omy15 and Omy28), but they explained each only  
478 a limited proportion of genetic variation (0.2% – 0.8%). It suggests a highly polygenic nature  
479 of hypoxia tolerance controlled by many loci of small effect (Houston et al., 2020). In previous  
480 studies, multiple significant and suggestive SNPs associated with hypoxia tolerance explained  
481 between 4.22 – 12.44% (four linkage groups – LGs) of phenotypic variation in catfish strains  
482 and hybrids (Zhong et al., 2017; Wang et al., 2017), 6.6 – 14.7% (five LGs) in Nile tilapia (Li et  
483 al., 2017), up to 32% (four LGs) in golden pompano (San et al., 2021) and 5.5 – 18% (four LGs)  
484 in large yellow croaker (Ding et al., 2022). Thus, identified SNPs had a significantly stronger  
485 effect on hypoxia tolerance than in our study. However, those studies were focused on a  
486 limited number of tolerant/sensitive fish from a few number of different families used for  
487 GWAS, which may explain this difference – 208 fish (Zhong et al., 2017), 376 fish (Wang et al.,  
488 2017), 45 fish (Li et al. 2017), 100 fish (San et al., 2021) and 398 fish (Ding et al., 2022) in  
489 contrast to our study population – 1,320 fish from hundreds of families. Consequently,  
490 comparing these studies might be difficult and partially irrelevant. In any case, all studies  
491 reinforce the evidence of the polygenic architecture of hypoxia tolerance regardless of the  
492 studied species and populations, statistical models or marker densities. However, HD imputed  
493 genotypes significantly refined our QTL regions by filling the gaps of missing genotypes on the  
494 MD SNP panel. This is consistent with previous studies that also led to refining QTL and  
495 candidate genes associated with various performance traits of dairy cattle (Höglund et al.,

496 2014; Wu et al., 2015; Nayeri et al., 2016), pigs (Yan et al., 2017; Xu et al., 2019) and fishes  
497 (Palaiokostas et al., 2018; Fraslin et al., 2020a; Yoshida and Yáñez, 2021) using imputed HD  
498 genotypes or sequence data.

499         Although each QTL does not explain a large part of the genetic variance at the  
500 population level, significant phenotypic differences between individuals of different  
501 genotypes at peak SNPs across all significant and putative QTLs were observed. The  
502 association of favourable homozygous genotypes significantly prolonged TLE in the range of  
503 17% to 30% (Supplementary Table S2). Thus, these SNPs might be used in marker-assisted  
504 selection for producing juveniles with tolerance for hypoxia in rainbow trout. Similarly,  
505 significant individual differences among different genotypes of the main SNPs associated with  
506 hypoxia tolerance were previously observed in golden pompano (San et al., 2021) and large  
507 yellow croaker (Ding et al., 2022).

508         Within the list of 113 genes annotated in our 5 identified QTL regions (see  
509 Supplementary Table S1), we proposed sixteen functional candidate genes with a meaningful  
510 biological association to hypoxia tolerance (*ids*, *fmr1*, *arx*, *lonrf3*, *commd5*, *map4k4*, *smu1*,  
511 *b4galt1*, *re1*, *abca1*, *noa1*, *igfbp7*, *noxo1*, *bcl2a*, *trim25*, *mylk3*). The potential candidate genes  
512 were firstly assessed with the database of Fish Hypoxia Responsive Genes (HRGFish) (Rashid  
513 et al., 2017). Furthermore, we also focused on the functional information given in the Mouse  
514 Genome Informatics (MGI) resource studied phenotypes in mutant mice (Eppig, 2017) or the  
515 zebrafish database (ZFIN) related to phenotypes in mutant zebrafish (Sprague et al., 2006).

516         The peak SNP of the main QTL (Omy31\_1) was located in the intergenic region between  
517 *aff2* (AF4/FMR2 family member 2) and *ids* (iduronate 2-sulfatase) genes. Interestingly, a  
518 mutation of the *ids* gene in a zebrafish line (*ids*<sup>ia200/ia200</sup>) led to a significantly larger liver  
519 (Bellesso et al., 2018). The liver is a significant source of glycogen which is the first mobilized

520 energy reserve during acute hypoxia leading to the glycogenolysis process and immediate  
521 increase of plasma glucose and glycogen, further increasing blood lactate concentration  
522 (Omlin and Weber, 2010; Li et al., 2018; Abdel-Tawwab et al., 2019; Léger et al., 2021).  
523 Therefore, liver size and glycogen level play a significant role in fish survival cope with acute  
524 hypoxia (Li et al., 2018), physiological stress response (Crespel et al., 2011) and energy  
525 mobilization during starvation (Crespel et al., 2013; Zhao et al., 2021). Similarly, male knockout  
526 mice (IdS-KO, ID MGI:259752) phenotypes for the *ids* gene exhibited significantly larger liver,  
527 spleen, and lungs and were hypoactive compared to wild mice (Garcia et al., 2007). This agrees  
528 with typical behaviour under hypoxia when some fish species remain static at the tank bottom  
529 to save their energy for facing the hypoxic condition (Abdel-Tawwab et al., 2019). Therefore,  
530 the *ids* gene might be suggested as a potential functional candidate. Besides, the *fmr1* gene  
531 (synaptic functional regulator FMR1) was annotated in the close vicinity of the peak SNP. The  
532 *fmr1* zebrafish mutant lines showed hyperactivity and abnormal swimming behaviour (Hu et  
533 al., 2020) and decreased startle reflex in case of loss of function of *fmr1* in mice (Chen and  
534 Toth, 2001; Pietropaolo et al., 2011; Rossignol et al., 2014). Abnormal behaviour under  
535 hypoxia conditions has been previously observed in fishes manifesting as rapid swimming in a  
536 circular motion with a wide mouth gape (Domenici et al., 2013; Bowyer et al., 2014) or as  
537 decreased startle reflex known as antipredator behaviour (Domenici et al., 2007). Moreover,  
538 *arx* gene (homeobox protein ARX-like) might also be proposed as an interesting functional  
539 candidate belonging to this QTL. Loss of function of this gene may cause hypoglycemia in mice  
540 (Collombat et al., 2003) and thus may block the natural response to acute hypoxia by upsetting  
541 the metabolism of carbohydrates (Polakof et al., 2012; Abdel-Tawwab et al., 2019). Let us also  
542 mention genes *lonrf3* (LON peptidase N-terminal domain and RING finger protein 3), *commd5*  
543 (COMM Domain-Containing Protein 5), and *fam199x* (protein FAM199X), which are in our QTL



544 region as well as in the vicinity of the main SNP associated to hypoxia tolerance in golden  
545 pompano (San et al., 2021). Interestingly, mouse mutant genotype hm1 of *lonrf3* (Dickinson  
546 et al., 2016) and *commd5* gene in rats (Matsuda et al., 2014) are linked to calcium influx, which  
547 is strongly modified under hypoxia conditions and inhibits the expression of myoglobin  
548 (Kanatous et al., 2009). Myoglobin is an oxygen-binding hemoprotein that is widely thought  
549 to be expressed exclusively in oxidative skeletal and cardiac myocytes, where it plays a crucial  
550 role in coping with both acute and chronic hypoxia by the supply of oxygen to the fish heart  
551 (Rashid et al., 2017), as previously reported, for example, in the Tibetan Plateau fish  
552 (*Glyptosternum maculatum*) (Qi et al., 2018), Japanese medaka (*Oryzias latipes*) (Wawrowski  
553 et al., 2011) and zebrafish (Jaspers et al., 2014). Besides, MAPK (mitogen-activated protein  
554 kinases) signalling pathway have been shown to be involved in low oxygen tolerance in fishes  
555 (Wang et al., 2017; Tian et al., 2019; Yu et al., 2021). Thus, the candidate gene *map4k4*  
556 (mitogen-activated protein kinase 4) may also play an important role in response to hypoxia  
557 challenges.

558         The second significant QTL was also detected on chromosome 31 (Omy31\_2) between  
559 33.43 Mb and 33.97 Mb, with the peak SNP positioned within the *smu1* gene, a member of  
560 the WD40-repeat protein family. Loss of *smu1* function leads to multiple cellular defects,  
561 including chromosomal instability, aberrant DNA replication and alternative RNA splicing  
562 events. Accordingly, the DNA damage is also closely linked to hypoxia, as was previously  
563 described in common carp and Nile tilapia (Poon et al., 2007; Mahfouz et al., 2015; Mustafa  
564 et al., 2015). Besides, the *smu1* gene in zebrafish was upregulated under acute hypoxia  
565 (Ragsdale et al., 2020). Thus, the *smu1* gene might be proposed as an interesting candidate  
566 gene. We also identified three other potential candidate genes linked to hypoxia: *b4galt1*, *re1*  
567 and *abca1*. *B4galt1* (beta-1,4-galactosyltransferase 1) and its exclusive function in the Golgi

568 compartment is a prerequisite for full catalytic activity. In contrast, a lack of this function is  
569 associated with cancers and intracellular hypoxia in human (Hassinen et al., 2019; Khoder-  
570 Agha et al., 2019). *Re1* was responsible for regulating approximately 20% of the hypoxia-  
571 repressed genes in human embryonic kidney cells. Hence, this gene was proposed as a key  
572 mediator of gene repression in hypoxia (Liang et al., 2014; Cavadas et al., 2016). *Abca1*  
573 (Phospholipid-Transporting ATPase ABCA1) is closely linked with hypoxia inducible factor  $\alpha$   
574 (HIF- $\alpha$ ) which increases *abca1* promoter activity. Thus, an expression of *abca1* might further  
575 stimulate the regulation of other hypoxia-inducible genes (Zhu et al., 2013; Pelster and Egg,  
576 2018; Dzhililova and Makarova, 2020). Moreover, not far from the peak SNP, an interesting  
577 gene *noa1* encodes nitric oxide-associated protein 1. It is involved in regulating mitochondrial  
578 protein translation and respiration, thereby controlling mitochondrial metabolism under  
579 acute and long-term hypoxia as previously described in goldfish (*Carassius auratus*) (Farhat et  
580 al., 2021), mice (Heidler et al., 2011) and humans (Tang et al., 2009). Mitochondria play a  
581 crucial role in coordinating responses to low oxygen because of their ability to detect  
582 physiological changes and supply the body with ATP through oxidative phosphorylation  
583 (OXPHOS) (Thomas et al., 2019). Moreover, the nitric oxide synthase (NOS) expression was  
584 observed to enhance the expression of HIF-1 $\alpha$  that activates several pivotal genes (Zhu et al.,  
585 2013; Dzhililova and Makarova, 2020) linked to hypoxia tolerance in goldfish and crucian carp  
586 (*Carassius carassius*). We thus hypothesize that *noa1* gene may have a complex role in  
587 rainbow trout response to hypoxia. In addition, the *igfbp7* gene (insulin-like growth factor-  
588 binding protein 7 precursor) might also be considered a potential candidate gene. Genes  
589 belonging to IGFs group are hypoxia-inducible genes involved in response to hypoxia and may  
590 have an important impact on fish growth during hypoxia conditions (Kajimura et al., 2006;  
591 Olsvik et al., 2013; Rashid et al., 2017; Hou et al., 2020; Aksakal and Ekinci, 2021). Moreover,

592 the *igfbp7* gene may inhibit the stimulatory effect of vascular endothelial growth factor (VEGF)  
593 (Tamura et al., 2014; Ma et al., 2016), a key regulator of angiogenesis, hematopoiesis and  
594 vascular density under low-oxygen conditions that activate genes involved in a series of  
595 responses to hypoxia (Rehn et al., 2011; Dzhililova and Makarova, 2020). In line with our  
596 finding, Yu et al. (2021) annotated a candidate gene (*igf1ra*) belonging to the IGFs group in a  
597 genome-wide association analysis of adaptation to long-term oxygen stress in Nile tilapia.

598         Within the two main QTLs on Omy31, seven and five genes are thus very convincing  
599 functional candidates whose interplay may lead to behavioural, morphological, and  
600 physiological responses to acute hypoxia stress. The response to acute hypoxia in rainbow  
601 trout could then be explained through the supergene concept (Schwander et al., 2014).  
602 Supergenes are tight clusters of multiple neighbouring genes inherited together because of  
603 close genetic linkage, each affecting a different developmental, physiological or behavioural  
604 characteristic. In combination, they provide integrated control of complex adaptive  
605 phenotypes segregating within species (Schwander et al., 2014). Recently, a supergene was  
606 identified in rainbow trout as a 55-Mb double-inversion region on Omy5 that mediates sex-  
607 specific migratory tendency (Pearse et al., 2019). Here we suggest that two supergenes on  
608 Omy31 may explain such enrichment of the QTL regions for involved hypoxia-responsive  
609 genes.

610         On Omy20, the peak SNP was located in the intergenic region of the *noxo1* gene  
611 (NADPH oxidase organizer 1), which is associated with NADPH oxidase — the enzyme  
612 responsible for the production of reactive oxygen species (ROS) (Fu et al., 2000; Sahoo et al.,  
613 2016). ROS can lead to oxidative stress during periods of environmental stress for fish  
614 (Wischhusen et al., 2020). However, ROS are usually balanced by an antioxidant (redox)  
615 defence and are considered a potent bactericide that actively destroys invading pathogens

616 (Birnie-Gauvin et al., 2017; Chowdhury and Saikia, 2020). But acute hypoxia commonly results  
617 in a disbalance between the production of oxidant and antioxidant components (oxidative  
618 stress) and thus significantly damages cell structures and DNA. In addition, hypoxia-tolerant  
619 fish appear to have an anticipatory response during low-oxygen availability by enhancing their  
620 ability to eliminate ROS production upon return to normoxia (Birnie-Gauvin et al., 2017; Qi et  
621 al., 2020). Hence, the *nox01* gene seems to be a promising functional candidate, playing an  
622 essential role in oxidative response pathways.

623         The peak SNP of the putative QTL on Omy15 was located within the gene *bcl2a*  
624 (apoptosis regulator Bcl-2). Bcl-2 proteins are a family of regulatory proteins that regulate cell  
625 apoptosis by either inducing or repressing cell death and are involved in several biological  
626 processes related to immune responses (Kratz et al., 2006). *Bcl-2a* gene was repressed after  
627 acute hypoxia stress in zebrafish (Cai et al., 2018) and channel catfish (Yuan et al., 2016),  
628 indicating that this gene is potentially involved in the hypoxia response by reducing cell  
629 apoptosis or autophagy, and could be considered a potential candidate gene. We may also  
630 propose *trim25* as a convincing candidate gene because the peak SNP is located just before  
631 this E3 ubiquitin/ISG15 ligase protein, with a major role in the initiation of intracellular  
632 antiviral response to herpesviruses (Gack et al., 2008). It is well known that hypoxia has  
633 substantial effects on fish's physiological and immune responses, which can depress their  
634 immune system leading to greater susceptibility to diseases (Giomi et al., 2016; Esteve et al.,  
635 2017; Sae-Lim et al., 2017; Abdel-Tawwab et al., 2019). Hence, loss of function of this gene  
636 may block an appropriate antiviral response as previously observed in common carp  
637 susceptible to koi herpesvirus (Palaiokostas et al., 2018).

638         Concerning the second putative QTL on Omy28, the candidate gene *mylk3* (myosin  
639 light chain kinase 3) was found close to the peak SNP. MGI mutant phenotype for the *mylk3*

640 gene was associated with several heart-related changes such as the increased response of the  
641 heart to induce stress, abnormal heart morphology, cardiac hypertrophy and necrosis (Ding  
642 et al., 2010; Bischof and Krishnan, 2016; Yuan et al., 2016) or influenced smooth blood  
643 pressure and angiogenesis in humans (Gordeuk et al., 2012). We thus hypothesise that acute  
644 hypoxia stress might lead to severe heart and blood changes causing limited oxygen transport  
645 to tissues and organs (Gattuso et al., 2018; Abdel-Tawwab et al., 2019).

646

## 647 **5 Conclusions**

648 In the present study, a newly designed HD chip was used to impute MD to HD markers  
649 in fish exposed to acute hypoxia. The GWAS showed clearly refined QTL regions and SNP peak  
650 position using HD-imputed markers but confirmed that hypoxia tolerance is of highly  
651 polygenic nature. Three significant and two putative QTLs explaining between 0.2% and 0.8%  
652 of the genetic variance, were identified for acute hypoxia tolerance, defined as TLE. However,  
653 moderate heritability suggests a solid potential to improve TLE in the studied rainbow trout  
654 population by genetic selection. As the variance explained by the five QTLs altogether is less  
655 than 5% of the total genetic variance, genomic selection should be promoted rather than  
656 marker-assisted selection to improve the selection efficiency of the broodstock population.  
657 However, marker-assisted selection may help to multiply broodstock producing offspring with  
658 high tolerance to hypoxia for production purposes. Finally, sixteen genes (*ids, fmr1, arx, lonrf3,*  
659 *commd5, map4k4, smu1, b4galt1, re1, abca1, noa1, igfbp7, noxo1, bcl2a, trim25, mylk3*) were  
660 proposed as potential functional candidates involved in hypoxia tolerance. Taking all proposed  
661 candidate genes within two main QTLs on Omy31 (12 out of 16 genes), we may hypothesize  
662 that the complex response to acute hypoxia in rainbow trout is encoded by two supergenes.

663 However, further functional validation of their effects may help to specify the biological  
664 mechanisms triggering a response to acute hypoxia in diploid rainbow trout.

665

666

667

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672

## 673 **References**

674 Abdel-Tawwab, M., Monier, M.N., Hoseinifar, S.H., Faggio, C., 2019. Fish response to hypoxia stress:  
675 growth, physiological, and immunological biomarkers. *Fish Physiol. Biochem.* 45, 997-1013.  
676 <http://dx.doi.org/10.1007/s10695-019-00614-9>

677

678 Aksakal, E., Ekinci, D., 2021. Effects of hypoxia and hyperoxia on growth parameters and transcription  
679 levels of growth, immune system and stress related genes in rainbow trout. *Comp. Biochem.*  
680 *Physiol. Part A Mol. Integr. Physiol.* 262, 111060. <http://dx.doi.org/10.1016/j.cbpa.2021.111060>

681

682 Allal, F., Nguyen, N.H., 2022. Genomic Selection in Aquaculture Species. in: Ahmadi, N., Bartholomé, J.  
683 (Eds.), *Genomic Prediction of Complex Traits: Methods and Protocols*. Springer US, New York, NY,  
684 pp. 469-491. [http://dx.doi.org/10.1007/978-1-0716-2205-6\\_17](http://dx.doi.org/10.1007/978-1-0716-2205-6_17)

685

686 Anttila, K., Dhillon, R.S., Boulding, E.G., Farrell, A.P., Glebe, B.D., Elliott, J.A., Wolters, W.R., Schulte,  
687 P.M., 2013. Variation in temperature tolerance among families of Atlantic salmon (*Salmo salar*) is  
688 associated with hypoxia tolerance, ventricle size and myoglobin level. *J. Exp. Biol.* 216, 1183-1190.  
689 <http://dx.doi.org/10.1242/jeb.080556>

690

691 Bellesso, S., Salvalaio, M., Lualdi, S., Tognon, E., Costa, R., Braghetta, P., Giraud, C., Stramare, R.,  
692 Rigon, L., Filocamo, M., 2018. FGF signaling deregulation is associated with early developmental  
693 skeletal defects in animal models for mucopolysaccharidosis type II (MPSII). *Hum. Mol. Genet.* 27,  
694 2262-2275. <http://dx.doi.org/10.1093/hmg/ddy131>

695

696 Bernard, M., Dehaullon, A., Gao, G., Paul, K., Lagarde, H., Charles, M., Prchal, M., Danon, J., Jaffrelo, L.,  
697 Poncet, C., Patrice, P., Haffray, P., Quillet, E., Dupont-Nivet, M., Palti, Y., Lallias, D., Phocas, F., 2022.  
698 Development of a High-Density 665 K SNP Array for Rainbow Trout Genome-Wide Genotyping.  
699 *Front. Genet.* 13, 941340. <http://dx.doi.org/10.3389/fgene.2022.941340>

700

701 Birnie-Gauvin, K., Costantini, D., Cooke, S.J., Willmore, W.G., 2017. A comparative and evolutionary  
702 approach to oxidative stress in fish: a review. *Fish Fish.* 18, 928-942.  
703 <http://dx.doi.org/10.1111/faf.12215>  
704

705 Bischof, C., Krishnan, J., 2016. Exploiting the hypoxia sensitive non-coding genome for organ-specific  
706 physiologic reprogramming. *Biochim Biophys Acta Mol Cell Res.* 1863, 1782-1790.  
707 <http://dx.doi.org/10.1016/j.bbamcr.2016.01.024>  
708

709 Blanc, J., 2002. Effects of egg size differences on juvenile weight between and within lots in rainbow  
710 trout *Oncorhynchus mykiss*. *J. World Aquacult. Soc.* 33, 278-286. [http://dx.doi.org/10.1111/j.1749-  
711 7345.2002.tb00504.x](http://dx.doi.org/10.1111/j.1749-7345.2002.tb00504.x)  
712

713 Boerner, V., Tier, B., 2016. BESSiE: a software for linear model BLUP and Bayesian MCMC analysis of  
714 large-scale genomic data. *Genet. Sel. Evol.* 48, 1-5. <http://dx.doi.org/10.1186/s12711-016-0241-x>  
715

716 Borey, M., Paroissin, C., Quillet, E., Terrier, F., Maunas, P., Burel, C., Lauga, B., 2018. Acute hypoxia  
717 reveals diverse adaptation strategies to fully substituted plant-based diet in isogenic lines of the  
718 carnivorous rainbow trout. *Aquaculture.* 490, 288-296.  
719 <http://dx.doi.org/10.1016/j.aquaculture.2018.02.005>  
720

721 Boudry, P., Allal, F., Aslam, M.L., Bargelloni, L., Bean, T.P., Brard-Fudulea, S., Briec, M.S., Calboli, F.C.,  
722 Gilbey, J., Haffray, P., 2021. Current status and potential of genomic selection to improve selective  
723 breeding in the main aquaculture species of International Council for the Exploration of the Sea  
724 (ICES) member countries. *Aquac. Rep.* 20, 100700. <http://dx.doi.org/10.1016/j.aqrep.2021.100700>  
725

726 Bowyer, J.N., Booth, M.A., Qin, J.G., D'Antignana, T., Thomson, M.J., Stone, D.A., 2014. Temperature  
727 and dissolved oxygen influence growth and digestive enzyme activities of yellowtail kingfish *Seriola*  
728 *lalandi* (Valenciennes, 1833). *Aquacult. Res.* 45, 2010-2020. <https://doi.org/10.1111/are.12146>  
729

730 Cai, X., Zhang, D., Wang, J., Liu, X., Ouyang, G., Xiao, W., 2018. Deletion of the fih gene encoding an  
731 inhibitor of hypoxia-inducible factors increases hypoxia tolerance in zebrafish. *J. Biol. Chem.* 293,  
732 15370-15380. <http://dx.doi.org/10.1074/jbc.RA118.003004>  
733

734 Cavadas, M.A., Mesnieres, M., Crifo, B., Manresa, M.C., Selfridge, A.C., Keogh, C.E., Fabian, Z., Scholz,  
735 C.C., Nolan, K.A., Rocha, L., 2016. REST is a hypoxia-responsive transcriptional repressor. *Sci. Rep.*  
736 6, 1-13. <http://dx.doi.org/10.1038/srep31355>  
737

738 Collombat, P., Mansouri, A., Hecksher-Sørensen, J., Serup, P., Krull, J., Gradwohl, G., Gruss, P., 2003.  
739 Opposing actions of Arx and Pax4 in endocrine pancreas development. *Genes Dev.* 17, 2591-2603.  
740 <http://dx.doi.org/10.1101/gad.269003>  
741

742 Correa, K., Bangera, R., Figueroa, R., Lhorente, J.P., Yáñez, J.M., 2017. The use of genomic information  
743 increases the accuracy of breeding value predictions for sea louse (*Caligus rogercresseyi*) resistance  
744 in Atlantic salmon (*Salmo salar*). *Genet. Sel. Evol.* 49, 1-5. [http://dx.doi.org/10.1186/s12711-017-  
745 0291-8](http://dx.doi.org/10.1186/s12711-017-0291-8)  
746

747 Crespel, A., Bernatchez, L., Garant, D., Audet, C., 2011. Quantitative genetic analysis of the  
748 physiological stress response in three strains of brook charr *Salvelinus fontinalis* and their hybrids.  
749 J. Fish. Biol. 79, 2019-2033. <https://doi.org/10.1111/j.1095-8649.2011.03149.x>  
750

751 Crespel, A., Bernatchez, L., Garant, D., Audet, C., 2013. Genetically based population divergence in  
752 overwintering energy mobilization in brook charr (*Salvelinus fontinalis*). *Genetica* 141, 51-64.  
753 <https://doi.org/10.1007/s10709-013-9705-x>  
754

755 D'Ambrosio, J., Morvezen, R., Brard-Fudulea, S., Bestin, A., Perez, A.A., Guéméné, D., Poncet, C.,  
756 Haffray, P., Dupont-Nivet, M., Phocas, F., 2020. Genetic architecture and genomic selection of  
757 female reproduction traits in rainbow trout. *BMC Genom.* 21, 1-14.  
758 <http://dx.doi.org/10.1186/s12864-020-06955-7>  
759

760 Diaz, R.J., 2001. Overview of hypoxia around the world. *J. Environ. Qual.* 30, 275-281.  
761 <http://dx.doi.org/10.2134/jeq2001.302275x>  
762

763 Dickinson, M.E., Flenniken, A.M., Ji, X., Teboul, L., Wong, M.D., White, J.K., Meehan, T.F., Weninger,  
764 W.J., Westerberg, H., Adissu, H., 2016. High-throughput discovery of novel developmental  
765 phenotypes. *Nature.* 537, 508-514. <http://dx.doi.org/10.1038/nature19356>  
766

767 Ding, J., Zhang, Y., Wang, J., Liu, C., Gao, X., Wu, Y., Wang, J., Wu, X., Zhu, J., Shen, W., 2022. Genome-  
768 wide association study identified candidate SNPs and genes associated with hypoxia tolerance in  
769 large yellow croaker (*Larimichthys crocea*). *Aquaculture*, 738472.  
770 <http://dx.doi.org/10.1016/j.aquaculture.2022.738472>  
771

772 Ding, P., Huang, J., Battiprolu, P.K., Hill, J.A., Kamm, K.E., Stull, J.T., 2010. Cardiac myosin light chain  
773 kinase is necessary for myosin regulatory light chain phosphorylation and cardiac performance in  
774 vivo. *J. Biol. Chem.* 285, 40819-40829. <http://dx.doi.org/10.1074/jbc.M110.160499>  
775

776 Domenici, P., Lefrançois, C., Shingles, A., 2007. Hypoxia and the antipredator behaviours of fishes.  
777 *Philos. Trans. R. Soc. Lond., B, Biol. Sci.* 362, 2105-2121. <http://dx.doi.org/10.1098/rstb.2007.2103>  
778

779 Domenici, P., Herbert, N., Lefrançois, C., Steffensen, J., McKenzie, D., 2013. The effect of hypoxia on  
780 fish swimming performance and behaviour, In: Palstra, A., Planas, J. (eds) *Swimming Physiology of*  
781 *Fish*. Springer, Berlin, Heidelberg. pp. 129-159. [https://doi.org/10.1007/978-3-642-31049-2\\_6](https://doi.org/10.1007/978-3-642-31049-2_6)  
782

783 Dudchenko, A.M., Chernobaeva, G.N., Belousova, V.V., Vlasova, I.G., Luk'yanova, L.D., 1993.  
784 Bioenergetic parameters of the brain in rats with different resistance to hypoxia. *Bull. Exp. Bio.l*  
785 *Med.* 115, 263-267.  
786 <http://dx.doi.org/10.1007/BF00836406>  
787

788 Dzhaliilova, D., Makarova, O., 2020. Differences in tolerance to hypoxia: physiological, biochemical, and  
789 molecular- biological characteristics. *Biomedicine.* 8, 428. <http://dx.doi.org/10.3390/biomedicine8100428>  
790

791

792 Eppig, J.T., 2017. Mouse genome informatics (MGI) resource: genetic, genomic, and biological  
793 knowledgebase for the laboratory mouse. *ILAR J.* 58, 17-41. <http://dx.doi.org/10.1093/ilar/ilx013>  
794



795 Esteve, C., Merchán, R., Alcaide, E., 2017. An outbreak of *Shewanella putrefaciens* group in wild eels  
796 *Anguilla anguilla* L. favoured by hypoxic aquatic environments. *J. Fish. Dis.* 40, 929-939.  
797 <https://doi.org/10.1111/jfd.12574>  
798

799 Fago, A., Jensen, F.B., 2015. Hypoxia tolerance, nitric oxide, and nitrite: lessons from extreme animals.  
800 *Physiology.* 30, 116-126. <http://dx.doi.org/10.1152/physiol.00051.2014>  
801

802 FAO, 2020. The State of World Fisheries and Aquaculture 2020 – Sustainability in Action. Rome  
803 <https://www.fao.org/3/ca9229en/ca9229en.pdf> (accessed 14 February 2022).  
804

805 Farhat, E., Cheng, H., Romestaing, C., Pamenter, M., Weber, J.-M., 2021. Goldfish Response to Chronic  
806 Hypoxia: Mitochondrial Respiration, Fuel Preference and Energy Metabolism. *Metabolites.* 11, 187.  
807 <http://dx.doi.org/10.3390/metabo11030187>  
808

809 Farrell, A.P., Richards, J.G., 2009. Chapter 11 Defining hypoxia: an integrative synthesis of the  
810 responses of fish to hypoxia, *Fish Physiol.* 27, 487-503. [https://doi.org/10.1016/S1546-](https://doi.org/10.1016/S1546-5098(08)00011-3)  
811 [5098\(08\)00011-3](https://doi.org/10.1016/S1546-5098(08)00011-3)  
812

813 Fishback, A.G., Danzmann, R.G., Ferguson, M.M., Gibson, J.P., 2002. Estimates of genetic parameters  
814 and genotype by environment interactions for growth traits of rainbow trout (*Oncorhynchus*  
815 *mykiss*) as inferred using molecular pedigrees. *Aquaculture.* 206, 137-150.  
816 [http://dx.doi.org/10.1016/S0044-8486\(01\)00707-4](http://dx.doi.org/10.1016/S0044-8486(01)00707-4)  
817

818 Fraslin, C., Phocas, F., Bestin, A., Charles, M., Bernard, M., Krieg, F., Dechamp, N., Ciobotaru, C., Hozé,  
819 C., Petitprez, F., 2020a. Genetic determinism of spontaneous masculinisation in XX female rainbow  
820 trout: new insights using medium throughput genotyping and whole-genome sequencing. *Sci. Rep.*  
821 10, 1-13. <http://dx.doi.org/10.1038/s41598-020-74757-8>  
822

823 Fraslin, C., Quillet, E., Rochat, T., Dechamp, N., Bernardet, J.-F., Collet, B., Lallias, D., Boudinot, P.,  
824 2020b. Combining multiple approaches and models to dissect the genetic architecture of resistance  
825 to infections in fish. *Front. Genet.* 11, 677. <http://dx.doi.org/10.3389/fgene.2020.00677>  
826

827 Friedrich, J., Janssen, F., Aleynik, D., Bange, H.W., Boltacheva, N., Çagatay, M., Dale, A.W., Etiope, G.,  
828 Erdem, Z., Geraga, M., 2014. Investigating hypoxia in aquatic environments: diverse approaches to  
829 addressing a complex phenomenon. *Biogeosciences.* 11, 1215-1259. [http://dx.doi.org/10.5194/bg-](http://dx.doi.org/10.5194/bg-11-1215-2014)  
830 [11-1215-2014](http://dx.doi.org/10.5194/bg-11-1215-2014)  
831

832 Fu, X.W., Wang, D., Nurse, C.A., Dinauer, M.C., Cutz, E., 2000. NADPH oxidase is an O<sub>2</sub> sensor in airway  
833 chemoreceptors: evidence from K<sup>+</sup> current modulation in wild-type and oxidase-deficient mice.  
834 *Proc. Natl. Acad. Sci.* 97, 4374-4379. <https://doi.org/10.1073/pnas.97.8.4374>  
835

836 Gack, M.U., Kirchhofer, A., Shin, Y.C., Inn, K.S., Liang, C., Cui, S., Myong, S., Ha, T., Hopfner, K.P., Jung,  
837 J.U., 2008. Roles of RIG-I N-terminal tandem CARD and splice variant in TRIM25-mediated antiviral  
838 signal transduction. *Proc. Natl. Acad. Sci.* 105, 16743-16748.  
839 <http://dx.doi.org/10.1073/pnas.0804947105>  
840

841 Gallage, S., Katagiri, T., Endo, M., Futami, K., Endo, M., Maita, M., 2016. Influence of moderate hypoxia  
842 on vaccine efficacy against *Vibrio anguillarum* in *Oreochromis niloticus* (Nile tilapia). *Fish Shellfish*  
843 *Immunol.* 51, 271-281. <http://dx.doi.org/10.1016/j.fsi.2016.02.024>  
844

845 Gao, G., Magadan, S., Waldbieser, G.C., Youngblood, R.C., Wheeler, P.A., Scheffler, B.E., Thorgaard,  
846 G.H., Palti, Y., 2021. A long reads-based de-novo assembly of the genome of the Arlee homozygous  
847 line reveals chromosomal rearrangements in rainbow trout. *G3-Genes Genom. Genet.* 11, jkab052.  
848 <http://dx.doi.org/10.1093/g3journal/jkab052>  
849

850 Garcia, A., Pan, J., Lamsa, J., Muenzer, J., 2007. The characterization of a murine model of  
851 mucopolysaccharidosis II (Hunter syndrome). *J. Inherited Metab. Dis.* 30, 924-934.  
852 <http://dx.doi.org/10.1007/s10545-007-0641-8>  
853

854 Gattuso, A., Garofalo, F., Cerra, M.C., Imbrogno, S., 2018. Hypoxia tolerance in teleosts: implications  
855 of cardiac nitrosative signals. *Front. Physiol.* 9, 366. <http://dx.doi.org/10.3389/fphys.2018.00366>  
856

857 Genz, J., Jyde, M., Svendsen, J.C., Steffensen, J.F., Ramløv, H., 2013. Excess post-hypoxic oxygen  
858 consumption is independent from lactate accumulation in two cyprinid fishes. *Comp. Biochem.*  
859 *Physiol. Part A Mol. Integr. Physiol.* 165, 54-60. <http://dx.doi.org/10.1016/j.cbpa.2013.02.002>  
860

861 Giomi, F., Rinaldi, A., Mandich, A., Fuentes, V., Mirto, S., Sara, G., Piraino, S., 2016. Concurrent  
862 environmental stressors and jellyfish stings impair caged European sea bass (*Dicentrarchus labrax*)  
863 physiological performances. <https://doi.org/10.1038/srep27929>  
864

865 Gjedrem, T., Rye, M., 2016. Selection response in fish and shellfish: a review. *Rev. Aquac.* 0, 1–12.  
866 <https://doi.org/10.1111/raq.12154>.  
867

868 Gordeuk, V.R., Zhang, X., Zhang, W., Ma, S.-F., Sable, C., Miasniakova, G., Sergueeva, A., Ammosova,  
869 T., Xu, M., Nekhai, S., 2012. Novel Putative Polymorphism in SERPINC1 Encoding Antithrombin III Is  
870 Implicated in Elevated Estimated Systolic Pulmonary Pressure in Patients with Chuvash  
871 Polycythemia. *Blood.* 120, 2869. <http://dx.doi.org/10.1182/blood.V120.21.2869.2869>  
872

873 Griot, R., Allal, F., Brard-Fudulea, S., Morvezen, R., Haffray, P., Phocas, F., Vandeputte, M., 2020. APIS:  
874 An auto-adaptive parentage inference software that tolerates missing parents. *Mol. Ecol. Resour.*  
875 20, 579-590. <http://dx.doi.org/10.1111/1755-0998.13103>  
876

877 Griot, R., Allal, F., Phocas, F., Brard-Fudulea, S., Morvezen, R., Haffray, P., François, Y., Morin, T., Bestin,  
878 A., Bruant, J.-S., 2021. Optimization of Genomic Selection to Improve Disease Resistance in Two  
879 Marine Fishes, the European Sea Bass (*Dicentrarchus labrax*) and the Gilthead Sea Bream (*Sparus*  
880 *aurata*). *Front. Genet.* 1294. <http://dx.doi.org/10.3389/fgene.2021.665920>  
881

882 Gunia, M., Saintilan, R., Venot, E., Hoze, C., Fouilloux, M.N., Phocas, F., 2014. Genomic prediction in  
883 French Charolais beef cattle using high-density single nucleotide polymorphism markers. *J. Anim.*  
884 *Sci.* 92, 3258-3269. <https://doi.org/10.2527/jas.2013-7478>  
885

886 Habier, D., Fernando, R.L., Kizilkaya, K., Garrick, D.J., 2011. Extension of the Bayesian alphabet for  
887 genomic selection. *BMC Bioinform.* 12, 1-12. <http://dx.doi.org/10.1186/1471-2105-12-186>  
888

889 Haffray, P., Vandeputte, M., Petit, V., Pincent, C., Chatain, B., Chapuis, H., Mériaux, J.-C., Coudurier, B.,  
890 Quillet, E., Dupont-Nivet, M., 2012. Minimizing maternal effect in salmonid families mixed since  
891 eyed stages and a posteriori DNA-pedigree. *Livest. Sci.* 150, 170-178.  
892 <http://dx.doi.org/10.1016/j.livsci.2012.08.017>  
893

894 Hassinen, A., Khoder-Agha, F., Khosrowabadi, E., Mennerich, D., Harrus, D., Noel, M., Dimova, E.Y.,  
895 Glumoff, T., Harduin-Lepers, A., Kietzmann, T., 2019. A Golgi-associated redox switch regulates  
896 catalytic activation and cooperative functioning of ST6Gal-I with B4GalT-I. *Redox Biol.* 24, 101182.  
897 <http://dx.doi.org/10.1016/j.redox.2019.101182>  
898

899 Heidler, J., Al-Furoukh, N., Kukat, C., Salwig, I., Ingelmann, M.-E., Seibel, P., Krüger, M., Holtz, J., Wittig,  
900 I., Braun, T., 2011. Nitric oxide-associated protein 1 (NOA1) is necessary for oxygen-dependent  
901 regulation of mitochondrial respiratory complexes. *J. Biol. Chem.* 286, 32086-32093.  
902 <http://dx.doi.org/10.1074/jbc.M111.221986>  
903

904 Ho, D.H., Burggren, W.W., 2012. Parental hypoxic exposure confers offspring hypoxia resistance in  
905 zebrafish (*Danio rerio*). *J. Exp. Biol.* 215, 4208-4216. <http://dx.doi.org/10.1242/jeb.074781>  
906

907 Höglund, J.K., Sahana, G., Brøndum, R.F., Guldbandsen, B., Buitenhuis, B., Lund, M.S., 2014. Fine  
908 mapping QTL for female fertility on BTA04 and BTA13 in dairy cattle using HD SNP and sequence  
909 data. *BMC Genom.* 15, 1-10. <http://dx.doi.org/10.1186/1471-2164-15-790>  
910

911 Hou, Z.-S., Wen, H.-S., Li, J.-F., He, F., Li, Y., Qi, X., 2020. Environmental hypoxia causes growth  
912 retardation, osteoclast differentiation and calcium dyshomeostasis in juvenile rainbow trout  
913 (*Oncorhynchus mykiss*). *Sci. Total Environ.* 705, 135272. <http://dx.doi.org/10.1016/j.scitotenv.2019.135272>  
914

915

916 Houston, R.D., Bean, T.P., Macqueen, D.J., Gundappa, M.K., Jin, Y.H., Jenkins, T.L., Selly, S.L.C., Martin,  
917 S.A., Stevens, J.R., Santos, E.M., 2020. Harnessing genomics to fast-track genetic improvement in  
918 aquaculture. *Nat. Rev. Genet.* 21, 389-409. <http://dx.doi.org/10.1038/s41576-020-0227-y>  
919

920 Hu, J., Chen, L., Yin, J., Yin, H., Huang, Y., Tian, J., 2020. Hyperactivity, memory defects, and craniofacial  
921 abnormalities in zebrafish *fmr1* mutant larvae. *Behav. Genet.*, 1-9.  
922 <http://dx.doi.org/10.1007/s10519-020-09995-7>  
923

924 Chang, C.C., Chow, C.C., Tellier, L.C., Vattikuti, S., Purcell, S.M., Lee, J.J., 2015. Second-generation  
925 PLINK: rising to the challenge of larger and richer datasets. *Gigascience.* 4, 7. <http://dx.doi.org/10.1186/s13742-015-0047-8>  
926

927

928 Chavanne, H., Janssen, K., Hofherr, J., Contini, F., Haffray, P., Komen, H., Nielsen, E.E., Bargelloni, L.,  
929 Consortium, A., 2016. A comprehensive survey on selective breeding programs and seed market in  
930 the European aquaculture fish industry. *Aquacult. Int.* 24, 1287-1307.  
931 <http://dx.doi.org/10.1007/s10499-016-9985-0>  
932

933 Chen, L., Toth, M., 2001. Fragile X mice develop sensory hyperreactivity to auditory stimuli.  
934 *Neuroscience.* 103, 1043-1050. [http://dx.doi.org/10.1016/S0306-4522\(01\)00036-7](http://dx.doi.org/10.1016/S0306-4522(01)00036-7)  
935

936 Chowdhury, S., Saikia, S., 2020. Oxidative stress in fish: a review. J. Sci. Res. 12, 145-160.  
937 <http://dx.doi.org/10.3329/jsr.v12i1.41716>  
938

939 Janssen, K., Chavanne, H., Berentsen, P., Komen, H., 2017. Impact of selective breeding on European  
940 aquaculture. Aquaculture. 472, 8-16. <https://doi.org/10.1016/j.aquaculture.2016.03.012>.  
941

942 Jaspers, R.T., Testerink, J., Della Gaspera, B., Chanoine, C., Bagowski, C.P., van der Laarse, W.J., 2014.  
943 Increased oxidative metabolism and myoglobin expression in zebrafish muscle during chronic  
944 hypoxia. Biol. Open. 3, 718-727. <http://dx.doi.org/10.1242/bio.20149167>  
945

946 Kajimura, S., Aida, K., Duan, C., 2006. Understanding hypoxia-induced gene expression in early  
947 development: in vitro and in vivo analysis of hypoxia-inducible factor 1-regulated zebra fish insulin-  
948 like growth factor binding protein 1 gene expression. Mol. Cell. Biol. 26, 1142-1155.  
949 <http://dx.doi.org/10.1128/MCB.26.3.1142-1155.2006>  
950

951 Kanatous, S.B., Mammen, P.P., Rosenberg, P.B., Martin, C.M., White, M.D., DiMaio, J.M., Huang, G.,  
952 Muallem, S., Garry, D.J., 2009. Hypoxia reprograms calcium signaling and regulates myoglobin  
953 expression. Am. J. Physiol. Cell Physiol. 296, 393-402. <http://dx.doi.org/10.1152/ajpcell.00428.2008>  
954

955 Khoder-Agha, F., Harrus, D., Brysbaert, G., Lensink, M.F., Harduin-Lepers, A., Glumoff, T., Kellokumpu,  
956 S., 2019. Assembly of B4GALT1/ST6GAL1 heteromers in the Golgi membranes involves lateral  
957 interactions via highly charged surface domains. J. Biol. Chem. 294, 14383-14393.  
958 <http://dx.doi.org/10.1074/jbc.RA119.009539>  
959

960 Kratz, E., Eimon, P., Mukhyala, K., Stern, H., Zha, J., Strasser, A., Hart, R., Ashkenazi, A., 2006. Functional  
961 characterization of the Bcl-2 gene family in the zebrafish. Cell Death Differ. 13, 1631-1640.  
962 <http://dx.doi.org/10.1038/sj.cdd.4402016>  
963

964 Kriaridou, C., Tsairidou, S., Houston, R.D., Robledo, D., 2020. Genomic prediction using low density  
965 marker panels in aquaculture: performance across species, traits, and genotyping platforms. Front.  
966 Genet. 11, 124. <http://dx.doi.org/10.3389/fgene.2020.00124>  
967

968 Lagarde, H., Prchal, M., Pouil, S., Goardon, L., Bideau, M., Guyvarc'h, F., Labbé, L., Dechamp, N., Phocas,  
969 D., Dupont-Nivet, M., Lallias, D., Submitted. Are resistances to acute hyperthermia or hypoxia stress  
970 similar and consistent between early and late ages in rainbow trout using isogenic lines?  
971

972 Léger, J.A., Athanasio, C.G., Zhera, A., Chauhan, M.F., Simmons, D.B., 2021. Hypoxic responses in  
973 *Oncorhynchus mykiss* involve angiogenesis, lipid, and lactate metabolism, which may be triggered  
974 by the cortisol stress response and epigenetic methylation. Comp. Biochem. Physiol. Part D  
975 Genomics Proteomics. 39, 100860. <http://dx.doi.org/10.1016/j.cbd.2021.100860>  
976

977 Li, H.L., Gu, X.H., Li, B.J., Chen, C.H., Lin, H.R., Xia, J.H., 2017. Genome-wide QTL analysis identified  
978 significant associations between hypoxia tolerance and mutations in the GPR132 and ABCG4 genes  
979 in Nile tilapia. Mar. Biotechnol. 19, 441-453. <http://dx.doi.org/10.1007/s10126-017-9762-8>  
980

981 Li, M., Wang, X., Qi, C., Li, E., Du, Z., Qin, J.G., Chen, L., 2018. Metabolic response of Nile tilapia  
982 (*Oreochromis niloticus*) to acute and chronic hypoxia stress. Aquaculture. 495, 187-195.  
983 <http://dx.doi.org/10.1016/j.aquaculture.2018.05.031>

984 Liang, H., Studach, L., Hullinger, R.L., Xie, J., Andrisani, O.M., 2014. Down-regulation of RE-1 silencing  
985 transcription factor (REST) in advanced prostate cancer by hypoxia-induced miR-106b~ 25. *Exp. Cell*  
986 *Res.* 320, 188-199. <http://dx.doi.org/10.1016/j.yexcr.2013.09.020>  
987

988 Liu, J., Peng, W., Yu, F., Shen, Y., Yu, W., Lu, Y., Lin, W., Zhou, M., Huang, Z., Luo, X., 2022. Genomic  
989 selection applications can improve the environmental performance of aquatics: a case study on the  
990 heat tolerance of abalone. *Evol. Appl.* 5, 992-1001 <http://dx.doi.org/10.1111/eva.13388>  
991

992 Luk'yanova, L.D., Chernobaeva, G.N., Romanova, V.E., 1995. Effects of adaptation to intermittent  
993 hypoxia on oxidative phosphorylation in brain mitochondria of rats with different sensitivities  
994 toward oxygen deficiency. *Bull. Exp. Biol. Med.* 120, 1189-1192.  
995 <http://dx.doi.org/10.1007/BF02445567>  
996

997 Ma, X., Dai, W., Kang, J., Yang, L., He, S., 2016. Comprehensive transcriptome analysis of six catfish  
998 species from an altitude gradient reveals adaptive evolution in Tibetan fishes. *G3-Genes Genom.*  
999 *Genet.* 6, 141-148. <https://doi.org/10.1534/g3.115.024448>  
1000

1001 Mahfouz, M.E., Hegazi, M.M., El-Magd, M.A., Kasem, E.A., 2015. Metabolic and molecular responses  
1002 in Nile tilapia, *Oreochromis niloticus* during short and prolonged hypoxia. *Mar. Freshwat. Behav.*  
1003 *Physiol.* 48, 319-340. <https://doi.org/10.1080/10236244.2015.1055915>  
1004

1005 Matsuda, H., Hamet, P., Tremblay, J., 2014. Hypertension-related, calcium-regulated gene  
1006 (HCaRG/COMMD5) and kidney diseases: HCaRG accelerates tubular repair. *J. Nephrol.* 27, 351-360.  
1007 <http://dx.doi.org/10.1007/s40620-014-0054-3>  
1008

1009 McBryan, T., Anttila, K., Healy, T., Schulte, P., 2013. Responses to temperature and hypoxia as  
1010 interacting stressors in fish: implications for adaptation to environmental change. *Integr. Comp.*  
1011 *Biol.* 53, 648-659. <http://dx.doi.org/10.1093/icb/ict066>  
1012

1013 McCarty, A.J., Allen Jr, S.K., Plough, L.V., 2022. Genome-wide analysis of acute low salinity tolerance in  
1014 the eastern oyster *Crassostrea virginica* and potential of genomic selection for trait improvement.  
1015 *G3-Genes Genom. Genet.* 12, jkab368. <http://dx.doi.org/10.1093/g3journal/jkab368>  
1016

1017 Michenet, A., Barbat, M., Saintilan, R., Venot, E., Phocas, F., 2016. Detection of quantitative trait loci  
1018 for maternal traits using high-density genotypes of Blonde d'Aquitaine beef cattle. *BMC Genet.* 17,  
1019 1-13. <http://dx.doi.org/10.1186/s12863-016-0397-y>  
1020

1021 Misztal, I., Tsuruta, S., Strabel, T., Auvray, B., Druet, T., Lee, D., 2002. BLUPF90 and related programs  
1022 (BGF90). 7<sup>th</sup> WCGALP 28, 743.  
1023

1024 Misztal, I., Tsuruta, S., Lourenco, Masuda, Y., Aguilar, I., Legarra, A., Vitezica, Z., 2014. Manual for  
1025 BLUPF90 family of programs., University of Georgia, Athens, USA. pp. 142. [http://nce.ads.uga.edu](http://nce.ads.uga.edu/wiki/lib/exe/fetch.php?media=blupf90_all7.pdf)  
1026 [/wiki/lib/exe/fetch.php?media=blupf90\\_all7.pdf](http://nce.ads.uga.edu/wiki/lib/exe/fetch.php?media=blupf90_all7.pdf) (accessed 4 April 2021).  
1027

1028 Mustafa, S.A., Karieb, S.S., Davies, S.J., Jha, A.N., 2015. Assessment of oxidative damage to DNA,  
1029 transcriptional expression of key genes, lipid peroxidation and histopathological changes in carp  
1030 *Cyprinus carpio* L. following exposure to chronic hypoxic and subsequent recovery in normoxic  
1031 conditions. *Mutagenesis.* 30, 107-116. <https://doi.org/10.1093/mutage/geu048>

1032 Nagy, A., Csanyi, V., Bakos, J., Horvath, L., 1980. Development of a short-term laboratory system for  
1033 the evaluation of carp growth in ponds. *Bamidgeh*. 32, 6-15.  
1034  
1035 Nayeri, S., Sargolzaei, M., Abo-Ismael, M.K., May, N., Miller, S.P., Schenkel, F., Moore, S.S., Stothard, P.,  
1036 2016. Genome-wide association for milk production and female fertility traits in Canadian dairy  
1037 Holstein cattle. *BMC Genet.* 17, 1-11. <https://doi.org/10.1186/s12863-016-0386-1>  
1038  
1039 Nikinmaa, M., Rees, B.B., 2005. Oxygen-dependent gene expression in fishes. *Am. J. Physiol. Regul.*  
1040 *Integr. Comp. Physiol.* 288, 1079-1090. <http://dx.doi.org/10.1152/ajpregu.00626.2004>  
1041  
1042 Olsvik, P.A., Vikeså, V., Lie, K.K., Hevrøy, E.M., 2013. Transcriptional responses to temperature and low  
1043 oxygen stress in Atlantic salmon studied with next-generation sequencing technology. *BMC Genom.*  
1044 14, 1-21. <https://doi.org/10.1186/1471-2164-14-817>  
1045  
1046 Omlin, T., Weber, J.-M., 2010. Hypoxia stimulates lactate disposal in rainbow trout. *J. Exp. Biol.* 213,  
1047 3802-3809. <http://dx.doi.org/10.1242/jeb.048512>  
1048  
1049 Palaikostas, C., Robledo, D., Vesely, T., Prchal, M., Pokorova, D., Piackova, V., Pojezdal, L., Kocour, M.,  
1050 Houston, R.D., 2018. Mapping and Sequencing of a Significant Quantitative Trait Locus Affecting  
1051 Resistance to Koi Herpesvirus in Common Carp. *G3-Genes Genom. Genet.* 8, 3507-3513.  
1052 <http://dx.doi.org/10.1534/g3.118.200593>  
1053  
1054 Palti, Y., Gao, G., Liu, S., Kent, M., Lien, S., Miller, M., Rexroad III, C., Moen, T., 2015. The development  
1055 and characterization of a 57 K single nucleotide polymorphism array for rainbow trout. *Mol. Ecol.*  
1056 *Resour.* 15, 662-672. <https://doi.org/10.1111/1755-0998.12337>  
1057  
1058 Pearse, D.E., Barson, N.J., Nome, T., Gao, G., Campbell, M.A., Abadía-Cardoso, A., Anderson, E.C.,  
1059 Rundio, D.E., Williams, T.H., Naish, K.A., 2019. Sex-dependent dominance maintains migration  
1060 supergene in rainbow trout. *Nat. Ecol. Evol.* 3, 1731-1742. <http://dx.doi.org/10.1038/s41559-019-1044-6>  
1061  
1062  
1063 Pelster, B., Egg, M., 2018. Hypoxia-inducible transcription factors in fish: expression, function and  
1064 interconnection with the circadian clock. *J. Exp. Biol.* 221, jeb163709. [http://dx.doi.org/10.1371/jo](http://dx.doi.org/10.1371/journal.pone.0017073)  
1065 [urnal.pone.0017073](http://dx.doi.org/10.1371/journal.pone.0017073)  
1066  
1067 Pietropaolo, S., Guilleminot, A., Martin, B., d'Amato, F.R., Crusio, W.E., 2011. Genetic-background  
1068 modulation of core and variable autistic-like symptoms in Fmr1 knock-out mice. *PLoS ONE.* 6,  
1069 e17073. <http://dx.doi.org/10.1371/journal.pone.0017073>  
1070  
1071 Piferrer, F., Beaumont, A., Falguière, J.-C., Flajšhans, M., Haffray, P., Colombo, L., 2009. Polyploid fish  
1072 and shellfish: production, biology and applications to aquaculture for performance improvement  
1073 and genetic containment. *Aquaculture.* 293, 125-156.  
1074 <http://dx.doi.org/10.1016/j.aquaculture.2009.04.036>  
1075  
1076 Polakof, S., Skiba-Cassy, S., Kaushik, S., Seiliez, I., Soengas, J.L., Panserat, S., 2012. Glucose and lipid  
1077 metabolism in the pancreas of rainbow trout is regulated at the molecular level by nutritional status  
1078 and carbohydrate intake. *J. Comp. Physiol. B.* 182, 507-516. [https://doi.org/10.1007/s00360-011-](https://doi.org/10.1007/s00360-011-0636-5)  
1079 [0636-5](https://doi.org/10.1007/s00360-011-0636-5)

1080 Poon, W., Hung, C., Nakano, K., Randall, D., 2007. An in vivo study of common carp (*Cyprinus carpio* L.)  
1081 liver during prolonged hypoxia. *Comp. Biochem. Physiol. Part D Genomics Proteomics* 2, 295-302.  
1082 <http://dx.doi.org/10.1016/j.cbd.2007.05.002>  
1083

1084 Qi, D., Chao, Y., Zhao, Y., Xia, M., Wu, R., 2018. Molecular evolution of myoglobin in the Tibetan Plateau  
1085 endemic schizothoracine fish (Cyprinidae, Teleostei) and tissue-specific expression changes under  
1086 hypoxia. *Fish Physiol. Biochem.* 44, 557-571. <http://dx.doi.org/10.1007/s10695-017-0453-1>  
1087

1088 Qi, M., Wu, Q., Liu, T., Hou, Y., Miao, Y., Hu, M., Liu, Q., 2020. Hepatopancreas transcriptome profiling  
1089 analysis reveals physiological responses to acute hypoxia and reoxygenation in juvenile Qingtian  
1090 paddy field carp *Cyprinus carpio* var *qingtianensis*. *Front. Physiol.* 11. <http://dx.doi.org/10.3389/fphys.2020.01110>  
1091

1092

1093 R2D2 Consortium, Fugerey-Scarbel, A., Bastien, C., Dupont-Nivet, M., Lemarié, S., 2021. Why and how  
1094 to switch to genomic selection: lessons from plant and animal breeding experience. *Front Genet.*  
1095 12, 629737. <https://doi.org/10.3389/fgene.2021.629737>  
1096

1097 Ragsdale, A., Ortega-Recalde, O., Dutoit, L., Besson, A.A., Chia, J.H., King, T., Nakagawa, S., Hickey, A.,  
1098 Gemmell, N.J., Hore, T., 2020. Paternal hypoxia exposure primes offspring for increased hypoxia  
1099 resistance. *bioRxiv*. <https://doi.org/10.1101/2020.12.09.416727>  
1100

1101 Rashid, I., Nagpure, N.S., Srivastava, P., Kumar, R., Pathak, A.K., Singh, M., Kushwaha, B., 2017.  
1102 HRGFish: A database of hypoxia responsive genes in fishes. *Sci. Rep.* 7, 1-9.  
1103 <http://dx.doi.org/10.1038/srep42346>  
1104

1105 Rehn, M., Olsson, A., Reckzeh, K., Diffner, E., Carmeliet, P., Landberg, G., Cammenga, J., 2011. Hypoxic  
1106 induction of vascular endothelial growth factor regulates murine hematopoietic stem cell function  
1107 in the low-oxygenic niche. *Blood.* 118, 1534-1543. <https://doi.org/10.1182/blood-2011-01-332890>  
1108

1109 Reid, G.K., Gurney-Smith, H.J., Flaherty, M., Garber, A.F., Forster, I., Brewer-Dalton, K., Knowler, D.,  
1110 Marcogliese, D.J., Chopin, T., Moccia, R.D., 2019. Climate change and aquaculture: considering  
1111 adaptation potential. *Aquac. Environ. Interact.* 11, 603-624. <http://dx.doi.org/10.3354/aei00333>  
1112

1113 Richards, J.G., 2011. Physiological, behavioral and biochemical adaptations of intertidal fishes to  
1114 hypoxia. *J. Exp. Biol.* 214, 191-199. <http://dx.doi.org/10.1242/jeb.047951>  
1115

1116 Robledo, D., Palaikostas, C., Bargelloni, L., Martínez, P., Houston, R., 2017. Applications of genotyping  
1117 by sequencing in aquaculture breeding and genetics. *Rev. Aquac.* 0, 1-13.  
1118 <http://dx.doi.org/10.1111/raq.12193>  
1119

1120 Rossignol, R., Ranchon-Cole, I., Pâris, A., Herzine, A., Perche, A., Laurenceau, D., Bertrand, P., Cercy, C.,  
1121 Pichon, J., Mortaud, S., 2014. Visual sensorial impairments in neurodevelopmental disorders:  
1122 evidence for a retinal phenotype in Fragile X Syndrome. *PLoS ONE.* 9, e105996.  
1123 <http://dx.doi.org/10.1371/journal.pone.0105996>  
1124

1125 Royer, E., Faccenda, F., Pastres, R., 2021. Estimating oxygen consumption of rainbow trout  
1126 (*Oncorhynchus mykiss*) in a raceway: A Precision Fish Farming approach. *Aquacult. Eng.* 92, 102141.  
1127 <http://dx.doi.org/10.1016/j.aquaeng.2020.102141>

1128  
1129 Sae-Lim, P., Kause, A., Mulder, H., Olesen, I., 2017. Breeding and genetics symposium: Climate change  
1130 and selective breeding in aquaculture. *J. Anim. Sci.* 95, 1801-1812.  
1131 <http://dx.doi.org/10.2527/jas2016.1066>  
1132  
1133 Sahoo, S., Meijles, D.N., Pagano, P.J., 2016. NADPH oxidases: key modulators in aging and age-related  
1134 cardiovascular diseases? *Clin. Sci.* 130, 317-335. <http://dx.doi.org/10.1042/CS20150087>  
1135  
1136 San, L.-Z., Liu, B.-S., Liu, B., Zhu, K.-C., Guo, L., Guo, H.-Y., Zhang, N., Jiang, S.-G., Zhang, D.-C., 2021.  
1137 Genome-wide association study reveals multiple novel SNPs and putative candidate genes  
1138 associated with low oxygen tolerance in golden pompano *Trachinotus ovatus* (Linnaeus 1758).  
1139 *Aquaculture*. 544, 737098. <https://doi.org/10.1016/j.aquaculture.2021.737098>  
1140  
1141 Sargolzaei, M., Chesnais, J.P., Schenkel, F.S., 2014. A new approach for efficient genotype imputation  
1142 using information from relatives. *BMC Genom.* 15, 1-12. [http://dx.doi.org/10.1186/1471-2164-15-](http://dx.doi.org/10.1186/1471-2164-15-478)  
1143 [478](http://dx.doi.org/10.1186/1471-2164-15-478)  
1144  
1145 Scott, M.A., Dhillon, R.S., Schulte, P.M., Richards, J.G., 2015. Physiology and performance of wild and  
1146 domestic strains of diploid and triploid rainbow trout (*Oncorhynchus mykiss*) in response to  
1147 environmental challenges. *Can. J. Fish. Aquat. Sci.* 72, 125-134. [http://dx.doi.org/10.1139/cjfas-](http://dx.doi.org/10.1139/cjfas-2013-0450)  
1148 [2013-0450](http://dx.doi.org/10.1139/cjfas-2013-0450)  
1149  
1150 Schwander, T., Libbrecht, R., Keller, L., 2014. Supergenes and complex phenotypes. *Curr. Biol.* 24, 288-  
1151 294. <http://dx.doi.org/10.1016/j.cub.2014.01.056>  
1152  
1153 Sonesson, A.K., Meuwissen, T.H., 2009. Testing strategies for genomic selection in aquaculture  
1154 breeding programs. *Genet. Sel. Evol.* 41, 1-9. <http://dx.doi.org/10.1186/1297-9686-41-37>  
1155  
1156 Song, H., Ye, S., Jiang, Y., Zhang, Z., Zhang, Q., Ding, X., 2019. Using imputation-based whole-genome  
1157 sequencing data to improve the accuracy of genomic prediction for combined populations in pigs.  
1158 *Genet. Sel. Evol.* 51, 1-13. <http://dx.doi.org/10.1186/s12711-019-0500-8>  
1159  
1160 Sprague, J., Bayraktaroglu, L., Clements, D., Conlin, T., Fashena, D., Frazer, K., Haendel, M., Howe, D.G.,  
1161 Mani, P., Ramachandran, S., 2006. The Zebrafish Information Network: the zebrafish model  
1162 organism database. *Nucleic Acids Res.* 34, 581-585. <http://dx.doi.org/10.1093/nar/gkj086>  
1163  
1164 Strowbridge, N., Northrup, S.L., Earhart, M.L., Blanchard, T.S., Schulte, P.M., 2021. Acute measures of  
1165 upper thermal and hypoxia tolerance are not reliable predictors of mortality following  
1166 environmental challenges in rainbow trout (*Oncorhynchus mykiss*). *Conserv. Physiol.* 9, 1-16.  
1167 <https://doi.org/10.1093/conphys/coab095>  
1168  
1169 Tamura, K., Yoshie, M., Hashimoto, K., Tachikawa, E., 2014. Inhibitory effect of insulin-like growth  
1170 factor-binding protein-7 (IGFBP7) on in vitro angiogenesis of vascular endothelial cells in the rat  
1171 corpus luteum. *J. Reprod. Dev.* 60, 447-453. <https://doi.org/10.1262/jrd.2014-069>  
1172  
1173 Tang, T., Zheng, B., Chen, S.-h., Murphy, A.N., Kudlicka, K., Zhou, H., Farquhar, M.G., 2009. hNOA1  
1174 interacts with complex I and DAP3 and regulates mitochondrial respiration and apoptosis. *J. Biol.*  
1175 *Chem.* 284, 5414-5424. <http://dx.doi.org/10.1074/jbc.M807797200>



- 1176 Thomas, Y., Flye-Sainte-Marie, J., Chabot, D., Aguirre-Velarde, A., Marques, G.M., Pecquerie, L., 2019.  
1177 Effects of hypoxia on metabolic functions in marine organisms: Observed patterns and modelling  
1178 assumptions within the context of Dynamic Energy Budget (DEB) theory. *J. Sea Res.* 143, 231-242.  
1179 <https://doi.org/10.1016/j.seares.2018.05.001>  
1180
- 1181 Tian, Y., Wen, H., Qi, X., Zhang, X., Li, Y., 2019. Identification of mapk gene family in *Lateolabrax*  
1182 *maculatus* and their expression profiles in response to hypoxia and salinity challenges. *Gene.* 684,  
1183 20-29. <http://dx.doi.org/10.1016/j.gene.2018.10.033>  
1184
- 1185 Townhill, B.L., van der Molen, J., Metcalfe, J.D., Simpson, S.D., Farcas, A., Pinnegar, J.K., 2017.  
1186 Consequences of climate-induced low oxygen conditions for commercially important fish. *Mar.*  
1187 *Ecol. Prog. Ser.* 580, 191-204. <http://dx.doi.org/10.3354/meps12291>  
1188
- 1189 Tsai, H.-Y., Hamilton, A., Tinch, A.E., Guy, D.R., Bron, J.E., Taggart, J.B., Gharbi, K., Stear, M., Matika, O.,  
1190 Pong-Wong, R., 2016. Genomic prediction of host resistance to sea lice in farmed Atlantic salmon  
1191 populations. *Genet. Sel. Evol.* 48, 1-11. <http://dx.doi.org/10.1186/s12711-016-0226-9>  
1192
- 1193 Vallejo, R.L., Leeds, T.D., Gao, G., Parsons, J.E., Martin, K.E., Evenhuis, J.P., Fragomeni, B.O., Wiens,  
1194 G.D., Palti, Y., 2017. Genomic selection models double the accuracy of predicted breeding values  
1195 for bacterial cold water disease resistance compared to a traditional pedigree-based model in  
1196 rainbow trout aquaculture. *Genet. Sel. Evol.* 49, 1-13. [http://dx.doi.org/10.1186/s12711-017-0293-](http://dx.doi.org/10.1186/s12711-017-0293-6)  
1197 [6](http://dx.doi.org/10.1186/s12711-017-0293-6)  
1198
- 1199 Vallejo, R.L., Silva, R.M., Evenhuis, J.P., Gao, G., Liu, S., Parsons, J.E., Martin, K.E., Wiens, G.D., Lourenco,  
1200 D.A., Leeds, T.D., 2018. Accurate genomic predictions for BCWD resistance in rainbow trout are  
1201 achieved using low-density SNP panels: Evidence that long-range LD is a major contributing factor.  
1202 *J. Anim. Breed. Genet.* 135, 263-274. <http://dx.doi.org/10.1111/jbg.12335>  
1203
- 1204 Van Binsbergen, R., Calus, M.P., Bink, M.C., van Eeuwijk, F.A., Schrooten, C., Veerkamp, R.F., 2015.  
1205 Genomic prediction using imputed whole-genome sequence data in Holstein Friesian cattle. *Genet.*  
1206 *Sel. Evol.* 47, 1-13. <http://dx.doi.org/10.1186/s12711-015-0149-x>  
1207
- 1208 Van Raaij, M., Van den Thillart, G., Vianen, G., Pit, D., Balm, P., Steffens, A., 1996. Substrate mobilization  
1209 and hormonal changes in rainbow trout (*Oncorhynchus mykiss*, L.) and common carp (*Cyprinus*  
1210 *carpio*, L.) during deep hypoxia and subsequent recovery. *J. Comp. Physiol. B.* 166, 443-452.  
1211 <https://doi.org/10.1007/BF02337889>  
1212
- 1213 VanRaden, P.M., 2008. Efficient methods to compute genomic predictions. *J. Dairy Sci.* 91, 4414-4423.  
1214 <http://dx.doi.org/10.3168/jds.2007-0980>  
1215
- 1216 Wang, X., Liu, S., Jiang, C., Geng, X., Zhou, T., Li, N., Bao, L., Li, Y., Yao, J., Yang, Y., 2017. Multiple across-  
1217 strain and within-strain QTLs suggest highly complex genetic architecture for hypoxia tolerance in  
1218 channel catfish. *Mol. Genet. Genomics.* 292, 63-76. <http://dx.doi.org/10.1007/s00438-016-1256-2>  
1219
- 1220 Wawrowski, A., Gerlach, F., Hankeln, T., Burmester, T., 2011. Changes of globin expression in the  
1221 Japanese medaka (*Oryzias latipes*) in response to acute and chronic hypoxia. *J. Comp. Physiol. B.*  
1222 181, 199-208. <http://dx.doi.org/10.1007/s00360-010-0518-2>

1223 Wischhusen, P., Larroquet, L., Durand, T., Oger, C., Galano, J.-M., Rocher, A., Vigor, C., Prabhu, P.A.J.,  
1224 Véron, V., Briens, M., 2020. Oxidative stress and antioxidant response in rainbow trout fry exposed  
1225 to acute hypoxia is affected by selenium nutrition of parents and during first exogenous feeding.  
1226 Free Radical Biol. Med. 155, 99-113. <http://dx.doi.org/10.1016/j.freeradbiomed.2020.05.006>  
1227

1228 Wolf, J.B., Wade, M.J., 2009. What are maternal effects (and what are they not)? Philosophical Philos.  
1229 Trans. R. Soc. Lond., B, Biol. Sci. 364, 1107-1115. <http://dx.doi.org/10.1098/rstb.2008.0238>  
1230

1231 Wu, X., Lund, M.S., Sahana, G., Guldbbrandtsen, B., Sun, D., Zhang, Q., Su, G., 2015. Association analysis  
1232 for udder health based on SNP-panel and sequence data in Danish Holsteins. Genet. Sel. Evol. 47,  
1233 1-14. <http://dx.doi.org/10.1186/s12711-015-0129-1>  
1234

1235 Xu, W., Chen, D., Yan, G., Xiao, S., Huang, T., Zhang, Z., Huang, L., 2019. Rediscover and refine QTLs for  
1236 pig scrotal hernia by increasing a specially designed F3 population and using whole-genome  
1237 sequence imputation technology. Front. Genet. 10, 890.  
1238 <http://dx.doi.org/10.3389/fgene.2019.00890>  
1239

1240 Yan, G., Qiao, R., Zhang, F., Xin, W., Xiao, S., Huang, T., Zhang, Z., Huang, L., 2017. Imputation-based  
1241 whole-genome sequence association study rediscovered the missing QTL for lumbar number in  
1242 Suta pigs. Sci Rep. 7, 1-10. <http://dx.doi.org/10.1038/s41598-017-00729-0>  
1243

1244 Yáñez, J.M., Houston, R.D., Newman, S., 2014. Genetics and genomics of disease resistance in salmonid  
1245 species. Front. Genet. 5, 415. <https://doi.org/10.3389/fgene.2014.00415>  
1246

1247 Yoshida, G.M., Banger, R., Carvalheiro, R., Correa, K., Figueroa, R., Lhorente, J.P., Yáñez, J.M., 2018a.  
1248 Genomic prediction accuracy for resistance against *Piscirickettsia salmonis* in farmed rainbow trout.  
1249 G3-Genes Genom. Genet. 8, 719-726. <http://dx.doi.org/10.1534/g3.117.300499>  
1250

1251 Yoshida, G.M., Carvalheiro, R., Lhorente, J.P., Correa, K., Figueroa, R., Houston, R.D., Yáñez, J.M.,  
1252 2018b. Accuracy of genotype imputation and genomic predictions in a two-generation farmed  
1253 Atlantic salmon population using high-density and low-density SNP panels. Aquaculture. 491, 147-  
1254 154. <http://dx.doi.org/10.1016/j.aquaculture.2018.03.004>  
1255

1256 Yoshida, G.M., Lhorente, J.P., Correa, K., Soto, J., Salas, D., Yáñez, J.M., 2019. Genome-wide association  
1257 study and cost-efficient genomic predictions for growth and fillet yield in Nile tilapia (*Oreochromis*  
1258 *niloticus*). G3-Genes Genom. Genet. 9, 2597-2607. <http://dx.doi.org/10.1534/g3.119.400116>  
1259

1260 Yoshida, G.M., Yáñez, J.M., 2021. Multi-trait GWAS using imputed high-density genotypes from whole-  
1261 genome sequencing identifies genes associated with body traits in Nile tilapia. BMC Genom. 22, 57.  
1262 <http://dx.doi.org/10.1186/s12864-020-07341-z>  
1263

1264 Yoshida, G.M., Yáñez, J.M., 2022. Increased accuracy of genomic predictions for growth under chronic  
1265 thermal stress in rainbow trout by prioritizing variants from GWAS using imputed sequence data.  
1266 Evol. Appl. 15, 537-552 <http://dx.doi.org/10.1111/eva.13240>  
1267

1267 Yu, X., Megens, H.-J., Mengistu, S.B., Bastiaansen, J.W., Mulder, H.A., Benzie, J.A., Groenen, M.A.,  
1268 Komen, H., 2021. Genome-wide association analysis of adaptation to oxygen stress in Nile tilapia  
1269 (*Oreochromis niloticus*). BMC Genom. 22, 1-13. <http://dx.doi.org/10.1186/s12864-021-07486-5>

1270 Yuan, Z., Liu, S., Yao, J., Zeng, Q., Tan, S., Liu, Z., 2016. Expression of Bcl-2 genes in channel catfish after  
1271 bacterial infection and hypoxia stress. *Dev. Comp. Immunol.* 65, 79-90.  
1272 <http://dx.doi.org/10.1016/j.dci.2016.06.018>  
1273

1274 Zhao, J., Prchal, M., Kause, A., Vandeputte, M., Gela, D., Kroupová, H.K., Piačková, V., Šauer, P.,  
1275 Steinbach, C., Allamellou, J.-M., 2021. The role of energy reserves in common carp performance  
1276 inferred from phenotypic and genetic parameters. *Aquaculture.* 541, 736799. [http://dx.doi.org/10](http://dx.doi.org/10.1016/j.aquaculture.2021.736799)  
1277 [.1016/j.aquaculture.2021.736799](http://dx.doi.org/10.1016/j.aquaculture.2021.736799)  
1278

1279 Zhong, X., Wang, X., Zhou, T., Jin, Y., Tan, S., Jiang, C., Geng, X., Li, N., Shi, H., Zeng, Q., 2017. Genome-  
1280 wide association study reveals multiple novel QTL associated with low oxygen tolerance in hybrid  
1281 catfish. *Mar. Biotechnol.* 19, 379-390. <http://dx.doi.org/10.1007/s10126-017-9757-5>  
1282

1283 Zhu, C.-D., Wang, Z.-H., Yan, B., 2013. Strategies for hypoxia adaptation in fish species: a review.  
1284 *J. Comp. Physiol. B.* 183, 1005-1013. <http://dx.doi.org/10.1007/s00360-013-0762-3>  
1285  
1286

1287 **Table 1.** Summary statistics of average body weight and hypoxia tolerance phenotypes and  
 1288 dissolved oxygen levels during hypoxia challenge trials

Trial	Phenotypes			Dissolved oxygen level mg/L <sup>-1</sup>			
	N <sup>a</sup>	BW (g) <sup>b</sup>	TLE (min.) <sup>c</sup>	Initial	First fish	50%	Last fish
1.	202	46.1±11.1	100.1±65.0	9.4	2.2	1.6	1.2
2.	205	47.3±10.6	65.3±60.1	9.6	2.1	1.5	1.3
3.	202	47.4±10.7	111.3±80.0	9.1	2.3	1.6	1.2
4.	203	52.9±11.8	115.7±64.4	9.3	4.4	1.7	1.2
5.	206	52.5±10.7	74.6±71.0	8.8	2.4	1.7	1.5
6.	217	55.3±11.5	129.3±75.2	8.7	5.9	1.9	1.3
7.	85	57.4±12.7	89.8±44.2	9.6	5.1	2.2	1.5
Mean	189	50.8±11.8	98.9±71.8	9.2	3.5	1.7	1.3

1289 <sup>a</sup> number of fish

1290 <sup>b</sup> body weight

1291 <sup>c</sup> time to loss of equilibrium

1292 **Table 2.** Variance components for acute hypoxia tolerance in rainbow trout estimated with  
 1293 three models (BLUP, GBLUP, Bayes C $\pi$ ) and two genotype densities (MD, HD)

<b>Model</b>	<b><math>\sigma_a^2</math></b>	<b><math>\sigma_m^2</math></b>	<b><math>\sigma_e^2</math></b>	<b><math>h^2</math></b>	<b><math>m^2</math></b>
BLUP	1332	416	2984	0.28 (0.09)	0.09 (0.04)
GBLUP_MD	1126	481	3081	0.24 (0.05)	0.10 (0.03)
GBLUP_HD	1118	471	3100	0.24 (0.05)	0.10 (0.03)
Bayes C $\pi$ _MD	1054	502	3218	0.22 (0.04)	0.11 (0.03)
Bayes C $\pi$ _HD	1084	484	3215	0.23 (0.04)	0.10 (0.03)

1294  $\sigma_a^2$ : additive genetic variance;  $\sigma_m^2$ : maternal variance;  $\sigma_e^2$ : residual variance;  $h^2$ : heritability (standard  
 1295 error);  $m^2$ : proportion of the phenotypic variance explained by maternal effect (standard error)

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1297 **Table 3.** Mean  $\pm$  standard deviation over 100 replicates of the selection accuracy (r) and  
1298 inflation coefficient (b) for BLUP and GBLUP evaluation for hypoxia tolerance using MD or HD  
1299 genotypes

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	<b>r</b>	<b>b</b>
<b>BLUP</b>	0.594 $\pm$ 0.079	1.456 $\pm$ 0.260
<b>GBLUP_MD</b>	0.656 $\pm$ 0.072	1.121 $\pm$ 0.174
<b>GBLUP_HD</b>	0.653 $\pm$ 0.071	1.202 $\pm$ 0.172

1302 **Table 4.** Summary statistics for GWAS for acute hypoxia tolerance in rainbow trout using Bayes  
 1303  $\pi$  method

Chr.	Peak SNP	Peak position (pb)	MAF	logBF	QTL start (Mb)	QTL end (Mb)	Var (%)	Homozygote standardized TLE difference <sup>A</sup>	Standardized TLE dominance effect <sup>B</sup>
Omy15	Affx-1237442714	17668393	0.32	5.16	17.58	18.32	0.22	82.07	6.99
Omy20	Affx-569843766	22880000	0.42	6.14	22.62	22.99	0.19	59.88	-1.67
Omy28	Affx-1237683396	15091174	0.49	5.13	14.92	15.09	0.17	90.27	38.15
Omy31_1	Affx-569844518	20875730	0.22	7.37	19.93	21.56	0.78	118.54	-9.73
Omy31_2	Affx-1237632075	33812474	0.49	6.27	33.43	33.97	0.33	56.53	13.37

1304 **Chr.:** Chromosome; **logBF:** Twice the natural logarithm of the Bayes Factor; **MAF:** Minor allele  
 1305 frequency; **Var(%):** The percentage of genetic variance explained by the QTL  
 1306 <sup>A</sup> the performance difference between the two homozygous genotypes (BB-AA) expressed in  
 1307 % of genetic standard deviation  
 1308 <sup>B</sup> difference between the performance of the heterozygote genotype (AB) and the average  
 1309 performance of the two homozygous genotypes (AA+BB)/2 expressed in % of genetic  
 1310 standard deviation

1311 **Figure Captions**

1312 **Fig. 1.** The cumulative loss of equilibrium in response to hypoxia challenge across seven trials.  
1313 Time represents the time since the first fish lost its equilibrium.

1314 **Fig. 2.** Boxplot of accuracy difference of GBLUP versus BLUP of hypoxia tolerance for each of  
1315 100 replicates.

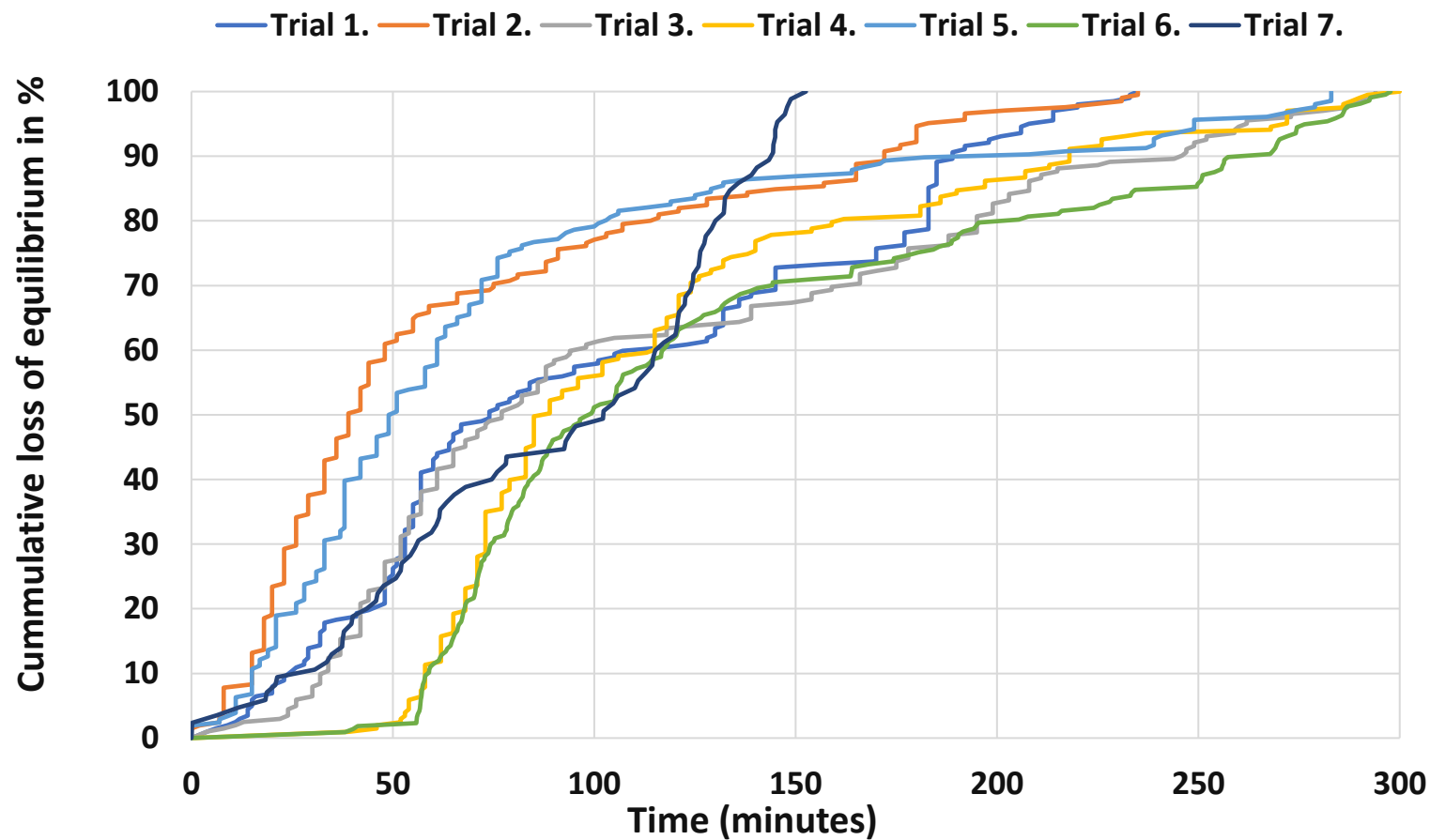
1316 **Fig. 3.** Manhattan plots of QTL detected under Bayesian GWAS for hypoxia tolerance. The  
1317 dashed red line corresponds to the threshold  $\log_{BF} > 6$  for defining evidence for a significant  
1318 QTL and dashed blue line corresponds to the threshold  $\log_{BF} > 5$  for defining evidence for a  
1319 putative QTL.

1320 **Fig. 4.** Annotated genes within the main QTL region on Omy31\_1. The red dots correspond to  
1321 significant SNPs that have  $\log_{BF} > 6$  (dashed red line). The positions of the five genes located  
1322 within the QTL region are figured by rectangles of a different colour: *slitrk4* (SLIT and NTRK-  
1323 like protein 4), *slitrk2* (SLIT and NTRK-like protein 2), *fmr1* (synaptic functional regulator  
1324 FMR1), *aff2* (AF4/FMR2 family member 2), *ids* (iduronate 2-sulfatase), *cracd1* (CRACD-like  
1325 protein isoform), *lonrf3* (LON peptidase N-terminal domain and RING finger protein 3),  
1326 *commd5* (COMM domain-containing protein 5), *fam199x* (protein FAM199X), *arx* (homeobox  
1327 protein ARX-like), *il1rapl2* (X-linked interleukin-1 receptor accessory protein-like 2), *map4k4*  
1328 (*mitogen-activated protein kinase 4*).

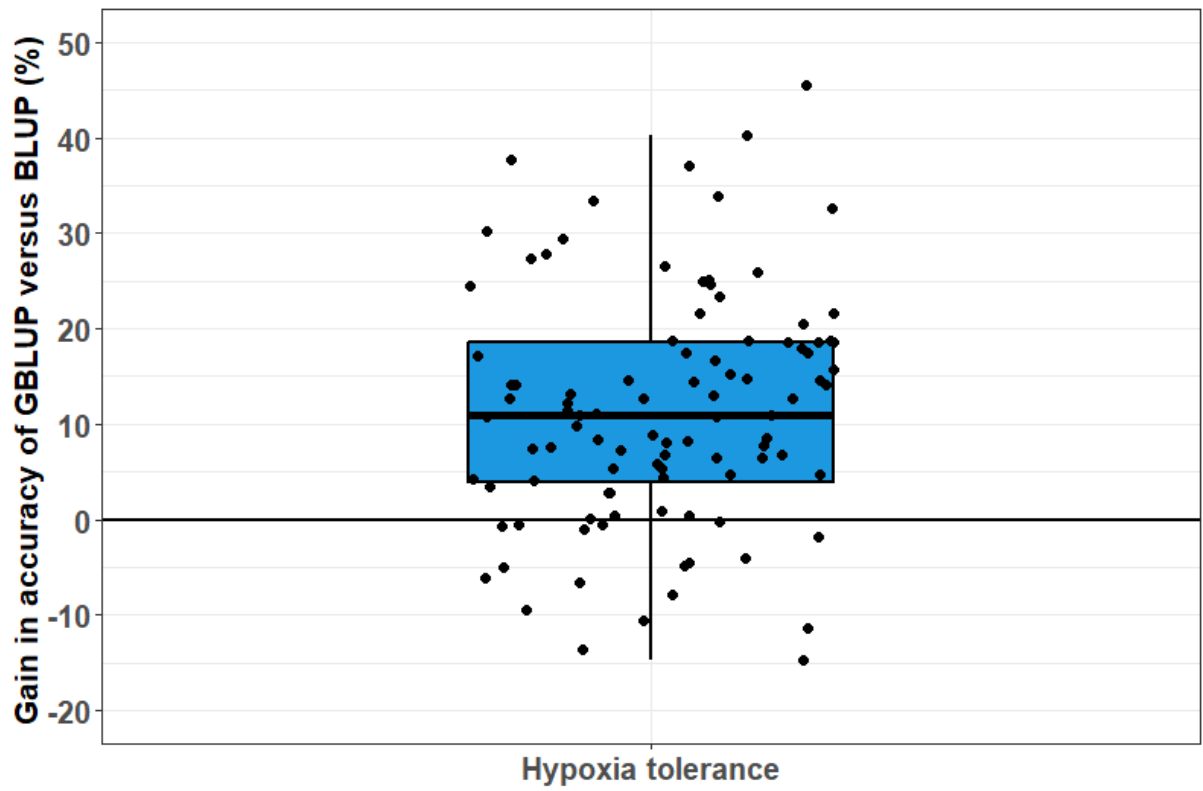
1329 **Fig. 5** Violin box plots of time to loss of equilibrium (TLE) with different genotypes of peak  
1330 SNPs at QTLs located on Omy31\_1 (A) and Omy28 (B). The vertical axis is the TLE of the  
1331 individuals, and different colours represent different genotypes.

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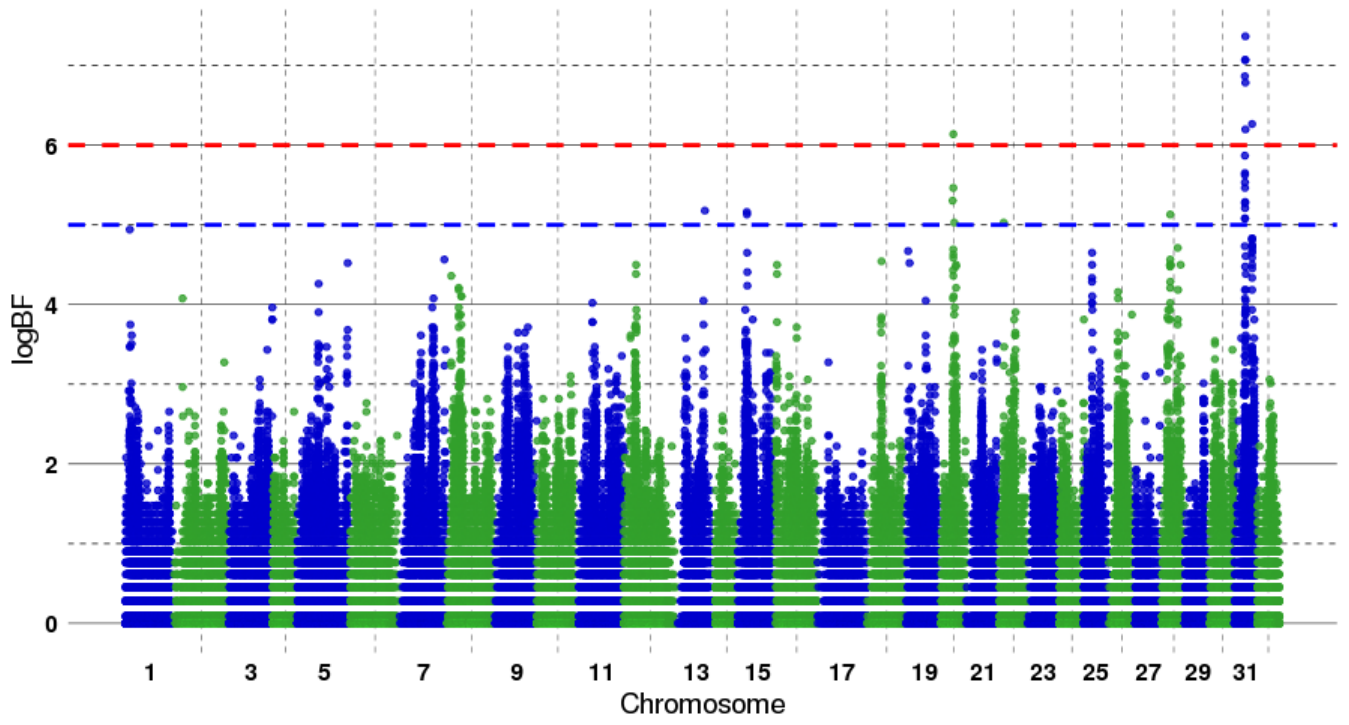




1335 Fig 2.

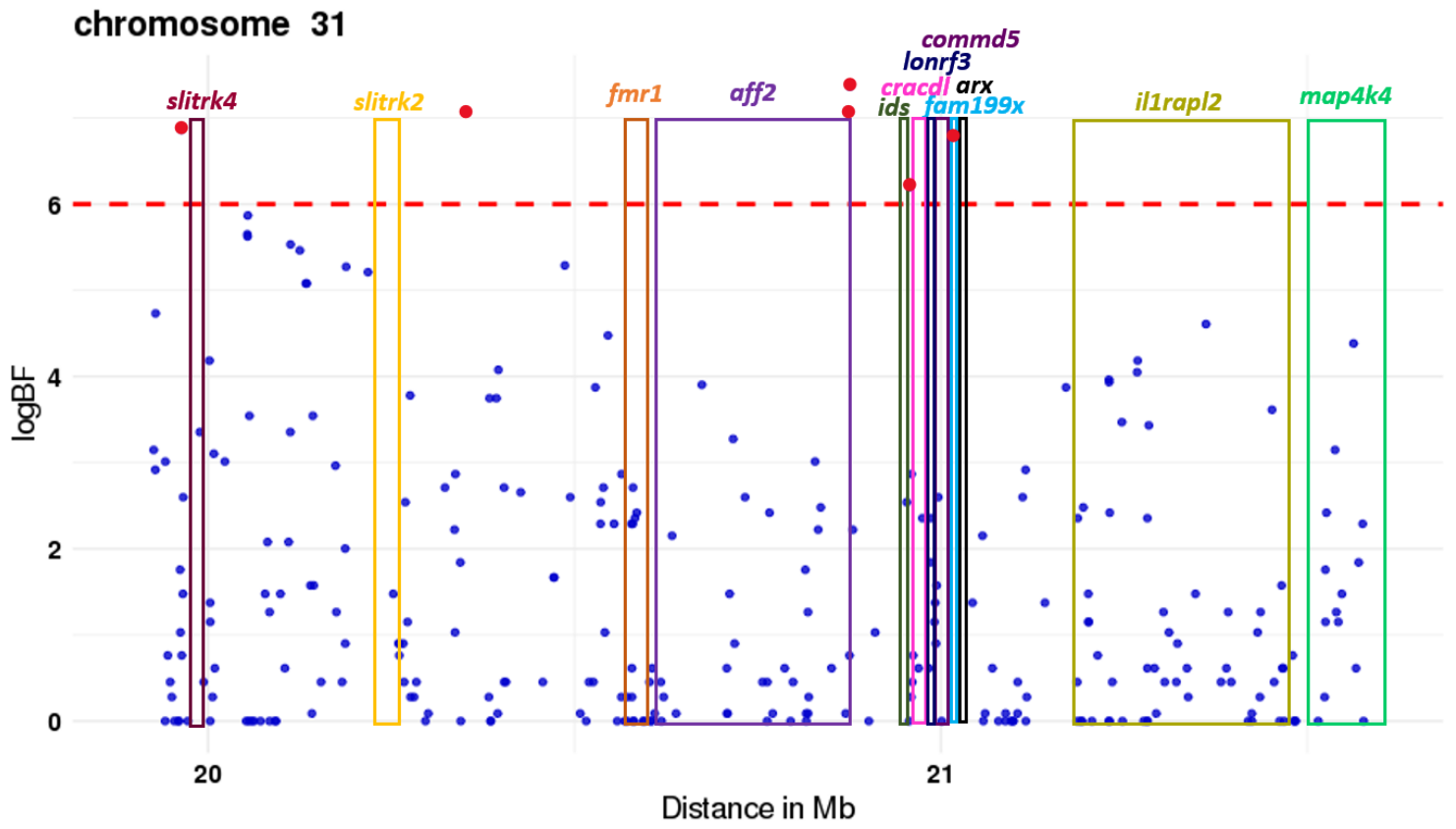


1336 Fig. 3



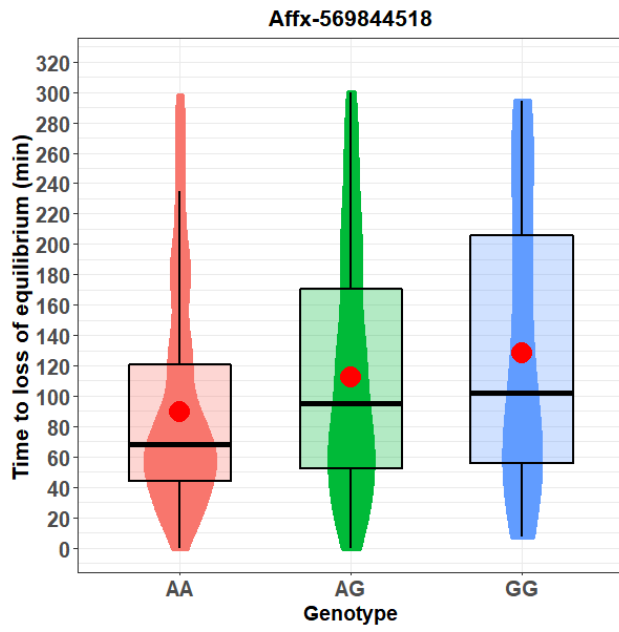
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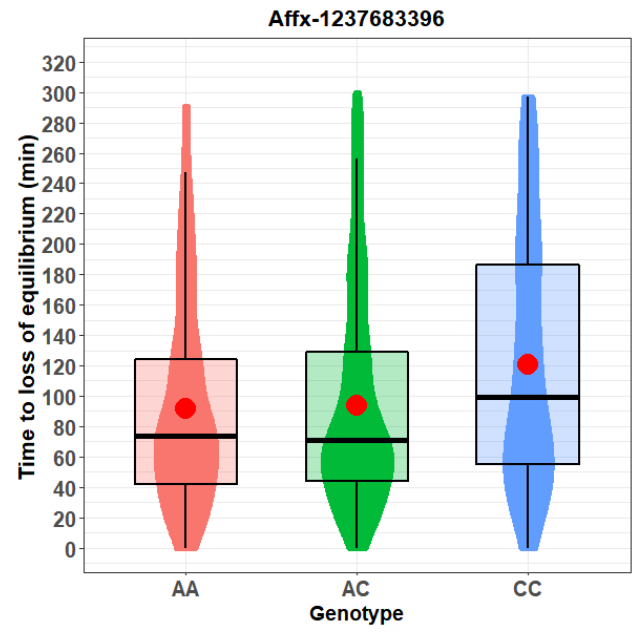


1341 Fig. 5

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B)



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