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Spontaneous polyploidization in critically endangered *Acipenser mikadoi*

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Abstract We investigated the source of spontaneous polyploidization in the critically endangered *Acipenser mikadoi*. Fourteen sib-progeny of *A. mikadoi* and eleven hybrids between an *A. mikadoi* female and *Huso dauricus* male, all showing atypically high ploidy, were analysed. Parent assignment based on five highly polymorphic microsatellite markers confirmed spontaneous duplication of maternal chromosome sets via retention of the second polar body to be the source of spontaneous polyploidization. To our knowledge, this provides the first evidence of maternal origin of spontaneous polyploidization in *A. mikadoi*. Factorial correspondence analysis of the multilocus microsatellite genotypes placed the parent fish of spontaneous polyploids in clearly delineated clusters of *A. mikadoi* and *H. dauricus* and parent fish had mitochondrial control region haplotypes corresponding to their presumed species. Thus, parent fish were confirmed to be of pure genetic origin and hybridization did not promote observed spontaneous polyploidization.

Keywords: Acipenseriformes; DNA content; unreduced gamete; genome duplication; second polar body; meiosis

Introduction

Most sturgeon species are currently classified as critically endangered, mainly due to habitat degradation, poaching, and overexploitation of wild populations, and all sturgeon species were listed in the Appendices to CITES (Convention on International Trade in Endangered Species of Wild Fauna and Flora) in 1998.

Sturgeon evolution has been characterised by several polyploidization and hybridization events [1] producing chromosome numbers of ~120, ~240, or ~360 and genome size from 2.44 in the beluga *Huso huso* [2] to 13.78 pgDNA nucleus⁻¹ in shortnose sturgeon *Acipenser brevirostrum* [3]. Presently, two scales of ploidy level are recognized: i) the evolutionary scale, which presumes tetraploid (4n) – octaploid (8n) – dodecaploid – (12n) relationships [4], referring to ancient ploidy levels and ii) the functional scale, which presumes diploid (2n) – tetraploid (4n) – hexaploid (6n) relationships [5] arising from significant functional genome re-diploidization in sturgeon evolution [6, 7]

Mikado sturgeon *Acipenser mikadoi* is classified as a functional tetraploid having 250–270 chromosomes [8, 9] and genome size of 8.2 pg DNA nucleus⁻¹ [10]. It is one of the most endangered of Acipenseriformes. The species was historically distributed in the Sea of Okhotsk and Sea of Japan, small coastal rivers of the former Soviet Union [11, 12], and the Ishikari and Teshio Rivers of Hokkaido, Japan [13]. Consistent illegal poaching led to the rapid decrease of natural populations of *A. mikadoi* over the past century, and the Tumnin River in Russia is currently its only known natural spawning area, with no more than 30 documented spawners [12, 14].

Recently, several individuals with ploidy levels and chromosome numbers atypical for this species were identified among artificially propagated *A. mikadoi* [9, 10]. The authors suggested that this was probably caused by spontaneous

67 duplication of one or more complete sets of chromosomes (spontaneous
68 polyploidization). Spontaneous polyploidization has been previously reported in
69 artificially propagated sturgeon [15-21], with retention of the second polar body
70 confirmed by molecular analysis to be the cause in white sturgeon *Acipenser*
71 *transmontanus* [16] and Siberian sturgeon *Acipenser baerii* [17]

72 Spontaneous polyploids of functionally tetraploid sturgeon species (i.e.
73 hexaploids) produce triploid gametes [18, 21]. Backcross individuals of such
74 spontaneous polyploids are functionally pentaploid ($2n + 3n = 5n$) [18] and are
75 possibly sterile or semi-sterile (Schreier A and Van Eenennaam J, pers. comm. 2017).
76 Thus, spontaneous polyploidization may have serious repercussions for success of
77 reintroduction and conservation efforts. The number of available broodstock of
78 functionally tetraploid *A. mikadoi* is severely limited. Therefore, confirmation of the
79 source of spontaneous polyploidization in this critically endangered sturgeon species
80 is highly desirable.

81 The goal of this study was to identify the mechanisms underlying unusual ploidy
82 level, and hence genome size and chromosome number, in *A. mikadoi* and in hybrids
83 between *A. mikadoi* and kaluga *Huso dauricus*. All studied individuals were progeny
84 of an *A. mikadoi* dam, to the best of our knowledge the only female of the species to
85 be successfully artificially bred in Japan. Considering that number of available
86 broodstock of *A. mikadoi* is very limited, finding the cause of increase of ploidy levels
87 in this critically endangered sturgeon species may have significance for its controlled
88 reproduction, restoration, and conservation.

90 **Material and Methods**

91 *Fish and Samples*

Fourteen *A. mikadoi* sib-progeny with DNA content of 13.6 pgDNA nucleus⁻¹ and eleven hybrids between an *A. mikadoi* female and *H. dauricus* male with DNA content of 13.1 pgDNA nucleus⁻¹ were used in the study. These individuals showed atypically high ploidy levels, and hence genome size, and were presumed to be spontaneous polyploids [10]. The analysis also included 14 *A. mikadoi* and 11 hybrid specimens of the same parentage as the spontaneous polyploids but with typical ploidy: DNA content of 9.0 pg DNA nucleus⁻¹ in *A. mikadoi* and 8.6 pg DNA nucleus⁻¹ in *H. dauricus*. Detailed description of measurement of DNA content may be found in Zhou et al. [10]. All individuals originated from crossbreeding of *A. mikadoi* female with male of *A. mikadoi* (purebreds) and that of the same female with male of *H. dauricus* (hybrids). Parent fish were captured on the coast of Hokkaido, Japan [22] and held at the Nanae Fresh-Water Station, Field Science Center for Northern Biosphere, Hokkaido University. The male *Huso dauricus* was kept in the Shibetsu Salmon Museum, Hokkaido Japan. Ovulation in females and spermiation in males, fertilization, and incubation of embryos were essentially in accordance with the procedure described by Omoto et al. [20]

To confirm species origin and genetic purity of the parent fish, we included 183 specimens of eight sturgeon species that naturally occur together with *A. mikadoi* and *H. dauricus* or may potentially be co-cultured in facilities along with *A. mikadoi* and *H. dauricus*. The mtDNA haplotypes and microsatellite genotypes of these specimens were clustered together with the mtDNA haplotypes and microsatellite genotypes of parent fish. Assignment of parent fish to particular cluster revealed their genetic origin. These species additionally included in the analyses were: sterlet *A. ruthenus*; Amur sturgeon *A. schrenckii*; Russian sturgeon *A. gueldenstaedtii*; Siberian

sturgeon *A. baerii*; beluga *H. huso*; white sturgeon *A. transmontanus*; *A. mikadoi* and *H. dauricus* (Table 1).

Mitochondrial DNA analyses

Genomic DNA was extracted from fin clips of parent fish and 183 specimens of 8 sturgeon species using NucleoSpin® Tissue Kit (MACHEREY-NAGEL). Standard polymerase chain reaction (PCR) was used to amplify a 574-bp mtDNA control region [23]. The PCR reaction was carried out on 30 µl as described by Havelka et al. [19]. The PCR amplicons were purified and sequenced by Macrogen Inc. (Seoul, Korea).

Microsatellite DNA analyses

Microsatellite genotyping was conducted to determine the species origin of parent fish and the source of the additional set of chromosomes in spontaneous polyploids. Genomic DNA of individuals with atypical ploidy and their typical-ploidy full siblings was extracted using a phenol-chloroform extraction method in 2009 and stored at -30 °C until analysis in 2015. Genomic DNA of other specimens was extracted as described above. Following initial testing of 15 microsatellite markers, 10 markers including *AciG 35* [24]; *Afu 19*, *Afu 68* [25]; *Aox 27*, *Aox 45* [26]; and *Spl 101*, *Spl 105*, *Spl 107*, *Spl 163*, and *Spl 173* [27] were selected for analysis based on the success of their cross-amplification. Amplification was carried out according to the protocol described by Havelka et al. [6]. Microsatellite fragment analysis was performed on a 3130xl ABI Genetic Analyzer (Applied Biosystems, TM) using a GeneScan LIZ 500 size standard (Applied Biosystems, TM), and genotypes were scored in GENEIOUS 6.1.8 [28], using Microsatellite Plugin 1.4. The complexity of duplicated sturgeon

genome and the nature of current microsatellite genotyping make it impossible to reliably determine allele dosage behind a specific peak. Hence, peak patterns were treated as dominant data and interpreted as “allele phenotypes” [29].

Genetic purity of parent fish

As suitable markers for identification of *A. mikadoi* and *H. dauricus* were not available, an alternative approach was used to investigate genetic makeup of parent fish based on clustering of the mtDNA control region and nuclear markers (microsatellites).

Partial sequences of the mtDNA control region were aligned by MAFFT v.7.017 [30], implemented in GENEIOUS 6.1.8. [28]. Bayesian inference and maximum likelihood (ML) were applied for phylogenetic reconstruction and clustering of parent specimens based on mtDNA. The American paddlefish *Polyodon spathula* was included as an outgroup taxon. To determine the best fit models of nucleotide substitution, jModelTest 2.1.7 [31] was applied, and HKY85 model [32] was selected and used in Bayesian and maximum likelihood analyses. MRBAYES 3.1 [33] was employed to conduct Metropolis-coupled Markov chain Monte Carlo (MCMC) analyses for 1 100 000 generations: two simultaneous runs of four MC chains each, chain temperature of 0.2, sample frequency of 200 generations, with burn-in of 100 000 generations. Maximum likelihood analysis was performed in MEGA 6 [34] with probability for each node tested by 1000 bootstrap replications.

To discriminate species and identify origin of parent fish by nuclear DNA, genetic relationships among individuals based on the multilocus genotypes at nine microsatellite loci (locus AciG 35 was not included) were visualized by factorial correspondence analysis (FCA) in GENETIX v.4.05 [35]. This enabled visualization

of data in multidimensional space, with no a priori assumptions on grouping, using each allele as an independent variable.

Origin of spontaneous polyploidy

The origin of the spontaneous increase in ploidy level and chromosome numbers was investigated as an increase of private maternal and/or paternal microsatellite alleles at five microsatellite loci (AciG 35, Aox 45, Spl 101, Spl 105, Spl 107) in spontaneous polyploid individuals compared to their full siblings with typical ploidy according to Havelka et al. [17]. Briefly, if an allele was unique to sire or dam, it was designated code 1. Alleles present in both parent genotypes were not taken into account, since they were not informative for the analyses. All genotypes of analysed progeny were assessed by this approach, and the number of dam and sire private alleles was determined for each microsatellite locus and each individual. The critical factor was the number of private sire and dam microsatellite alleles at each locus in polyploid individuals and in their full siblings with typical ploidy. The total number of private sire and dam microsatellite alleles at all loci was calculated for each ploidy group. The increase of private alleles in spontaneous polyploids was calculated according to the following model: $\Delta a_i = (\sum x_i) / (\sum y_i)$, where Δa_i = increase of dam or sire private alleles at all loci, $\sum x_i$ = sum of private alleles of dam i or sire i at all loci in spontaneous polyploids, and $\sum y_i$ = sum of private alleles of dam i or sire i at all loci in individuals with typical ploidy. Subsequently, the following hypotheses were tested: i) $\Delta a_{dam} = 2$ while $\Delta a_{sire} \approx 1$, duplicated part of the genome originated from clonal duplication of the dam genome; ii) $\Delta a_{dam} = 1 - 2$ while $\Delta a_{sire} \approx 1$, the duplicated part of the genome originated from the dam by retention of second polar body; iii) $\Delta a_{dam} \approx$

1 while $\Delta a_{sire} = 1 - 2$, duplicated part of the genome originated from the sire by dispermic fertilization (see also Supplementary material Figure S1).

Results

Identification of origin of parent fish

Analyses of partial sequences of the mtDNA control region revealed well-supported clusters corresponding to each analysed species. Trees produced with Bayesian (Fig. 1) and ML (not shown) methods were similar in topology. The parents of *A. mikadoi* clustered with other *A. mikadoi* individuals and represented a sister cluster to *H. dauricus*, in which the male *H. dauricus* was found. One specimen of *A. gueldenstaedtii* (Ag_13) had a haplotype highly similar to haplotypes of *A. baerii* (Pairwise identity = 99.8%).

The multidimensional FCA analysis was used to reveal the origin of the parent fish based on nuclear markers (Fig. 2). The most informative was axis 1, which explained 20.45% of the total genetic variation, while axes 2 and 3 represented 16.07% and 14.15% of the total genetic variation, respectively. Separate clusters were determined for *H. huso*, *A. ruthenus*, *A. schrenckii*, *H. dauricus*, and *A. transmontanus*. Two slightly mixed clusters were created by *A. baerii* and *A. gueldenstaedtii* with one *A. gueldenstaedtii* (Ag_13) specimen placed in the *A. baerii* cluster. *Acipenser mikadoi* were clustered separately between *A. gueldenstaedtii* and *A. schrenckii*, with one individual placed on the edge of the *A. gueldenstaedtii* cluster. Male and female *A. mikadoi* and male *H. dauricus* broodfish were placed clearly in *A. mikadoi* and *H. dauricus* cluster, respectively (Fig. 2).

Spontaneous polyploidization

Three loci used in this study (*Aox 45*, *Spl 101*, *Spl 107*) exhibited maximally tetraploid allelic band patterns in parent fish as well as in all offspring with typical ploidy. The remaining loci (*AciG 35*, *Spl 105*) displayed maximally octaploid allelic band patterns in parent fish and in all offspring with typical ploidy. In spontaneous polyploids, loci *AciG 35*, *Aox 45*, *Spl 101*, and *Spl 105* (in hybrids) had higher numbers of allelic bands than did these loci in fish of typical ploidy. The additional alleles in spontaneous polyploids were of maternal origin; however, spontaneous polyploid individuals did not display microsatellite genotypes completely identical to the dam genotypes (Supplementary material: Table S1 and Supplementary material: Table S2).

In both analysed groups, a greater number of private dam alleles were found in spontaneous polyploids compared to their full siblings with typical ploidy (Fig. 3). This was most pronounced at loci *Spl 101*, *Spl 107*, *AciG 35*, in which the number of private dam alleles in polyploids of *A. mikadoi* was 1.68 to 2.08-fold that of typical ploidy siblings and, in hybrid polyploids was 1.73 to 1.97-fold that of typical ploidy siblings. At loci *Aox 45* and *Spl 105*, the increase of private dam alleles reached 1.31-fold in polyploid *A. mikadoi* and 1.32-fold in polyploid hybrids. The increase of private dam alleles across all analysed loci was 1.64-fold in polyploid *A. mikadoi* and 1.63-fold in polyploid hybrids. In contrast, the total number of private sire alleles in spontaneous polyploid *A. mikadoi* was 1.04-fold, and in polyploidy hybrid was 0.96-fold, that of their full siblings with typical ploidy (Fig. 3).

Discussion

Origin of parent specimens

Interspecific hybrids of sturgeon species have been documented in nature [36-39] and are commonly bred in aquaculture [40, 41]. Interspecific hybrids of *A. mikadoi* have not been reported in the literature, but hybrids of *H. dauricus* are common in aquaculture [41, 42], and were observed in wild [43]. Because hybrids may produce unreduced gametes (mostly eggs) [44], confirmation of genetic purity of the parent fish was a necessary prerequisite for investigation of the source of the observed spontaneous polyploidization.

All parent fish had mtDNA haplotypes corresponding to their species, showing no evidence of maternal gene introgression from other sturgeon species. Phylogenetic relationships of analysed species were in agreement with those generally observed in Acipenseriformes [45, 46]. Admixture of *A. baerii* and *A. gueldenstaedtii* haplotypes agreed with Birstein et al. [47], who described two mtDNA lineages of *A. gueldenstaedtii*, one of which clustered with *A. baerii*.

The nuclear markers placed parent fish in clusters of *A. mikadoi* and *H. dauricus*. Hence, they were assumed to be pure species based on nuclear markers, which, together with results of mtDNA analysis, showed parent fish to be pure specimens of *A. mikadoi* and *H. dauricus*.

The *A. gueldenstaedtii* individual (Ag_13) with *A. baerii* haplotype and strong evidence of *A. baerii* genome was probably morphologically misidentified or was of hybrid-backcross origin.

Origin of the spontaneous polyploidy

Acipenser mikadoi and *H. dauricus* are classified as functional tetraploid species having 250–270 chromosomes [8]. Zhou et al. [10] identified 47% of spontaneous polyploids in the purebred family of *A. mikadoi* and 57% of spontaneous polyploids in

the hybrid family of *A. mikadoi* and *H. dauricus*. The reported DNA content was indicative of functional hexaploid genome organization and the authors referred to these individuals as genetic triploids, based on the DNA content 1.5-fold that of typical individuals. They also described a karyotype of the previously observed genetic triploid *A. mikadoi* with the chromosome number of 402 as a representative metaphase [9]. In the present study, we found molecular evidence for the origin of those individuals with atypical ploidy levels originally reported in Zhou et al. [10]. For clarity, we related all ploidy levels to functional scales, i.e. parent fish and individuals with typical ploidy to tetraploidy (4n) and individuals with atypical ploidy to hexaploidy (6n).

Hexaploid individuals exhibited a higher number of alleles at analysed loci compared to their full siblings with typical ploidy. The additional alleles originated from the maternal genome, as evidenced by the greater number of private dam alleles observed in hexaploid individuals compared to their tetraploid full siblings. No such increase was observed in private sire alleles. This clearly demonstrated that increase of ploidy level, DNA content, and chromosome number originated from maternal genome through production of unreduced oocytes. Among fish, production of unreduced oocytes via spontaneous duplication of maternal chromosome sets (SDM) is not rare [48]. Spontaneous duplication of maternal chromosome sets can be caused by apomixis, premeiotic endomitosis, or retention of the second polar body in meiosis II. The first two have been shown to provide clone individuals genetically identical to mothers [49-54]. Retention of the second polar body in meiosis II does not result in genetic clones, as the first meiotic division is not affected, and cross-over normally occurs in meiotic prophase I. No hexaploid individuals under study were genetically identical to the mother (Supplementary material Table S1 and

Supplementary material Table S2); hence, they most plausibly originated from unreduced oocytes formed by retention of second polar body in meiosis II. Similar cases have been observed in spontaneous polyploids of *A. transmontanus* [16] and *A. baerii* [17].

Spontaneous polyploidy originating from retention of the second polar body has been observed in rainbow trout *Oncorhynchus mykiss* [55], tench *Tinca tinca* [56], and Japanese eel *Anguilla japonica* [57] with over ripening eggs considered the cause. Omoto et al. [20] showed that spontaneous polyploidy in bester (*H. huso* × *A. ruthenus*) resulted from a prolonged interval between hormonal stimulation and ovulation and was associated with lower egg quality and lower hatching rate. We also strongly suggest that the high incidence of spontaneous polyploidization in eggs of the *A. mikadoi* dam from the Nanae Fresh-Water Station, first documented by Zhou et al. [10] and more detailed analysed in the present study, resulted from a prolonged interval between hormonal stimulation and ovulation. Water does not usually reach the optimal temperature for sturgeon reproduction in the Nanae Fresh-Water Station during the sturgeon reproduction season. After hormone injection, females are monitored for the spontaneous release of eggs, indicating the initiation of ovulation. The interval between stimulation and ovulation may be prolonged, as latency is temperature dependent [58]. This may increase the incidence of spontaneous polyploidy in *A. mikadoi*, similarly as observed in bester [20].

In diploids, the presence of a spontaneously added chromosome set results in triploidy and in infertility or semi-sterility. In contrast, spontaneous polyploidization in tetraploid sturgeon species produces fertile hexaploid individuals, as has been shown in *A. baerii* [18] and *A. transmontanus* [16, 21]. Backcrossing of these spontaneous hexaploids to tetraploid individuals has been reported to produce fully viable

pentaploid progeny [16, 18, 21]. Although the reproductive potential of such pentaploids ($5n$) is unconfirmed, they likely exhibit significantly reduced fertility, as their chromosomes cannot pair during the zygotene stage of meiosis prophase I due to the odd number of chromosome sets. Therefore, an incidence of spontaneous polyploids in populations intended for reintroduction might represent a serious threat. Sturgeon re-introduction programmes are of considerable interest, and the presence of spontaneous polyploids originating from release programs is possible in wild sturgeon populations. Thus, the potential for interbreeding of spontaneous polyploids and individuals in the wild might exist. As in captivity, fertile spontaneous polyploids may spawn, resulting in progeny with reduced fertility, leading to significant decrease of fitness of wild populations and producing a negative impact on critically endangered sturgeon species, such as *A. mikadoi*, that have low abundance of natural spawners. To reduce the potential for spontaneous polyploidy in critically endangered sturgeon and to avoid stocking of spontaneous polyploids into wild populations, ploidy level of all fish should be carefully determined before their inclusion in a program of reproduction or reintroduction. Determination of the optimal latency period in sturgeon should be seriously considered in sturgeon controlled reproduction.

Competing interests

The authors declare that they have no competing interests.

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Table legends

Table 1. Origin of specimens of eight acipenseriform species used in this study.

Supplementary material Table S1. Genotypes with private dam and sire alleles observed at all analysed loci for parent *Acipenser mikadoi*, spontaneous polyploids, and their full siblings with typical ploidy level of purebred *Acipenser mikadoi*.

Supplementary material Table S2. Genotypes with private dam and sire alleles observed at all analysed loci for parent *Acipenser mikadoi* dam and *Huso dauricus* sire, spontaneous polyploids and their full siblings with typical ploidy level of hybrids *Acipenser mikadoi* × *Huso dauricus*.

Figure captions

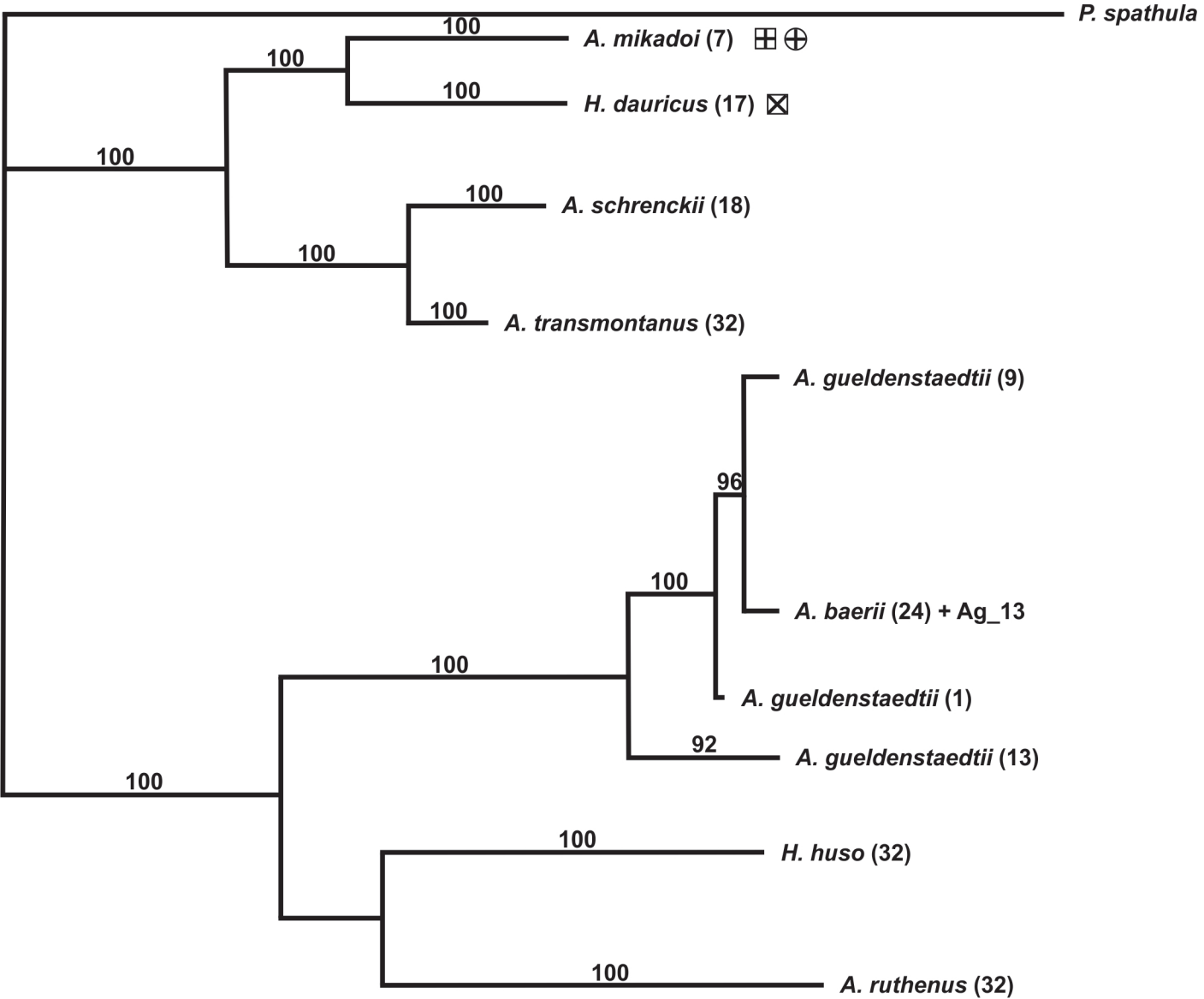
Fig. 1. Phylogenetic tree based on partial sequence of the mitochondrial control region (574-bp) produced by Bayesian inference. Numbers above each branch indicate posterior probability (%). The parents of spontaneous polyploids are ⊕ *Acipenser mikadoi* dam; ⊞ *Acipenser mikadoi* sire; ⊠ *Huso dauricus* sire. Scale bar 0.6; number of individuals in each cluster in brackets.

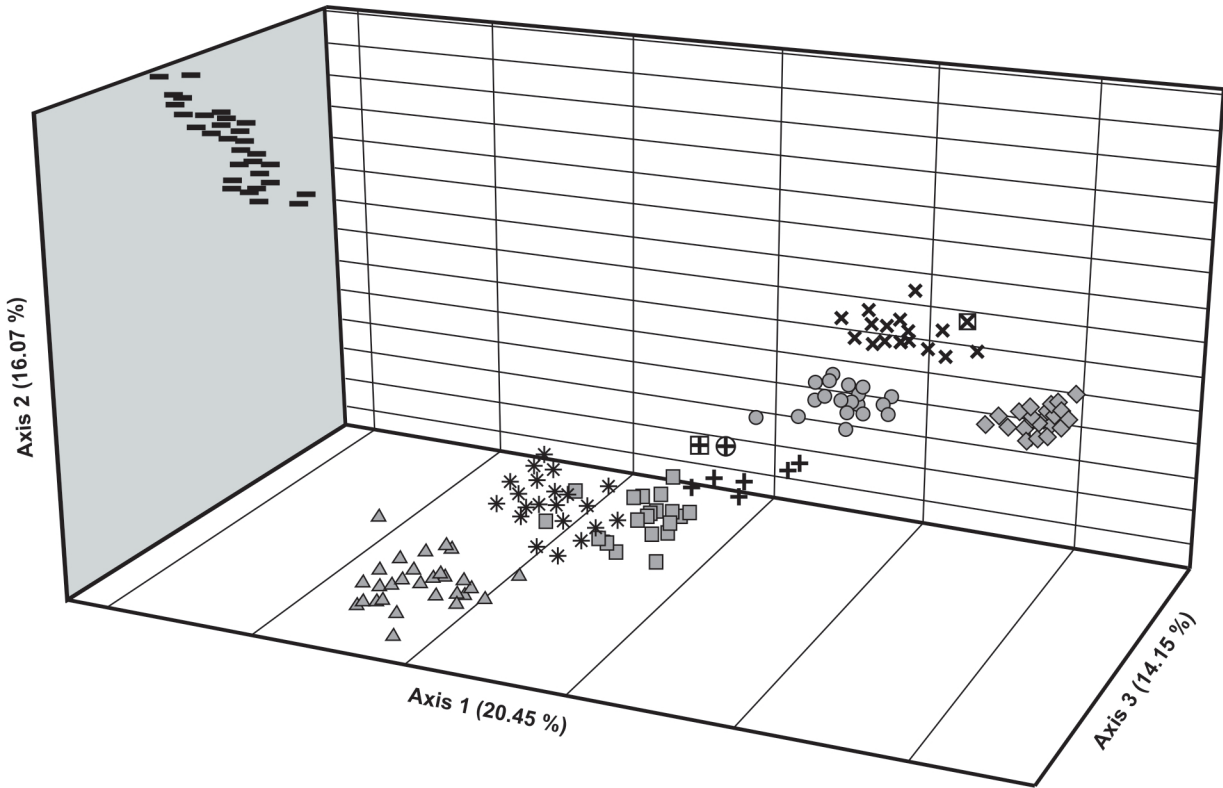
Fig. 2. Factorial correspondence analysis (FCA) based on nine microsatellite loci in eight sturgeon species: △ *Acipenser ruthenus*; – *Huso huso*; ◆ *Acipenser transmontanus*; ● *Acipenser schrenckii*; ✕ *Huso dauricus*; * *Acipenser baerii*; ■ *Acipenser gueldenstaedtii*; + *Acipenser mikadoi*. The parents of spontaneous polyploids are ⊕ *Acipenser mikadoi* dam; ⊞ *Acipenser mikadoi* sire; ⊠ *Huso dauricus* sire. Mark for population consensus, owning the same symbol as corresponding species, is included in each cluster.

534 **Fig. 3.** Fold increase of private dam and sire microsatellite alleles in spontaneous
535 polyploids compared to their full siblings with typical ploidy level. A = purebred of
536 *Acipenser mikadoi*; B = crossbreed of *Acipenser mikadoi* × *Huso dauricus*. See also
537 Table S1 and Table S2.

538 **Supplementary material Fig. S1.** Scheme of investigation of the increase of private
539 alleles in spontaneous polyploids.

Species	Common name	Sample size	Geographic origin of analyzed species
<i>Acipenser ruthenus</i>	sterlet	32	Danube River
<i>Huso huso</i>	beluga	32	Caspian sea / Aquaculture Germany
<i>Acipenser transmontanus</i>	white sturgeon	32	Aquaculture, Japan
<i>Acipenser schrenckii</i>	Amur sturgeon	18	Aquaculture, Japan
<i>Huso dauricus</i>	kaluga	17	Coast of Hokkaido, Japan
<i>Acipenser baerii</i>	Siberian sturgeon	24	Aquaculture Russia / Aquaculture Germany
<i>Acipenser gueldenstaedtii</i>	Russian sturgeon	24	Caspian sea / Aquaculture Germany
<i>Acipenser mikadoi</i>	Mikado sturgeon	7	Coast of Hokkaido / Coast of Ibaraki, Japan





Title: Spontaneous polyploidization in critically endangered *Acipenser mikadoi*

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Journal: Fisheries Science

Note: For better orientation, the genotypes at particular loci are presented on separate sheets of this document

Allele phenotypes

Pop	Sample name	Marker	Allele_1	Allele_2	Allele_3	Allele_4	Allele_5	Pop
			Sire private	Dam privat		Dam privat	Dam privat	
F	Ussu_A. mikadoi	Spl101		330	336	340	360	F
M	Hiro-o_A. mikadoi	Spl101	326		336			M
4n	08/05/28_10	Spl101	326	330	336		360	4n
	08/05/28_3	Spl101	326	330		340		
	08/05/28_5	Spl101	326	330		340		
	08/05/28_6	Spl101	326	330	336			
	08/05/28_7	Spl101	326	330	336	340		
	08/05/28_8	Spl101	326	330	336		360	
	08/05/28_9	Spl101	326	330	336		360	
	08/06/10_1	Spl101	326	330	336			
	08/06/10_2	Spl101	326	330	336		360	
	08/06/10_3	Spl101	326	330		340		
	08/06/10_4	Spl101	326			340	360	
	08/06/10_5	Spl101	326	330		340		
	08/06/10_6	Spl101	326			340	360	
	08/06/10_8	Spl101	326	330	336			
6n	08/05/28_4	Spl101	326	330	336	340	360	6n
	08/06/06_14	Spl101	326	330	336	340	360	
	08/06/06_16	Spl101	326	330	336	340	360	
	08/06/06_18	Spl101	326	330	336	340	360	
	08/06/06_2	Spl101	326	330	336	340	360	
	08/06/06_20	Spl101	326	330	336	340	360	
	08/06/06_22	Spl101	326	330	336	340	360	
	08/06/06_24	Spl101	326	330	336	340	360	
	08/06/06_25	Spl101	326	330	336	340	360	
	08/06/06_3	Spl101	326	330	336	340	360	
	08/06/06_6	Spl101	326	330	336	340	360	
	08/06/10_10	Spl101	326	330	336	340	360	
	08/06/10_7	Spl101	326	330	336	340	360	
	08/06/10_9	Spl101	326	330	336	340	360	

Binary codes

Sample name	Marker	Allele_1	Allele_2	Allele_3	Allele_4	Allele_5
		Sire private	Dam privat		Dam privat	Dam privat
Ussu_A. mikadoi	Spl101	–	1		1	1
Hiro-o_A. mikadoi	Spl101	1	–		–	–
08/05/28_10	Spl101	1	1		–	1
08/05/28_3	Spl101	1	1		1	–
08/05/28_5	Spl101	1	1		1	–
08/05/28_6	Spl101	1	1		–	–
08/05/28_7	Spl101	1	1		1	–
08/05/28_8	Spl101	1	1		–	1
08/05/28_9	Spl101	1	1		–	1
08/06/10_1	Spl101	1	1		–	–
08/06/10_2	Spl101	1	1		–	1
08/06/10_3	Spl101	1	1		1	–
08/06/10_4	Spl101	1	–		1	1
08/06/10_5	Spl101	1	1		1	–
08/06/10_6	Spl101	1	–		1	1
08/06/10_8	Spl101	1	1		–	–
Total		14	12		7	6
08/05/28_4	Spl101	1	1		1	1
08/06/06_14	Spl101	1	1		1	1
08/06/06_16	Spl101	1	1		1	1
08/06/06_18	Spl101	1	1		1	1
08/06/06_2	Spl101	1	1		1	1
08/06/06_20	Spl101	1	1		1	1
08/06/06_22	Spl101	1	1		1	1
08/06/06_24	Spl101	1	1		1	1
08/06/06_25	Spl101	1	1		1	1
08/06/06_3	Spl101	1	1		1	1
08/06/06_6	Spl101	1	1		1	1
08/06/10_10	Spl101	1	1		1	1
08/06/10_7	Spl101	1	1		1	1
08/06/10_9	Spl101	1	1		1	1
Total		14	14		14	14

Summary of private alleles				
Spl101	4n		6n	
Total alleles	Dam 25	Sire 14	Dam 42	Sire 14

Allele phenotypes

E

Pop	Sample name	Marker	Allele_1	Allele_2	Allele_3
			Sire private	Dam privat	
F	Ussu_A. mikadoi	Spl107		308	312
M	Hiro-o_A. mikadoi	Spl107	300		312
4n	08/05/28_10	Spl107	300	308	
	08/05/28_3	Spl107	300		312
	08/05/28_5	Spl107	300		312
	08/05/28_6	Spl107	300	308	
	08/05/28_7	Spl107		308	312
	08/05/28_8	Spl107	300		312
	08/05/28_9	Spl107			312
	08/06/10_1	Spl107		308	312
	08/06/10_2	Spl107		308	312
	08/06/10_3	Spl107		308	312
	08/06/10_4	Spl107		308	312
	08/06/10_5	Spl107	300		312
	08/06/10_6	Spl107			312
	08/06/10_8	Spl107	300		312
	08/05/28_4	Spl107		308	312
	08/06/06_14	Spl107	300	308	312
6n	08/06/06_16	Spl107	300	308	312
	08/06/06_18	Spl107	300	308	312
	08/06/06_2	Spl107	300	308	312
	08/06/06_20	Spl107	300	308	312
	08/06/06_22	Spl107		308	312
	08/06/06_24	Spl107	300	308	312
	08/06/06_25	Spl107		308	312
	08/06/06_3	Spl107	300	308	312
	08/06/06_6	Spl107	300	308	312
	08/06/10_10	Spl107		308	312
	08/06/10_7	Spl107		308	312
	08/06/10_9	Spl107	300	308	312

Pop	Sample name
F	Ussu_A. mikadoi
M	Hiro-o_A. mikadoi

4n	08/05/28_10
	08/05/28_3
	08/05/28_5
	08/05/28_6
	08/05/28_7
	08/05/28_8
	08/05/28_9
	08/06/10_1
	08/06/10_2
	08/06/10_3
	08/06/10_4
	08/06/10_5
	08/06/10_6
	08/06/10_8
	Total

6n	08/05/28_4
	08/06/06_14
	08/06/06_16
	08/06/06_18
	08/06/06_2
	08/06/06_20
	08/06/06_22
	08/06/06_24
	08/06/06_25
	08/06/06_3
	08/06/06_6
	08/06/10_10
	08/06/10_7
	08/06/10_9
	Total

	S
Spl107	
Total alleles	

Binary codes

Marker	Allele_1	Allele_2	Allele_3
	Sire private	Dam privat	
Spl107	–	1	
Spl107	1	–	
Spl107	1	1	
Spl107	1	–	
Spl107	1	–	
Spl107	1	1	
Spl107	–	1	
Spl107	1	–	
Spl107	–	–	
Spl107	–	1	
Spl107	–	1	
Spl107	–	1	
Spl107	–	1	
Spl107	1	–	
Spl107	–	–	
Spl107	1	–	
	7	7	
Spl107	–	1	
Spl107	1	1	
Spl107	1	1	
Spl107	1	1	
Spl107	1	1	
Spl107	1	1	
Spl107	–	1	
Spl107	1	1	
Spl107	–	1	
Spl107	1	1	
Spl107	1	1	
Spl107	–	1	
Spl107	–	1	
Spl107	1	1	
	9	14	

Summary of private alleles			
4n		6n	
Dam 7	Sire 7	Dam 14	Sire 9

Allele phenotypes

Pop	Sample name	Marker	Allele_1	Allele_2	Allele_3	Allele_4	Allele_5	Allele_6
			Sire private		Dam privat	Sire private	Sire private	Dam privat
F	Ussu_A. mikadoi	Aox45		155	164			185
M	Hiro-o_A. mikadoi	Aox45	143	155		173	182	
4n	08/05/28_10	Aox45	143	155	164		182	
	08/05/28_3	Aox45		155			182	
	08/05/28_5	Aox45		155			182	
	08/05/28_6	Aox45		155		173	182	185
	08/05/28_7	Aox45		155		173		
	08/05/28_8	Aox45				173	182	185
	08/05/28_9	Aox45	143	155				185
	08/06/10_1	Aox45	143	155				185
	08/06/10_2	Aox45	143	155	164		182	
	08/06/10_3	Aox45	143	155		173		185
	08/06/10_4	Aox45		155	164		182	
	08/06/10_5	Aox45	143	155		173		185
	08/06/10_6	Aox45		155	164		182	
	08/06/10_8	Aox45		155			182	
	08/05/28_4	Aox45	143	155				185
	08/06/06_14	Aox45		155	164	173		
6n	08/06/06_16	Aox45	143	155	164	173		185
	08/06/06_18	Aox45	143	155	164			
	08/06/06_2	Aox45	143		164	173		185
	08/06/06_20	Aox45	143	155		173		185
	08/06/06_22	Aox45		155			182	
	08/06/06_24	Aox45	143	155			182	
	08/06/06_25	Aox45		155	164		182	
	08/06/06_3	Aox45	143		164	173		185
	08/06/06_6	Aox45		155		173	182	185
	08/06/10_10	Aox45		155	164		182	185
	08/06/10_7	Aox45		155		173	182	
	08/06/10_9	Aox45	143	155			182	

Binary codes

Allele_7	Pop	Sample name	Marker	Allele_1	Allele_2	Allele_3	Allele_4
Dam privat				Sire private		Dam privat	Sire private
194	F	Ussu_A. mikadoi	Aox45	–		1	–
	M	Hiro-o_A. mikadoi	Aox45	1		–	1
				–			–
		08/05/28_10	Aox45	1		1	–
194		08/05/28_3	Aox45	–		–	–
194		08/05/28_5	Aox45	–		–	–
		08/05/28_6	Aox45	–		–	1
194		08/05/28_7	Aox45	–		–	1
194		08/05/28_8	Aox45	–		–	1
194	4n	08/05/28_9	Aox45	1		–	–
		08/06/10_1	Aox45	1		–	–
		08/06/10_2	Aox45	1		1	–
		08/06/10_3	Aox45	1		–	1
194		08/06/10_4	Aox45	–		1	–
		08/06/10_5	Aox45	1		–	1
194		08/06/10_6	Aox45	–		1	–
194		08/06/10_8	Aox45	–		–	–
		Total		6		4	5
194		08/05/28_4	Aox45	1	–	–	–
194		08/06/06_14	Aox45	–	–	1	1
		08/06/06_16	Aox45	1	–	1	1
194		08/06/06_18	Aox45	1	–	1	–
		08/06/06_2	Aox45	1	–	1	1
		08/06/06_20	Aox45	1	–	–	1
194	6n	08/06/06_22	Aox45	–	–	–	–
194		08/06/06_24	Aox45	1	–	–	–
194		08/06/06_25	Aox45	–	–	1	–
		08/06/06_3	Aox45	1	–	1	1
		08/06/06_6	Aox45	–	–	–	1
194		08/06/10_10	Aox45	–	–	1	–
194		08/06/10_7	Aox45	–	–	–	1
194		08/06/10_9	Aox45	1	–	–	–
		Total		8		7	7

Summary of private alleles				
Aox45	4n		6n	
Total alleles	Dam 18	Sire 20	Dam 23	Sire 22

Allele_5	Allele_6	Allele_7
Sire	Dam	Dam
private	privat	privat
–	1	1
1	–	–
–	–	–
1	–	–
1	–	1
1	–	1
1	1	–
–	–	1
1	1	1
–	1	1
–	1	–
1	–	–
–	1	–
1	–	1
–	1	–
1	–	1
1	–	1
9	6	8

–	1	1
–	–	1
–	1	–
–	–	1
–	1	–
–	1	–
1	–	1
1	–	1
1	–	1
–	1	–
1	1	–
1	1	1
1	–	1
1	–	1
7	7	9

Allele phenotypes

Pop	Sample name	Marker	Allele_1	Allele_2	Allele_3	Allele_4	Allele_5	Allele_6
				Dam privat	Dam privat		Dam privat	Sire private
F	Ussu_A. mikadoi	Spl105	140	144	152	164	168	
M	Hiro-o_A. mikadoi	Spl105	140			164		172
4n	08/05/28_10	Spl105	140	144	152	164		
	08/05/28_3	Spl105	140	144		164		172
	08/05/28_5	Spl105	140	144		164		172
	08/05/28_6	Spl105	140			164		172
	08/05/28_7	Spl105	140	144		164		172
	08/05/28_8	Spl105	140		152		168	172
	08/05/28_9	Spl105	140	144		164		
	08/06/10_1	Spl105	140	144	152	164		
	08/06/10_2	Spl105	140	144		164		
	08/06/10_3	Spl105	140		152	164		172
	08/06/10_4	Spl105	140			164	168	
	08/06/10_5	Spl105	140			164	168	
	08/06/10_6	Spl105	140	144	152	164		
	08/06/10_8	Spl105	140			164		172
6n	08/05/28_4	Spl105	140		152	164		
	08/06/06_14	Spl105	140	144		164	168	
	08/06/06_16	Spl105	140			164	168	172
	08/06/06_18	Spl105	140	144		164	168	
	08/06/06_2	Spl105	140		152	164	168	172
	08/06/06_20	Spl105	140	144	152	164		172
	08/06/06_22	Spl105	140	144		164		
	08/06/06_24	Spl105	140			164		172
	08/06/06_25	Spl105	140	144		164	168	
	08/06/06_3	Spl105	140	144	152		168	172
	08/06/06_6	Spl105	140	144	152	164		172
	08/06/10_10	Spl105	140		152	164		
	08/06/10_7	Spl105	140	144		164		172
	08/06/10_9	Spl105	140	144		164		

Binary codes

Pop	Sample name	Marker	Allele_1	Allele_2	Allele_3	Allele_4	Allele_5
				Dam privat	Dam privat		Dam privat
	Ussu_A. mikadoi	Spl105		1	1		1
	Hiro-o_A. mikadoi	Spl105		-	-		-
4n	08/05/28_10	Spl105		1	1		-
	08/05/28_3	Spl105		1	-		-
	08/05/28_5	Spl105		1	-		-
	08/05/28_6	Spl105		-	-		-
	08/05/28_7	Spl105		1	-		-
	08/05/28_8	Spl105		-	1		1
	08/05/28_9	Spl105		1	-		-
	08/06/10_1	Spl105		1	1		-
	08/06/10_2	Spl105		1	-		-
	08/06/10_3	Spl105		-	1		-
	08/06/10_4	Spl105		-	-		1
	08/06/10_5	Spl105		-	-		1
	08/06/10_6	Spl105		1	1		-
	08/06/10_8	Spl105		-	-		-
	Total			8	5		3
6n	08/05/28_4	Spl105		-	1		-
	08/06/06_14	Spl105		1	-		1
	08/06/06_16	Spl105		-	-		1
	08/06/06_18	Spl105		1	-		1
	08/06/06_2	Spl105		-	1		1
	08/06/06_20	Spl105		1	1		-
	08/06/06_22	Spl105		1	-		-
	08/06/06_24	Spl105		-	-		-
	08/06/06_25	Spl105		1	-		1
	08/06/06_3	Spl105		1	1		1
	08/06/06_6	Spl105		1	1		-
	08/06/10_10	Spl105		-	1		-
	08/06/10_7	Spl105		1	-		-
	08/06/10_9	Spl105		1	-		-
	Total			9	6		6

Summary of private alleles				
Spl105	4n		6n	
Total alleles	Dam 16	Sire 7	Dam 21	Sire 7

Allele_6

Sire

private

-

1

-

1

1

1

1

1

-

-

-

1

-

-

-

1

7

-

-

1

-

1

1

-

1

-

1

1

-

1

-

7

Allele phenotypes

Pop	Sample name	Marker	Allele_1	Allele_2	Allele_3	Allele_4	Allele_5	Allele_6
			Dam privat		Sire private		Dam privat	Dam privat
F	Ussu_A. mikadoi	AciG35	244	256		278	286	294
M	Hiro-o_A. mikadoi	AciG35		256	268	278		
4n	08/05/28_10	AciG35	244	256	268			294
	08/05/28_3	AciG35		256	268	278		294
	08/05/28_5	AciG35		256	268	278		294
	08/05/28_6	AciG35			268	278		294
	08/05/28_7	AciG35			268	278		294
	08/05/28_8	AciG35		256	268	278		294
	08/05/28_9	AciG35		256	268	278		294
	08/06/10_1	AciG35		256	268	278	286	
	08/06/10_2	AciG35	244	256		278		294
	08/06/10_3	AciG35	244	256		278	286	
	08/06/10_4	AciG35	244		268	278	286	
	08/06/10_5	AciG35			268	278		294
	08/06/10_6	AciG35		256	268	278		294
	08/06/10_8	AciG35		256	268	278		294
6n	08/05/28_4	AciG35	244	256	268	278	286	294
	08/06/06_14	AciG35	244	256	268	278	286	294
	08/06/06_16	AciG35	244	256	268	278	286	294
	08/06/06_18	AciG35	244	256		278	286	294
	08/06/06_2	AciG35	244	256	268	278	286	294
	08/06/06_20	AciG35	244	256	268	278	286	294
	08/06/06_22	AciG35	244	256	268	278	286	294
	08/06/06_24	AciG35	244	256	268	278	286	294
	08/06/06_25	AciG35	244	256	268	278	286	294
	08/06/06_3	AciG35	244	256		278	286	294
	08/06/06_6	AciG35	244	256	268	278	286	294
	08/06/10_10	AciG35	244	256		278		294
	08/06/10_7	AciG35	244	256	268	278	286	294
	08/06/10_9	AciG35	244	256	268	278	286	294

Allele_7	Allele_8	Allele_9	Allele_10	Allele_11	Allele_12	Pop	Sample name
Sire private	Sire private	Dam privat		Sire private			
		326	330		338	F	Ussu_A. mikadoi
298	306		330	334	338	M	Hiro-o_A. mikadoi
298			330	334	338		08/05/28_10
	306	326	330				08/05/28_3
	306	326	330				08/05/28_5
298		326	330	334			08/05/28_6
	306	326	330		338		08/05/28_7
298			330	334	338		08/05/28_8
298			330	334	338		08/05/28_9
298		326	330	334		4n	08/06/10_1
	306				338		08/06/10_2
298					338		08/06/10_3
	306	326	330	334			08/06/10_4
	306	326	330		338		08/06/10_5
	306	326	330				08/06/10_6
298			330	334	338		08/06/10_8
							Total
	306	326	330	334	338		08/05/28_4
	306	326	330		338		08/06/06_14
298		326	330	334	338		08/06/06_16
298		326	330				08/06/06_18
	306	326	330	334	338		08/06/06_2
	306	326	330		338		08/06/06_20
	306	326	330	334	338		08/06/06_22
298		326	330	334	338	6n	08/06/06_24
	306				338		08/06/06_25
	306	326	330	334	338		08/06/06_3
	306	326	330		338		08/06/06_6
	306	326	330	334	338		08/06/10_10
298		326	330		338		08/06/10_7
	306	326	330		338		08/06/10_9
							Total

	Su
AciG35	
Total alleles	

Binary codes

Marker	Allele_1	Allele_2	Allele_3	Allele_4	Allele_5	Allele_6	Allele_7	Allele_8
	Dam privat		Sire private		Dam privat	Dam privat	Sire private	Sire private
AciG35	1		–		1	1	–	–
AciG35	–		1		–	–	1	1
AciG35	1		1		–	1	1	–
AciG35	–		1		–	1	–	1
AciG35	–		1		–	1	–	1
AciG35	–		1		–	1	1	–
AciG35	–		1		–	1	1	–
AciG35	–		1		–	1	1	–
AciG35	1		–		–	1	–	1
AciG35	1		–		1	–	1	–
AciG35	1		1		1	–	–	1
AciG35	–		1		–	1	–	1
AciG35	–		1		–	1	–	1
AciG35	–		1		–	1	–	1
AciG35	–		1		–	1	1	–
	4		12		3	11	7	7
AciG35	1		1		1	1	–	1
AciG35	1		1		1	1	–	1
AciG35	1		1		1	1	1	–
AciG35	1		–		1	1	1	–
AciG35	1		1		1	1	–	1
AciG35	1		1		1	1	–	1
AciG35	1		1		1	1	–	1
AciG35	1		1		1	1	1	–
AciG35	1		1		1	1	–	1
AciG35	1		–		1	1	–	1
AciG35	1		1		1	1	–	1
AciG35	1		–		–	1	–	1
AciG35	1		1		1	1	1	–
AciG35	1		1		1	1	–	1
	14		11		13	14	4	10

Summary of private alleles			
4n		6n	
Dam	Sire	Dam	Sire
26	33	54	32

Allele_9	Allele_10	Allele_11	Allele_12
Dam		Sire	
privat		private	
1		-	
-		1	
-		1	
1		-	
1		-	
1		1	
1		-	
-		1	
-		1	
1		1	
-		-	
-		-	
1		1	
1		-	
1		-	
-		1	
8		7	
1		1	
1		-	
1		1	
1		-	
1		1	
1		-	
1		1	
1		1	
-		-	
1		1	
1		-	
1		1	
1		-	
1		-	
13		7	

All loci

Summary of private alleles				
Locus	4n		6n	
	y_{dam}	y_{sire}	x_{dam}	x_{sire}
Spl_101	25	14	42	14
Spl_107	7	7	14	9
Aox_45	18	20	23	22
Spl_105	16	7	21	7
AciG 35	26	33	54	32
Total (Σ)	92	81	154	84

Fold increase of private alleles		
Locus	Alleles	
	Dam	Sire
Spl_101	17	0
Spl_107	7	2
Aox_45	5	2
Spl_105	5	0
AciG 35	28	-1
Total	62	3

$$\Delta a_{dam} = (\Sigma x_{dam})$$

$$\Delta a_{sire} = (\Sigma x_{sire}) /$$

Captions

x_{dam} = sum of private alleles of dam in spontaneous polyploids

x_{sire} = sum of private alleles of sire in spontaneous polyploids

y_{dam} = sum of private alleles of dam in individuals with typical ploidy

y_{sire} = sum of private alleles of sire in individuals with typical ploidy

Δa_{dam} = increase of dam private alleles at all loci

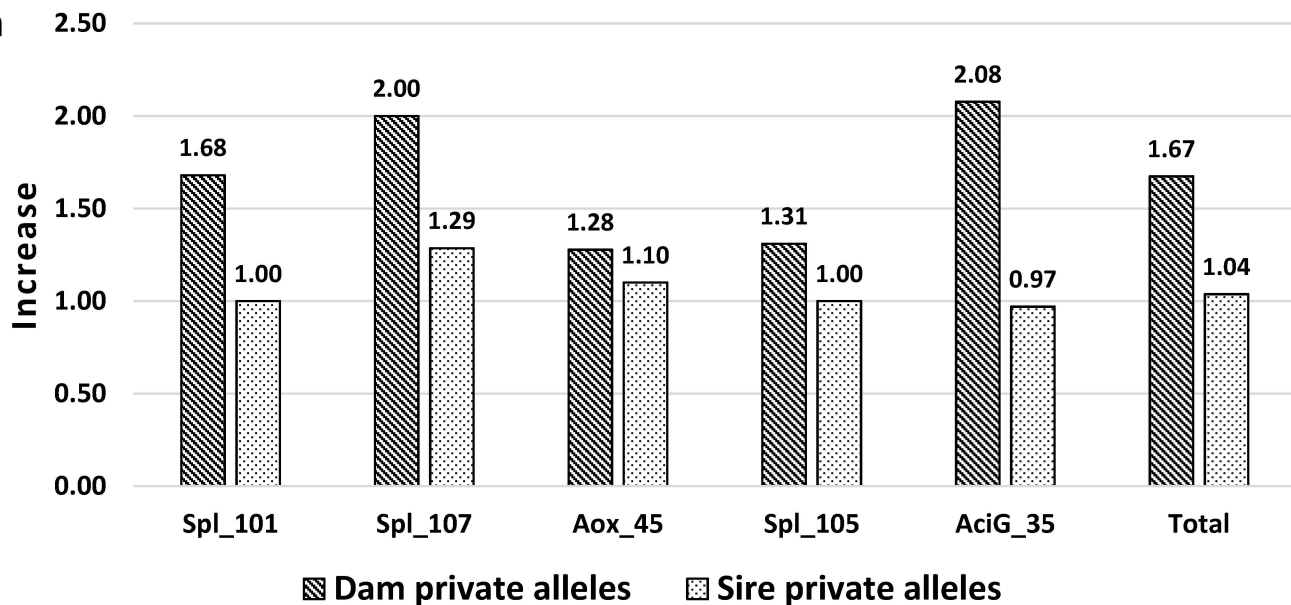
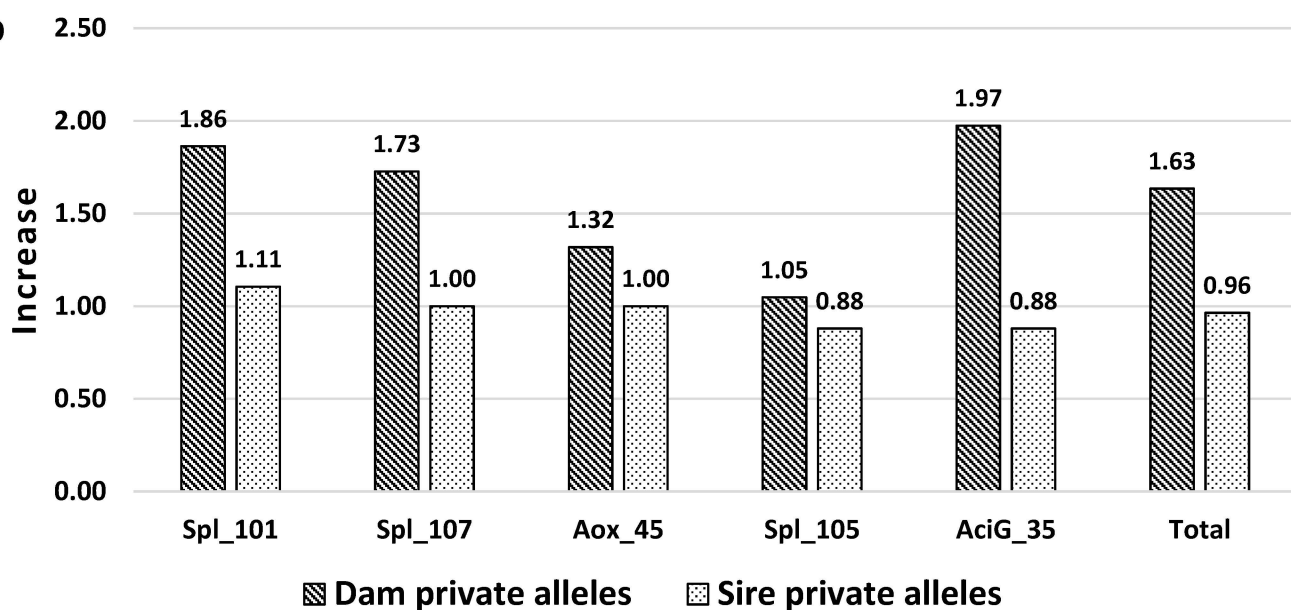
Δa_{sire} = increase of sire private alleles at all loci

as in 6n individuals
x times

Dam	Sire
1.68	1.00
2.00	1.29
1.28	1.10
1.31	1.00
2.08	0.97
1.67	1.04

Δa_{dam} Δa_{sire}

$/ (\sum y_{dam})$
 $(\sum y_{sire})$

a**b**

Title: Spontaneous polyploidization in critically endangered *Acipenser mikadoi*

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Journal: Fisheries Science

Note: For better orientation, the genotypes at particular loci are presented on separate sheets of this document

			Allele phenotypes					
Pop	Sample name	Marker	Allele_1	Allele_2	Allele_3	Allele_4	Allele_5	Allele_6
			Sire private	Sire private	Dam privat	Dam privat	Dam privat	Sire private
F	Ussu_A. mikadoi	Spl101			330	336	340	
M	Uppi_H. dauricus	Spl101	300	316				346
4n	08/07/23_1	Spl101	300	316		336	340	
	08/07/23_2	Spl101		316	330		340	346
	08/07/23_8	Spl101	300			336	340	346
	08/07/29_1	Spl101	300		330	336		346
	08/07/29_16	Spl101	300		330		340	
	08/07/29_17	Spl101	300		330	336		346
	08/07/29_3	Spl101	300		330	336		346
	08/07/29_4	Spl101		316	330		340	346
	08/07/29_7	Spl101	300		330			
	08/07/29_8	Spl101	300		330			
	08/08/29_9	Spl101	300	316	330		340	
6n	08/07/23_3	Spl101	300		330	336	340	346
	08/07/23_4	Spl101		316	330		340	346
	08/07/23_5	Spl101	300	316	330	336	340	
	08/07/23_6	Spl101		316	330	336	340	346
	08/07/23_7	Spl101	300	316	330	336	340	
	08/07/29_10	Spl101	300	316	330	336	340	
	08/07/29_13	Spl101	300		330	336	340	346
	08/07/29_14	Spl101	300	316	330		340	
	08/07/29_2	Spl101	300		330	336	340	
	08/07/29_5	Spl101	300	316	330	336	340	
	08/07/29_6	Spl101	300	316	330		340	

Binary codes

Allele_7	Pop	Sample name	Marker	Allele_1	Allele_2	Allele_3	Allele_4
Dam privat				Sire private	Sire private	Dam privat	Dam privat
360	F	Ussu_A. mikadoi	Spl101	–	–	1	1
	M	Uppi_H. dauricus	Spl101	1	1	–	–
		08/07/23_1	Spl101	1	1	–	1
		08/07/23_2	Spl101	–	1	1	–
		08/07/23_8	Spl101	1	–	–	1
		08/07/29_1	Spl101	1	–	1	1
		08/07/29_16	Spl101	1	–	1	–
	4n	08/07/29_17	Spl101	1	–	1	1
		08/07/29_3	Spl101	1	–	1	1
		08/07/29_4	Spl101	–	1	1	–
360		08/07/29_7	Spl101	1	–	1	–
360		08/07/29_8	Spl101	1	–	1	–
		08/08/29_9	Spl101	1	1	1	–
		Total		9	4	9	5
360		08/07/23_3	Spl101	1	–	1	1
360		08/07/23_4	Spl101	–	1	1	–
360		08/07/23_5	Spl101	1	1	1	1
360		08/07/23_6	Spl101	–	1	1	1
360		08/07/23_7	Spl101	1	1	1	1
360	6n	08/07/29_10	Spl101	1	1	1	1
360		08/07/29_13	Spl101	1	–	1	1
360		08/07/29_14	Spl101	1	1	1	–
360		08/07/29_2	Spl101	1	–	1	1
360		08/07/29_5	Spl101	1	1	1	1
360		08/07/29_6	Spl101	1	1	1	–
		Total		9	8	11	8

Summary of private alleles				
Spl101	4n		6n	
Total alleles	Dam 22	Sire 19	Dam 41	Sire 21

Allele_5	Allele_6	Allele_7
Dam privat	Sire private	Dam privat

1	–	1
–	1	–

1	–	–
1	1	–
1	1	–
–	1	–
1	–	–
–	1	–
–	1	–
1	1	–
–	–	1
–	–	1
1	–	–

6	6	2
---	---	---

1	1	1
1	1	1
1	–	1
1	1	1
1	–	1
1	–	1
1	1	1
1	–	1
1	–	1
1	–	1
1	–	1
1	–	1

11	4	11
----	---	----

Allele phenotypes

Pop	Sample name	Marker	Allele_1	Allele_2	Allele_3	Allele_4	Allele_5	Allele_6
			Dam privat	Dam privat	Sire private	Sire private	Sire private	Sire private
F	Ussu_A. mikadoi	Spl107	308	312				
M	Uppi_H. dauricus	Spl107			328	332	336	344
4n	08/07/23_1	Spl107		312	328			344
	08/07/23_2	Spl107	308		328	332		
	08/07/23_8	Spl107		312	328			344
	08/07/29_1	Spl107	308			332		344
	08/07/29_16	Spl107		312	328		336	
	08/07/29_17	Spl107	308		328	332		
	08/07/29_3	Spl107		312		332		344
	08/07/29_4	Spl107		312	328			344
	08/07/29_7	Spl107		312	328			344
	08/07/29_8	Spl107	308			332	336	
	08/08/29_9	Spl107		312		332		344
	08/07/23_3	Spl107	308	312			336	344
	08/07/23_4	Spl107	308			328		344
6n	08/07/23_5	Spl107	308	312		332		344
	08/07/23_6	Spl107	308	312		332	336	
	08/07/23_7	Spl107	308	312	328		336	
	08/07/29_10	Spl107		312	328		336	
	08/07/29_13	Spl107	308	312	328			344
	08/07/29_14	Spl107	308	312			336	344
	08/07/29_2	Spl107	308			332	336	
	08/07/29_5	Spl107	308	312	328		336	
	08/07/29_6	Spl107	308	312			336	344

Binary codes

Pop	Sample name	Marker	Allele_1	Allele_2	Allele_3	Allele_4	Allele_5
			Dam privat	Dam privat	Sire private	Sire private	Sire private
F	Ussu_A. mikadoi	Spl107	1	1	–	–	–
M	Uppi_H. dauricus	Spl107	–	–	1	1	1
4n	08/07/23_1	Spl107	–	1	1	–	–
	08/07/23_2	Spl107	1	–	1	1	–
	08/07/23_8	Spl107	–	1	1	–	–
	08/07/29_1	Spl107	1	–	–	1	–
	08/07/29_16	Spl107	–	1	1	–	1
	08/07/29_17	Spl107	1	–	1	1	–
	08/07/29_3	Spl107	–	1	–	1	–
	08/07/29_4	Spl107	–	1	1	–	–
	08/07/29_7	Spl107	–	1	1	–	–
	08/07/29_8	Spl107	1	–	–	1	1
	08/08/29_9	Spl107	–	1	–	1	–
	Total		4	7	7	6	2
6n	08/07/23_3	Spl107	1	1	–	–	1
	08/07/23_4	Spl107	1	–	–	1	–
	08/07/23_5	Spl107	1	1	–	1	–
	08/07/23_6	Spl107	1	1	–	1	1
	08/07/23_7	Spl107	1	1	1	–	1
	08/07/29_10	Spl107	–	1	1	–	1
	08/07/29_13	Spl107	1	1	1	–	–
	08/07/29_14	Spl107	1	1	–	–	1
	08/07/29_2	Spl107	1	–	–	1	1
	08/07/29_5	Spl107	1	1	1	–	1
	08/07/29_6	Spl107	1	1	–	–	1
	Total		10	9	4	4	8

Summary of private alleles				
Spl107	4n		6n	
Total alleles	Dam	Sire	Dam	Sire
	11	22	19	22

Allele_6
Sire
private
-
1
1
-
1
1
-
-
1
1
1
-
1
7
1
1
1
-
-
-
1
1
-
-
1
6

Allele phenotypes

Pop	Sample name	Marker	Allele_1	Allele_2	Allele_3	Allele_4	Allele_5	Allele_6
			Sire private	Dam privat	Dam privat	Sire private	Sire private	Dam privat
F	Ussu_A. mikadoi	Aox45		155	164			185
M	Uppi_H. dauricus	Aox45	152			179	182	
4n	08/07/23_1	Aox45	152		164	179		
	08/07/23_2	Aox45			164	179		185
	08/07/23_8	Aox45			164	179		
	08/07/29_1	Aox45	152		164			
	08/07/29_16	Aox45			164		182	
	08/07/29_17	Aox45		155		179		185
	08/07/29_3	Aox45	152				182	185
	08/07/29_4	Aox45	152	155	164		182	
	08/07/29_7	Aox45		155		179	182	185
	08/07/29_8	Aox45	152		164	179		185
	08/08/29_9	Aox45		155	164		182	
	08/07/23_3	Aox45	152					185
	08/07/23_4	Aox45					182	185
6n	08/07/23_5	Aox45	152	155	164	179		185
	08/07/23_6	Aox45	152		164			185
	08/07/23_7	Aox45	152	155			182	
	08/07/29_10	Aox45		155	164	179		185
	08/07/29_13	Aox45	152		164			185
	08/07/29_14	Aox45	152	155			182	185
	08/07/29_2	Aox45		155			182	
	08/07/29_5	Aox45	152		164	179		185
	08/07/29_6	Aox45	152	155			182	185

Binary codes

Allele_7	Allele_8	Pop	Sample name	Marker	Allele_1	Allele_2	Allele_3
Sire private	Dam privat				Sire private	Dam privat	Dam privat
	194	F	Ussu_A. mikadoi	Aox45	–	1	1
188		M	Uppi_H. dauricus	Aox45	1	–	–
	194		08/07/23_1	Aox45	1	–	1
188			08/07/23_2	Aox45	–	–	1
188	194		08/07/23_8	Aox45	–	–	1
188	194		08/07/29_1	Aox45	1	–	1
188	194		08/07/29_16	Aox45	–	–	1
188		4n	08/07/29_17	Aox45	–	1	–
	194		08/07/29_3	Aox45	1	–	–
			08/07/29_4	Aox45	1	1	1
			08/07/29_7	Aox45	–	1	–
			08/07/29_8	Aox45	1	–	1
188			08/08/29_9	Aox45	–	1	1
			Total		5	4	8
188	194		08/07/23_3	Aox45	1	–	–
188	194		08/07/23_4	Aox45	–	–	–
	194		08/07/23_5	Aox45	1	1	1
188	194		08/07/23_6	Aox45	1	–	1
	194		08/07/23_7	Aox45	1	1	–
188	194	6n	08/07/29_10	Aox45	–	1	1
188			08/07/29_13	Aox45	1	–	1
	194		08/07/29_14	Aox45	1	1	–
188	194		08/07/29_2	Aox45	–	1	–
			08/07/29_5	Aox45	1	–	1
	194		08/07/29_6	Aox45	1	1	–
			Total		8	6	5

Summary of private alleles				
Aox45	4n		6n	
Total alleles	Dam	Sire	Dam	Sire
	22	22	29	22

Allele_4	Allele_5	Allele_6	Allele_7	Allele_8
Sire private	Sire private	Dam privat	Sire private	Dam privat
–	–	1	–	1
1	1	–	1	–
1	–	–	–	1
1	–	1	1	–
1	–	–	1	1
–	–	–	1	1
–	1	–	1	1
1	–	1	1	–
–	1	1	–	1
–	1	–	–	–
1	1	1	–	–
1	–	1	–	–
–	1	–	1	–
6	5	5	6	5
–	–	1	1	1
–	1	1	1	1
1	–	1	–	1
–	–	1	1	1
–	1	–	–	1
1	–	1	1	1
–	–	1	1	–
–	1	1	–	1
–	1	–	1	1
1	–	1	–	–
–	1	1	–	1
3	5	9	6	9

Allele phenotypes

Pop	Sample name	Marker	Allele_1	Allele_2	Allele_3	Allele_4	Allele_5	Allele_6
			Sire private		Dam privat	Sire private	Dam privat	Sire private
F	Ussu_A. mikadoi	Spl105		140	144		152	
M	Uppi_H. dauricus	Spl105	116	140		148		160
4n	08/07/23_1	Spl105		140			152	160
	08/07/23_2	Spl105	116		144		152	160
	08/07/23_8	Spl105	116		144	148	152	160
	08/07/29_1	Spl105	116	140	144			160
	08/07/29_16	Spl105	116		144		152	160
	08/07/29_17	Spl105	116		144		152	160
	08/07/29_3	Spl105	116	140		148		160
	08/07/29_4	Spl105	116	140	144	148		160
	08/07/29_7	Spl105	116	140		148	152	160
	08/07/29_8	Spl105		140			152	160
	08/08/29_9	Spl105	116	140		148		160
	08/07/23_3	Spl105	116		144	148	152	160
	08/07/23_4	Spl105	116		144		152	160
6n	08/07/23_5	Spl105		140			152	160
	08/07/23_6	Spl105	116	140	144		152	
	08/07/23_7	Spl105	116	140	144	148		
	08/07/29_10	Spl105	116	140	144	148	152	
	08/07/29_13	Spl105	116	140	144	148	152	160
	08/07/29_14	Spl105	116	140	144	148	152	160
	08/07/29_2	Spl105	116	140				160
	08/07/29_5	Spl105		140	144		152	
	08/07/29_6	Spl105	116	140	144	148	152	160

Binary codes

Allele_7	Allele_8	Pop	Sample name	Marker	Allele_1	Allele_2	Allele_3
	Dam privat				Sire private		Dam privat
164	168	F	Ussu_A. mikadoi	Spl105	–		1
164		M	Uppi_H. dauricus	Spl105	1		–
164	168		08/07/23_1	Spl105	–		–
	168		08/07/23_2	Spl105	1		1
164	168		08/07/23_8	Spl105	1		1
164	168		08/07/29_1	Spl105	1		1
164	168		08/07/29_16	Spl105	1		1
164		4n	08/07/29_17	Spl105	1		1
164	168		08/07/29_3	Spl105	1		–
164	168		08/07/29_4	Spl105	1		1
164			08/07/29_7	Spl105	1		–
164			08/07/29_8	Spl105	–		–
164	168		08/08/29_9	Spl105	1		–
			Total		9		6
164			08/07/23_3	Spl105	1		1
164			08/07/23_4	Spl105	1		1
164			08/07/23_5	Spl105	–		–
164			08/07/23_6	Spl105	1		1
164			08/07/23_7	Spl105	1		1
164	168	6n	08/07/29_10	Spl105	1		1
	168		08/07/29_13	Spl105	1		1
164			08/07/29_14	Spl105	1		1
164	168		08/07/29_2	Spl105	1		–
164	168		08/07/29_5	Spl105	–		1
164			08/07/29_6	Spl105	1		1
			Total		9		9
Summary of private alleles							
Spl105				4n		6n	
Total alleles				Dam 21	Sire 25	Dam 22	Sire 22

Allele_4	Allele_5	Allele_6	Allele_7	Allele_8
Sire private	Dam privat	Sire private		Dam privat
–	1	–		1
1	–	1		–
–	1	1		1
–	1	1		1
1	1	1		1
–	–	1		1
–	1	1		1
–	1	1		–
1	–	1		1
1	–	1		1
1	1	1		–
–	1	1		–
1	–	1		1
5	7	11		8

1	1	1		–
–	1	1		–
–	1	1		–
–	1	–		–
1	–	–		–
1	1	–		1
1	1	1		1
1	1	1		–
–	–	1		1
–	1	–		1
1	1	1		–
6	9	7		4

			Allele phenotypes					
Pop	Sample name	Marker	Allele_1	Allele_2	Allele_3	Allele_4	Allele_5	Allele_6
			Dam privat	Dam privat	Sire private	Sire private	Dam privat	Dam privat
F	Ussu_A. mikadoi	AciG35	244	256			278	286
M	Uppi_H. dauricus	AciG35			260	272		
4n	08/07/23_1	AciG35		256		272		286
	08/07/23_2	AciG35		256		272	278	
	08/07/23_8	AciG35	244		260	272	278	286
	08/07/29_1	AciG35		256		272	278	
	08/07/29_16	AciG35		256	260	272	278	
	08/07/29_17	AciG35	244		260			286
	08/07/29_3	AciG35		256	260			286
	08/07/29_4	AciG35	244		260	272	278	
	08/07/29_7	AciG35		256	260		278	
	08/07/29_8	AciG35	244			272	278	
	08/08/29_9	AciG35		256	260		278	
6n	08/07/23_3	AciG35	244	256	260	272	278	286
	08/07/23_4	AciG35	244	256		272	278	286
	08/07/23_5	AciG35	244	256	260		278	286
	08/07/23_6	AciG35	244	256		272	278	286
	08/07/23_7	AciG35	244	256	260		278	286
	08/07/29_10	AciG35	244	256		272	278	286
	08/07/29_13	AciG35	244	256	260	272	278	286
	08/07/29_14	AciG35	244	256			278	286
	08/07/29_2	AciG35	244	256			278	286
	08/07/29_5	AciG35	244	256		272	278	286
	08/07/29_6	AciG35	244	256			278	286

Allele_7	Allele_8	Allele_9	Allele_10	Allele_11	Allele_12	Pop	Sample name
	Sire private	Sire private	Dam privat	Dam privat	Dam privat		
294			326	330	338	F	Ussu_A. mikadoi
294	302	306				M	Uppi_H. dauricus
294		306	326	300			08/07/23_1
294		306	326	330			08/07/23_2
294		306			338		08/07/23_8
294	302				338		08/07/29_1
294					338		08/07/29_16
294		306	326	330		4n	08/07/29_17
294	302	306	326	330			08/07/29_3
294					338		08/07/29_4
294		306			338		08/07/29_7
294		306			338		08/07/29_8
294	302	306	326	330			08/08/29_9
							Total
294			326	330	338		08/07/23_3
294		306	326	330	338		08/07/23_4
294		306	326	330	338		08/07/23_5
294		306	326	330	338		08/07/23_6
294	302		326	330	338		08/07/23_7
294		306	326	330	338	6n	08/07/29_10
294			326	330	338		08/07/29_13
294	302	306	326	330	338		08/07/29_14
294	302	306	326	330	338		08/07/29_2
294		306	326	330	338		08/07/29_5
294	302	306	326	330	338		08/07/29_6
							Total

	Si
AciG35	
Total alleles	

Binary codes

Marker	Allele_1	Allele_2	Allele_3	Allele_4	Allele_5	Allele_6	Allele_7	Allele_8
	Dam privat	Dam privat	Sire private	Sire private	Dam privat	Dam privat		Sire private
AciG35	1	1	–	–	1	1		–
AciG35	–	–	1	1	–	–		1
AciG35	–	1	–	1	–	1		–
AciG35	–	1	–	1	1	–		–
AciG35	1	–	1	1	1	1		–
AciG35	–	1	–	1	1	–		1
AciG35	–	1	1	1	1	–		–
AciG35	1	–	1	–	–	1		–
AciG35	–	1	1	–	–	1		1
AciG35	1	–	1	1	1	–		–
AciG35	–	1	1	–	1	–		–
AciG35	1	–	–	1	1	–		–
AciG35	–	1	1	–	1	–		1
	4	7	7	7	8	4		3
AciG35	1	1	1	1	1	1		–
AciG35	1	1	–	1	1	1		–
AciG35	1	1	1	–	1	1		–
AciG35	1	1	–	1	1	1		–
AciG35	1	1	1	–	1	1		1
AciG35	1	1	–	1	1	1		–
AciG35	1	1	1	1	1	1		–
AciG35	1	1	–	–	1	1		1
AciG35	1	1	–	–	1	1		1
AciG35	1	1	–	1	1	1		–
AciG35	1	1	–	–	1	1		1
	11	11	4	6	11	11		4

Summary of private alleles			
4n		6n	
Dam	Sire	Dam	Sire
39	25	77	22

Allele_9	Allele_10	Allele_11	Allele_12
Sire private	Dam privat	Dam privat	Dam privat
–	1	1	1
1	–	–	–
1	1	1	–
1	1	1	–
1	–	–	1
–	–	–	1
–	–	–	1
1	1	1	–
1	1	1	–
–	–	–	1
1	–	–	1
1	–	–	1
1	1	1	–
8	5	5	6
–	1	1	1
1	1	1	1
1	1	1	1
1	1	1	1
–	1	1	1
1	1	1	1
–	1	1	1
1	1	1	1
1	1	1	1
1	1	1	1
1	1	1	1
8	11	11	11

All loci

Summary of private alleles

Locus	4n		6n	
	y_{dam}	y_{sire}	x_{dam}	x_{sire}
Spl_101	22	19	41	21
Spl_107	11	22	19	22
Aox_45	22	22	29	22
Spl_105	21	25	22	22
AciG 35	39	25	77	22
Total	115	113	188	109

Fold increase of private alleles

Locus	Alleles	
	Dam	Sire
Spl_101	19	2
Spl_107	8	0
Aox_45	7	0
Spl_105	1	-3
AciG 35	38	-3
Total	73	-4

$$\Delta a_{dam} = (\sum x_{dam})$$

$$\Delta a_{sire} = (\sum x_{sire}) /$$

Captions

x_{dam} = sum of private alleles of dam in spontaneous polyploids

x_{sire} = sum of private alleles of sire in spontaneous polyploids

y_{dam} = sum of private alleles of dam in individuals with typical ploidy

y_{sire} = sum of private alleles of sire in individuals with typical ploidy

Δa_{dam} = increase of dam private alleles at all loci

Δa_{sire} = increase of sire private alleles at all loci

as in 6n individuals
x times

Dam	Sire
1.86	1.11
1.73	1.00
1.32	1.00
1.05	0.88
1.97	0.88
1.63	0.96

Δa_{dam} Δa_{sire}

$/ (\sum y_{dam})$
 $(\sum y_{sire})$

Supplementary material Figure S1

Spontaneous polyploidization in critically endangered *Acipenser mikadoi*

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Yamaha, Shinji Adachi, Katsutoshi Arai

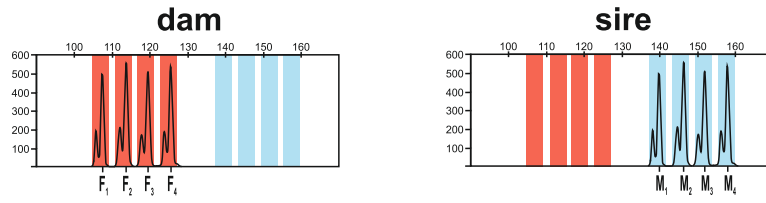
Affiliations: Hokkaido University, Faculty and Graduate School of Fisheries Sciences, 3-
1-1 Minato, Hakodate, Hokkaido 041-8611, Japan

University of South Bohemia in Ceske Budejovice, Faculty of Fisheries and Protection of
Waters, South Bohemian Research Center of Aquaculture and Biodiversity of
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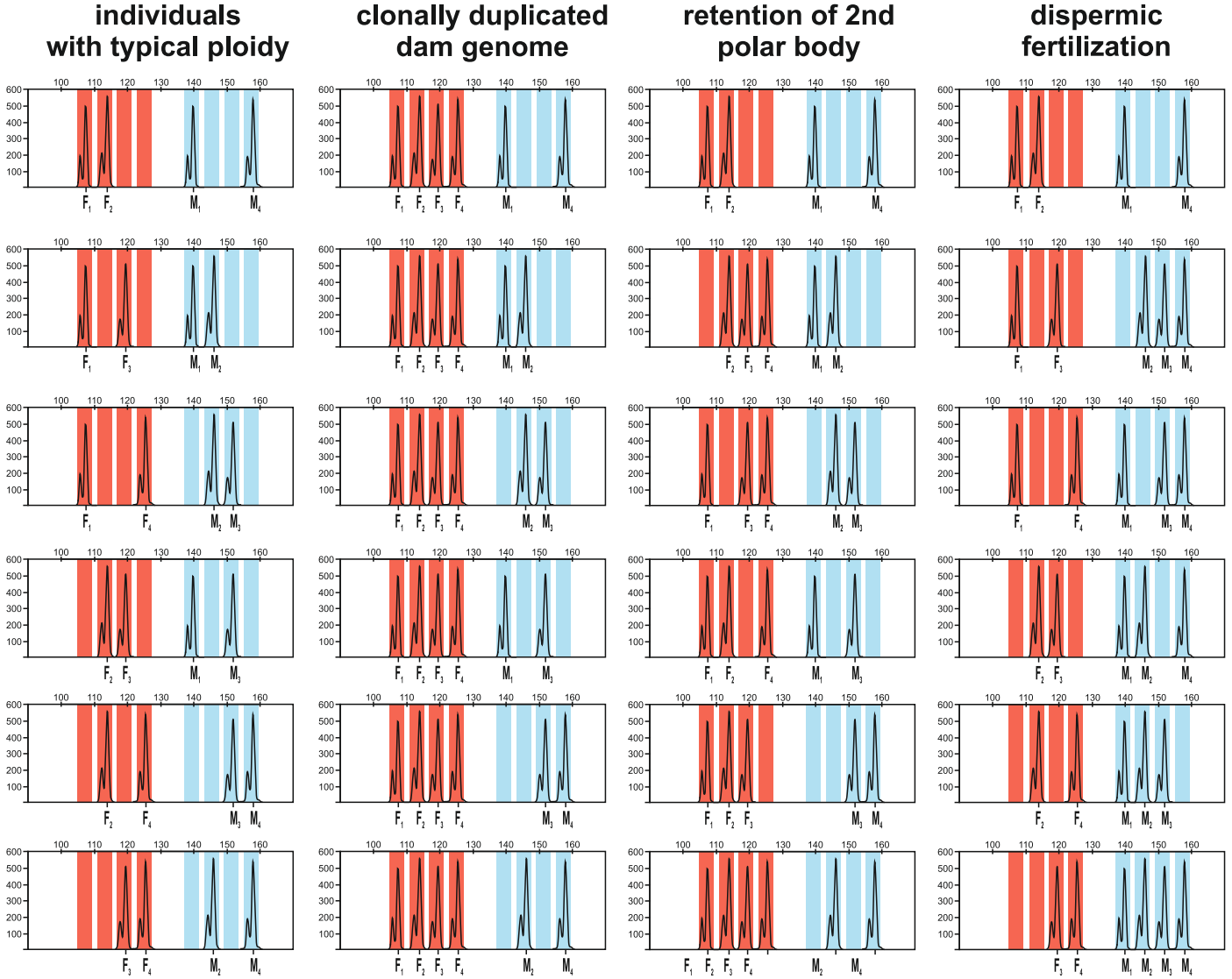
Email: miloshavelka@seznam.cz

Journal: Fisheries Science

P:



F1:



Σ

$$\begin{aligned}\Sigma y_{dam} &= 12 \\ \Sigma y_{sire} &= 12\end{aligned}$$

$$\begin{aligned}\Sigma x_{dam} &= 24 \\ \Sigma x_{sire} &= 12\end{aligned}$$

$$\begin{aligned}\Sigma x_{dam} &= 18^* \\ \Sigma x_{sire} &= 12\end{aligned}$$

$$\begin{aligned}\Sigma x_{dam} &= 12 \\ \Sigma x_{sire} &= 18^*\end{aligned}$$

Δa_i

$$\Delta a_i = (\Sigma x_i) / (\Sigma y_i) \rightarrow \text{where } i = \text{dam or sire}$$

$$\begin{aligned}\Delta a_{dam} &= 24 / 12 = \underline{\underline{2}} \\ \Delta a_{sire} &= 12 / 12 = \underline{\underline{1}}\end{aligned}$$

$$\begin{aligned}\Delta a_{dam} &= 18 / 12 = \underline{\underline{1.67^*}} \\ \Delta a_{sire} &= 12 / 12 = \underline{\underline{1}}\end{aligned}$$

$$\begin{aligned}\Delta a_{dam} &= 12 / 12 = \underline{\underline{1}} \\ \Delta a_{sire} &= 18 / 12 = \underline{\underline{1.67^*}}\end{aligned}$$

Figure S1. Scheme of investigation of the increase of private alleles (Δa_i) in spontaneous polyploids using the following model: $\Delta a_i = (\Sigma x_i) / (\Sigma y_i)$, where Δa_i = increase of private alleles for dam or sire at all loci, Σx_i = sum of private alleles of dam i or sire i at all loci in spontaneous polyploids, and Σy_i = sum of private alleles of dam i or sire i at all loci in individuals with typical ploidy. For simplicity, the scheme includes only one fully heterozygote locus with no allele sharing between parents. The calculation procedure is same for not fully heterozygote loci or in the case when parents share one or more alleles at the locus. Only alleles that are not shared by parents (private alleles) are informative for the analysis. *the number of alleles in spontaneous polyploids and thus Δa_i depend on frequency of recombination as genome duplication occurs after crossing-over.