Genetic tools for traceability of sturgeon aquaculture products, for population assignment in wild sturgeons and for for rehabilitation programs.

Nikolai Mugue

VNIRO, Moscow, Russia

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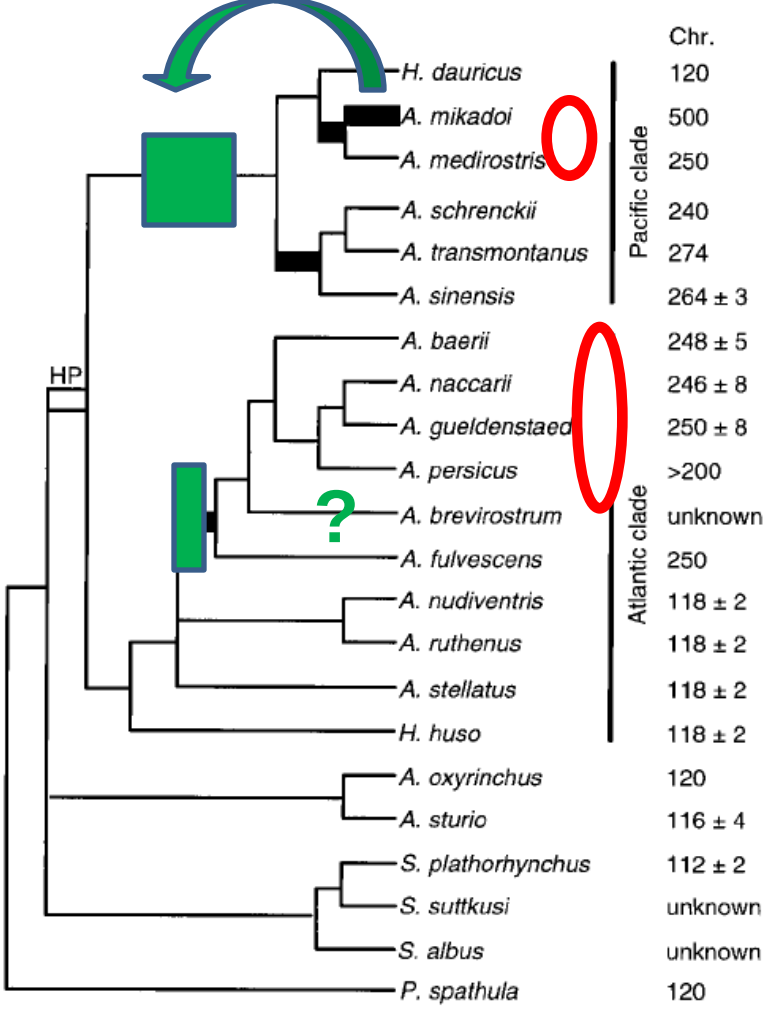
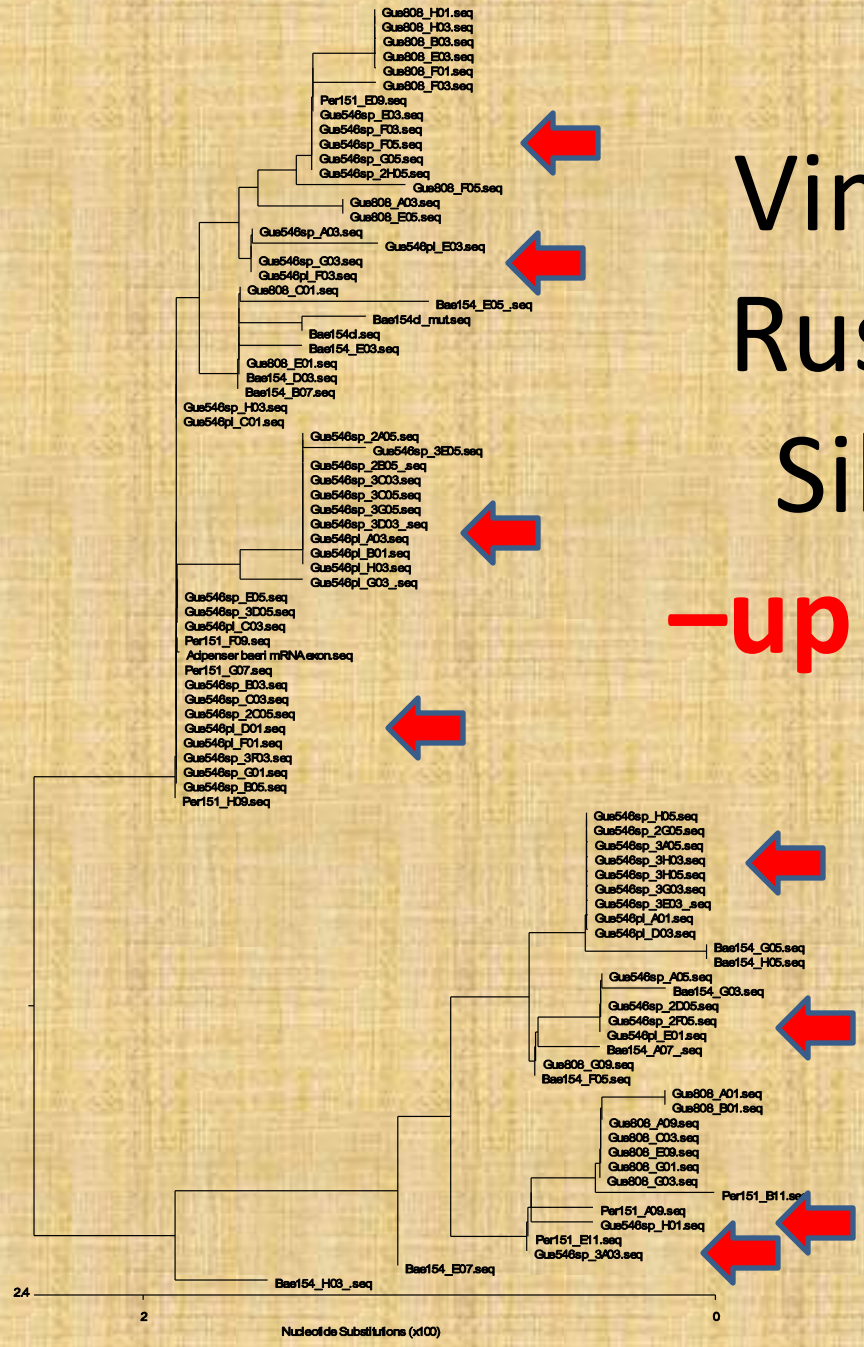


FIGURE 4.—Ploidy evolution in the Acipenseridae, including some related Acipenseriform taxa for comparison. The tree represents the topology, calculated using maximum parsimony, of the entire cytochrome-*b* gene. Polyploidization events are mapped on the tree using black boxes. The existence of heteroplasmy resulting from length variations of the mtD-loop is shown by a white box (data were taken from LUDWIG *et al.* 2000; *A. schrenckii*, *A. sinensis*, and *H. dauricus*; A. LUDWIG, unpublished data; ZHANG 1998). The number of chromosomes is reviewed in BIRSTEIN *et al.* (1997). See also Table 3.

nome Duplication Events and Functional Reduction of Ploidy Levels in Sturgeon (Acipenser, Huso and Scaphirhynchus)

e Ludwig,* Natalia M. Belfiore,† Christian Pitra,‡ Victor Svirsky§ and Ingo Jenneckens**

itute of Freshwater Ecology and Inland Fisheries, 12561 Berlin, Germany, †Department of Animal Science, University of California, Davis, California 95616, ‡Institute for Zoo Biology and Wildlife Research, 10252 Berlin, Germany, §Pacific Research Institute of Fisheries and Oceanography, Vladivostock 690600, Russia and **Institute for Animal Breeding and Genetics, University of Göttingen, 37075 Göttingen, Germany



Vimentin (exon 1) in Russian, Persian and Siberian sturgeons

–up to 8 gene copies in one individual

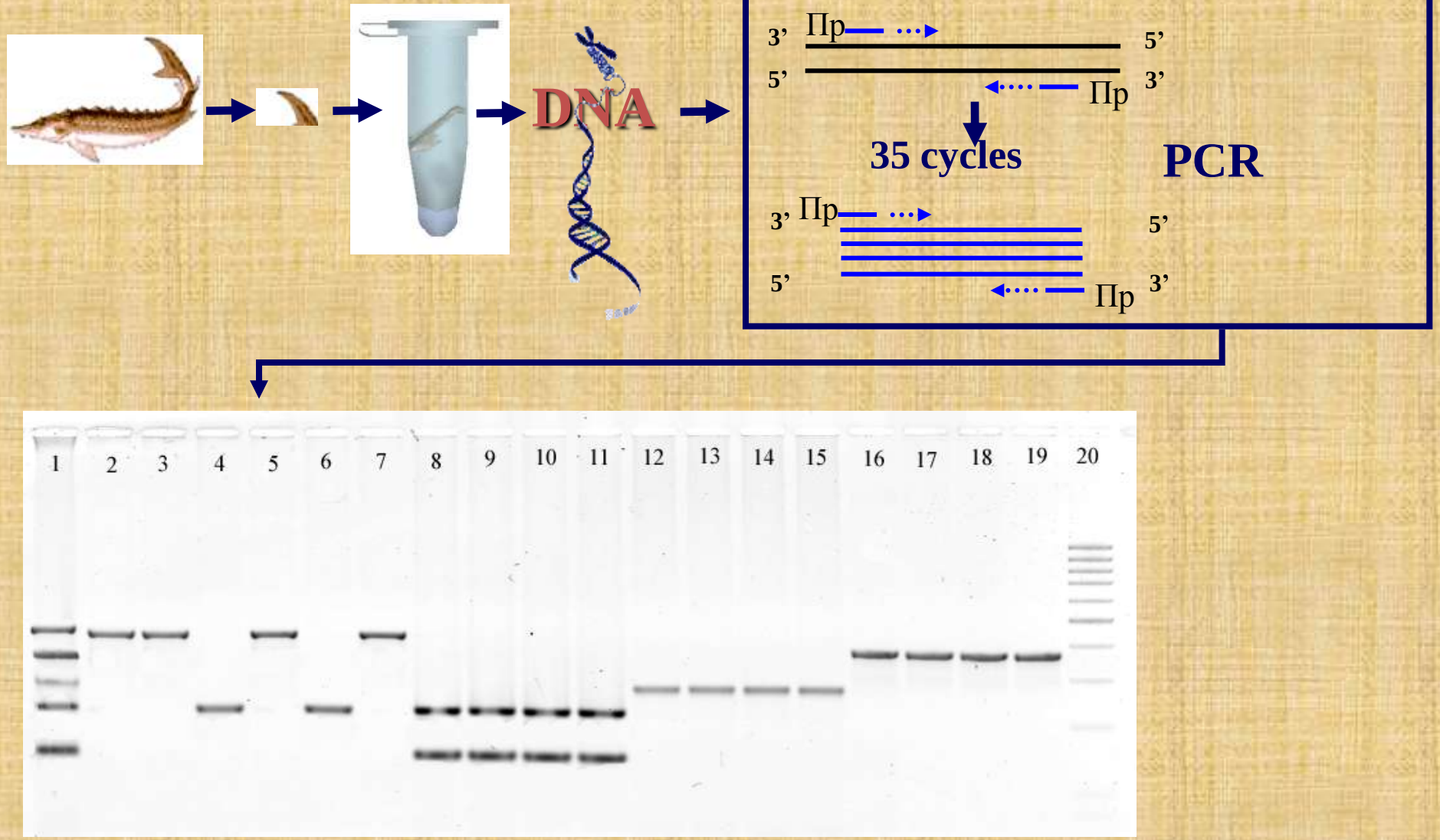
Natural hybridization in Acipenseridae (after Berg, 1948)

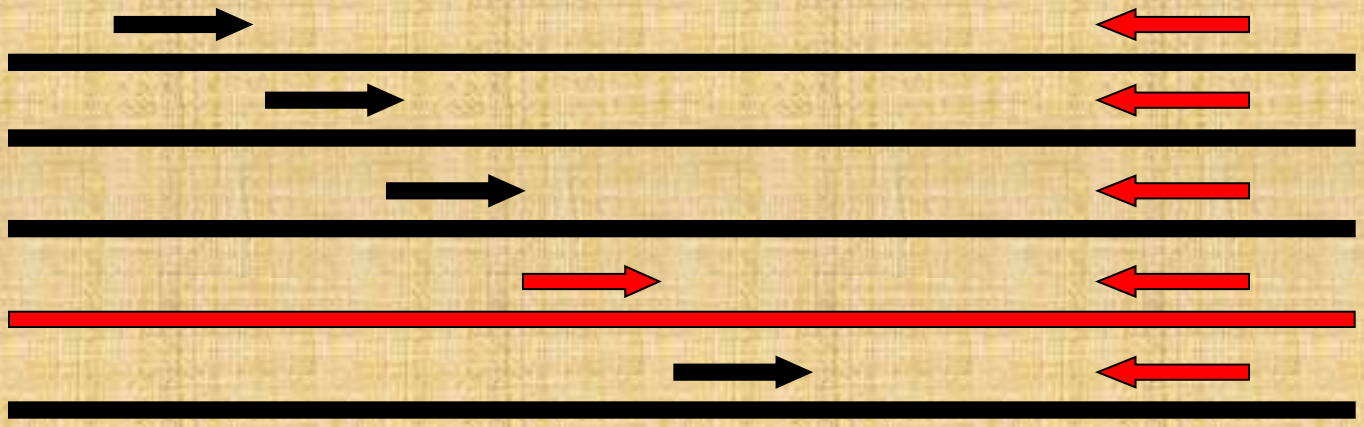
Sturgeons (Acipenseridae) are known to interbreed under natural conditions, giving rise to viable and sometimes fertile interspecific and intergeneric hybrids. Hybrids have been described from crosses of various combinations of almost all species of the family. L.S. Berg in his well-known book "Freshwater fishes of the USSR and the neighboring countries" (1948) enumerates the hybrid forms of this family from the following crossings:

- Huso dauricus × Acipenser schrencki (kaluga × Amur sturgeon)
- H. huso × A. nudiventris (beluga × spiny sturgeon)
- H. huso × A. güldenstädti (beluga × sturgeon)
- H. huso × A. stellatus (beluga × stellate sturgeon)
- A. nudiventris × A. stellatus (spiny sturgeon × stellate sturgeon)
- A. ruthenus × A. güldenstädti (sterlet × sturgeon)
- A. ruthenus × A. stellatus (sterlet × stellate sturgeon)
- A. güldenstädti × A. stellatus (sturgeon × stellate sturgeon)
- A. baeri × A. ruthenus (Siberian sturgeon × sterlet)

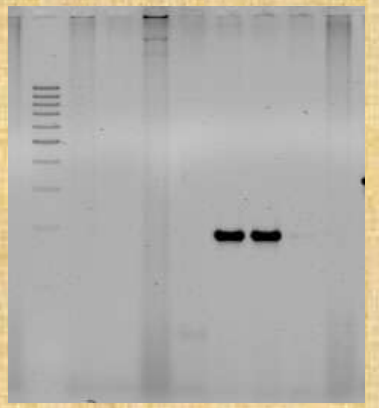
Sturgeon hybrids in industrial aquaculture

- Bester (*Huso huso* x *A. ruthenus*) (3 strains)
- “Rolik” (*A. gueldenstaedtii* x *A. baerii*)
- “AL” (*A. naccarii* x *A. baerii*)
- *Huso dauricus* x *A schrenckii*


DNA
3’ Пр 5’
5’ Пр 3’
35 cycles
PCR
3’ Пр 5’
5’ Пр 3’
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20



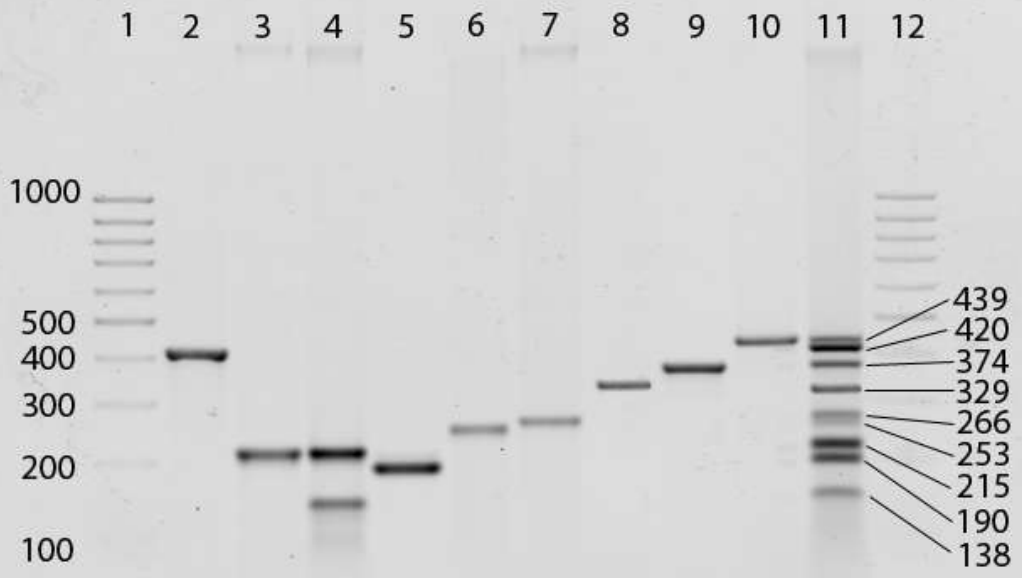
- All PCR products have different length
- Forward primer is species specific, reverse primer is universal
- PCR-identifications can be performed as a set of individual reactions or as a multiplex PCR



Eight species can be unambiguously identified (Mugue *et al*., 2008)

2009 - *Polyodon and Scaphyrhinchus*.

- A. gueldenstaedtii (2,3)
- A. baerii (4)
- A. ruthenus (5)
- A. schrenkii (6)
- A.stellatus (7)
- A. nudiventris (8)
- Huso huso (9)
- Huso dauricus (10)



GENETIC PASSPORTS

Every speciman in the farm ponds:

- Sexed, measured, photographed
- Pit-taged
- Fin-clipped for DNA analysis

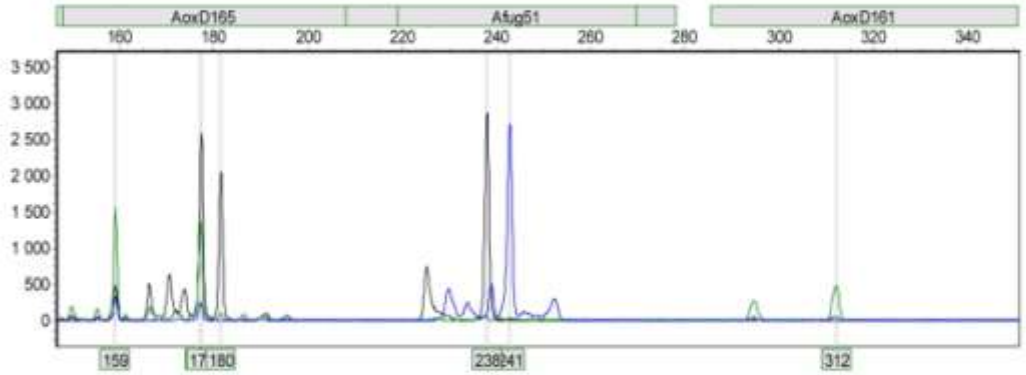
Specimen from wild:

- Location of catch
- Species identification
- Fin-clipped for DNA analysis

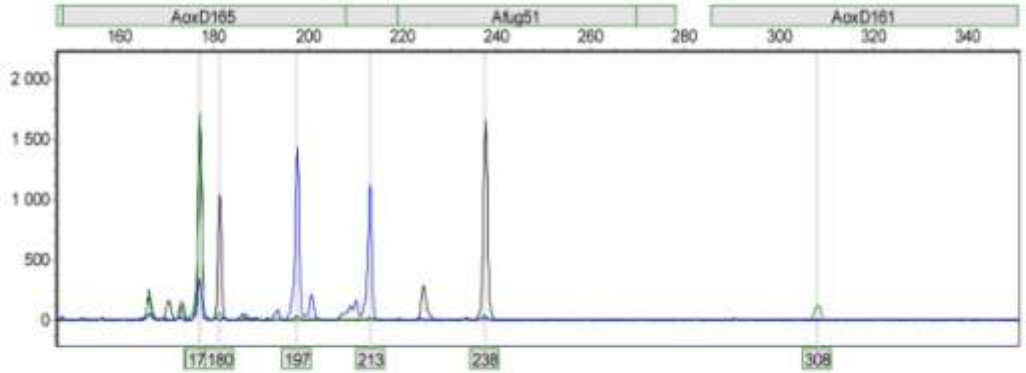
Five highly polymorphic msat loci with 4bp repeat.

Локус	Праймеры 5’-3’
An20	F:AATAACAATCATTACATGAGGCT R:TGGTCAGTTGTTTTTTTATTGAT
AfuG41	F:TGACGCACAGTAGTATTATTTATG R:TGATGTTTGCTGAGGCTTTTC
AoxD165	F: TTTGACAGCTCCTAAGTGATACC R: AAAGCCCTACAACAAATGTCAC
Afug 51	F:ATAATAATGAGCGTGCTTTCTGTT R:ATTCCGCTTGCGACTTATTTA
AoxD161	F:CATTCAGTATGAGACAGACACTC R:ATCTCAGGGACTGCTGTGATTGG

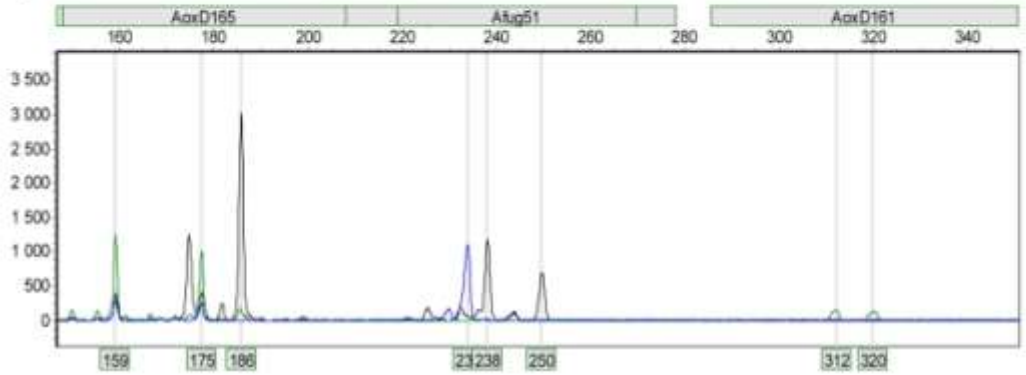
Sample 65



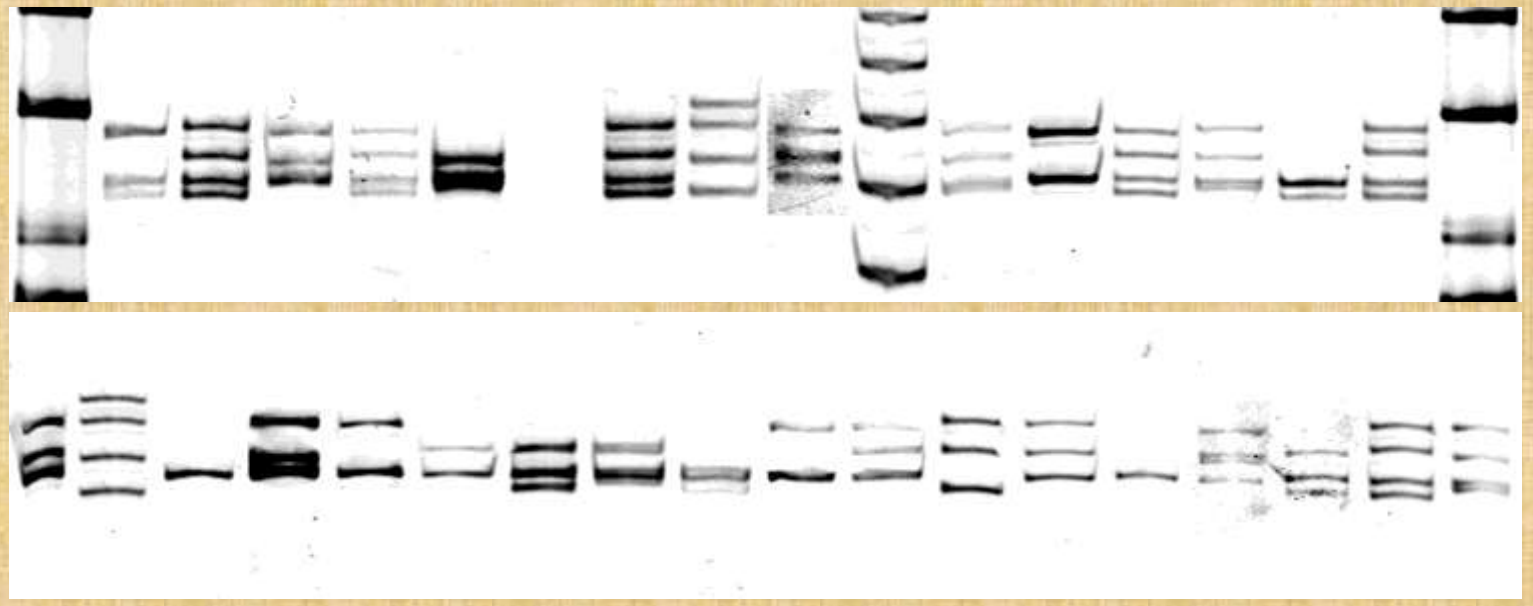
Sample 66



Sample 67



- Five microsatellite loci were screened.
- Four loci (Afug41, Afug51, AoxD165, and An20) demonstrate very high polymorphism and number of alleles.
- Genotyping by set of these loci unambiguously identify each specimen and eggs it produced.



Microsatellite analysis (locus AoxD165) in PAAG

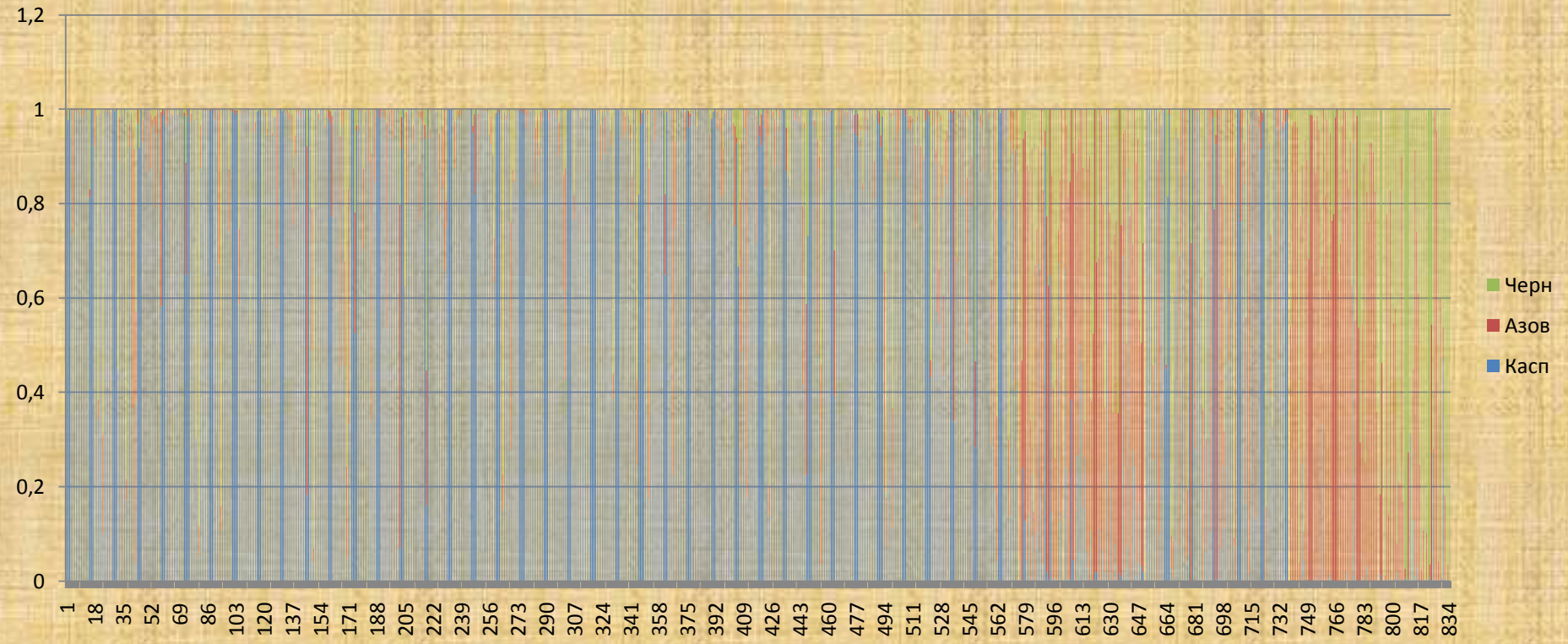
Microsattelite profile for specimen GUE2629 based on four loci:

Locus	An 20	Afug 41	AoxD165	Afug 51
Allele	PCR product length (b.p.)			
A	-	-	200	270x2
B	178x2	-	188	-
C	174	-	180x2	262
D	170	-	-	-
E	-	232		-
F	-	-		226
G	-	-		-
H		216		-
I		-		-
J		-		
K		204x2		
L		-		

Traceability of aquacultured (vs. wild) caviar

- All females in fish farm MUST have genetic passports (pit-tagged and genotyped for 5 ms loci)
- Caviar for testing is provided along with list of female tags used as a caviar source (easy test fo positive IDs)
- More complicated – assignment test for given fishfarm populations (no data on females provided, or forensic request

Population assignment by microsattelites



Assessment of Sturgeon plant efficacy via parental assignment of sturgeons collected in the sea

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Assessment of national input in sturgeon restocking activity

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Mechanism of sturgeon homing

- Very little knowledge of mechanisms
- Age of imprinting of chemical stimuli?
- Role of genetics?

Restocking: where fish should be taken from?

- **Best – from the local spawning populations!!!**
- **But:**
- Shortage of both male and female specimens at the same time
- Loss of genetic variation if founders number is too small due to inbreeding

Specimens caught in the coastal waters –valuable genetic reference

AND

source of boodstock for restocking

But: It is feeding migrations, not a spawning migration (origin is unknown).

Importance of Aquaculture

- Good experience and knowledge of biology
- Live genebank (broodstock) handy
- Choice of matured producers

Pitfalls:

- Selection and domestication vs. maintenance of natural genetic polymorphism
- Threat of genetic pollution (escapees, release)