Sturgeon Species and Hybrids: Can Hybrids Produce Caviar?

by Vadim J. Birstein*

Four years ago, on 1 April 1998, the listing of all sturgeon species became effective under the Convention on the International Trade in Endangered Species of Wild Fauna and Flora (CITES). As the author of the draft proposal for the listing, I have kept my eye on the development of events with great interest. Not everything went the way it was discussed by the environmental community before the listing (in brief see, for instance, DeSalle and Birstein, 1996). Also, it became clear that the question of the Caspian Sea is a political, not scientific, issue (UNEP, 2002). But, still, political decisions regarding the caviar trade inevitably need to be based on biological characteristics of sturgeon. In this article I would like to make several comments on scientific aspects of the sturgeon law enforcement.

I have already tried to attract the attention of conservation biologists and environmentalists to the following problems. First, the CITES Secretariat based its own evaluation of the sturgeon situation and caviar production on official governmental data. In the meantime, evaluation by independent observers such as TRAFFIC Europe showed that, for instance, the official data provided by the Russian government is far from reality (TRAFFIC Europe-Russia, 1999). Second, the CITES listing has created a serious problem for sturgeon scientists. Since the CITES implementation covers tissue and egg samples necessary for any DNA research, it has become impossible to study the most endangered sturgeon species by independent scientists not working in governmental institutions. In many countries the local CITES authorities do not issue CITES permits for scientific samples (Birstein, 1999; Birstein and Doukakis, 2001). Third, the CITES Secretariat accepted the situation when governmental environmental law enforcement institutions used DNA methods that had not been peer reviewed for law implementation. For instance, for four years the US Fish and Wildlife Service (FWS) has been using its own DNA caviar identification method that had never been scientifically proven and reviewed by independent experts (Birstein, 1999, 2000; Birstein and Doukakis, 2001). This is absolutely unacceptable from a scientific point of view.

The most disturbing factor is that the CITES listing and its implementation are not based on current scientific knowledge. In this article I want to discuss two examples that illustrate this point. First, the CITES listing does not consider a change in the number of sturgeon species due to new genetic data. Second, as I know from my Russian colleagues, the CITES Secretariat adopted a theory in its everyday work that fertile female hybrid sturgeon capable of producing caviar are thriving in the Caspian Sea.

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How many sturgeon species are there in the wild?

While working with different international environmental institutions, environmental lawyers, etc., I realized that most people who were not trained as professional biologists have a problem with understanding that the concept of species changes with time and, as a result, the number of species within a particular animal group can also change. In fact, this number depends on current scientific knowledge and the opinion of specialists working with the group (Wheeler and Meir, 2000). Unfortunately, CITES was written as a very rigid legal document without the flexibility necessary to include new scientific discoveries or points of view. The sturgeon listing is a good example of this problem.

In 1997, my colleague, Dr Willy Bemis, and I discussed how professional opinion on the number of sturgeon species has been changing since the first description of sturgeon species in the 18th century (Birstein and Bemis, 1997). Based on morphological differences, in the mid-19th century several scores of sturgeon species living in Eurasia were described by specialists. However, at the beginning of the 20th century, the Russian ichthyologist Leo Berg sorted out the previously known data and concluded that there were only about 17 sturgeon species worldwide (Berg, 1904). According to his opinion, there were twelve *Acipenser*, two *Huso* and three *Pseudoscaphirhynchus* species in Eurasia. Of these species, later two of Berg's species, *A. stenorhynchus* and *A. baerii*, were merged into one, the Siberian sturgeon (Ruban, 1991). The existence of another species, *A. kikuchii*, was not confirmed. The existence of one more species, *A. mikadoi*, was recently genetically confirmed, contrary to the current opinion of ichthyologists who used only morphological data (Birstein *et al.*, 1993; 2002; Birstein and DeSalle, 1998; Ludwig *et al.*, 2001). Dr Bemis and I suggested adopting 25 extant sturgeon species with the understanding that new data would introduce corrections in this number.

The morphological studies published since then have shown that the traditional division of some of sturgeon species into subspecies was not scientifically strongly supported. Thus, Dr Georgii Ruban did not confirm the existence of three subspecies within the Siberian sturgeon, *A. baerii* (Ruban, 1991). According to his results, *A. baerii* is a highly morphologically variable species without subspecies.

According to the new molecular data, more corrections in the sturgeon systematics are needed. For example, our genetic data do not support the recent elevation of the Persian sturgeon to a species level, *A. persicus* (Birstein, 1999; Birstein *et al.*, 2000a). It appears that the European sturgeon, *A. sturio*, consists of two genetic forms

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(Birstein *et al.*, 1998; Birstein and Doukakis, 2000). The most recent molecular study has changed the previous understanding of the relationships between the European and American Atlantic sturgeon, *A. sturio* and *A. oxyrinchus* (Ludwig *et al.*, 2002b). Moreover, there are problems with molecular discrimination between three North American species of the genus *Scaphirhynchus* (Campton *et al.*, 2000).

All these data demonstrate that there is no clear opinion on how many sturgeon species there are in the wild. Evidently, from the beginning of the implementation, the CITES listing of sturgeon was outdated and did not reflect present scientific knowledge. A simple and efficient procedure to make changes in the listing based on new scientific evidence should be introduced. The existing procedure of a change in the species status through the Animals Commission takes years and is not efficient enough for immediate inclusion of new data.

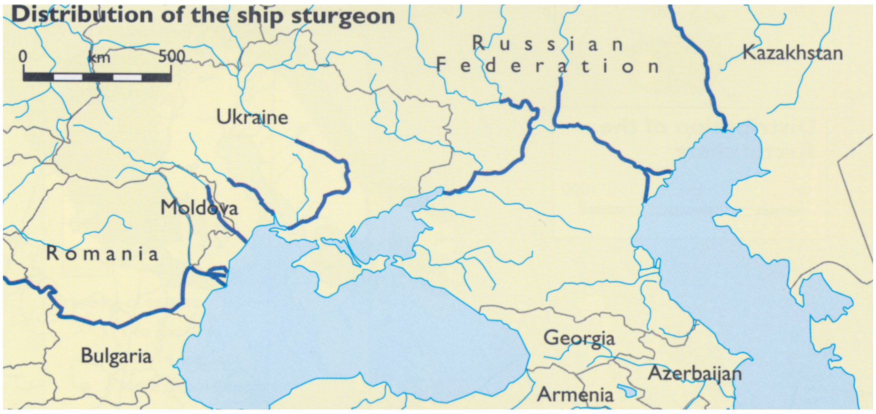
An unexpected discovery

In 2000, two independent groups of scientists working on the mitochondrial (mt) DNA discovered two genetic forms within the Caspian Sea population of the Russian sturgeon, *A. gueldenstaedtii* (Birstein *et al.*, 2000a; Jennikens *et al.*, 2000). Individuals of the "typical" form have mtDNA similar to that of the Russian sturgeon from different locations – the Black, Azov and Caspian seas, while sturgeon of the other form have mtDNA similar to that of Siberian sturgeon. However, some characteristics of mtDNA of the second form and of Siberian sturgeon living in Siberia are different. We named the second form the "Siberian sturgeon-like" or "*A. baerii*-like." Amazingly, the sturgeon with these two different types of mtDNA are morphologically indistinguishable (we now have scientific proof of this) and the local Caspian Sea fishermen do not discriminate between them. In our study we demonstrated that the "*A. baerii*-like" form is present in both the northern (the Volga River basin) and southern (Iranian waters) parts of the Caspian Sea and it comprises approximately 30 per cent of the population of the Russian sturgeon.

We suggested that the "*A. baerii*-like" Russian sturgeon represents a form that is closely related to the ancestors of the Siberian sturgeon and, possibly, is a cryptic, *i.e.*, morphologically similar to the Russian sturgeon, unknown species. In our opinion, the Siberian sturgeon may be a "young" species that populated the Siberian basins when they were interconnected approximately 8,000 years ago (Birstein and DeSalle, 1998). During that geological period, a system of continuing glacial lakes connected all Siberian rivers with the Caspian Sea basin. Therefore, it is possible that a form closely related to the ancestral form of *A. baerii* that populated Siberian rivers still lives in the Caspian Sea (Birstein *et al.*, 2000a).

Instead of discussing this or any other scientifically-based hypothesis, the US Fish and Wildlife Service (FWS) interpreted the presence of the *A. baerii*-like mtDNA in some Russian sturgeon individuals as proof of the existence of fertile *A. baerii*/*A. gueldenstaedtii* hybrids produced from a cross between *A. baerii* females and *A. gueldenstaedtii* males in the northern part of the Caspian Sea. Currently, the US FWS confiscates caviar shipments if it identifies "the Siberian sturgeon caviar" by means of its mtDNA-based method. Its reason for this action is that the Russian, and not Siberian, sturgeon was included in the CITES documentation.

Such actions have nothing to do with the legal imple-



Distribution of the ship sturgeon (*Acipenser nudiventris*)
Source: P. S. Maitland, personal communication

The ship sturgeon (*Acipenser nudiventris*) is one of several species of sturgeons found in Europe. All sturgeon have a similar life history, all are important economically (both for caviar and for their flesh) and all are severely threatened. The ship sturgeon is found in the Black, Azov and Caspian seas and formerly migrated into most of the larger, northern rivers associated with these seas. Now, due to overfishing, pollution and the construction of river barriers, its numbers and distribution are much reduced.

Fishing for ship sturgeon, Volga
Source: W. Fisher, WWF
Courtesy: Europe's Environment

mentation. Even according to a common-sense approach, fishermen and caviar businessmen should not be punished for what they cannot know – that caviar was taken from females that had the "*A. baerii*-like" type of mtDNA. This characteristic can be discovered only by specially trained scientific staff in a specially equipped laboratory, not by fishermen who catch sturgeon in primitive conditions in the wilderness of the Volga River Delta.

The hypothesis of "fertile *A. baerii/A. gueldenstaedtii* hybrids" contradicts important biological principles and scientific data. To show how "revolutionary" the theory of these fertile hybrids is, let me describe what "hybrids" mean in biological and genetic terms.

On hybrids and fertility

In general, interspecies hybridization between vertebrate species is a rare event due to the genetic incompatibility of parental genomes (Arnold, 1997). In other words, an overwhelming number of interspecies hybrids is not viable. Even if the interspecies vertebrate hybrids survive, they are sterile because genetic incompatibility prevents normal development of their gonads. The hybrids that survive manifest a mixture of morphological characteristics of both parental species. A mule, the sterile offspring of a female horse and a male donkey, is a good example of a viable hybrid.

Natural interspecies hybridization happens more frequently between closely related fish species and some amphibians than in other groups of vertebrates. Due to the unusual genetic structure of sturgeon (all are polyploids, *i.e.* they have four or eight chromosome sets instead of two), sturgeon species hybridize more easily than other fish (Birstein *et al.*, 1997). According to the morphological description, hybrids between many sturgeon species can survive in the wild (see Table 5 in Birstein *et al.*, 1997).

However, a morphological description is not enough to prove that a particular individual is a hybrid. Only a genetic study can provide the necessary proof that nuclear genes from both parental species are present in the hybrid. Using genetic methods, natural hybridization between the beluga (*Huso huso*) and Russian sturgeon in the Danube River has been demonstrated (Radu Suciu, personal communication) Also, natural hybrids between female Russian sturgeon and male stellate and sterlet were found in the Volga River, and between the Russian or Persian (*A. persicus*) female sturgeon and male ship sturgeon (*A. nudiventris*) in the Iranian waters of the Caspian Sea (Ludwig *et al.*, 2002a). These three types of hybrid are sterile because their parental species have different numbers of chromosome sets. On the whole, no mature adult sturgeon hybrid that could potentially produce caviar has ever been found in the wild.

The natural hybrids between *A. baerii* females and *A. gueldenstaedtii* males have never been caught, simply because geographic areas of these species do not overlap. Siberian sturgeon live only in the main Siberian rivers and in Lake Baikal (Ruban, 1991). There is no physical way for female Siberian sturgeon to reach the Caspian Sea and spawn there with male Russian sturgeon because the Caspian Sea is far away from Siberia, and the Ural Mountains divide these two distant areas. Moreover, the Siberian sturgeon is a freshwater species that never enters the sea, whereas the Russian sturgeon is an anadromous sturgeon, *i.e.* it lives in the sea and only spawns in rivers.

In special laboratory conditions the viability of different sturgeon interspecies hybrids is higher than in the wild (Burtsev, 1997). As a rule, artificially obtained sturgeon female hybrids of the first generation between the parental species of the same ploidy (*i.e.* those with the same number of chromosomes, four or eight sets) can produce eggs, while hybrids from the crosses of parental species of different ploidy (four and eight sets of chromosomes), are sterile (Arefyev, 1997, 1998). Some second-generation females of one of the artificial hybrids, bester (a laboratory-produced hybrid between the beluga females and male *A. ruthenus* sterlets) are fertile (Burtsev, 1997). However, the Russian scientist who created this hybrid, Dr Nikolyukin, always stressed the fact that besters had never been found in the wild (Nikolyukin, 1964).

The artificial hybrids between female Siberian sturgeon and male Russian sturgeon have never been studied in the laboratory and it is not even known if they can survive in the wild. Only hybrids from the cross between female Russian sturgeon and male Siberian sturgeon (*A. gueldenstaedtii/A. baerii)* were obtained artificially and studied (Arefyev, 1997). These hybrids can mature and produce eggs, but the second generation of hybrids is not viable. Therefore, any discussion of the *A. baerii/A. gueldenstaedtii* viable and fertile hybrids that can produce caviar is pure speculation.

Could numerous female Siberian sturgeon appear in the Caspian Sea?

Since mtDNA is maternally inherited and only sturgeon hybrids of the first generation can theoretically be fertile, the presumption that, contrary to all scientific data, there are numerous fertile *A. baerii/A. gueldenstaedtii* in the Caspian Sea that produce caviar, suggests the existence of female Siberian sturgeon that cross with male Russian sturgeon. These females should also be numerous because, as we now know, the "*A. baerii*-like" form comprises up to 30 per cent of the population of the Russian sturgeon in this sea. However, no adult female Siberian sturgeon has ever been recorded in this area.

As I have already mentioned, Siberian sturgeon cannot move from Siberia to the Caspian Sea. It is true that small numbers of Siberian sturgeon fry were released several times in the 1960s and 1970s into the reservoirs of the upper reaches of the Volga River. However, the release of these fry could not result in the creation of a huge population of Siberian sturgeon spawners in the lower reaches of the Volga River. There are three reasons for this conclusion. First, according to the available data of the Ichthyologic Commission in Moscow, the total number of Siberian sturgeon fry released between 1961 and 1975 was approximately 650,000. The fry were released into the Gorky, Saratov and Volgograd reservoirs above the Volgograd Dam. This number is small compared with the hundreds of millions of Russian sturgeon fry released in the lower part of the Volga River during the same period

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(Khodorevskaya *et al.*, 1997). Second, no Siberian sturgeon could ever get through the turbines of the hydroelectric stations on the lower Volga River. In other words, these young Siberian sturgeon were physically cut off from the spawning sites of the Russian sturgeon located below the Volgograd Dam. And third, there was no record of the catch of even one mature female Siberian sturgeon in any of the Volga River reservoirs.

Theoretically, there is a very low possibility that a fertile interspecies fish hybrid of the first generation can produce viable eggs that would be fertilized by the sperm of the second parental species. In such a case the number of nuclear genes of the first parental female will decrease with each subsequent cross, and as a result of this long process eggs of one fish species can have mtDNA of another species, coming from the egg of the first cross. This process is called "the genetic introgression". However, this process has never been reported for sturgeon and it could not create 30 per cent of individuals of the "*A. baerii*-like" form within the population of Russian sturgeon in two or three generations.

There was not enough time for the appearance of such a high number of "*A. baerii*-like" individuals through recent introgression. Even if one imagines that against all odds several female Siberian sturgeon could mature above the Volgogard Dam, their maturation would have taken six to eight years. If to continue speculating that these several females managed to sneak through the Volgogard Dam turbines and breed with male Russian sturgeon, and that these rare hybrids survived and matured in the Caspian Sea (another speculation), their maturation would have taken six to eight more years. And so on. Thirty per cent of the Russian sturgeon population could not acquire mtDNA from several Siberian sturgeon females in only 30–40 years.

Since the "*A. baerii*-like" form was found in the Volga River and in the Iranian waters, to follow the hybrid hypothesis, the female Siberian sturgeon should also exist in the rivers of the Iranian coast. No Siberian sturgeon has ever been released into Iranian rivers.

If one were to suggest that the Siberian sturgeon is an "old" species which originated before the last period of glaciation, the introgression could have happened 8,000 years ago during the postglacial period when Siberian rivers, as I have already mentioned, were connected with the Caspian Sea basin. But such an event had nothing to do with the Siberian sturgeon that, 30–40 years ago, were released into the Volga River basin and that supposedly "hybridized" with the local Russian sturgeon.

Therefore, there is no current scientific basis for speculations about mysterious fertile hybrids between female Siberian sturgeon and male Russian sturgeon in the Caspian Sea that produce caviar. Only a special genetic study of the nuclear, not mitochondrial, DNA can prove that the "*A. baerii*-like" individuals are hybrids of the first generation if they have in fact genes inherited from both parental species, the Siberian and Russian sturgeon. Unfortunately, technically it is not easy to conduct such a study, and these experiments need to wait for the future.

In science, only data professionally described and accepted by the scientific community through evaluation by independent experts and publication becomes scientific fact. According to this rule, any conclusion about the "fertile sturgeon hybrids" should be based on the results of at least the following types of research:

1. a genetic study of the maternal and paternal nuclear genes in the "*A. baerii*-like" individuals;
2. a study of the ability of Siberian/Russian sturgeon hybrids to survive in seawater; and
3. a study of the ability of Siberian/Russian sturgeon hybrids to mature at all and produce eggs.

Any conclusion on the existence of fertile hybrids made by the US FWS and the CITES Secretariat *before* these studies are carried out is ridiculous and makes no scientific sense.

Conclusions

As a concerned biologist, I would like to summarize my recommendations for the CITES implementation as follows:

(1) An efficient mechanism of changes in the CITES listing is needed to follow rapid developments in scientific data on the number of sturgeon and other species. I realize that the creation of such a mechanism will take a lot of time and thinking. But this mechanism is urgently needed if the environmental community wants to have a real impact of its laws on the fate of endangered species. There is little time left for endangered species such as sturgeon for long bureaucratic procedures that take many years to change the species status.

(2) The work of the CITES Secretariat should not be based on pure speculation that contradicts basic scientific knowledge in biology and genetics. Numerous fertile hybrid sturgeon will not appear in the Caspian Sea because the US FWS or the CITES Secretariat have decided that they should exist. If such a practice of the environmental law institutions to misinterpret scientific data continues, there will be a huge gap between science and environmental law institutions.

(3) The CITES Secretariat must not support the US FWS actions against the "Siberian sturgeon caviar" shipments, since at present there is no scientific data on the existence or even potential possibility of the existence of fertile *A. baerii/A. gueldenstaedtii* hybrids in the Caspian Sea.

References

Arefyev, V.A. (1997) Sturgeon hybrids: Natural reality and practical prospects. *Aquaculture Magazine* 23: 53–58.

Arefyev, V.A. (1998) Sturgeon hybrids: Natural reality and practical prospects. *Aquaculture Magazine* 24: 44–50.

Arnold, M.L. (1997) *Natural Hybridization and Evolution*. Oxford University Press, NY.

Berg, L. (1904) Zur Systematik der Acipenseriden. *Zoologische Anzeiger* 27: 665–667.

Birstein, V. (1999) New perspectives for monitoring migratory animals – improving knowledge for conservation. *Environmental Policy and Law* 29(5): 235–238.

Birstein, V.J. (2000) Sturgeon and Paddlefishes (Acipenseriformes). In

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R.P. Reading and B. Miller (eds) *Endangered Animals: A Reference Guide to Conflicting Issues*. Greenwood Press, Westport, CT, pp. 269–278.

Birstein, V.J. and Bemis, W.E. (1997) How many species are there within the genus *Acipenser*? In V.J. Birstein, J.R. Waldman and W.E. Bemis (eds) *Sturgeon Biodiversity and Conservation*. Kluwer Academic Publishers, Dordrecht, pp. 157–163.

Birstein, V.J. and DeSalle, R. (1998) Molecular phylogeny of Acipenserinae. *Molecular Phylogenetics and Evolution* 9: 141–151.

Birstein, V.J., Betts, J. and DeSalle, R. (1998) Molecular identification of *Acipenser sturio* specimens: a warning note for recovery plans. *Biological Conservation* 84: 97–101.

Birstein, V.J. and Doukakis, P. (2000) Molecular analysis of *Acipenser sturio,* L., 1758 and *A. oxyrinchus* Mitchill, 1815: A review. *Boletín Instituto Español de Oceanografia* 16(1–4): 61–73.

Birstein, V.J. and Doukakis, P. (2001) Molecular identification of sturgeon species: science, bureaucracy, and the impact of environmental agreements. In K. Riede (ed.) *New Perspectives for Monitoring for Migratory Animals – Improving Knowledge for Conservation.* Landwirtschaftsverlag GmbH, Munster, Germany, pp. 47–63.

Birstein, V.J., Doukakis, P. and DeSalle, R. (2000a) Polyphyletic genetic structure of the Russian sturgeon and caviar species identification. *Conservation Genetics* 1(1): 81–88.

Birstein, V.J., Doukakis, P., Ruban, G.I. and DeSalle, R. (2000b) A new cryptic species within the Russian sturgeon in the Caspian Sea: The first molecular evidence. In *Proceedings of the International Conference "Sturgeon on the Boundary of the 21st Century",* Astrakhan, Russia, 11–15 September, p. 124 (in Russian).

Birstein, V.J., Hanner, R. and DeSalle, R. (1997) Phylogeny of the Acipenseriformes: cytogenetic and molecular approaches. In V.J. Birstein, J.R. Waldman and W.E. Bemis (eds) *Sturgeon Biodiversity and Conservation*. Kluwer Academic Publishers, Dordrecht, pp. 127–155.

Birstein, V.J., Doukakis, P. and DeSalle, R. (2002) Molecular phylogeny of Acipenseridae: Nonmonophyly of Scaphirhynchinae. *Copeia* 2: 287–301.

Birstein, V.J., Poletaev, A.I. and Goncharov, B.F. (1993) The DNA content in Eurasian sturgeon species determined by flow cytometry. *Cytometry* 14: 377–383.

Burtsev (1997) Bester in aquaculture. In V.J. Birstein, A. Bauer and A. Kaiser-Pohlmann (eds) *Sturgeon Stocks and Caviar Trade Workshop*. IUCN, Gland, Switzerland and Cambridge, UK, pp. 71–88.

Campton, D.E., Bass, A.L., Chapman, F.A. and Bowen, B.W. (2000) Genetic distribution of pallid, shovelnose and Alabama sturgeon: Emerging species and the US Endangered Species Act. *Conservation Genetics* 1: 17–32.

DeSalle, R. and Birstein, V.J. (1996) PCR identification of black caviar. *Nature* 381: 197–198.

Jennikens, I., Meyer, J.-N., Debus, L., Pitra, C. and Ludwig, A. (2000) Evidence of mitochondrial DNA clones of Siberian sturgeon, *Acipenser baerii*, within Russian sturgeon, *Acipenser gueldenstaedtii*. *FEBS Letters* 3: 503–508.

Khodorevskaya, R.P., Dovgopol, G.F., Zhuravleva, O.L. and Vlasenko, A.D. (1997) Present status of commercial stocks of sturgeon in the Caspian Sea basin. In V.J. Birstein, J.R. Waldman and W.E. Bemis (eds) *Sturgeon Biodiversity and Conservation*. Kluwer Academic Publishers, Dordrecht, pp. 209–219.

Ludwig, A., Belfiore, N.M., Pitra, C., Svirsky, V. and Jenneckens, I. (2001) Genome duplication events and functional reduction of ploidy levels in sturgeon (*Acipenser*, *Huso* and *Scaphirhynchus*). *Genetics* 158: 1203–1215.

Ludwig, A., Debus, L. and Jenneckens, I. (2002a) Molecular methods of discriminating between caviar of different sturgeon species. In F. Kirschbaum, R. Billard, J. Gessner and M. Wirth (eds) *Caviar Production and Breeding, Conservation and Product Processing of Sturgeon* (in press).

Ludwig, A., Debus, L., Lieckfeldt, D., Wirgin, I., Benecke, N., Jenneckens, I., Williot, P., Waldman, P.J. and Pitra, C. (2002b) American Atlantic sturgeon went East. *Nature* (submitted).

Nikolyukin, N.I. (1964) Some observations on the histological structure of the gonads of sturgeon hybrids. *Trudy VNIRO* **55**: 145–157 (in Russian).

Ruban, G.I. (1991) *The Siberian Sturgeon,* Acipenser baerii *Brandt: The Structure of the Species and Its Ecology* (in Russian).

TRAFFIC Europe-Russia (1999) *Estimation of Stock and Population Conditions of Sturgeon in Russia and Monitoring of Domestic Trade in Sturgeon Products* (the first view pilot project). A report prepared for the Sea Shepherd Conservation Society (USA), Moscow.

UNEP (2002) Caspian Sea countries set to resume caviar trade. Press release by Daniel Dubois (UNEP) posted on 6 March.

Wheeler, Q.D. and Meier, R. (eds) (2000) *Species Concepts and Phylogenetic Theory. A Debate*. Columbia University Press, NY.