A review on genetic studies in sturgeons and their trade control in China

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Summary

Eight native species of Acipenseriformes mainly inhabit three geographical regions in China: the Yangtze system, the Amur system and the Xinjiang area. All sturgeon populations in China have suffered from various anthropogenic impacts, pushing them to rarity or extirpation in most of their historical range. Sturgeon aquaculture in China has become popular since 1998 with eleven species and hybrids presently being cultured. In this paper, we firstly review the development and current status of aquaculture with sturgeons and paddlefish in China. Secondly, we present some of the main conclusions that can be drawn from previous conservation genetic and phylogenetic studies in native sturgeon species. The sturgeon trade control in China is also reported in this review. Considering the outcome of all previous studies, we conclude, firstly, in sturgeon aquaculture, capture breeding program is the key to establish a sustainable sturgeon culture industry in China, which will also help sturgeon conservation. Secondly, in conservation genetics, the influence of restocking and release programmes on native populations need to be assessed much more comprehensively in order to avoid admixture and hybridization among genetically different spawning stocks. This holds especially for Chinese sturgeon and Amur sturgeon. Thirdly, in trade control, considering the current endangered status of this species, catch quotas should be based on the results of scientific investigations rather than on official statistics of harvest records. Finally, species identification methods based on the combination of mitochondrial and nuclear markers are still insufficiently developed in China and in need of rapid attention. They are the first step in structuring and implementing a successful conservation programme and provide the necessary tools for effective international trade control.

Introduction

Once widely distributed and highly abundant in the northern Hemisphere (Billard and Lecointre, 2001), sturgeon species today exist as fragmented populations occupying limited geographic areas, showing an alarming decline in adult abundance. According to the International Union for the Conservation of Nature and Natural Resources (IUCN), most of the species are threatened or endangered because of various human activities. Some populations are rare or even extinct in their original habitats. Therefore, due to the critical status of these species, as well as their high economic value and significance in phylogenetic evolution, Acipenseriformes have been of critical concern to these involved in species conservation (Rosenthal et al., 2006).

Eight species of Acipenseriformes are native in China, mainly distributed in the Xinjiang Area and the rivers Yangtze and Amur. Huso dauricus and Acipenser schrenckii are shared with Russia in the Amur River system, and A. baerii and A. ruthenus with Kazakhstan and Russia in the Irtysh River. A. nudoventris occurs in the Lii River, which is a tributary of Lake Balkhash in Kazakhstan. The remaining three species, A. sinensis, A. dabryanus and Psephurus gladius are endemic to China; particularly distributed in the Yangtze River and Sea of China (Wei et al., 1997). Like other sturgeon species, all stocks of sturgeon populations in China have suffered from overfishing, poaching, barriers to migration, loss of spawning habitat and the deterioration of water quality. However, the combination of the various anthropogenic factors have different impacts for each species, and are also particular to each river system. Overexploitations and poaching for caviar production are responsible for the significant decline in Amur stocks of H. dauricus and A. schrenckii during the last 20 years. For instance, the maximum output was 452 tonnes year$^{-1}$ and its average value was 346 tonnes during the 1980s, while the maximum output was 243 tonnes year$^{-1}$ and its average value was 181 tonnes during the 1990s, which was only half of that in the 1980s (Qu et al., 1997; Feng et al., 2004). Acipenser sinensis, A. dabryanus and P. gladius were once the major commercial fishes in the Yangtze, but habitat alteration and the closing of Gezhouba Dam have led to a collapse of their abundance during the last decades. Although the capture of these species are strictly prohibited since 1983; they were listed as a state protected species class I in 1988; their survival in the wild is further threatened by the construction of Jinhaijiang First Hydroelectric Project and the Three Gorges Project, which will disrupt spawning grounds and alter hydrological conditions dramatically. Few studies have been conducted on A. baerii, A. ruthenus and A. nudoventris in China because the wild populations are believed to be very small and their catches are rare (Wei et al., 2004).

Sturgeon aquaculture can be used as a tool not only for economic development to meet the demand for products from these species, but also for restocking (Burtsev et al., 2002; Chebanov et al., 2002). Scientific research on sturgeon aquaculture in China was in its infancy before 1970s. Only a few fragmentary scientific studies were performed on the reproductive biology. Due to market demands and the economic benefits, sturgeon aquaculture was aggressively promoted from the beginning of 1990s, mainly for restocking programmes in the Yangtze and Amur river systems (Chang and Cao, 1999; Sun et al., 2003). For example, A. schrenckii was firstly artificially reproduced in China in 1957 (Zhang, 1985), A. sinensis in 1971, and A. dabryanus in 1976 (Anonymous, 1988). Meanwhile, a
large number of fertilized sturgeon eggs or of prolarvae of other non-indigenous species, including P. spathula, A. stellatus, A. guledenstaedtii and a hybrid called Bester (H. huso × A. ruthenus), were also imported from Russia, Germany, France and Hungary (Sun et al., 2003). It is believed that China has become one of the largest sturgeon aquaculture country in the world by the year 2000 (Wei and Yang, 2003). Hopefully, artificial breeding and aquaculture will contribute to a reduction of fishing pressure and will be able to effectively support rehabilitation programmes for wild stocks.

Sturgeons were and are of large interest to many researchers worldwide because of their special position in vertebrate evolution and the great public interest in these species. Modern molecular genetic techniques play a key role as tools in sturgeon preservation strategies and in trade control (Wolf et al., 1999; Ludwig et al., 2002a and Ludwig, 2006, 2008). An increasing number of studies relate to molecular phylogenetics and evolution (Krieger et al., 2000, 2005; Ludwig et al., 2000, 2001, 2003; Krieger and Fuerst, 2002), and to the importance of genetic variation for conservation management (May et al., 1997; Wirgin et al., 1997; Mcquown et al., 2000; Jenneckens et al., 2001; Ludwing et al., 2002b; Schrey and Heist, 2007). The genetic studies on native sturgeon species of China were initiated in the 1980s. Most of these studies were focused on species preservation, population stabilizing and controlled propagation (Anonymous, 1988). After the 1990s, with the development of molecular markers, studies concentrated on conservation genetics and phylogenetics. Important results on genetics were derived for A. sinensis, A. dabryanus, P. gladius, A. schrenckii and H. dauricus, and were applied in conservation management (Deng and Deng, 1997; Wei et al., 1997; Zhuang et al., 1997; Zhu et al., 2006; Peng et al., 2007). Due to the rareness of A. baerii, A. ruthenus and A. nudiventris, few genetic data are available for these species from the region. Genetic studies on sturgeons in China have until recently still been in the initial state. Numerous problems, such as population structure and species identification, have not yet been resolved adequately or have not been employed until now. In this paper, we review the development and current status of sturgeon aquaculture and paddlefish in China, as well as the main genetic and phylogenetic results for native species. The trade control in sturgeon specimens and caviar products in China is also addressed.

Status of sturgeon aquaculture in China

The conflict between limited natural population abundances and the great economic interest in sturgeon products is one of the driving forces to push the development of the sturgeon aquaculture in China. After the first attempt of controlled reproduction, the successful culture of A. schrenckii in 1957 (Zhang, 1985), promoted the development of sturgeon aquaculture techniques. Cultivation was based on the collection of mature broodfish in relevant spawning grounds and the injection of pituitary hormones for final maturation and these methods were gradually improved during the 1990s and applied for other species from China (Chen and Zhou, 1992; Wei et al., 1997; Chang and Cao, 1999; Zhuang et al., 2002; Sun et al., 2003). Late in the 1990s, fry and fertilized eggs of adventive sturgeons and hybrids were imported and introduced to farms in different provinces. Techniques for mass rearing of juveniles were tested on a commercial scale and were gradually developed until 1996, while sturgeon farming in China increased in popularity since 1998. As a result, there were 11 sturgeon species/hybrids being cultured in China around the year 2000 (Sun et al., 2003). Besides the Chinese native species, P. spathula, H. huso, A. stellatus, A. guledenstaedtii and Bester hybrids (H. huso × A. ruthenus) were also raised (Wei and Yang, 2003). At present, sturgeon aquaculture operations in China occur in about 70% of the provinces, including Anhui, Chongqing, Guangxi, Guizhou, Hainan, Heilongjiang, Henan, Hunan, Jiangxi, Liaoning, Shandong, Shanxi, Sichuan and Yunnan. A series of culture experiments showed that aquaculture of sturgeons could be successfully in the warmer waters of South China with great economic potential (Zhuang et al., 2002). Therefore, most sturgeon farms are located in the Pearl River delta and the Yangtze delta. Investigations in 39 major sturgeon farms producing fingerlings in China estimated an overall number in standing stock of 17.86 million for all sturgeon species and hybrids (Wei et al., 2004). The authors also predicted from the farms surveyed that approximately 6000 tonnes of sturgeons with an average size of 0.75 kg should be ready for the markets, assuming a 50% survival rate of farmed fingerlings.

Yangtze River

As Chinese sturgeon, Chinese paddlefish and Dabry’s sturgeon are listed nationally as Class I state protected animals, people who are permitted to culture them have also the obligation to restock these to natural waters or preserve them in captivity. Commercial use of these species is strictly prohibited. The populations of Chinese paddlefish and Dabry’s sturgeon dramatically declined since the Gezhouba Dam construction. The latest catch of a Chinese paddlefish was reported in the Nanjing branch of the Yangtze River in January 2003. Several attempts on artificial reproduction of Chinese paddlefish were not successful. Meantime, very few Dabry’s sturgeon are being held in captivity and are maintained by artificial breeding as a tool for conservation. Acipenser sinensis cultured at the Chinese Sturgeon Institute (CSI) and the Yangtze River Fisheries Research Institute (YFI) originated from captive broodstock artificially fertilized eggs from wild broodfish (Wei and Yang, 1998; Chang and Cao, 1999). The two institutes received special licenses on a yearly basis to capture a few specimens of multiple spawners for use in restocking programmes and for specific research projects. With improvements in cultivation techniques, facility design and rearing techniques, the number of wild spawners actively caught for restocking purposes has decreased, while the fry production has noticeable increased. Two institutes (CSI and YFI) received special licenses from the Fisheries Bureau of Ministry of Agriculture (FB of MOA) to capture Chinese sturgeon spawners for artificial propagation with a quota of 20 spawners in 1999, 26 spawners in 2000 and 14 spawners in 2001. Fry production was about 1 000 000, 2 000 000 and 800 000 in 1999, 2000 and 2001, respectively (Wei et al., 2004). In recent years, the central government (MOA) granted permits to several hatcheries and companies to raise Chinese sturgeons to try to complete the life cycle in captivity in case the species becomes extinct in the wild. This threat is real because of the anticipated additional impact of the Three Gorges Project on the remaining population. Some fry were sent to farms that had been licensed for grow-out of the species. Since 1998, a total of eleven farms and companies in China have been granted permits for aquaculture of the Chinese sturgeon. Although there have been many discussions on the potential exploitation and commercial use of Chinese
sturgeon because of successful rearing in experimental aquaculture facilities, commercial utilization of these reared fish has not been permitted.

**Amur River**

In the Heilongjiang Province, *A. schrenckii* and *H. dauricus* are the only two commercially important species from which caviar is still produced. Historically, landings of these two species have not been recorded separately from others so that only the total yield can be reported. From 1957 to 1977, the landings ranged 13–100 tonnes year⁻¹, with an average of 43.3 tonnes. In 1978, the industry expanded the sturgeon fisheries as foreign traders came to China to purchase caviar at high prices. Sturgeon catches for all species reached a peak of 452 tonnes in 1987 and averaged 322.2 tonnes annually between 1987 and 1991. Sturgeon landings declined thereafter and averaged 176.2 tonnes year⁻¹ between 1992 and 1996. From 1997 to 1999, landings were recorded with 136, 149 and 141 tonnes respectively (Feng et al., 2004; Wang and Chang, 2006). There is a lack of scientific information on the current status of the sturgeons in the Chinese part of the Amur River. Many data on fisheries were collected by the Fisheries Department of the local government, however, these data are usually scattered and insufficient for a rigid scientific analysis. Nevertheless, available catch figures indicate a significant decline in stocks of *H. dauricus* and *A. schrenckii* over the last decade. It is anticipated that *H. dauricus* may soon be near extinction.

Two major sturgeon propagation stations on the Chinese side of the Amur Rivers were set up at Qingdeli in 1988 and at Fuyuan in 1998. As of 2005, about 8.45 million fry (according to the FB of Heilongjiang Province) have been released into the Heilongjiang River. From 1988 to 1998, the Qingdeli Farm released a total of 3.6 million fry of *A. schrenckii*. In 1998, the Fuyuan station released about 80,000 fry. In 2001, 20,000 Kaluga and 130,000 Amur sturgeon fingerlings were released into the Amur River by the Fuyuan Sturgeon Propagation Station. It was the first time that *H. dauricus*, a critically endangered species, was released into the Amur. Although the captive breeding of Amur sturgeon was successful in 2002 (Qu et al., 2002), the majority of the cultured specimens of *A. schrenckii* and *H. dauricus* have still been obtained under controlled propagation using wild spawners caught in the Amur River. Survey data revealed that 25.8 million and 43.3 million fertilized eggs and fry of Amur sturgeon or Kaluga/Amur hybrids were produced in Heilongjiang in 2001 and 2002 respectively (Wei et al., 2004). Most fertilized eggs were quickly transported to provinces in the South of China for incubation. Some fry in the southern hatcheries were maintained for grow-out and some were sold to other fish farms. The author also indicated that the Amur sturgeon is now the most commonly farmed sturgeon species in China, accounting for 70.8% of the total number of cultured specimens.

**Exotic species**

Most of the introduced (non-native) sturgeon species in China are owned by private companies and organizations. There is unfortunately no official data reporting on their production figures or total output except the data for sturgeon meat produced for export. All we know is that seven and a half tonnes of Siberian sturgeon and Russian sturgeon were exported to Singapore and Hong Kong in 2000. The exported volume to Hong Kong and Japan increased to 123 tonnes in 2001, which included Siberian sturgeon, Russian sturgeon and sterlet. Besides these species, only the cultivation of *P. schrenkii* has also widely developed (Wu and Lin, 1999), the output depending on the successful breeding since 2002 (Wang et al., 2004). In recent years, aquaculture of Russian sturgeon has declined because the yellow colour and coarse meat of this species is not well accepted by consumers in China and worldwide.

**Trade control of sturgeon products in China**

Sturgeon aquaculture is a new developing industry in China. The cultured sturgeons have been used for many diversified products, such as fins, pharmaceutical products and leather, but have also been sold as whole live fish. Generally, caviar and meat are the major products derived and marketed from sturgeon processing in China. The caviar is exported and the meat is mainly consumed domestically. In order to protect the sturgeon resources in China, the Centre Government has taken measures to restrict sturgeon production. According to the ‘Wild Animals Protection Law’ promulgated in 1988, the permits for the export and import of sturgeons, their caviar, specimens and parts thereof, are issued by the China CITES Management Authority after approval by the FB of MOA in China (Wang and Chang, 2006). Harvest quotas for sturgeons (for caviar production and for specimens destined for propagation) are based on the sturgeon quota established for caviar production. In accordance with the provisions of CITES, China has implemented effective management measures over caviar trade since 1998. The export quotas for Chinese caviar were 6.27, 5.94, and 5.65 tonnes for the years 1998, 1999 and 2000 respectively. Generally, the quotas declined gradually and have been maintained at the level around 5.9 tonnes since 2001 (Wei et al., 2004).

**Domestic trade**

Live sturgeons are sold in most provincial capitals and in large cities of China, with plenty of huge fish markets, such as in Guangdong, Shanghai, Xinjiang and Beijing. Among all the cultured sturgeons, *A. schrenckii* dominates the domestic markets. For instance, its amount in Guangzhou was 20,000 individuals day⁻¹, while it was 10,000–15,000 individuals day⁻¹ in Shanghai and 12,000–16,000 individuals day⁻¹ in Beijing (Wang et al., 2004) during the breeding seasons.

**International trade**

Sturgeons and paddlefish are highly prized for their caviar; one of the most valuable wildlife products in international trade (De Meulenaer and Raymaker, 1996; Ludwig, 2008). There is little caviar consumed on the domestic markets, although caviar processing in China started in the early 1950s. Caviar is mainly exported to Germany, the USA and Japan. Although there are eight native species in China, only caviar from wild Amur populations of *A. schrenckii* and *H. dauricus* are traded (exported) legally. Six companies were licensed to export caviar from these species, which are located along the Amur (Wang and Chang, 2006). The caviar trade is strictly executed by the quotas from China CITES Management Authority. According to official statistics, the volume of exported caviar reached a peak of 26 metric tonnes (mt) in 1987. Since 1992, the caviar production has declined gradually, fluctuating...
between 6.46 and 12.7 mt, with an annual average production of 9.16 mt. The export reached the minimum of about 2000 kg of A. schrenkii and H. dauricus in 2003, but the export rose slightly up to about 2600 kg in 2005 (Cui et al., 2006). Furthermore, some private corporations also have got the caviar from cultured sturgeons in China, for instance, 500 kg caviar was successfully produced by Tianxia Sturgeons Cultivating Corporation in 2006; and thereafter, 343 kg caviar were obtained from the culture of A. gueldenstaedtii and A. baerii and this was exported to the Occident, Japan and Hongkong by Tianhai Aquiculture Food Corporation in 2007 (pers. comm.).

**Status of sturgeon genetic studies in China**

The important results on sturgeon genetic research in China were achieved during two time periods. The first period happened before the 1980s, focusing on ecological genetics. During this period, certain preliminary outcomes were achieved, such as reproductive ecology and hatchery operations providing the basic knowledge for implementing advanced genetic research and programmes. Research during the period after 1980s mainly focused on conservation biology, especially on conservation genetics addressing the endangered status of native sturgeons. Many important results have also been achieved on the genetic structure of variation within wild stocks (Zhang et al., 1999a,b, 2000a; Zhu et al., 1999, 2002, 2005; Liang et al., 2002), controlled reproduction and hybridization (Zhuang et al., 1997; Chang and Cao, 1999; Xiao et al., 1999; Pan et al., 2001) and on molecular phylogenetics and evolution (Song et al., 1997; Zhang et al., 1999c,d, 2001; Wang et al., 2005, Peng et al., 2007). During this period, the studies identified a lower genetic variation in sturgeons compared to other bony fishes, yet little influence on the natural stocks by controlled reproduction and release and some basic phylogenetic relationships within Acipenseriformes. In this part, we will focus in detail on the native species which are now frequently studied in genetics.

**Chinese paddlefish and Daibry’s sturgeon**

The rarity of the Chinese paddlefish makes genetic studies very difficult. Cytogenetic studies revealed that P. gladius is possibly tetraploid, which is a similar ploidy level as in Polyodon spathula (Dingerkus and Howell, 1976). In contrast, A. dabryanus, A. sinensis and A. schrenkii are octoploid species (Zhang et al., 1999d). However, these results are still not helping to resolve the unclear ploidy status of Acipenseriformes (Birstein and Vasiliev, 1987; Blacklidge and Bidwell, 1993; Fontana, 1994; Song et al., 1997; Ludwig et al., 2001). Although the wild populations are very rare, several genetic studies were performed focusing on the phylogenetic relationships of these species. Zhang et al. (1999d) used 13 RAPD primers to probe the relationship among seven species including P. gladius and A. dabryanus. The result revealed that P. gladius is the most closely related species to P. spathula, and A. dabryanus can be considered phylogenetically to be the sister species of A. sinensis. Other studies based on mtDNA supported these results (Zhang et al., 2000a, 2001; Peng et al., 2007).

**Amur sturgeon and Kaluga**

Quite a few (cyto)genetic reports dealing with the Amur sturgeon were published previously (Song et al., 1997; Zhang et al., 1999a,b, 2000a; Zhu et al., 1999, 2002, 2005; Shao et al., 2002). Four microsatellites were employed to evaluate the recruitment effects of controlled propagation and release programmes in nature. The parentage analysis revealed a low proportion (5–10%) of artificially propagated individuals among juveniles in the estuary of the Yangtze River (Zhu et al., 2002). The present rate of release of artificially produced larvae may not be adequate to maintain the population of A. sinensis, as the natural spawning area has been dramatically reduced in size. Zhu et al. (2006) detected the genetic variation within and among three annual spawning runs of the Chinese sturgeon by six microsatellites. In this study, a new method called unsupervised self-organizing mapping was employed to extract information of the genetic structure of the population. A complex genetic structure was detected in the annual spawning runs. Meanwhile these genetically differentiated spawning stocks may occur sympatrically on a small geographic scale. These results further suggested that the current supportive breeding of A. sinensis may require a careful identification of broodstocks for the artificial propagation in order to avoid admixture and hybridization among these genetically different spawning stocks.
et al., 1999a; Ludwig et al., 2001; Robles et al., 2004, 2005). Chen et al. (2004) reported that the Sox 9 gene has been widely expressed in the early eight stages, which indicated a conserved function of Sox 9 in the cartilage development among vertebrates. Very recently, sex specific cDNA expression differences between female and male gonads in adult individuals of *A. schrenckii* were obtained (Chen et al., 2006). However, it is not clear whether the difference also exist in sex development and polarization at the beginning. No gender specific genetic differences were observed in European sturgeon, *Acipenser sturio* (Hett and Ludwig, 2005; Hett et al., 2005). Due to the different outcome by different species of Acipenseriformes, it is valuable to promote research about sex specific cDNA expression. Only unsatisfactory knowledge has yet been gained for *H. dauricus*. Propagation biology of *H. dauricus* was analysed by Pan et al. (2001), who reported that ovaries grew big as weight increased. However, egg production was less than in *A. schrenckii*. In addition, Liang et al. (2002) performed microsatellite investigations on *H. dauricus* in samples from the Heilongjiang River. The authors found relatively low genetic variation in this species. One hundred and sixty-nine DNA bands were amplified using eleven microsatellites. Overall 47.93% of the loci were polymorphic.

**Discussion and conclusion**

There are eight indigenous species of Acipenseriformes in China, all of which are endangered resulting from various human activities. All these species are protected as class I or class II animals under the Chinese endangered species act. Of all native species so far only five have been often used in genetic studies. These studies focused on genetic variation, phylogenetic and evolution aspects and breeding programmes. Not only traditional methods but also molecular methods such as RAPD, mtDNA and microsatellites were widely used in genetic studies in sturgeon. However, a scientific stock assessment for the Amur sturgeon or Chinese sturgeon based on genetic diversity and on the population structure is still lacking. Little was known about the genetic influence or risks associated with restocking/rehabilitation programmes on the wild populations. In comparison to the identification trials on the origin of caviar products in European and North American countries, our molecular identification efforts on native reared stocks presently commercially utilized in agriculture is far behind, and this will impede progress in sturgeon aquaculture, trade control of black caviar and also the development of adequate strategies in conservation genetics.

Commercial farming of sturgeons has become popular since 1998. There are eleven species and hybrids presently cultured in China, including imported species and hybrids. *Acipenser schrenckii* and Amur hybrids are presently the major objects of commercial culture. Sturgeon aquaculture is practiced in about 20 provinces, with major centres in the region of the Pearl River delta, the Yangtze delta as well as in the Fujian and Hubei provinces. The quantity of sturgeons produced by aquaculture in China appears to be the largest in the world as of the year 2000. However, there have not been any national or governmental programmes with a certain guaranteed investment to preserve native sturgeons via aquaculture as a tool and very limited sturgeon culture facilities are employed for conservation purposes in China. Successful breeding of sturgeon in captivity is the key to establishing a sustainable sturgeon culture industry, which will also help sturgeon conservation. Unfortunately, most sturgeon farms in China could not establish sustained captive breeding programmes to built consistent brood stocks because of economic restrictions. At present, none of the sturgeon species complete their life cycle in a hatchery except for Amur sturgeon. Development of sustainable sturgeon aquaculture for both commercial and conservation purposes is hindered by a shortage of higher-age parental stock that can serve as a founder population in captivity. Preserving sufficient higher-age fish annually and developing captive breeding populations of *A. sinensis, A. schrenckii* and *H. dauricus* should become a national priority in fisheries science and management. Furthermore, the introduction of millions of fry of exotic sturgeon species and their hybrids to China will negatively impact native sturgeon species because little efforts are presently undertaken to prevent escapement. There is an urgent need to improve management to maintain sturgeon biodiversity and sturgeon ecosystems. We believe that hatcheries should be very cautious with open-water rearing of sturgeons in net cages because of the high risk for escapement and biological pollution and alternative and save culture methods should be employed even if cage culture greatly reduces rearing costs.

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