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First specimen of the white sturgeon (*Acipenser transmontanus* Richardson, 1836) in coastal waters of Mexico with data on its genetic identity

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The chondrostean fishes of the family Acipenseridae are composed of four genera (*Acipenser*, *Pseudoscaphirhynchus*, *Scaphirhynchus*, and *Huso*) and almost 25 extant species in the marine and continental waters of the world (Birstein 1993, Nelson et al. 2006). The genus *Acipenser* is the most diverse taxon within the family, with 17 valid, extant species (Birstein 1993, Birstein and Bemis 1997), of which eight occur in waters of North America (Vladykov and Greeley 1963, Nelson et al. 2004). All species of chondrosteans face problems of conservation due to overfishing and destruction of habitat (Birstein and Bemis 1997), including those in North America (Jelks et al. 2008).

Along the Pacific Coast of North America, the white sturgeon (*Acipenser transmontanus*) and the green sturgeon (*A. medirostris*) reach their respective southernmost ranges in northwestern Baja California, Mexico (Love et al.1995, Rosales-Casian and Almeda-Jauregui 2009). The white sturgeon is known to occur from Northern Gulf of Alaska (Mecklenburg et al. 2002) to Ensenada, Baja California (Hubbs 1967, Miller and Lea 1972). This anadromous chondrostean is commonly found in estuaries and the sea as deep as 122 m, migrating into large rivers to spawn, and spending much of

its life in freshwater in some areas (Mecklenburg et al. 2002, Moyle 2002). Its known maximum length (FL) and age are 3.2 m and 82 years, respectively (Valdykov and Greeley 1963), although specimens over 6 m fork length have also been reported (Moyle 2002).

The first report of the white sturgeon in Mexico was provided by Rosales-Casian and Ruz-Cruz (2005), who observed a male (123 cm total length) in a seafood market at Ensenada, which had been collected on 15 March 2003 at Todos Santos Bay, Baja California. Unfortunately, this individual was not secured as a voucher specimen.

On 5 May 2010, a female white sturgeon (126 cm total length) was captured with a bottom gillnet (20.3 cm mesh size) at 1.5 km off coast of Todos Santos Bay (31º 52' 48.4" N, 116º 42' 05.4" W). The capture site was 25 m deep of sandy bottom. Other fishes captured along with the white sturgeon were scorpion fish (*Scorpaena guttata*) and California halibut (*Paralichthys californicus*). The specimen (Figure 1, top) was identified on the basis of the following combination of diagnostic characteristics (Miller and Lea 1972, Mecklenburg et al. 2002): barbels closer to tip of snout than to mouth (Figure 1, bottom); 11-14 predorsal scutes; no scutes posterior to dorsal fin; 38-48 scutes in lateral row; 9-12 in ventrolateral row; 34-36 gillrakers; body gray dorsally and white in ventral surface; and, viscera black.

Measurements and counts of the specimen were as follows: head length 23.0 cm, snout length 7.22 cm, distance between tip of snout and central edge of the mouth 7.82 cm, 38 lateral scutes, 13 predorsal scutes, 12 ventrolateral scutes, and weight of 9.13 kg.

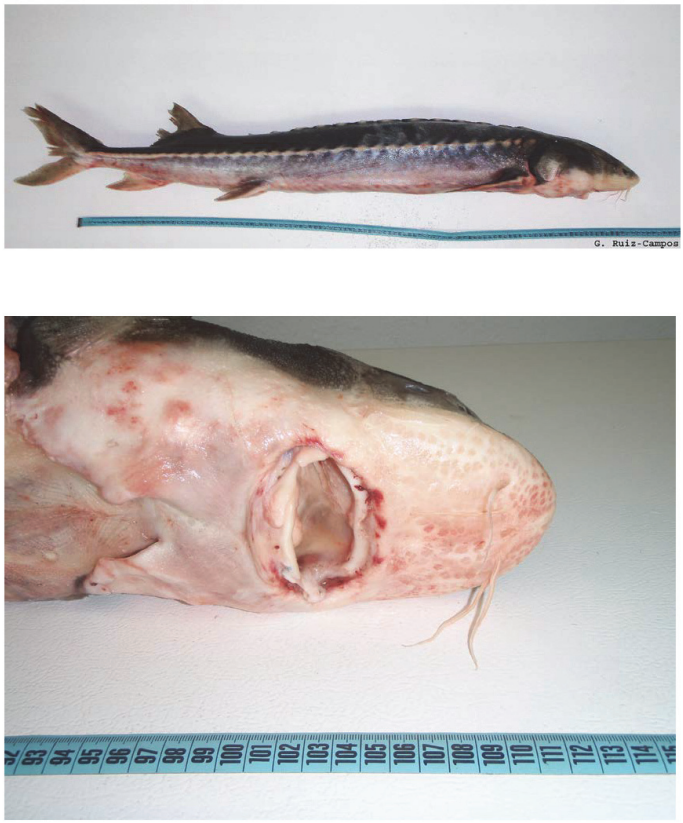

Figure 1.—(Top) Female of white sturgeon, *Acipenser transmontanus*, 126 cm total length, collected at Todos Santos Bay, Baja California, Mexico, on 5 May 2010. (Bottom) Ventral cephalic view of the specimen showing the insertion of barbels closer to the tip of the snout than to the mouth. Photographs by Gorgonio Ruiz-Campos.

The examination of gonadic volume determined a stage of III type (Lagler 1978) and the presence of ovocites was noted with the aim of a stereoscopic microscope. The stomach was empty. A fin clip was obtained and fixed with 95% ethanol. Finally, the specimen was fixed with 10% formalin and then deposited in the Fish Collection of the Universidad Autonoma de Baja California (UABC-2328) at Ensenada. Therefore, our individual represents the first voucher specimen for the species in Mexican waters. The unusual presence of white sturgeon as far south as Todos Santos Bay in northern Baja California might be associated with La Nina conditions that have prevailed in the California Current System since the summer of 2007 (McClatchie et al. 2009).

Also, we also corroborated the morphological identification using Deoxyribonucleic Acid (DNA) barcoding system (with a fragment of Cytochrome Oxidase [COI]) gene of mitochondrial DNA). DNA was extracted as described by Aljanabi and Martinez (1997). Polymerase Chain Reaction (PCR) amplification for a 710 bp fragment of the COI was performed with primers FishF1 (52 - TCA ACC AAC CAC AAA GAC ATT GGC AC-32) and FishR1 (52- TAG ACT TCT GGG TGG CCA AAG AAT CA-3) described by Ward et al. (2005) and Hubert et al. (2008). PCR reactions were carried out in a solution containing 50 microliters of approximately 50 nanograms of total DNA, 0.40 micromoles of each primer, 2.5 millimoles of $MgCl_2$, 0.2 millimoles of each of the dNTP, 1×PCR buffer (Invitrogen, #Y02028, Carlsbad, California), and 0.5 U Taq polymerase (Invitrogen, #18038-042). PCR was performed in a thermocycler (BioRad Laboratories, Hercules, California) under the following conditions: initial denaturing step at 94°C for 4 min, 35 cycles of denaturing at 94°C for 45 s, annealing (for 45 s) at 52°C, followed by an extension at 72°C for 45 s, and ending with a final extension at 72°C for 10 min. DNA sequencing reactions were carried out following the manufacturer's instructions (Big Dye Terminator cycle sequencing kit, PerkinElmer, Waltham, MA). We bi-directionally sequenced using the same primers for PCR. Capillary electrophoresis was performed with a genetic analyzer (ABI PRISM 3730XL, Macrogen, Seoul, Korea). To determine the sequence identity, we retrieved homologous sequences of white sturgeon registered in GenBank (EU523887, EU523888, EU523889, from Nechako reservoir at Washington; EU523890, EU523891 from Fraser river at Washington reported by Hubert et al. (2008); and AB042837 reported by Inoue et al. (2003), which came from Sacramento River (J. Inoue, personal communication).

These sequences were aligned (without gaps) with the sequence obtained from our specimen collected in Todos Santos Bay. Two haplotypes were observed; haplotype A formed for all sequences reported by Hubert et al. (2008) and that constitutes the northern haplotype. Haplotype B formed for the sequences reported by Inoue et al. (2003), and our sample from Todos Santos Bay representing the southernmost haplotype. The difference between both hapolotypes was a punctual mutation CT (G/A) in the 248 position on the 3rd codon position corresponding to 82 amino acid; the Kimura two parameters genetic distance between both haplotypes was 0.0016.

In order to reconstruct the neighbor-joining cladogram, we retrieved sequences for eleven species of sturgeon (Table 1). The neighbor-joining analysis was conduced in MEGA using the Kimura two parameter genetic distances (Tamura et al. 2007). The statistical robustness was tested with 10,000 bootstrap replicates (Felsenstein 1985). We confirmed that the haplotype sequence of the specimen from Todos Santos Bay was grouped with that of the white sturgeons retrieved from GenBank with support of bootstrapping (99 %), while the specimens with hapolotype A formed the same clade

Table 1.—List of accession numbers of homologous sequences retrieved from GenBank for COI gene of mitochondrial DNA for ten species of the genus *Acipenser*.

Species	Accession number of GenBank
A. dabryanus	AY510085
A. medirostris	EU523885
A. medirostris	EU523883
A. stellatus	FJ809739
A. stellatus	FJ809737
A. nudiventris	FJ809730
A. nudiventris	FJ809733
A. brevirostrum	EU523872
A. brevirostrum	EU523870
A. fulvescens	EU524397
A. fulvescens	EU524396
A. fulvescens	EU524395
A. baerii-baerii	FJ205562
A. baerii-baerii	FJ205560
A. gueldenstaedtii	FJ809728
A. gueldenstaedtii	FJ809725
A. persicus	FJ809724
A. persicus	FJ809721
A. oxyrinchus	EU524400
A. oxyrinchus	EU524401

with the sturgeons sharing the haplotype B (Figure 2). These results confirm that the sturgeon caught at Todos Santos Bay was a white sturgeon. However, the interspecific relationships among the closest species to white sturgeon are less clear. Hubert et al. (2008) using a similar fragment to our analysis, reported that white sturgeon and green sturgeon are monophyletic, and both species share some part of their ranges. In this study, the analysis included Dabry's sturgeon (*A. dabryanus*), a species with disjunct distribution and inhabiting the Yangtze River in China. Unexpectedly, this species formed a group with white sturgeon. In addition, Peng et al. (2007) using cytochrome *b* sequences and more species of sturgeons, reported that white sturgeon formed a monophyletic group with disjunct Asian species but not with the sympatric North Eastern Pacific species

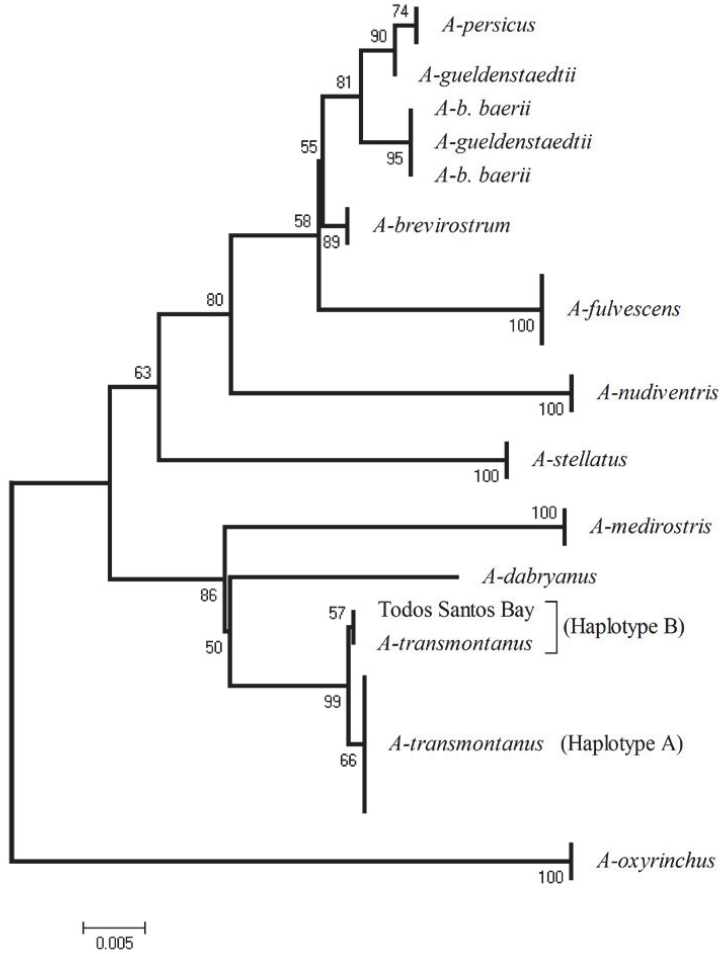

Figure 2.—Topology of neighbor-joining cladogram (Kimura-2 parameters distance; 10,000 replicates of bootstrap) of COI mtDNA for species of sturgeon obtained from GenBank and a sample taken at Todos Santos Bay at Baja California, Mexico. The number in nodes indicates the statistical support of branches.

(green sturgeon). It is clear that a global, genetic study is needed to clarify phylogenetic relationships and geographic distribution patterns.

On the other hand, the cladogram also demonstrates that the two specimens of Russian sturgeon (*A. gueldenstaedtii*) do not form a natural group, because one of them is grouped with the Persian sturgeon (*A. persicus*), while the other is more closely aligned with Siberian sturgeon (*A. baerii baerii*). This topology seems to confirm the inclusion of the most diverse sturgeon linage ("*gueldenstaedtii*-complex") that could contain cryptic species (Birstein et al. 2005). Based on the outcome of this study, the fragment of COI showed more polymorphism than suspected; therefore, the analysis of an adequate number

of species and the high level of polymorphism for this DNA fragment should be used with caution to identify sturgeons to species.

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Literature Cited

Aljanabi, S. M., and I. Martinez. 1997. Universal and rapid salt-extraction of high quality genomic DNA for PCR-based techniques. Nucleic Acids Research 25:4692-4693.

Birstein, V. J. 1993. Sturgeons and paddlefishes: threatened fishes in need of conservation. Conservation Biology 7:773-787.

Birstein, V. J., G. Ruban, A. Ludwig, P. Doukakis, and R. DeSalle. 2005. The enigmatic Caspian Sea Russian sturgeon: how many cryptic forms does it contain? Systematics and Biodiversity 2:203-218.

Birstein, V. J., and W. E. Bemis. 1997. How many species are there within the genus Acipenser? Pages 157-163 *in* V. J .Birstein, J. R. Waldman, and W. E. Bemis, editors. Sturgeon Biodiversity and Conservation. Kluwer Academic Publishers, Dordrecht, The Netherlands.

Felsenstein, J. 1985. Confidence on phylogenies: an approach using bootstrap. Evolution 39:783-791.

Hubbs, C. L. 1967. Occurrence of the Pacific lamprey, *Entosphenus tridentatus,* off Baja California and in streams of southern California, with remarks on its nomenclature. Transactions of the San Diego Society of Natural History 14:301–312.

Hubert, N., R. Hanner, E. Holm, N. E. Mandrak, E. Taylor, M. Burridge, D. Watkinson, P. Dumont, A. Curry, P. Bentzen, J. Zhang, J. April, and L. Bernatchez. 2008. Identifying Canadian freshwater fishes through DNA barcodes. PLoS ONE 3(6), e2490.

Inoue J. G., M. Miya, K. Tsukamoto, and M. Nishida. 2003. Basal actinopterygian relationship: a mitogenomic perspective on the phylogeny of the “ancient fish”. Molecular Phylogenetics and Evolution 26:110-120.

Lagler, K. F. 1978. Freshwater fishery biology. 2nd edition. W. C. Brown Company, Dubuque, Iowa, USA.

Love, M. S., C. W. Mecklenburg, T. A. Mecklenburg, and L. K. Thorsteinson. 2005. Resource inventory of marine and estuarine fishes of the west coast and Alaska: a checklist of North Pacific and Arctic Ocean species from Baja California to the Alaska-Yukon Border. OCS Study MMS 2005-030 and USGS/NBII 2005-001. U. S. Department of the Interior, U. S. Geological Survey, Biological Resources Division, Seattle, Washington, USA.

McClatchie, S., R. Goericke, F. B. Schwing, S.J. Bograd, W. T. Peterson, R. Emmett, R. Charter, W. Watson, N. Lo, K. Hill, C. Collins, M. Kuhru, B. G. Mitchell, J. A. Koslow, J. Gomez-Valdez, B. E. Lavaniegos, G. Gaxiola-Castro, J. Gottschalk, M. L'Heureux, Y. Xue, M. Manzano-Sarabia, E. Bjorkstedt, S. Ralston, J. Field, L. Rogers-Bennet, L. Munger, G. Campbell, K. Merkens, D. Camacho, A. Havron, A. Douglas, and J. Hildebrand . 2009. The state of the California Current, spring 2008-2009: cold conditions drive regional differences in coastal production. California Cooperative Oceanic Fisheries Investigations Reports 50:43-68.

Mecklenburg, C. W., T. A. Mecklenburg, and L. K. Thorsteinson. 2002. Fishes of Alaska. American Fisheries Society, Bethesda, Maryland, USA.

Moyle, P. B. 2002. Inland fishes of California. University of California Press, Berkeley, USA.

Miller, D. J., and R. N. Lea. 1972. Guide to the coastal marine fishes of California. California Department of Fish and Game, Fish Bulletin 157:1-249.

Nelson, J. S. 2006. Fishes of the world. 4th edition. Wiley and Sons, Inc., Hoboken, New Jersey, USA.

Nelson, J. S., E. J. Crossman, H. Espinosa-Perez, L. T. Findley, C. R. Gilbert, R. N. Lea, and J. D. Williams. 2004. Common and scientific names of fishes from the United States, Canada, and Mexico. 6th edition. Special Publication 29, American Fisheries Society, Bethesda, Maryland, USA.

Peng, Z., A. Ludwig, D. Wang, R. Diogo, Q. Wie, and S. He. 2007. Age and biogeography of major clades in sturgeon and paddlefishes (Pisces: Acipenseriformes). Molecular Phylogenetics and Evolution 42:854-862.

Rosales-Casian, J. A., and C. Almeda-Jauregui. 2009. Unusual occurrence of a green sturgeon, *Acipenser medirostris*, at El Socorro, Baja California, Mexico. California Cooperative Oceanic Fisheries Investigations Reports 50:169-171.

Rosales-Casian, J. A., and R. Ruz-Cruz. 2005. Record of a white sturgeon, *Acipenser transmontanus*, from Bahia de Todos Santos, Baja California, Mexico, found at the Ensenada seafood market. Bulletin of the Southern California Academy of Sciences 104:154-156.

Tamura, K., J. Dudley, M. Nei, and S. Kumar. 2007. MEGA4: Molecular Evolutionary Genetics Analisis (MEGA). Software version 4.0. Molecular Biology and Evolution 24:1596-1599.

Vladykov, V. D., and J. R. Greeley. 1963. Order Acipenseroidei. Memoirs of the Sears Foundation for Marine Research 1:24-60.

Ward, R. D., T. S. Zemlak, B. H. Innes, P. R. Last, and P. D. N. Hebert. 2005. DNA barcoding Australia's fish species. Philosophical Transactions of the Royal Society B 360:1847-1857.

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