**Appendix S1.** Catostomidae fossils, fossil and external calibration age priors, and additional discussion of phylogenetic relationships within the Catostominae.

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**1. Introduction**

It is important to use objective and repeatable methods for identifying minimum age justifications for fossil sampling dates or calibrations used in phylogenetic studies. By following published recommendations in Parham et al. (2012) and precedents set by previous empirical studies (e.g. Pyron, 2011; Near et al., 2011, 2012; Unmack et al., 2014), we were able to identify a suitable external calibration for our sucker tree based on a phylogenetic study. Subsequently, we used this external calibration to set the time of origin prior (*t*or) for the fossilized birth-death (FBD) process tree prior (Gavryushkina et al., 2013; Heath et al., 2014) that we used in all of our BEAST relaxed-clock divergence time analyses of sucker diversification in this study. Additionally, we were able to include dates for 19 extinct sucker taxa in our FBD analyses, most of which were derived from fossil information, with the only exception being the age assigned to the historically extinct species †*Moxostoma lacerum* (see discussion in main text; this species is omitted here because there are no fossil records of it).

In this Appendix, our objectives are four-fold. First, we present the results of a literature review of the sucker fossil record that we conducted considering 33 sucker fossils from Asia and North America. We present a table summarizing the taxonomy of extinct fossil sucker species that we are aware have been described to date. Also in table format, we review the spatial and temporal distributions of sucker fossils, while highlighting minimum age calibrations that we estimate can be obtained from each fossil, and attending geological and phylogenetic metadata. Our fossil review is not necessarily exhaustive, but it covers the most well-known and best-justified fossil specimens for the family, while excluding questionable fossils. It is worth reiterating that this review provided the raw data for determining suitable fossil ages and prior settings used in our BEAST analyses.

Second, we provide background information and justifications for the external root-age calibration point we used in our FBD BEAST runs mentioned above. We list details on this calibration, including characters and phylogenetic results on which the calibration is based, the minimum age estimate, and prior settings supplied to BEAST. Then, we list (1) the calibrated node, (2) the taxa that represent the first known occurrence of the lineage in the fossil record, and (3) available phylogenetic results supporting the clade in question, or linking the fossil to the group and aiding its placement in the sucker phylogeny. We also list (4) the location and age of the geological formation(s) for the fossil assigned to the node, as well as (5) our minimum age assessment based on the available phylogenetic and geological data. Calibration ID letters and numbers referred to here are the same as those given in the text.

Third, we briefly review why some of the fossils for which we were able to determine minimum age justifications were not used in our BEAST analyses.

Fourth, we here provide a supplemental presentation and discussion of the finer-scale patterns of relationships within and among clades 1–9 that we resolved for the subfamily Catostominae (e.g. Figs. 3 and S4). We present this material here due to space constraints: including this material in the main text would have rendered the Results and Discussion section too long, making it burdensome for the reader. In doing so, we avoid this issue while keeping the material peer-reviewed to ensure objectivity of presentation. However, in order to avoid duplicating content, we mainly present information here on clades that were not emphasized in Section 3.4 of the main text.

**2. Review: Sucker Fossils**

The first fossil sucker genus and species, †*Amyzon mentale* Cope 1872, was described by Cope (1872) from North American fossil beds at the Osino Oil Shales of Nevada, which date to ~37–34 million years ago (Ma) in the late Eocene–early Oligocene (Bruner, 1991; Swisher & Prothero, 1990). Since Cope’s groundbreaking systematic work on fossil suckers nearly 150 years ago, contributions from many workers clearly demonstrate that suckers possess a rich and relatively complete fossil record, especially when compared to other speciose North American freshwater fish clades for which little or no fossil information is available (e.g. darters within family Percidae; reviewed by Smith, 1981; Cavender, 1986, 1998; Smith et al., 2002). Presently, sucker fossils span a wide geographical area, ranging in Asia from Mongolia to central–eastern China and eastern Siberia, and dotting the western North American continent from British Columbia to Mexico, and from inland California to eastern Texas (e.g. Cavender, 1998: Fig. 3; Chang et al., 2001: Fig. 1). The sucker fossil record also covers a wide timespan, with fossilized specimens ranging in age from the Eocene up until the end-Pleistocene (~56–0.01 Ma). Thus, the available fossils demonstrate that suckers have been distributed over large areas in multiple Holarctic infraregions in at least western North America and eastern Asia throughout the Cenozoic, and that major ‘derived’ lineages of suckers (e.g. genera within subfamily Catostominae) having been undergoing speciation since at least the Miocene (Smith, 1975, 1981, 1992; Cavender, 1986, 1998).

For additional information on North American sucker fossils, we refer the reader to broad summaries comparing Catostomidae against other North American or European freshwater fish families (Smith, 1981; Cavender, 1986, 1998; Smith et al., 2002, 2013), as well as the recent, comprehensive review of the biogeography, systematics, ecology, and natural history of extant and fossil suckers by Harris et al. (2014). The taxonomy of North American fossil suckers has been relatively stable for decades, with the principal disagreements being the fluctuating number of species within †*Amyzon* (Grande et al., 1982; Bruner, 1991), and that some authors have recognized the elevation of subgenus *Pantosteus* to genus status (Minckley, 1973; Unmack et al., 2014), while others have not (Smith, 1966, 1992; Smith & Koehn, 1971; Harris & Mayden, 2001; Harris et al., 2002; Smith et al., 2013). Compared with North American specimens, the Asian fossil sucker assemblage is lesser known and represented by fewer collections, and it has traditionally exhibited more taxonomic confusion and instability, for example due to the use of some fragmentary bone material and inadequate comparisons with North American material (Chang & Chow, 1986; Chang et al., 2001). However, several reviews as well as empirical studies of fossil sucker systematics and biogeography provide excellent sources of information on the Asian fossil specimens, which include the most recent fossil sucker species to be described or redescribed (Chang & Chow, 1986; Sytchevskaya, 1986; Smith, 1992; Chang et al., 2001; Chang & Chen, 2008; Liu & Chang, 2009; Harris et al., 2014).

Fossil suckers have been noted, or formally taxonomically described, for both extant and extinct lineages of suckers in North America and Asia (Tables A1 and A2). Among the 15 species of extinct fossil suckers described from North American sites that we include here, there are 12 currently valid species, and these are assignable to the following five genera (subfamilies in parentheses): †*Amyzon* Cope 1872 (Ictiobinae), *Catostomus* Lesueur 1817 (Catostominae), *Chasmistes* Jordan 1878 (Catostominae), *Ictiobus* Rafinesque 1820 (Ictiobinae), and *Moxostoma* Rafinesque 1820 (Catostominae) (Table A1). While all 12 of these species are endemic to North America, no extinct fossil genera are endemic to the continent. The only extinct genus of suckers found in North America is †*Amyzon*, which is known from productive fossil beds of British Columbia, Washington, Nevada, and Wyoming (Table A2). Aside from †*Amyzon*, all North American sucker fossils are believed to correspond to extant lineages; indeed, fossils are available for seven extant sucker species, including *Ictiobus cyprinellus*, *I. niger*, *Catostomus ardens*, *C. commersoni*, *C. discobolus*, *Moxostoma duquesnei*, and *Xyrauchen texanus* (Table A2). We note that several fossil sucker collections from North America whose bones could possibly be placed with other species are not reviewed here. For example, we exclude “†*Deltistes ellipticus*” maxilla bones of Miller & Smith (1967) that were placed with †*Catostomus shoshonensis* material by Smith et al. (1982), and dentaries attributed to “†*Catostomus cristatus*” by Smith (1975) that were possibly identifiable as †*C. shoshonensis* (Smith et al., 1982).

Three genera and species of fossil suckers that have been described from Asia are discussed here, all of which are extinct (Table A1). Two of these taxa are widely agreed upon as valid and having clear morphological affinities or differences with other suckers; these are †*A. huanensis* (Cheng 1962) and †*Plesiomyxocyprinus arratiae* Liu & Chang 2009 (Table A1). A third taxon, the Eocene–Oligocene genus †*Vasnetzovia*, was described by Sytchevskaya (1986) from complete specimens collected north of Vladivostok, Primorsky Krai in eastern Siberia, Russia. †*Vasnetzovia* is also considered validly described, and is monotypic with one species, †*V. artemica* Sytchevskaya 1986 (e.g. Chang & Chen, 2008; Harris et al., 2014). However, we exclude from our analysis the first fossil sucker known from Asia, which was described from disarticulated opercles and vertebrae sampled from middle Eocene rocks of the Gobi Desert, Inner Mongolia, and identified as “*Catostomus*” (Hussakof, 1932). We exclude this record because Nelson (1949) showed that this specimen was misidentified as belonging to genus *Catostomus* and instead may be referable to another sucker genus, either *Carpiodes* or *Myxocyprinus*, or might not even be a catostomid. We also exclude records of 10 Asian fossil sucker species from Eocene–Oligocene deposits of Kazakhstan, Mongolia, and Siberia that Sytchevskaya (1986) described as belonging to †*A. gosiutensis* (a species only known from North America), two new species of †*Amyzon* (†*A. interruptus*,and †*A. zaissanicus* from the Zaissan Basin of eastern Kazakhstan), and seven new species allocated to extant sucker genera (*Carpiodes brevidens*, *Cycleptus robustus*, *Catostomus columnaris*, *Erimyzon luxus*, *Minytrema shevyrevi*, *Moxostoma fungidens*, and *Xyrauchen rotundus*). We exclude these taxa because they were described based on isolated specimens of pharyngeal teeth samples, without the context of complete specimens or adequate comparisons to North American material (Sytchevskaya, 1986). Smith (1992) dismissed all of Sytchevskaya’s species above based on pharyngeal teeth because he deemed the original materials as lacking sufficiently detailed information to warrant the descriptions she authored. Moreover, Smith (1992) argued that the Asian *Cycleptus* species Sytchevskaya proposed was based on misidentified materials when compared with Eastman (1977), and that her Asian *Carpiodes* specimen from Kazakhstan was insufficient to diagnose the form as being distinct from †*Amyzon*.

**Table A1.** Summary of the taxonomy and distributions of extinct sucker (Catostomidae) species known from the fossil record.

| No. | Described fossil species | Subfamily (Tribe) | Status | Geological epoch | Geographical distribution | Fossil references |
| --- | --- | --- | --- | --- | --- | --- |
| 1 | †*Amyzon aggregatum* (Wilson 1977) | Ictiobinae | Valid (Bruner, 1991) | Early Eocene | British Columbia, Pacific northwestern North America | Wilson (1977); Grande et al. (1982); Bruner (1991) |
| 2 | †*A. brevipinne* (Cope 1893) | Ictiobinae | Valid (Bruner, 1991) | Early Eocene | British Columbia, Pacific northwestern North America | Cope (1893); Grande et al. (1982); Bruner (1991) |
| 3 | †*A. commune* (Cope 1874) | Ictiobinae | Valid (Bruner, 1991) | Late Eocene | Colorado, western North America | Cope (1874); Bruner (1991) |
| 4 | †*A. fusiforme* Cope 1875 | Ictiobinae | Junior synonym of *A. commune* (Bruner, 1991) | Eocene | Colorado, western North America | Cope (1875); Bruner (1991) |
| 5 | †*A. gosiutensis* Grande, Eastman, & Cavender 1982 | Ictiobinae | Junior synonym of *A. aggregatum* (Bruner, 1991) | Early middle Eocene | Wyoming, western North America | Grande et al. (1982); Bruner (1991) |
| 6 | †*A. huanensis* (Cheng 1962) | Ictiobinae | Valid, described by Chang et al. (2001) based on older materials that were incorrectly referred to the cyprinid genus *Osteochilus* by Cheng (1962) | Eocene | Hunan Province, southern China | Cheng (1962); Chang et al. (2001) |
| 7 | †*A. mentale* (Cope 1872) | Ictiobinae | Valid (Bruner, 1991) | Eocene–Oligocene | Nevada, western North America | Cope (1872); Bruner (1991) |
| 8 | †*A. pandatum* Cope 1874 | Ictiobinae | Junior synonym of *A. commune* (Bruner, 1991) | Eocene | Colorado, western North America | Cope (1874); Bruner (1991) |
| 9 | †*Ictiobus aguilerai* Alvarado-Ortega et al. 2006 | Ictiobinae | Valid (Alvarado-Ortega et al., 2006) | Pliocene | Hidalgo, central Mexico | Alvarado-Ortega et al. (2006) |
| 10 | †*Vasnetzovia artemica* Sytchevskaya 1986 | Ictiobinae | Valid (Chang & Chen, 2008; Harris et al., 2014) | Eocene–early Oligocene | eastern Siberia, northeastern Russia | Sytchevskaya (1986) |
| 11 | †*Plesiomyxocyprinus arratiae* Liu & Chang 2009 | Myxocyprininae | Valid (Liu & Chang, 2009) | Early middle Eocene | various Huadian Formation coal mines, Jilin Province, northeastern China | Liu & Chang (2009) |
| 12 | †*Catostomus arenatus* Miller & Smith 1967 | Catostominae (Catostomini) | Valid (Smith et al., 1982) | Late Miocene–Pliocene | Idaho, western North America | Miller & Smith (1967); Smith (1975); Taylor & Smith (1981); Smith et al. (1982, 2013) |
| 13 | †*Catostomus asitus* Smith, Steward, & Carpenter 2013 | Catostominae (Catostomini) | Valid (Smith et al., 2013) | Early Pliocene | Nevada, western North America | Smith et al. (2013) |
| 14 | †*Catostomus hyomyzon* Smith, Steward, & Carpenter 2013 | Catostominae (Catostomini) | Valid (Smith et al., 2013) | Miocene | Oregon, western North America | Smith et al. (2013) |
| 15 | †*Catostomus oromyzon* Smith, Steward, & Carpenter 2013 | Catostominae (Catostomini) | Valid (Smith et al., 2013) | Pliocene | Idaho, western North America | Smith et al. (2013) |
| 16 | †*Catostomus owyhee* (Miller & Smith 1967) | Catostominae (Catostomini) | Valid (Smith, 1975; Smith et al., 2013); originally described as “*Deltistes owyhee*” by Miller & Smith (1967) | Late Miocene–Pliocene | Idaho, western North America | Miller & Smith (1967); Smith (1975) |
| 17 | †*Catostomus shoshonensis* Cope 1883 | Catostominae (Catostomini) | Valid (Smith et al., 1982, 2002) | Late Miocene–Pliocene | Idaho, western North America | Cope (1883); Armstrong et al. (1975); Smith (1975); Taylor & Smith (1981); Smith et al. (1982) |
| 18 | †*Chasmistes spatulifer* (Miller & Smith 1967) | Catostominae (Catostomini) | Valid (Smith, 1975; Smith et al., 2002) | Late Miocene–Pliocene | Idaho, western North America | Miller & Smith (1967); Smith (1975) |

Abbreviations: Co., county; No., number.

**Table A2.** Summary of Catostomidae fossils and potential derived minimum age justifications for phylogenetic fossil calibrations.

| No. | Fossil taxon | Age (Ma) | Gap analysis age at node (Ma) | Minimum age justification (Ma) | Location | Fossil references | Age references | Minimum age references |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| 1 | †*Amyzon aggregatum* | 49.42 ± 0.54, Ypresian-Lutetian or Bridgerian NALMA | – | 48.88 | Klondike Mountain Formation, British Columbia | Wilson (1977); Grande et al. (1982); Greenwood et al. (2005) | Wolfe et al. (2003) | Wolfe et al. (2003) |
| 2 | †*A. aggregatum* (*Amyzon gosiutensis*) | 48–45, early Middle Eocene (Bridgerian) | – | 47.09 | Laney Shale Member of the Green River formation, Wyoming | Grande et al. (1982); Bruner (1991) | Mauger (1977); Smith et al. (2010) | Smith et al. (2010) |
| 3 | †*A. brevipinne* | 49.42 ± 0.54, Ypresian-Lutetian or Bridgerian NALMA | – | 48.88 | Klondike Mountain Formation, British Columbia | Wilson (1977); Grande et al. (1982); Greenwood et al. (2005) | Wolfe et al. (2003) | Wolfe et al. (2003) |
| 4 | †*A. commune* | 34.7–33.7 Ma, Late Eocene–Early Oligocene | – | 33.7 | Florissant Formation, South Park, Colorado | Wilson (1977); Cope (1874); Bruner (1991) | Bruner (1991) | Prothero & Swisher (1992); Prothero & Sanchez (2004) |
| 5 | †*A. huanensis* | 47.8–37.8, Middle Eocene (taken as Lutetian to Bartonian) | – | 37.8 | Xiawanpu, Xiangxiang, Hunan Province, southern China | Cheng (1962); Chang et al. (2001) | Chang et al. (2001) | Walker et al. (2012) |
| 6 | †*A. mentale* | 37–34, Late Eocene–early Oligocene, Chadronian-Orellan NALMA | – | 34.0 | Osino Oil Shales, near Elko, Nevada | Cope (1872); Bruner (1991) | Bruner (1991); Cavender (1986, 1998); Harris et al. (2014) | Swisher & Prothero (1990) |
| 7 | †*Ictiobus aguilerai* | Pliocene  Blancan | – | 1.72 | Tula de Allende, Hidalgo, Mexico | Alvarado-Ortega et al. (2006) | Alvarado-Ortega et al. (2006) | Bell et al. (2004) |
| 8 | *Ictiobus cyprinellus* | 4.1–3.4a, Pliocene | 7.2 | 3.4 | Sand Draw local fauna, Brown Co., Nebraska | Smith & Lundberg (1972) | Bell et al. (2004); Near et al. (2005) | Bell et al. (2004); Near et al. (2005) |
| 9 | *Ictiobus niger* | Pleistocene Sheridanian | – | 0.3 | Kanopolis local fauna, Ellsworth Co., Kansas | Neff (1975) | Neff (1975); Repenning (1987) | Neff (1975); Repenning (1987) |
| 10 | *Ictiobus* sp. | Pleistocene Sangamonian interglacial | – | 0.055 | Lower Shuler member of the Pemberton Hill-Lewisville (T2 terrace) of the Trinity River, Dallas Co., Texas | Uyeno & Miller (1962) | Uyeno & Miller (1962); Markewich et al. (2011) | Uyeno & Miller (1962); Markewich et al. (2011) |
| 11 | †*Ictiobus* sp. cf. *bubalus* | 10–9.5 Ma, Tortonian (Miocene; Walker et al., 2012) | – | 9.5 | Laverne local fauna, Beaver Co., Oklahoma | Smith (1962) | Smith (1962); Tedford et al. (2004) | Tedford et al. (2004) |
| 12 | *Ictiobus* sp. | Pleistocene Illinoian glaciation | – | 0.135 | Mt. Scott fauna, Meade Co., Kansas | Smith (1963) | Hibbard (1963); Wood et al. (2010) | Wood et al. (2010) |
| 13 | *Ictiobus* sp. | 12.0a; medial Clarendonian | – | 10.0 | Wakeeney local fauna, Trego Co., Kansas | Wilson (1968) | Wilson (1968); Tedford et al. (1987, 2004) | Woodburne (2007) |
| 14 | †*Vasnetzovia artemica* | 37.8–28.1, Late Eocene–early Oligocene (taken as Priabonian to Rupelian) | – | 28.1 | Wuglovaya Formation, Artem lignite mine, Primorsky Krai, Russia | Sytchevskaya (1986) | Sytchevskaya (1986); Liu & Chang (2009) | Walker et al. (2012) |
| 15 | †*Plesiomyxocyprinus arratiae* | 39.74 ± 0.07, Early middle Eocene; correlated to the late Uintan NALMA | – | 39.67 | Huadian Formation, Jilin Province, China | Liu & Chang (2009) | Wang & Li (1990) | Prothero & Swisher (1992) |
| 16 | *Catostomus ardens* | 0.0112–0.01 Ma, end-Pleistocene–Holocene | – | 0.01 | Homestead Cave, Box Elder Co., Utah | Broughton (2000) | Broughton (2000) | Broughton (2000) |
| 17 | †*Catostomus arenatus* | 2.4–1.9 | – | 1.9 | Pliocene Sand Point local fauna, Glenns Ferry Formation, Idaho | Miller & Smith (1967); Smith (1975); Smith et al. (1982, 2013) | Miller & Smith (1967); Smith et al. (2013) | Miller & Smith (1967); Smith et al. (2013) |
| 18 | †*Catostomus asitus* | 4.7–4.5 | – | 4.5 | Starvation Flat area, Pliocene White Narrows formation, southern Nevada | Smith et al. (2013) | Schmidt et al. (1996); Reynolds & Lindsay (1999) | Schmidt et al. (1996); Reynolds & Lindsay (1999) |
| 19 | *Catostomus columbianus* | 2.4–1.9 | – | 1.9 | Pliocene Sand Point local fauna, Glenns Ferry Formation, Idaho | Miller & Smith (1967); Smith (1975); Smith et al. (1982, 2013) | Miller & Smith (1967); Smith et al. (2013) | Miller & Smith (1967); Smith et al. (2013) |
| 20 | *Catostomus commersoni* | Irvingtonian Land Mammal Age | – | 0.160 | Berends Fauna, Beaver Co., Oklahoma | Smith (1954) | Smith & Cifelli (2000) | Bell et al. (2004) |
| 21 | *Catostomus commersoni* | Pleistocene Illinoian glaciation | – | 0.135 | Mt. Scott fauna, Meade Co., Kansas | Smith (1963) | Hibbard (1963); Wood et al. (2010) | Wood et al. (2010) |
| 22 | *Catostomus discobolus* | 0.0112–0.01 Ma, end-Calabrian Pleistocene | – | 0.01 | Homestead Cave, Box Elder Co., Utah | Broughton (2000) | Broughton (2000) | Broughton (2000) |
| 23 | †*Catostomus hyomyzon* | 11.6 | – | 12.7b | Juntura Formation, Malheur Co., Oregon | Smith et al. (2013) | Smith et al. (2013) | Unmack et al. (2014) |
| 24 | †*Catostomus oromyzon* | 4.5, late Miocene | – | 5.5b | Glenns Ferry Formation, Owyhee Co., Idaho | Smith et al. (2013) | Smith et al. (2013) | Unmack et al. (2014) |
| 25 | †*Catostomus owyhee*c | 4.4–6.2, late Miocene–Pliocene | – | 3.0 | Glenns Ferry fauna, multiple counties, Idaho | Miller & Smith (1967); Smith (1975) | Armstrong et al. (1975); Ruez & Ginsler (2008) | Ruez & Ginsler (2008) |
| 26 | †*Catostomus shoshonensis* | 4.4–6.2, late Miocene–Pliocene | – | 3.0 | Glenns Ferry fauna, multiple counties, Idaho | Cope (1883); Armstrong et al. (1975); Smith (1975); Taylor & Smith (1981); Smith et al. (1982) | Armstrong et al. (1975); Ruez & Ginsler (2008) | Ruez & Ginsler (2008) |
| 27 | †*Catostomus* sp. cf*. tahoensis* | Pliocene | – | 2.588 | Mopung Hills, Churchill Co., Nevada | Taylor & Smith (1981) | Taylor & Smith (1981) | Taylor & Smith (1981); Gibbard et al. (2009); Walker et al. (2012) |
| 28 | *Catostomus* sp. | Pliocene | – | 2.588 Ma | Honey Lake, Lassen Co., California | Taylor & Smith (1981) | Taylor & Smith (1981) | Taylor & Smith (1981); Gibbard et al. (2009); Walker et al. (2012) |
| 29 | †*Chasmistes* sp. cf. *cujus* | Pliocene | – | 2.588 Ma | Honey Lake, Lassen Co., California | Taylor & Smith (1981) | Taylor & Smith (1981) | Taylor & Smith (1981); Gibbard et al. (2009); Walker et al. (2012) |
| 30 | †*Chasmistes spatulifer* | 4.4–6.2, late Miocene–Pliocene | – | 3.0 | Glenns Ferry fauna, multiple counties, Idaho | Miller & Smith (1967); Smith (1975) | Armstrong et al. (1975); Ruez & Ginsler (2008) | Ruez & Ginsler (2008) |
| 31 | *Moxostoma duquesnei* | Pleistocene Illinoian glaciation | – | 0.135 | Mt. Scott fauna, Meade Co., Kansas | Smith (1963) | Hibbard (1963); Wood et al. (2010) | Wood et al. (2010) |
| 32 | *Moxostoma* sp. | Miocene–Pliocene Blancan | – | 1.72 | Lake Chapala local fauna, Jalisco, Mexico | Smith et al. (1975) | Israde-Alcántara et al. (2010) | Bell et al. (2004) |
| 33 | *Xyrauchen texanus* | 5.3–3.6 Ma, Early Pliocene | – | 3.6 | Anza-Borrego Desert, San Felipe Hills, Imperial Co., California | Stewart & Roeder (1993); Hoetker & Gobalet (1999) | Stewart & Roeder (1993) | Stewart & Roeder (1993) |

a From Near et al. (2005).

b Corrected age for stem lineage branch based on method of Marshall (1990).

c Probably the sister lineage to *Deltistes*, a monotypic genus containing the species *D. luxatus*.

All fossil data are from localities within the United States, unless stated otherwise. Abbreviations: Co., county; Ma, millions of years ago; NALMA, North American Land Mammal Age; No., number; sp., species.

**3. Calibrations Used for Sucker Divergence Dating**

**Root (R).** *Node*: root node of tree containing five cypriniform outgroup taxa plus Catostomidae ingroup in our analysis. Catostomidae is placed within Cypriniformes, with parent taxon Cyprininae, by Betancur-R et al. (2013). *Character states*: catostomids share various similarities with other cypriniform groups indicating correct placement together in the order, as well as possible sister group relationships; among these, suckers share joined second and fourth pleural ribs of the Weberian apparatus with *Gyrinocheilus*; a single row of pharyngeal teeth and reduced first basibranchial with Cobitidae fishes; and lobed epibranchials, paired basipterygoid processes articulated with the first branchial arches, and enlarged, triangular supraneurals completely fused into the expanded neural complex of the Weberian apparatus with the cyprinids *Cyprinus* and *Carassius* (reviewed by Smith, 1992). *Resolution in phylogenetic analysis*:Smith’s (1992) analysis of 157 mostly morphological (but also biochemical and transition series) characters resolved Catostomidae as monophyletic relative to two cypriniform outgroup lineages, *Cyprinus* (Cyprinidae) and *Leptobotia* (Cobitidae). More recently, a phylogenetic analysis of a dataset of nine nuclear genes with a total of 7587 DNA nucleotides in the matrix resolved two species of Catostomidae in a Cypriniformes clade also containing two lineages of minnow fishes (Cyprinidae: *Danio*, *Opsariichthys*), with definitive (100%) maximum-likelihood bootstrap support (Near et al., 2012). Catostomidae is also placed in Cypriniformes along with members of the families Botiidae, Cobitidae, Cyprinidae, Gyrinocheilidae, and Nemacheilidae with high (100%) bootstrap support, based on phylogenetic analysis of 21 mitochondrial and nuclear genes for 1410 bony fish taxa by Betancur-R et al. (2013). *Absolute age estimate*: 94.9 Ma with Bayesian credible intervals of 113.3–78.9 Ma (Near et al., 2012). *Prior setting*: a normal distribution with the mean = 94.9, sigma = 9.0, and offset = 0. The mean and bounds are based on the *t*MRCA for Cypriniformes estimated by Near et al. (2012) using fossil-calibrated molecular divergence time analyses.

**4. Unsuitable Minimum Age Calibrations for Suckers**

Fossils should not be used as point estimates, because the oldest fossil available for a lineage will always underestimate the age of the true point of origin of that lineage; instead, fossils should be treated as minimum age estimates (Ho & Phillips, 2009; Parham et al., 2012). As shown above, we arrived at several reasonable minimum age priors, and some soft and hard upper bound priors, for our analyses of sucker diversification based on a synthesis of paleontological, geological, and phylogenetic information. However, given the fossil record is better for some sucker lineages (e.g. subfamily Ictiobinae, tribe Catostomini) than others (e.g. subfamily Cycleptinae or tribe Moxostomatini), and that there is tamphonomic bias in the record favoring the preservation of specimens from lacustrine or palustrine environments (Elder & Smith, 1988), fossils from relatively recent deposits (e.g. dating to <1 Ma or <0.5 Ma) should not be used to place priors on divergence times because they might vastly underestimate the timing of lineage divergence. These kinds of fossils are deceptive, because they are young in age, but they can represent lineages that have existed for several or many millions of years. In the case of the suckers, the fossils that appear most likely to underestimate the timing of lineage divergence include (1) the end-Pleistocene *C. ardens* and *C. discobolus* fossil from Homestead Cave (0.0112–0.01 Ma; Broughton, 2000), (2) late Pleistocene *C. commersoni* fossils dated to the Irvingtonian age and Pleistocene Illinoian glaciation (0.16–0.135 Ma; Smith, 1954, 1963), (3) *Ictiobus* fossils from the mid-late Pleistocene Sheridanian and Sangamonian (Neff, 1975; Uyeno & Miller, 1962), and (4) the Pleistocene Illinoian-aged *Moxostoma duquesnei* fossil from Mt. Scott fauna (Smith, 1963). As a result, we did not include these fossils in our divergence dating analyses. Other fossils reviewed above are not necessarily likely to underestimate the age of particular lineages because they are from very recent strata, but were deemed unsuitable because older age estimates were available from other fossils from the same lineage. For example, phylogenetic hypotheses indicate that the *Catostomus* subgenus *Pantosteus* forms a monophyletic lineage except for *C.* (*P.*) *columbianus* (Smith, 1992; Unmack et al., 2014), and the oldest fossil known from this lineage corresponds to a mid-Miocene †*C. hyomyzon* specimen. Thus, since several of the *Catostomus* fossils we review above (e.g. †*C. arenatus*, *C. discobolus*, and †*C. oromyzon*) are members of *Pantosteus* and dated to rocks younger than mid-Miocene in age, they were not used to calibrate *Pantosteus* nodes in BEAST.

**5. Additional Phylogenetic Relationships Within the Catostominae**

Within the Catostominae, ‘Clade 1’ (BPP = 1, most analyses) corresponded to the Thoburniini, including *Thoburnia* and *Hypentelium*. Relationships among *Moxostoma* lineages were generally unresolved; however, the concatenated mtDNA and mtDNA + morphology results congruently resolved *Moxostoma* sp. cf. *lachneri* from Surry County, in northwestern North Carolina (Great Pee Dee River), as sister to two clades with varying support that, with several exceptions, contained taxa associated with previously recognized subgenera *Scartomyzon* (‘Clade 2’; BPP = 0.74–1) and *Moxostoma* (‘Clade 3’; BPP = 0.55–0.96) (Figs. 1, S1 and S2). Alternatively, our four-locus and total-evidence topologies placed *M.* sp. cf. *lachneri* in Clade 3 with an unresolved position (Figs. 3 and S4). Subgenera *Moxostoma* and *Scartomyzon* were never resolved as monophyletic in Clade 2 or Clade 3. Within Clade 3, well-supported species groups included *M. anisurum* sister to *M. collapsum* (BPP = 1), *M. carinatum* 1 (Alabama) sister to *M.* sp. cf. *macrolepidotum* (BPP = 0.96–1), and *M. hubbsi* sister to the *M. breviceps* species group (BPP = 0.75–1) (Figs. 3, S1, S2 and S4). Alternative topologies placed *M. erythrurum* or *M. ariommum* sister to, or within, Clade 2, but with non-significant posterior support. With exceptions, relationships in Clade 2 generally collapse into a polytomy of four lineages: *M*. sp. cf. *poecilurum*; *M*. *duquesnei* sister to *M*. *poecilurum* + *M. congestum*; *M. carinatum* 2 (Minnesota) + *M. valenciennesi*; and the ‘*lachneri* group’, with *M. lachneri* sister to a clade of four Mexican suckers.

As an additional note on Clade 5, Unmack et al.’s (2014) analysis of *Pantosteus* also resolved *P. jordani*, *P. bondi*, and *P. lahontan* in a well supported clade based on mtDNA, and also showed that relationships in this clade appear not to have been influenced by introgressive hybridization events, based on comparing mtDNA and morphological characters. However, neither our study nor that of Doosey et al. (2010) sampled these lineages. Further sampling is thus required to see if these results would be supported by the nuclear loci we have examined, or by total-evidence dating analyses similar to those we employed herein.

Catostominae ‘Clade 6’ (BPP = 1;Fig. 3) contained species from basins on the eastern side of the Cascade and Sierra Mountain ranges in California, south-central Oregon, and northwestern Nevada, except for subspecies of *C. occidentalis* from the Central Valley and Pacific coast drainages of California. Subclade ‘6-a’ (BPP = 1) contained three species each restricted to a single drainage basin, including the Owens Valley sucker (*C. fumeiventris*) sister to a clade of Warner Valley (*C. warnerensis*) and Wall Canyon (*Catostomus* sp.) suckers (BPP = 1). In subclade ‘6-b’, four species from the Klamath Basin of south-central Oregon and northern California, including two species of *Catostomus*, *D. luxatus,* and *Chasmistes brevirostris*, formed a polytomy (BPP = 1). In subclade ‘6-c’, Modoc sucker (*C. microps*) were embedded within a clade containing four subspecies of *C. occidentalis* (BPP = 1).

Across analyses, we inferred a strongly supported sister relationship between a clade of *Catostomus catostomus* + *C. commersoni* (‘Clade 7’; BPP = 1) and a moderately well supported Catostominae ‘Clade 8’ + ‘Clade 9’ lineage composed of all remaining *Catostomus* (BPP = 0.71–91) (Figs. 3, S1, S2 and S4). Clade 8 contained species from the coastal rivers of Oregon, the Columbia River basin, the Great Basin, and Bonneville Basin resolved in a polytomy (BPP = 1). In the four-locus and IRBP analyses, which had the largest sample sizes we considered, *Chasmistes liorus mictus* and *C. ardens* from the Bonneville Basin were each inferred to be para-/polyphyletic in Clade 8 (Figs. S3 and S5). Also within Clade 8, *C. columbianus* from the Columbia River basin was sister to *C. tahoensis* from the Great Basin (BPP = 0.99–1). *Catostomus macrocheilus* from the Columbia River basin was resolved in a clade with *C. tsiltcoosensis* (Kettratad and Markle, 2010) and *Catostomus* sp. (Coquille River), both species from rivers of coastal Oregon; relationships among these taxa received definitive support (BPP = 1), except in the four-locus analysis where support varied (BPP = 0.79–0.99). ‘Clade 9’ contained species from the Colorado River basin, with *C. latipinnis* usually sister to a clade of *C. insignis* + *Xyrauchen texanus* in subclade ‘9-a’; a close affinity among members in this clade was strongly supported by analyses of the concatenated mtDNA, four-locus, and total-evidence datasets (BPP = 0.96–1) (Fig. 3). Four Mexican *Catostomus* species were placed in subclade ‘9-b’ with definitive support (BPP = 1); however, *C. bernardini* was polyphyletic in the concatenated mtDNA analysis, because specimens from the Río Batopilas formed a clade distinct from the Papagochic and Yecora rivers (Fig. S1).

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