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New phylogenetic insights into the African catfish families Mochokidae and Austroglanididae

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Abstract

Several hundred catfish species (order: Siluriformes) belonging to 11 families inhabit Africa, of which at least six families are endemic to the continent. Although four of those families are well-known to belong to the 'Big-Africa clade', no previous study has addressed the phylogenetic placement of the endemic African catfish family Austroglanididae in a comprehensive framework with molecular data. Furthermore, interrelationships within the 'Big-Africa clade', including the most diverse family Mochokidae, remain unclear. This study was therefore designed to help reconstruct inter- and intrarelationships of all currently valid mochokid genera, to infer their position within the 'Big Africa clade' and to establish a first molecular phylogenetic hypothesis of the relationships of the enigmatic Austroglanididae within the Siluriformes. We assembled a comprehensive mitogenomic dataset comprising all protein coding genes and representing almost all recognized catfish families ($N = 33$ of 39) with carefully selected species ($N = 239$). We recovered the monophyly of the previously identified multifamily clades 'Big Asia' and 'Big Africa' and determined Austroglanididae to be closely related to Pangasiidae, Ictaluroidea and Ariidae. Mochokidae was recovered as the sister group to a clade encompassing Auchenoglanididae, Claroteidae, Malapteruridae and the African Schilbeidae, albeit with low statistical support. The two mochokid subfamilies Mochokinae and Chiloglanidinae as well as the chiloglanid tribe Atopochilini were recovered as reciprocally monophyletic. The genus *Acanthocleithron* forms the sister group of all remaining Mochokinae, although with low support. The genus *Atopodontus* is the sister group of all remaining Atopochilini. In contrast to morphological reconstructions, the monophyly of the genus *Chiloglanis* was strongly supported in our analysis, with *Chiloglanis macropterus* nested within a *Chiloglanis* sublineage encompassing only other taxa from the Congo drainage. This is an important result because the phylogenetic relationships of *C. macropterus* have been controversial in the past, and because we and other researchers assumed that this species would be resolved as sister to most or all other members of *Chiloglanis*. The apparent paraphyly of *Synodontis* with

respect to *Microsynodontis* provided an additional surprise, with *Synodontis punu* turning out to be the sister group of the latter genus.

KEYWORDS
Acanthocleithron, *Atopodontus*, *Austroglanis*, Big Africa clade, Siluriformes

1 | INTRODUCTION

The order Siluriformes (Superorder: Ostariophysi; Series: Otophysi), commonly known as catfishes, represents one of the most diverse vertebrate groups on our planet, with currently 4019 valid described species and 500 genera assigned to 39 families (Fricke *et al*., 2021). However, the exact number of catfish families remains debated because of conflicting views about familial classifications (Betancur-R. *et al*., 2017; Diogo & Peng, 2010; Ferraris Jr., 2007; Nelson *et al*., 2016; Sullivan *et al*., 2006). Nevertheless, the monophyly of the Siluriformes is well supported by morphological and molecular data (Betancur-R. *et al*., 2017; Fink & Fink, 1981, 1996; Hughes *et al*., 2018). Systematists typically recognize three major clades within the siluriform catfishes, two of which (Loricarioidei and Diplomystoidei) are restricted to the freshwater systems of South America, whereas the third (Siluroidei) is globally distributed and contains some families that have secondarily adapted to brackish and even marine environments (Betancur-R, 2009; Covain & Fisch-Muller, 2007; Diogo, 2004; Stange *et al*., 2016). Although the order has a global distribution, many catfish families occur on only one continent, and are sometimes restricted to a comparatively small geographic area. For example, the family Diplomystidae is endemic to southern South America, the family Austroglanididae is confined to Southern Africa, and the recently described family Lacantuniidae inhabits only Central America (Armbruster, 2011; Rodiles-Hernandez *et al*., 2005; Skelton, 2001). Because of their Pangaean origin probably dating back to the Early Cretaceous, siluriform catfishes became important model systems for testing key biogeographic questions (Kappas *et al*., 2016; Rivera-Rivera & Montoya-Burgos, 2017; Sullivan *et al*., 2006). Considerable progress has been made over the past two decades to reconstruct the evolutionary history of the Siluriformes through use of different molecular markers (Arcila *et al*., 2017; Betancur-R. *et al*., 2017; Hardman, 2005; Kappas *et al*., 2016; Nakatani *et al*., 2011; Sullivan *et al*., 2006). These studies have helped to shed some light on relationships within some superfamily and multifamily clades, for example by providing consistent support for the monophyly of the 'Big Asia clade' and 'Big Africa clade' *sensu* Sullivan *et al*. (2006). However, many interfamilial relationships remain poorly resolved and hence controversial.

Uncertainties also still persist for intrafamilial relationships, even for some of the comparatively well-studied catfish families such as Mochokidae. This family is endemic to Africa and belongs to the 'Big Africa clade' (*sensu* Sullivan *et al*., 2006). It is the most species-rich catfish family on the continent and it is currently represented by 224 valid species (Fricke *et al*., 2021). Their largest density of species occurs in the Congo basin, but mochokids are ubiquitous in all tropical African river drainages and basins. *Synodontis* Cuvier, 1816 has radiated into several species flocks within the East African Great Lakes (Day *et al*., 2009, 2013; Koblmüller *et al*., 2006). The oldest mochokid fossil, which is tentatively assigned to *Synodontis*, dates to the Early Oligocene and was found in Oman, suggesting that the family's distribution was wider in the past (Otero & Gayet, 2001).

Following the most recent taxonomic revision by Vigliotta (2008) and the description of the genus *Atopodontus* Friel & Vigliotta, 2008, nine mochokid genera are currently recognized. Three of these, *Mochokiella* Howes, 1980, *Acanthocleithron* Nichols & Griscom, 1917 and *Atopodontus* are monotypic, whereas two, *Synodontis* and *Chiloglanis* Peters, 1868, are extremely species-rich. Two subfamilies are recognized. These are Chiloglanidinae Riehl & Baensch, 1990, which is characterized by species with lips and barbels modified into an oral disc (suckermouth), and Mochokinae Jordan, 1923, which comprises species without suckermouths (Seegers, 2008). A comprehensive morphological investigation of all mochokid genera revealed strong support for the monophyly of Chiloglanidinae, but recovered the Mochokinae as paraphyletic (Vigliotta, 2008). That study also recovered a monophyletic group encompassing the chiloglanidin genera *Microsynodontis* Boulenger, 1903 and *Synodontis*, and suggested the paraphyly of *Chiloglanis* and *Euchilichthys* Boulenger, 1900. Interestingly, the single representative of *Atopochilus* Sauvage, 1879 in this study nested phylogenetically within *Euchilichthys*, while *Chiloglanis macropterus* Poll & Stewart, 1975 was recovered as a sister group to all remaining chiloglanidin taxa.

Mochokids have also attracted considerable attention in biogeographical research because of their almost pan-African distribution. The species-rich genus *Synodontis* provides a particularly excellent model for testing the role of geological processes in promoting lineage diversification and shaping present-day biogeographic patterns on a continental scale (Day *et al*., 2009, 2013; Pinton *et al*., 2013). Furthermore, a growing number of studies focus on mochokid biogeography at a regional scale, most notably within the species-rich and widely distributed genus *Chiloglanis* (Chakona *et al*., 2018; Morris *et al*., 2016; Schmidt *et al*., 2014, 2016; Watson, 2020).

The discovery of many endemic and often cryptic candidate species of *Chiloglanis* underscores the fact that our knowledge of mochokid diversity and phylogenetic relationships is still incomplete. This gap persists mainly because previous phylogenetic studies focused only on *Synodontis* or *Chiloglanis*, or considered only higher-level relationships of Siluriformes and included relatively

few mochokid taxa. Day *et al*. (2013) achieved the most complete mochokid taxon sampling so far. This study of continental diversification within *Synodontis* used a molecular dataset with four genes and included representatives of most known mochokid genera as outgroups. Only *Atopodontus* and *Acanthocleithron* were missing. Interestingly, the generic interrelationships recovered from this study contradicted those inferred from the morphological data of Vigliotta (2008) in some respects. Most importantly, Day *et al*. (2013) recovered the monophyly of the Mochokinae, with *Mochokiella* being the sister group of *Synodontis*, whereas in Vigliotta's (2008) study *Mochokus* followed by *Mochokiella* were recovered as lineages originating at the oldest splits within Mochokidae.

To date, the phylogenetic positions of *Atopodontus* and *Acanthocleithron* remain unknown. To our knowledge no DNA sequences of *Atopodontus* have been published but there is a single *COX1* barcode sequence for *Acanthocleithron chapini* Nichols & Griscom, 1917 available. That sequence was generated in a barcoding project focusing on the ichthyological diversity of the north-eastern Congo basin (Decru *et al*., 2016) and has never been included in an evolutionary analysis.

Similarly, the South African family Austroglanididae remains one of the most enigmatic clades in terms of its phylogenetic placement because no comprehensive molecular phylogeny of the order has included it (Kappas *et al*., 2016; Rivera-Rivera & Montoya-Burgos, 2018; Sullivan *et al*., 2006). The only available genetic data for this family originated from a pair of studies that focused on population structure and the relationships among the three species of *Austroglanis*. Skelton *et al*. (1984) used mitochondrial markers (cytochrome b and 16 S) for this purpose while Cunningham, Bills & Swartz (unpublished data, as cited in Bills and Impson (2013)) employed allozymes. In contrast, several cladistic studies aimed to elucidate the placement of this catfish family using morphological data, but the resulting phylogenetic hypotheses were diverse and often contradictory. For example, Skelton *et al*. (1984) inferred that *Austroglanis* was closely related to the family Bagridae (which at that time included the family Claroteidae), whereas Mo (1991) inferred a close relationship with the family Cranoglanididae (cladogram II) and raised the genus to family level. Subsequently, de Pinna (1993) recovered a sister group relationship of Austroglanididae with a large clade encompassing several families, including Ariidae, Anchariidae, Bagridae, Claroteidae, Schilbeidae, Pangasiidae, Pimelodidae and the genus *Horabagrus* Jayaram, 1955. Finally, Diogo (2005) and Diogo and Bills (2006) suggested a close relationship between Austroglanididae and the families Ariidae, Claroteidae, Ictaluridae and Cranoglanididae based on four derived morphological characters.

This present study aimed to resolve the unsettled phylogenetic question regarding the placement of Austroglanididae within Siluriformes and to improve the understanding of the inter- and intrageneric relationships of Mochokidae. To achieve these tasks, we took advantage of the constantly growing body of mitochondrial genome data available for Siluriformes (*i.e*., Kappas *et al*., 2016; Ma *et al*., 2015; Moreira *et al*., 2017; Nakatani *et al*., 2011) which we combined with newly sequenced partial mitochondrial genomes of representative taxa of all mochokid genera as well as of all described *Austroglanis* species. Furthermore, we compiled a small nuclear dataset (rag 1 and rag 2) based on the dataset provided by Sullivan *et al*. (2006) and newly obtained sequence data of the family Austroglanididae. We used these data to reconstruct the most comprehensive phylogeny of Mochokidae to date, showing the intrageneric relationships of all currently valid mochokid genera, including the previously omitted *Atopodontus* and *Acanthocleithron*. The reconstruction also confirms Mochokidae's position within the 'Big Africa clade' and infers the placement of Austroglanididae within Siluriformes for the first time.

2 | MATERIALS AND METHODS

2.1 | Taxon sampling

We included mitogenome data of 256 specimens (approx. 239 species) composed of concatenated alignments of all mitochondrial protein coding genes (see Supporting Information Table S1; hereafter referred to as the 'all-catfish dataset'). The taxon sampling for newly generated sequences targeted mainly the catfish clade 'Big Africa' (*sensu* Sullivan *et al*., 2006), with a specific focus on Mochokidae (40 specimens, 32 species) and its two subfamilies Mochokinae (14 specimens/species) and Chiloglanidinae (26 specimens, 18 species). This is the first dataset to ever include representatives of all nine mochokid genera. Furthermore, the taxon sampling includes representative taxa of all nominal African catfish families associated with the 'Big Africa clade': Auchenoglanididae ($N = 2$), Amphiliidae ($N = 2$), Claroteidae ($N = 4$), Schilbeidae ($N = 4$) and Malapteruridae (N = 1). Unfortunately, we were unable include *Lacantunia enigmatica* Rodiles-Hernández, Hendrickson & Lundberg 2005 in our mitogenome taxon sampling, the only member of the Mesoamerican family Lacantuniidae and a member of the 'Big Africa clade' (Lundberg *et al*., 2007). In addition, we sequenced partial mitochondrial genomes of all three described species of the phylogenetically unplaced family Austroglanididae. A comprehensive overview of the newly sequenced catfish specimens for this study (52 specimens and 43 species, see Supporting Information Table S1), including photos of either of living or of preserved specimens and X-rays, is available in Supporting Information S1.

The all-catfish dataset was compiled by downloading all available mitochondrial genomes for the catfish sister group order Gymnotiformes ($N = 8$) and Siluriformes, including representative members of all three major catfish lineages: Loricarioidei ($N = 40$), Siluroidei ($N = 157$) and Diplomystoidei ($N = 1$). All taxon names were checked for current validity using Eschmeyer's Catalogue of Fishes (Fricke *et al*., 2021). In cases where more than one mitochondrial genome was available for a species, we randomly selected one representative sequence to be included in our data set (see Supporting Information Table S1 for GenBank accession numbers). Some sequences/taxa obtained from GenBank were identified as of doubtful quality or identity and excluded after a preliminary phylogenetic

analysis due to their highly unlikely phylogenetic position (for further details see Supporting Information S2).

Furthermore, we downloaded all available *COX1* sequence data for the family Mochokidae from GenBank (646 sequences, see Supporting Information Table S2) and extracted the *COX1* sequence data of the partial mitochondrial genomes of the newly sequenced mochokids. This second data set (hereafter referred to as the 'mochokid *COX1* dataset') included 688 specimens (representing approximately 105 mochokid species, including several undetermined and/or undescribed species) and covered much of the natural distribution of the family. This *COX1* dataset enabled us to investigate the taxonomic assignment of selected mochokid taxa, such as *Chiloglanis niloticus* Boulenger, 1900 and *Synodontis punu* Vreven & Milondo, 2009.

In addition to the mitochondrial data, we obtained partial sequences for the nuclear recombination activating genes *rag1* and *rag2* from GenBank. This nuclear dataset is mainly based on the data provided by Sullivan *et al*. (2006) and included 114 Siluriformes, to which we added new sequence data of all three *Austroglanis* species (see Supporting Information Table S2). The nuclear dataset includes two species of Gymnotiformes and one characiform as outgroups. This third DNA dataset provided an opportunity to investigate phylogenetic relationships of the family Austroglanididae with more slowly evolving nuclear markers. We refrained from constructing a combined dataset of nuclear and mitochondrial data because matching mitochondrial and nuclear data were available for only for 29 species, and thus the combined matrix would have contained a very high proportion of missing data.

2.2 | Sampling procedures

The tissues used in this study were collected on various field expeditions and sourced from available museum specimens. Fish were caught using various techniques (*i.e*., fish traps, frame and dip nets, gillnets and beach seines) depending on the sampled habitats and local conditions or were obtained from local fishermen. Freshly caught fish were sacrificed by an overdose of approved fish anaesthetic (*i.e*., MS222) and photographed in a cuvette (most specimens). Fin clips or muscle plugs were taken and immediatley preserved in 96% ethanol, and corresponding specimens were fixed in formalin (4% formaldehyde in aqueous solution) and later transferred into 75 % ethanol for long-term storage following Neumann (2010). All relevant ethical standards and applicable national laws for collecting, sampling and export of specimens were considered. Tissue samples and corresponding export documentation for most investigated specimens are stored at the SNSB-bavarian state collection of zoology (SNSB-ZSM). Those for *Atopodontis adriaensi*, *Atopochilus savorgnani* and *Synodontis punu* are available from oregon state university ichthyology collection (OS) and were collected with all appropriate permits following Animal Care and Use Protocol #4909 from Oregon State University. Samples and specimens of *Austroglanis* are deposited in the National Fish Collection Facility at the NRF-SAIAB. The sampling approaches that were used for collecting the corresponding *Austroglanis* samples were approved by the National research foundation – South African institute for aquatic biodiversity (NRF-SAIAB) animal ethics committee. Research permits were issued by Cape Nature and Northern Cape authorities and are deposited at the National Fish Collection Facility at the NRF-SAIAB. See Acknowledgements for more detailed permit information.

2.3 | Molecular methods and phylogenetic analysis

Partial mitochondrial genomes were sequenced by the Sequencing Service of the Ludwig Maximilian University of Munich on an Illumina MiSeq platform (MiSeq Reagent Kit v2; 2X250). Individual library preparations were based on two different approaches. First, total genomic DNA was extracted for all samples following a custom cetyltrimethylammonium bromide (CTAB) DNA extraction protocol and DNA concentrations were quantified using a spectrophotometer (NanoDrop ND-1000, Thermo Scientific, Wilmington, USA) and adjusted to 25 ng μl^{-1} per sample. Subsequently, the libraries were prepared following the first approach, which was based on the amplification of a large fragment of the mitochondrial genome (~13,000 bp, including all mitochondrial protein coding genes) using the TaKaRa LA Taq DNA (Takara Bio Inc., Shiga, Japan) polymerase kit and with the following primer pair (taken from Abwe *et al*., in prep): Amp2571F (TTC AAC GAT TAA AGT CCT ACG TGA TCT GAG) and Amp15543R (TTT AAC CTT CGA TCT CCG GAT TAC AAG AC). Furthermore, we used a modified thermal profile from Schedel *et al*. (2019) for the amplification reactions: initial denaturation at 98°C (60 s), followed by 35 cycles of denaturation 98°C (10 s), annealing at 58°C for 60s, elongation at 68°C (15 min) and a last extension step at 72°C (10 min). Successfully amplified PCR products were excised from agarose gels and purified using the GeneJET Gel Extraction Kit (Thermo Fisher Scientific) following the manufacturer's protocol. Subsequently, DNA concentrations of cleaned amplification products were adjusted to 0.21 ng μl^{-1} per sample and individual libraries were prepared using the Nextera XT DNA Sample Preparation Kit (Illumina Inc, San Diego, CA, USA) following the manufacturer's protocol until to the library normalization step. The latter was conducted by pooling all libraries equimolarly based on the DNA concentration and the size distribution of the individual libraries, resulting in a final pool containing the 'amplified partial mitochondrial genomes'.

The second library preparation approach was only applied for selected samples for which the long-range PCR failed. In those cases, we used directly the extracted DNA (adjusted to 0.21 ng μl^{-1} per sample) as input template for the Nextera XT DNA Sample Preparation Kit (Illumina), which was conducted as described above. The resulting shotgun libraries were pooled equimolarly as described above. Once sequenced we used Geneious v.11.0.4 (Kearse *et al*., 2012) and the plugin BBDuk Trimmer for quality control, and for adaptor trimming of the demultiplex reads. We *de novo* assembled the amplified partial mitochondrial genomes using the 'De Novo Assembly' function implemented in Geneious. Sequence reads derived from the shotgun libraries were first mapped against the complete mitochondrial genome of *Synodontis schoutedeni* (GenBank accession number: AP012023.1; Nakatani *et al*., 2011) and the resulting contigs were subsequently used for further analysis. The assembled contigs were finally annotated using the

S. schoutedeni mitochondrial genome as a reference and uploaded to GenBank (GenBank accession numbers: MZ930069-MZ930120; see Supporting Information Table S1).

2.4 | Mitochondrial analysis

To prepare a protein-coding gene alignment we extracted from newly generated and GenBank mitogenome data the sequence information of all mitochondrial protein-coding genes (*ND1*, *ND2*, *COX1*, *COX2*, *ATP8*, *ATP6*, *COX3*, *ND3*, *ND4L*, *ND4*, *ND5*, *ND6*, *CytB*) of all specimens included in our all-catfish dataset ($N = 256$). Subsequently, we aligned sequences of individual genes using the Geneious alignment tool (default settings). In a few cases where the sequence information for an individual gene was incomplete or missing (*i.e.*, due to poor sequence quality) a multi-N string was inserted in the respective position of the corresponding alignment. Finally, single gene alignments were concatenated resulting in an alignment of 11,400 bp with relative base frequencies of A = 28.8%, T = 28.6%, C = 15.1% and G = 27.4% and about 0.5% of missing data (excluding gaps and ambiguous sites). We assessed the extent of substitution saturation of our alignment using Xia's test (Xia *et al*., 2003; Xia & Lemey, 2009) and plotted the observed number of transitions and transversions against the corrected genetic distances using the program DAMBE v.7.2.152 (Xia, 2018). The program jModelTest (Posada, 2008), implemented on the CIPRES Science Gateway (Miller *et al*., 2010), was used to calculate the best substitution model based on the Akaike information criterion: GTR + I + G. RAxML v.8.2.12 (Stamatakis, 2014), as implemented on the CIPRES Science Gateway was used for maximum likelihood (ML) inference of phylogenetic relationships by further partitioning the dataset according to first, second and third codon positions, and assigning all included gymnotiform taxa as outgroups based on evidence from previous phylogenetic studies (Arcila *et al*., 2017; Betancur-R. *et al*., 2017; Betancur-R *et al*., 2019; Hughes *et al*., 2018). Since the third codon position of the all-catfish dataset showed signatures of saturation we also ran an ML analysis on the all-catfish dataset using the same parameters but excluding the third codon position to explore whether the saturation affected the inference.

Furthermore, a Bayesian Inference (BI) analysis was conducted for the all-catfish dataset using MrBayes 3.2.7a (Huelsenbeck & Ronquist, 2001; Ronquist *et al*., 2012). Two Markov Chain Monte Carlo (MCMC) runs were calculated simultaneously for 15 million generations, with tree space sampled every 1000th generation. We discarded the first 3.75 million generations as burn-in before checking the convergence of all parameters, that is, that all estimated sample size (ESS) values were above 200. The remaining trees were then used to calculate the consensus tree.

We also aligned all available *COX1* sequences of the family Mochokidae and representative taxa of the family Amphiliidae (mochokid *COX1* dataset) resulting in alignment of 1375 bp and 688 specimens. An ML analysis was conducted on this alignment in RAxML using the same substitution model and partition scheme as for the all-catfish dataset. Amphiliidae sequences were used as the outgroup taxa.

2.5 | Nuclear analysis

In a final step, we mapped the demultiplexed, trimmed reads of the four shotgun-sequenced *Austroglanis* libraries (~4.326.187 reads per specimen) against two nuclear markers which were previously used to infer phylogenetic relationships among Siluriformes (see Sullivan *et al*., 2006): *rag1* (exon 1, 2 and 3) and *rag2*. For doing so we used the 'Map to Reference' option in Geneious v.11.0.4 and, as references, sequences of the corresponding markers [GenBank accession numbers: *rag1* (exons 1 + 2): DQ492636, *rag1* (exons 3): JN020097, *rag2*: JN020130] derived from different *Pangasius* Valenciennes 1840 (in Cuvier & Valenciennes, 1840) species. This choice was based on the phylogenetic analysis conducted on our all-catfish dataset, which recovered Pangasiidae as sister to Austroglanididae (see below). The resulting contigs were extracted and aligned to the corresponding sequence data obtained from GenBank (see Supporting Information Table S2). For both gene alignments we first obtained neighbour-joining trees calculated using the Geneious tree builder. Then we concatenated both alignments and inferred phylogenetic relationships using RAxML. As in the other analyses we partitioned the dataset into first, second and third codon positions, and defined the included gymnotiform taxa as outgroup. The newly obtained *rag1* and *rag2* sequences are available on GenBank under the accession numbers MZ895066–MZ895072 (see Supporting Information Table S2).

3 | RESULTS

3.1 | Partial mitogenomes and nuclear makers

Amplification of partial mitochondrial genomes and subsequent sequencing on an Illumina MiSeq platform was successful for 44 of the 52 specimens considered herein, yielding for most samples a single contig sequence; in a few cases two sequences and in a single case three contigs were retrieved (mean coverage 400, mean sequence length 10,704 bp). The remaining eight samples, including four mochokids and all four studied *Austroglanis* samples, were shotgun-sequenced from total genomic DNA. These efforts yielded an average of 2.75 million raw reads per sample of which approximately 0.05% mapped against the mitochondrial reference genome, returning between two and four contig sequences per sample. Almost complete mitochondrial genomes were recovered for *Chiloglanis* sp. Nigeria and *Austroglanis gilli* (Barnard, 1943) (mean coverage 52, mean sequence length 5832 bp).

Mapping of shotgun-sequenced *Austroglanis* reads against the nuclear markers was only partially successful. All four *Austroglanis* samples reads could be mapped against the reference sequence of *rag1* (exon 1, 2) thereby yielding four contig sequences (mean coverage 3, mean sequence length 1374 bp) but no reads mapped against the reference sequence for *rag1* (exon 3). Therefore, we decided to exclude the *rag1* (exon 3) from downstream analysis. Mapping against the reference sequence of *rag2* was successful for three of the four

Austroglanis samples, yielding three contig sequences (mean coverage 3.4, mean sequence length 933 bp).

3.2 | Siluriform phylogenetic relationships based on partial mitochondrial genomes

The ML analysis (Figures 1 and S1) based on all mitochondrial protein coding genes (all-catfish dataset) strongly supported the monophyly of the Siluroidei (BS: 100) but the monophyly of the Loricarioidei was only moderately supported (BS: 80). Diplomystoidei was recovered as the sister group to the Siluroidei, albeit with low support (BS: 61). A weakly supported clade (BS: 31) composed of the families Plotosidae, Chacidae, Cetopsidae and Ritidae was recovered as originating at the earliest split within Siluroidei.

As in previous phylogenetic studies (*i.e.*, Arcila *et al.*, 2017; Betancur-R. *et al.*, 2017; Kappas *et al.*, 2016; Silva *et al.*, 2021; Sullivan *et al.*, 2006), several multifamily clades were recovered, including (a) a clade (BS: 73) composed of *Aspredinidae* + Doradoidea (Doradidae + Auchenipteridae), (b) Clarioidea (Clariidae + Heteropneustidae; BS: 100), (c) Pimelodoidea (Pimelodidae + Pseudopimelodidae; BS: 100, note that the family Heptapteridae and the genus *Conorhynchos* Bleeker, 1858 are missing in our analysis), (d) Ictaluroidea (Ictaluridae + Cranoglanididae (BS: 100), (e) the informal clade 'Big Asia' (BS: 100) and (f) the informal clade 'Big Africa' (BS: 78). Furthermore, we recovered a clade (BS: 70) encompassing Ictaluroidea, Ariidae, Pangasiidae and Austroglanididae as the sister group of the 'Big Africa clade' again with moderate support (BS: 71). Within this clade the clade comprising all Austroglanididae was recovered as sister to Pangasiidae with moderate support (BS: 72). Interrelationships of these siluroid multifamily clades were only poorly supported, as in previous studies focusing on the reconstruction of siluriform interfamily relationships (*i.e.*, Kappas *et al.*, 2016; Sullivan *et al.*, 2006).

Apart from the megadiverse family Loricariidae (BS: 38), the monophyly of most catfish families was highly supported (BS: >95). One unexpected discovery was that *Eutropiichthys vacha* (Hamilton, 1822) nests within the pangasid genus *Pangasianodon* Chevey, 1931, which contradicts the current taxonomic assignment of *Eutropiichthys* to the family Schilbeidae (Wang *et al.*, 2016). Furthermore, our analyses recovered paraphyletic or polyphyletic genera within the Loricariidae, Bagridae, Sisoridae, Siluridae, Ariidae, Pangasiidae, Schilbeidae and Mochokidae (see Supporting Information Figure S1).

3.3 | Results excluding saturated third codon positions

The topology of the ML analysis (see Supporting Information Figure S2) based on the all-catfish dataset but with the third codon position removed from the alignment was widely congruent with that of the full dataset. Nevertheless, several topological differences concerning interfamily and multifamily clade relationships are detectable between the two analyses. For example, the multifamily clade (including the Plotosidae, Chacidae, Cetopsidae and Ritidae) recovered as originating at the earliest divergence within Siluroidei based on the full dataset was not recovered in this analysis. Instead, Ritidae and Ariidae were reconstructed as successive sister groups to all remaining members of Siluroidei. Several traditionally recognized multifamily clades were recovered with lower support compared to the full data set, that is, the 'Big Africa clade' (BS: 42 *vs.* 71); others were not recovered at all, that is, the 'Big Asia clade'. Loricariidae, Doradidae and Auchenipteridae were recovered as paraphyletic or polyphyletic. Austroglanididae was recovered as sister group to Ictaluroidea, but with low support (BS: 32). The recovered intrageneric and intergeneric relationships within Mochokidae were generally identical to those of the full dataset but often with lower BS support (*i.e.*, that of *Chiloglanis niloticus*). However, see below for some important exceptions. These comparisons suggest that although third codon positions in the all-catfish dataset show signatures of saturation, those loci still carry critical phylogenetic information, especially at the intrafamilial level.

3.4 | Congruence of ML and BI analyses

Likewise, the topology of the BI analysis (see Supporting Information Figure S3) based on the all-catfish dataset was widely congruent with that of the ML analysis with respect to intrafamilial relationships. The monophyly of the three superfamilies Siluroidei, Diplomystoidei and Loricarioidei and their relationships to each other were identical to the results of the ML analysis. Both analyses recovered the major multifamily clades, but the interrelationships among these clades showed some differences. For example, the BI analysis obtained the 'Big Asia clade' as sister to a clade encompassing the 'Big Africa clade' and the multifamily clade encompassing Ictaluroidea, Ariidae, Pangasiidae and Austroglanididae.

3.5 | Phylogenetic relationships within Mochokidae

The monophyly of Mochokidae (see Figure 2) was highly supported (BS: 96) by our ML analysis based on the all-catfish dataset. While our analysis strongly supported the monophyly of the subfamily Chiloglanidinae (BS: 100), it only weakly supported that of the Mochokinae (BS: 64). Within Chiloglanidinae, the genus *Chiloglanis* was recovered as sister group to all remaining chiloglanidin genera (*Atopodontus*, *Euchilichthys*, *Atopochilus*). *Acanthocleithron* was recovered as sister group to a strongly supported clade encompassing all other mochokin taxa (BS: 100). Within this clade the first split separated *Mochokus* from all other lineages. Unexpectedly, *Synodontis punu* was recovered as sister to *Microsynodontis*, rendering the genus *Synodontis* polyphyletic. This finding was congruent with the ML analysis of the mochokid *COX1* dataset which included additional specimens of *S. punu* as well as of *Microsynodontis* (see Supporting Information Figure S4).

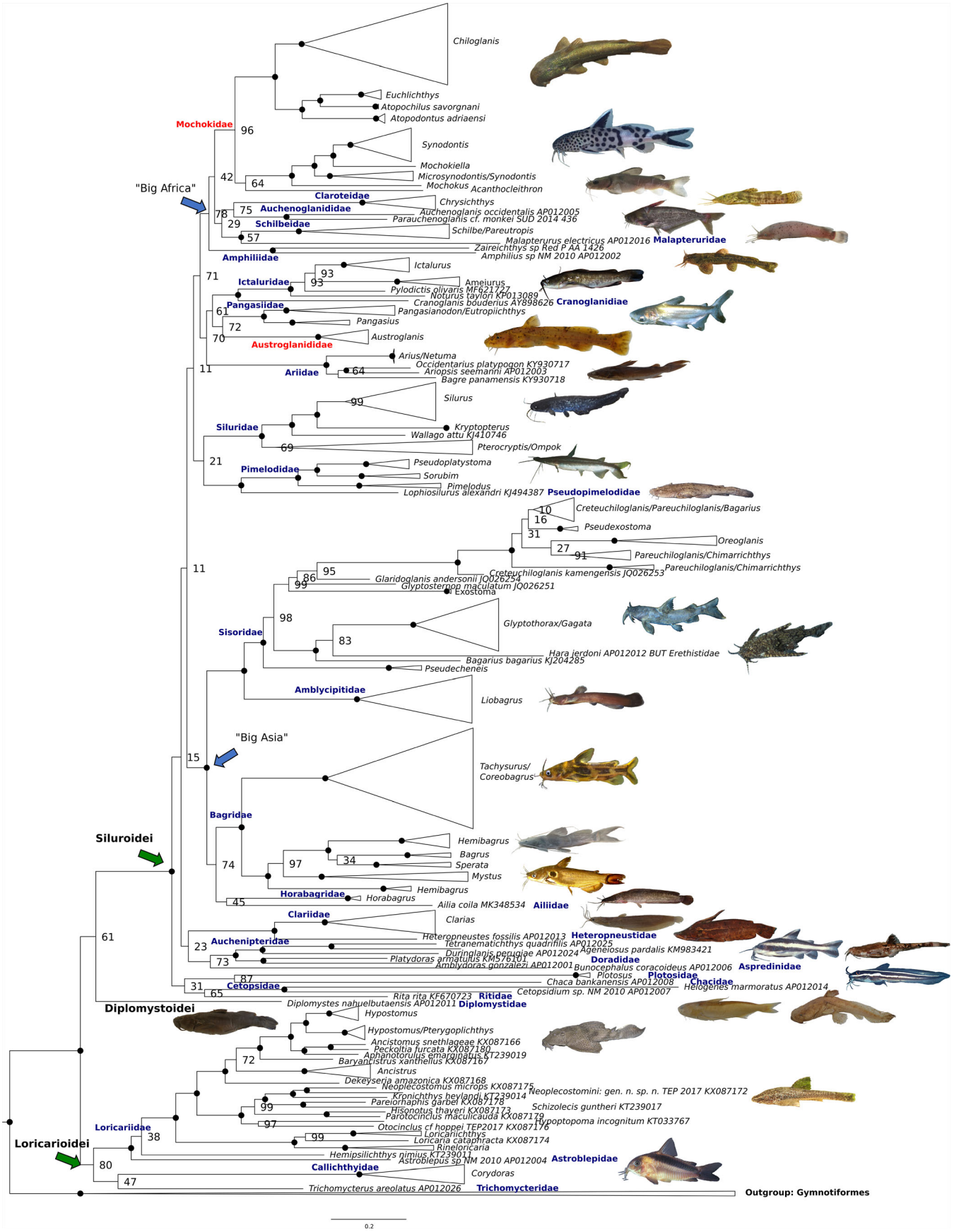

FIGURE 1 Legend on next page.

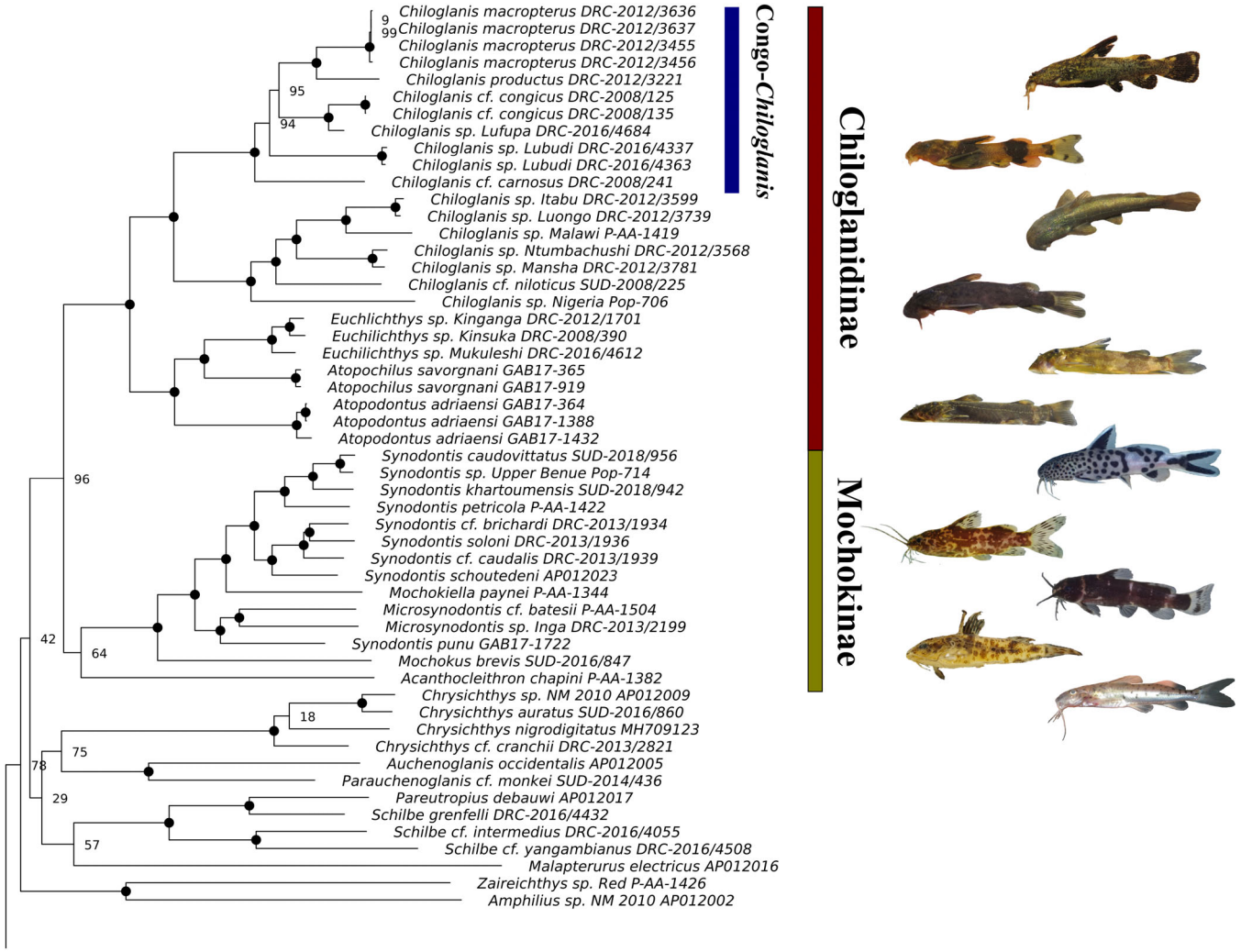

FIGURE 2 Detailed relationships among the 'Big African clade' (pruned from the ML phylogeny depicted in Figure 1). Bootstrap proportion based on 1000 BS replicates are indicated at nodes either by numbers or by black dots (BS = 100). Represented mochokid species depicted from top to bottom (photographers and corresponding sample ID in brackets): *Chiloglanis macropterus* (F.D.B. Schedel, DRC-2012/3637), Chiloglanis sp. 'Lufupa' (F.D.B. Schedel, DRC-2016/4684), *Chiloglanis* sp. 'Ntumbachushi' (F.D.B. Schedel, DRC-2012/3568), *Euchilichthys* sp. Mukuleshi (F.D.B. Schedel, DRC-2012-4612), *Atopochilus savorgnani* (B. Sidlauskas, GAB17-365), *Atopodontus adriaensi* (B. Sidlauskas, GAB17-364), *Synodontis petricola* (F.D.B. Schedel, P-AA-1422), *Mochokiella paynei* (F.D.B. Schedel, P-AA-1344), *Microsynodontis* cf. *batasii* (F.D.B. Schedel, P-AA-1504), *Mochokus brevis* (F.D.B. Schedel, SUD-2016-847), *Acanthocleithron chapini* (E.J.W.M.N. Vreven, P-AA-1382)

FIGURE 1 ML phylogeny (RAxML) of the order Siluriformes, including 30 catfish families (indicated either in blue or in red = target families), based on all mitochondrial protein coding genes (11,400 bp). Bootstrap proportion based on 1000 BS replicates are indicated at nodes either by numbers or by black dots (BS = 100). Genera including more than one taxon are collapsed (see Supporting Information Figure S1 for the noncollapsed tree). Representative species of most included catfish families/subfamilies are depicted from top to bottom (photographers in brackets, specimen/species not necessarily included in the dataset): Mochokidae; Chiloglanidinae: *Chiloglanis* sp. Ntumbachushi (F.D.B. Schedel), Mochokidae; Chiloglanidinae: *Synodontis petricola* (F.D.B. Schedel), Claroteidae: Chrysichthys (F.D.B. Schedel), Auchenoglanididae: *Parauchenoglanis* cf. *monkei* (F.D.B. Schedel), Schilbeidae: *Schilbe grenfelli* (F.D.B. Schedel), Malapteruridae: *Malapterurus* sp. (F.D.B. Schedel), Amphiliidae: *Amphilius* sp. (F.D.B. Schedel). Ictaluridae: *Ameiurus nebulosus* (F.D.B. Schedel), Pangasiidae: *Pangasius pangasius* (E. Schraml), Austroglanididae: *Austroglanis barnardi* (Roger Bills), Ariidae: *Ariopsis* cf. *guatemalensis* (preserved specimen; F.D.B. Schedel), Siluridae: *Silurus glanis* (E. Schraml), Pimelodidae: *Sorubim lima* (E. Schraml), Pseudopimelodidae: *Lophiosilurus alexanderi* (E. Schraml), Sisoridae: *Glyptothorax* cf. *sinensis* (E. Schraml), Sisoridae: *Hara jerdoni* (E. Schraml), Amblycipitidae: *Liobagrus reinii* (E. Schraml), Bagridae: *Tachysurus fulvidraco* (F. Schäfer), African Bagridae: *Bagrus* sp. (F.D.B. Schedel), Horabagridae: *Horabagrus brachysoma* (J. Geck), Clariidae: *Clarias gariepinus* (F.D.B. Schedel), Heteropneustidae: *Heteropneustes fossilis* (preserved specimen, F.D.B. Schedel), Auchenipteridae: *Tetranematichthys wallacei* (E. Schraml), Doradidae: *Platydoras armatulus* (E. Schraml), Aspredinidae: *Bunocephalus coracoideus* (E. Schraml), Plotosidae: *Plotusus lineatus* (E. Schraml), Chacidae: *Chaca bankanensis* (preserved specimen, F.D.B. Schedel), Cetopsidae: *Cetopsis coecutiens* (preserved specimen, F.D.B. Schedel), Diplomystidae: *Diplomystes* sp. (preserved specimen, F.D.B. Schedel) Loricariidae: Hypostominae: *Hypostomus* cf. *plecostomus* (E. Schraml) Loricariidae: Hypoptopomatinae: *Otocinclus* sp. (J. Geck), Callichthyidae: *Corydoras rabauti* (E. Schraml)

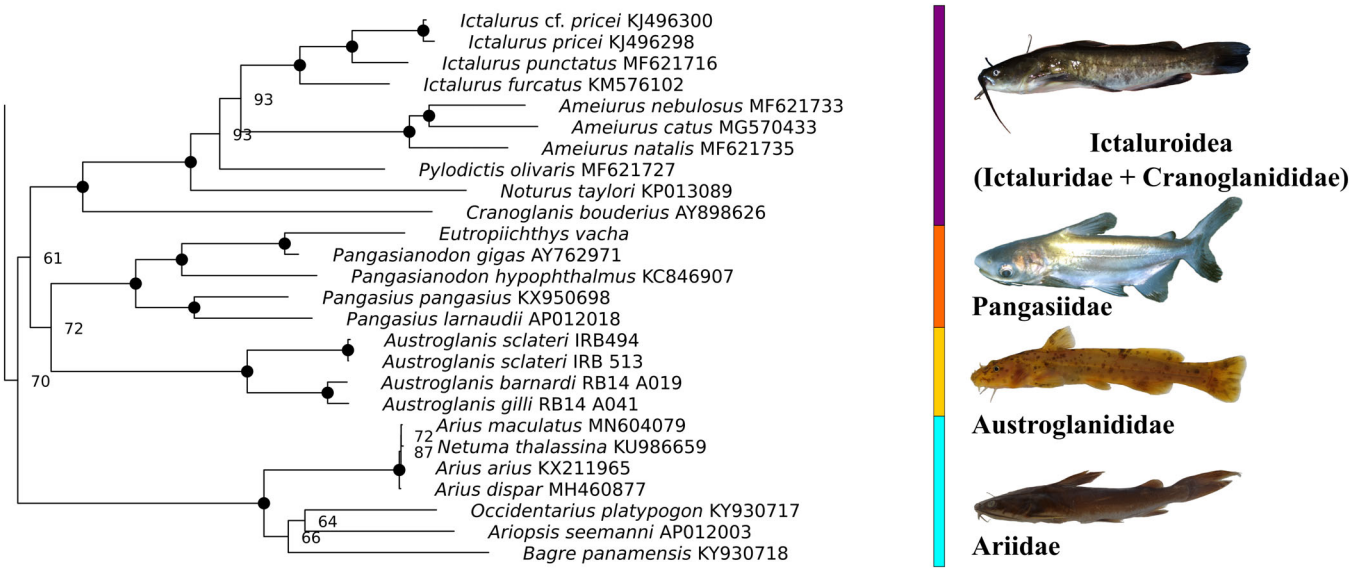

FIGURE 3 Detailed relationships of the family Austroglanididae and related families (pruned from the ML phylogeny depicted in Figure 1). Bootstrap proportion based on 1000 BS replicates are indicated at nodes either by numbers or by black dots (BS = 100)

3.6 | Phylogenetic relationships of Austroglanididae

The ML analysis based on the mitochondrial all-catfish dataset (Figure 3), as well as the one based on the nuclear dataset (Figure 4), recovered *Austroglanis sclateri* (Boulenger, 1901) as sister to the remaining species of Austroglanididae. *Austroglanis gilli* and *Austroglanis barnardi* (Skelton, 1981) were consistently recovered as sister taxa in all analyses with high support (BS: >98, see also Supporting Information Figures S1 and S2). The mitochondrial data supported a close relationship of Austroglanididae with Pangasiidae (Asia) and Ictaluroidea (North America & Asia). Together the three families form a weakly supported clade (BS: 61) which was recovered to be the sister group to Ariidae (BS: 70). The BI analysis (Supporting Information Figure S3) recovered Austroglanididae instead as sister group to Ariidae although with weak support (BPP: 0.62), and the Pangasiidae as sister group of this clade, again with low support (BPP: 0.7). Together with the Ictaluroidea these three families formed a well-supported clade (BPP: 1). These results contrast with our ML analysis of the nuclear dataset which recovered a close relationship between Austroglanididae and the southeast Asian family Ritidae, albeit with very weak support (BS: 32).

4 | DISCUSSION

4.1 | Mochokidae: Phylogenetic placement, intrageneric relationships and taxonomic implications

This study tentatively identifies the family Mochokidae as the sister to the clade containing the Claroteidae, Auchenoglanididae, (African) Schilbeidae and Malapteruridae. The Amphiliidae were recovered as originating at the earliest split within the 'Big Africa clade' (Figure 1). These placements partially conflict with the results of Kappas *et al.* (2016), which is the only other large-scale phylogenetic study of Siluriformes based on mitochondrial genomes. Kappas *et al.* (2016) recovered a sister relationship of Mochokidae to a clade encompassing the Schilbeidae and Auchenoglanididae (the later referred in the corresponding study as Claroteidae; BS: 60), while the Malapteruridae formed a sister group to the Amphiliidae (BS: 56). As in the present study, interfamilial relationships within the 'Big Africa clade' were rather weakly supported, indicating that the phylogenetic information carried by the mitochondrial genome might not suffice to resolve those relationships. However, the topological differences could also originate from the low overall taxon sampling for the 'Big Africa clade' in the study by Kappas *et al.* (2016), which lacked members of Claroteidae entirely. Likewise, the phylogeny obtained by Sullivan *et al.* (2006), based on two nuclear genes and including representatives of all catfish families currently associated with the 'Big Africa clade', differs in some aspects from our mitogenomic phylogeny. For example, Sullivan *et al.* (2006) recovered Mochokidae as sister group to a clade encompassing Malapteruridae and Amphiliidae (BS: 99), with the remaining members of the 'Big Africa clade' forming the sister to that clade (BS: 99). However, interfamily relationships were rather poorly supported in this study too. Though other published multilocus studies have reconstructed relationships using many more genes (*i.e.*, Arcila *et al.*, 2017; Betancur-R. *et al.*, 2017; Rivera-Rivera & Montoya-Burgos, 2018) and could potentially allow for further insights into interfamilial relationships, all these efforts included very few representatives of the 'Big Africa clade'. Thus, DNA-based results to date do not unambiguously favour any of the alternative hypotheses about the exact placement of Mochokidae within the order.

Neither has morphological evidence proved sufficient in resolving this conundrum. Several authors (*i.e.*, de Pinna, 1993, 1998;

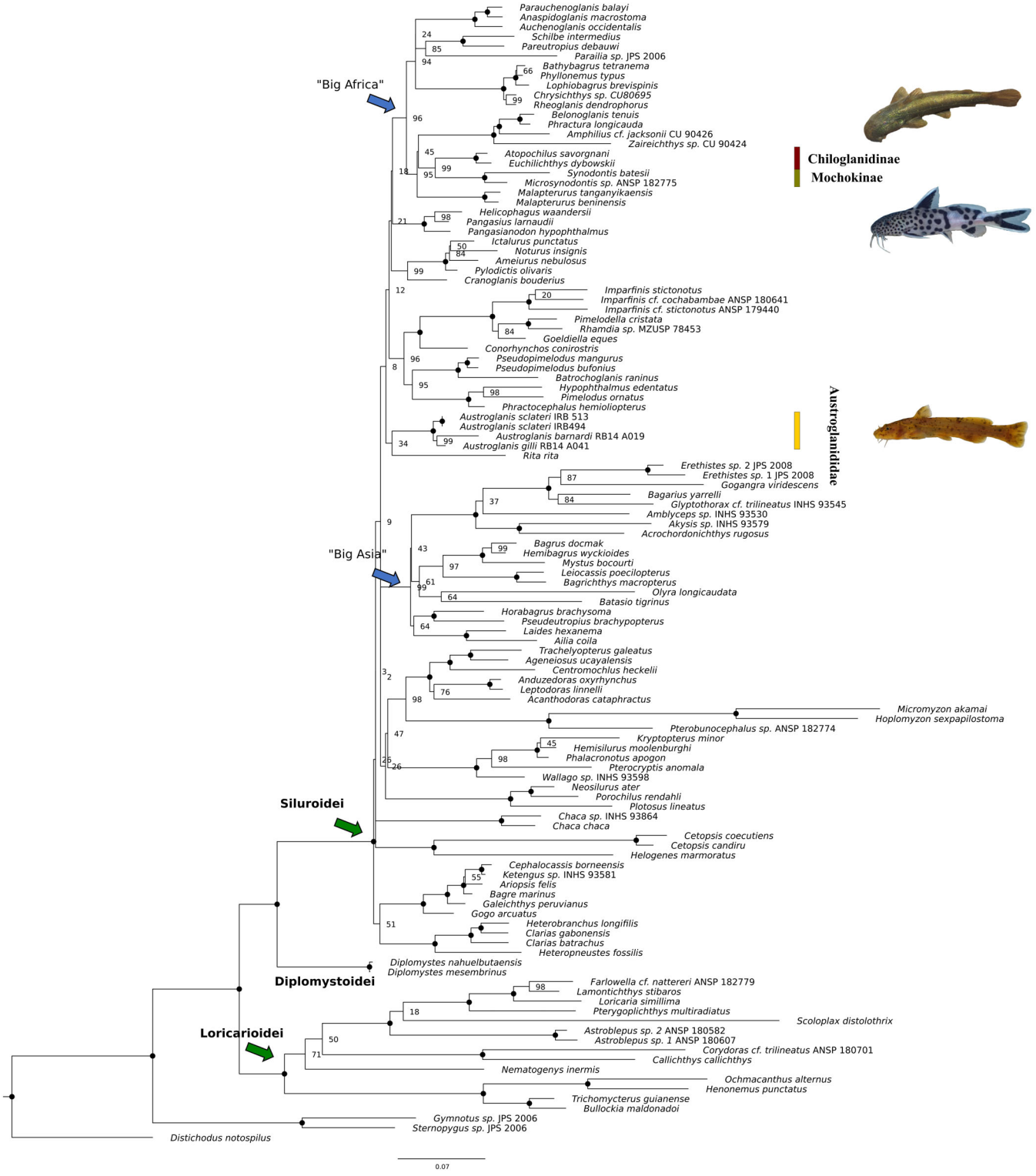

FIGURE 4 ML phylogeny (RAxML) of the order Siluriformes (focal families are indicated by pictures of representative species) based on the 'nuclear dataset', including the sequence data of rag1 and rag2 provided by the study of Sullivan *et al*. (2006) and the newly created sequences of four *Austroglanis* specimens (117 individuals, 1983 bp). Bootstrap proportions based on 1000 BS replicates are indicated at nodes either by numbers or by black dots (BS = 100)

Diogo, 2005; Vigliotta, 2008) have suggested a sister relationship between Mochokidae and Doradoidea based on morphological similarities of the compound centrum, elements at the junction between the dorsal fin and the neurocranium and the nuchal shield. However, Vigliotta (2008) argued that these hypothesized synapomorphies grouping Doradoidea and Mochokidae might represent striking examples of

homoplasies. Alternatively, Vigliotta (2008) tentatively suggested some synapomorphies for a clade encompassing Malapteruridae, Amphiliidae and Mochokidae, namely the absence of the ascending Meckel's cartilage (although present in the amphiliid subfamily Leptoglanidinae) and a characteristically shaped autopalatine. No study has yet rigorously tested these alternative hypotheses using morphological data alone or in a total evidence approach.

Given the conflicting reconstructions yielded by molecular and morphological data alike, it is premature at this point to draw any further conclusions about the interfamilial relationships within the 'Big Africa clade'. The best path forward will involve analysis of a more comprehensive dataset with deeper sampling of the nuclear genome and even broader taxon sampling. For example, it will be critical to include the Mesoamerican *Lacantunia*, which currently is assumed to be the only non-African representative in the 'Big Africa clade' (Lundberg *et al*., 2007). Likewise, revisiting the morphological data in light of and in combination with the molecular data has the potential to enhance tests of alternative hypotheses, reveal additional synapomorphies and test hypotheses about character evolution in this diverse clade of fishes.

Despite the need to investigate deep phylogenetic relationships with more encompassing nuclear and morphological data, analysis of the mitochondrial dataset still yielded substantial insight about intrafamilial relationships. Our phylogenetic reconstruction based on a large fraction of the mitochondrial genome included for the first time all mochokid genera, and tentatively supports the postulated monophyly of Mochokinae and Chiloglanidinae *sensu* Seegers (2008). Within subfamilies, intrageneric relationships were all strongly supported, except for the phylogenetic placement of *Acanthocleithron* at the earliest split within Mochokinae. Overall, our intergeneric topology is largely congruent with the results of Day *et al*. (2013), which specifically focused on the genus *Synodontis* and did not include *Acanthocleithron* and *Atopodontus*. The limited taxon sampling outside of *Synodontis*, might explain why Day *et al*. (2013) recovered *Microsynodontis* rather than *Mochokus* as the sister group of a clade encompassing *Mochokiella* and *Synodontis*.

4.2 | The genus *Acanthocleithron*

The phylogenetic placement of *Acanthocleithron* within Mochokidae remains elusive due to the weak support for its origination at the earliest split within Mochokinae (BS:64). The difficulty of this reconstruction might result from ancient incomplete lineage sorting during rapid early cladogenesis at the base of the Mochokidae. As such, it is possible that the *Acanthocleithron* lineage is even older, potentially originating at the earliest split within the entire family. Such a scenario would render the Mochokinae as currently defined (*sensu* Seegers, 2008) paraphyletic and further suggest a Congo basin origin for the family Mochokidae. However, this hypothesis has not yet been critically tested.

Specimens of *Acanthocleithron* are rare in ichthyological collections and apart from the few specimens known from the Ituri/Aruwimi drainage, a tributary of the Congo basin from which *A. chapini* was originally described (Seegers, 2008), specimens of *A. chapini* have been collected at Bamu Island (Pool Malebo) and at Yangambi (Poll, 1959; Poll & Gosse, 1963). Although *Acanthocleithron* is currently monotypic, Seegers (2008) speculated that specimens collected outside of the Ituri/Arwuimi drainage might not be conspecific with *A. chapini* but rather represent undescribed species of *Acanthocleithron*. He based that hypothesis on differences in coloration and morphological features, particularly differences in the serration of the pelvic fin spine. For a better understanding of the evolutionary history of this little-studied genus and to help stabilize the reconstruction of relationships within Mochokidae, it might be essential to include these enigmatic *Acanthocleithron* populations in upcoming molecular analysis.

4.3 | The genus *Chiloglanis*

The present study recovered a molecular hypothesis of intergeneric mochokid relationships that partially conflicts with those based on morphological data (Vigliotta, 2008). While both datasets support the monophyly of Chiloglanidinae and the chiloglanid tribe Atopochilini Vigliotta, 2008 (including *Atopodontus*, *Atopochilus* and *Euchilichthys*), the morphological analysis of Vigliotta (2008) does not support the monophyly of the subfamily Mochokinae (*sensu* Seegers, 2008), and the studies differ in other inferred relationships among mochokin genera.

Vigliotta (2008) also recovered the genus *Chiloglanis* as paraphyletic, with *C. macropterus* and *Chiloglanis* sp. 'Burundi' (subsequently described as *Chiloglanis kazumbei* Friel & Vigliotta, 2011) not recovered within his *Chiloglanis sensu stricto* clade. Rather, he inferred the lineages leading to those species as originating from splits closer to the base of Chiloglanidinae. That finding was perhaps not surprising, given that Balon and Stewart (1983) had noted that *C. macropterus* in many ways is an unusual *Chiloglanis* and rather might be a 'primitive sister species to all *Chiloglanis*'. A robust test of the monophyly of the genus *Chiloglanis* would therefore depend on inclusion of *C. kazumbei* and *C. macropterus*. We included four specimens of *C. macropterus* for the first time in a molecular analysis and recovered a deeply nested placement of this taxon within the genus (see Figure 2). Unfortunately, we were unable to obtain samples from *C. kazumbei*.

The four *C. macropterus* specimens were collected at two different locations, one of which expands the known distribution of this species. The first location was situated on the Luongo River (a right-hand tributary of the Luapula River), most likely at the type locality of *C. macropterus* which Poll and Stewart (1975) indicated to be situated on the Luongo River at the ferry crossing 53 km south of Kawambwa (Poll & Stewart, 1975) and which corresponds to Locality 2 in the study of Balon and Stewart (1983). The two other specimens were surprisingly caught above Kundabikwa Falls on the Kalungwishi River (a tributary of Lake Mweru), which appears to be a natural barrier for upstream migration of fish (Schedel, 2020). Previously, *C. macropterus* was only recorded from the Luongo River, although Balon and

Stewart (1983) speculated that a record by Malaisse (1968) of an undescribed *Chiloglanis* from the Luanza River (a left-hand tributary of the Luapula River) might be conspecific with *C. macropterus*. Our records expand the known distribution area of this species to the Kalungwishi River. This adds further support to the hypothesis of Balon and Stewart (1983) that the Luongo River and Kalungwishi had been once connected. Interestingly, it seems that genetic divergence between the populations from the Luongo and Kalungwishi River is quite low if not absent (see Figure 2). These findings suggest that the breakup of the Luongo–Kalungwishi connection took place rather recently, or alternatively that movement between these systems still occurs. For example, the extensive dambo wetland system situated between the two river systems (~25 km east of Kawamba) might permit occasional exchanges and gene flow.

Our phylogenetic data not only strongly supported the monophyly of *Chiloglanis* (BS: 100) but also revealed for the first time the presence of at least two major lineages (BS:100) within the genus (see Figure 2). One clade includes taxa from the Zambezi drainage (including Lake Malawi), Nile drainage, Niger drainage, and the Luapula and Chambeshi rivers (both part of the upper Congo drainage). The second clade includes only taxa from the Congo basin and its subdrainage systems (including Lake Tanganyika and the Luapula drainage systems) and it is hence referred to as the 'Congo-*Chiloglanis*' clade. Strikingly, *C. macropterus* was nested within the 'Congo-*Chiloglanis*' clade with *Chiloglanis productus* Ng & Bailey, 2006 as its sister group. This contrasts with Balon and Stewart (1983) suggestion that *C. macropterus* could be a primitive member of the genus *Chiloglanis*. Our findings also differ from those of Vigliotta (2008), who recovered the lineage leading to *C. macropterus* as originating at the earliest split within Chiloglanidinae. Morphological characters distinguishing *C. macropterus* from other *Chiloglanis* include comparatively large fins with elongated pectoral and dorsal fin spines, a roundish caudal fin and elongated mandibular barbels (see Poll & Stewart, 1975). These morphological features could be easily considered as secondarily derived from a more bottom-dwelling *Chiloglanis* mode of life because our own underwater observations in the Luongo River (F.D.B. Schedel) revealed that *C. macropterus* is epibenthic, that is, it tends to hover several centimetres above the substrate. This contrasts with other *Chiloglanis*, which are typically benthic.

The phylogeographic signal recovered from the major *Chiloglanis* subclades should be interpreted with great caution for the moment because our all-catfish dataset included only a small fraction of the impressive *Chiloglanis* diversity and highly likely missed potential additional lineages from ichthyological provinces not sampled in this study. Indeed, the first phylogenetic analysis based on the mochokid *COX1* dataset (Supporting Information Figure S4) suggests the presence of additional sublineages and implies that the 'Congo-*Chiloglanis*' clade does not exclusively includes taxa from the Congo basin but likely also species from Western Africa and the Nile drainage. Other recent studies have suggested a past linkage and exchange between the Congo and Nile drainages as well, such as the presence of syntopically collected *Garra* specimens in the Main Nile suggest (Moritz *et al*., 2019), and the sister relationship recovered between the cichlid species *Pseudocrenilabrus nicholsi* (Upper Congo drainage) and *Pseudocrenilabrus multicolor* (Lower Nile drainage), with an estimated divergence time of approximately 2.2 million years (Schedel *et al*., 2019).

Surprisingly, two specimens identified as *Chiloglanis niloticus* (GenBank accession number: HF565846; Day *et al*., 2013) from the Main Nile did not cluster together in the analysis of the COX1 dataset. Instead, the *C. niloticus* specimen clustered within the 'Congo-*Chiloglanis*' clade while our newly sequenced specimen of *C.* cf. *niloticus* clustered with a *Chiloglanis* from Niger drainage. The sequence similarity of these two individuals on COX1 therefore was only 86.7%. This result strongly suggests the presence of at least two *Chiloglanis* species in the Main Nile (see Neumann *et al*. (2016) for an overview of what is known about *Chiloglanis* populations in that system). Given that both specimens were collected hundreds of river kilometres upstream from the type locality, it is very possible that neither specimen is not conspecific with *C. niloticus*. This conjecture would have to be investigated further using both morphological and molecular data. Overall, these results underline our limited knowledge about the alpha-taxonomy and distribution of the genus *Chiloglanis*, starting with its alpha-taxonomy as well as its biogeographical history.

4.4 | The genus *Microsynodontis*

Contrasting with the morphological study of Vigliotta (2008) and the molecular study of Day *et al*. (2013), our analysis did not recover the monophyly of *Synodontis*, but rather obtained *Synodontis punu* as sister to *Microsynodontis*. Vreven and Milondo (2009) originally assigned this species to *Synodontis* based on a combination of characters diagnostic for the genus, including a forked caudal fin (see Vreven & Milondo, 2009). *Microsynodontis* is the only mochokid genus that includes species with a rounded caudal fin (Howes, 1980), which *Synodontid punu* clearly lacks. However, the latest revision of *Microsynodontis* and the description of several new species by Ng (2004) clarified that caudal fin shape ranges from emarginate to truncate among *Microsynodontis* species. In addition, Seegers (2008) reported that mochokid catfish imported by the ornamental fish trade to Germany had an overall '*Microsynodontis*-appearance', but with deeply forked caudal fins (potentially associated with sexual dimorphism). This apparent variability in caudal fin shape among *Microsynodontis* species in combination with phylogenetic results from our study suggests that *S. punu* should be transferred to *Microsynodontis*. Another small-sized species, *Synodontis acanthoperca* Friel & Vigliotta, 2006, was described from the Ogowe drainage in Gabon and found to be morphologically quite similar to *S. punu* (Vreven & Milondo, 2009). It might likewise represent a member of *Microsynodontis*, but so far no molecular data are available for that species.

4.5 | Phylogenetic placement of Austroglanididae

This study provides the first molecular reconstruction of the phylogenetic affinities of the African catfish family Austroglanididae within

Siluriformes. While our analysis resolved the intrageneric relationships of the three currently known *Austroglanis* species quite well, the phylogenetic placement of the family received only weak support. Our data suggest a closer relationship of Austroglanididae to Pangasiidae, Ictaluroidea and/or Ariidae. These four families were consistently recovered in a monophyletic group in all phylogenetic analyses based on mitochondrial data.

Substantial efforts over the past five decades have attempted to infer the relationship of this family using morphological characters (de Pinna, 1993; Diogo, 2005; Diogo & Bills, 2006; Mo, 1991; Skelton *et al*., 1984). Some of those studies have suggested Austroglanididae to be related closely to one or more of three clades with which it clusters in the molecular analysis, but close ties to Anchariidae, Bagridae, Claroteidae, Schilbeidae and Pimelodidae have also been obtained. Diogo and Bills (2006) provided a detailed discussion on potential autapomorphies of Austroglanididae as well as of morphological similarities between Austroglanididae and Ictaluroidea. However, to our knowledge no study has identified clear synapomorphies supporting a clade containing Austroglanididae and one or more of the families Ictaluroidea, Pangasiidae and Ariidae. Of the various catfish families proposed to be related to Austraoglanididae, our dataset was missing only Anchariidae, a family endemic to Madagascar and which has been generally considered to be closely related to Ariidae, with which it forms the superfamily Arioidea (Sullivan *et al*., 2006).

Despite all this uncertainty, our findings are congruent with the general morphological consensus that the closest relatives of Austroglanididae are not part of the 'Big Africa clade' of Sullivan *et al*. (2006). That consensus raises intriguing questions about the evolutionary and biogeographic origin of Austroglanididae, which is endemic to Southern Africa. A close relationship with Pangasiidae and Ictaluroidea would suggest an Asian origin for the family and imply that their ancestors colonized Africa without leaving any modern descendants in Asia or in remaining parts of Africa. Alternatively, the family might have derived from marine/brackish ancestors that ascended and colonized exclusively the Southern African river systems. This hypothesis is appealing given the fact that members of Ariidae (one of the clades to which Austroglanidiade appears to be closely related) are primarily marine but known to have colonized freshwater systems (Nelson *et al*., 2016). Undoubtedly, an in-depth phylogenomic approach will be needed to test the validity of a clade containing Austroglanididae, Ictaluroidea, Pangasiidae and potentially Arioidea, and to clarify the biogeographical origin of Austroglanididae.

5 | CONCLUSIONS

Our study provides 52 new partial mitochondrial genomes (*i.e*., covering all protein-coding genes) of representative taxa of all currently valid mochokid genera, all described *Austroglanis* species, and selected species of other catfish families belonging to the 'Big Africa clade' *sensu* Sullivan *et al*. (2006). Hence, this study substantially increases the mitochondrial genomic resources for African catfish available on public databases such as GenBank, which were limited previously to seven species. Our increased taxon sampling allowed reconstruction of the first robust phylogeny that resolves all intergeneric relationships within Mochokidae. It is also the first to infer the relationships of the family Austroglanididae, which is apparently not closely related to the 'Big Africa clade' but rather linked to the Asian Pangasiidae, the Asian/North American Ictaluroidea and perhaps even the marine Ariidae. As in previous studies based primarily on mitochondrial data, our analyses only weakly supported a reconstruction of the earliest divergences within Siluriformes. We encourage large-scale genomic studies and a renewed examination of the phylogenetic signal in their morphological characters in the future. Such studies would increase the robustness of the phylogenetic framework needed to resolve the spatiotemporal diversification of catfishes throughout Africa and beyond, further test the placement of the enigmatic family Austroglanididae, and help reconstruct the history of morphological and ecological evolution in this rich and diverse clade of fishes.

DISCLAIMER

Data on genetic material contained in this paper are exclusively published for noncommercial use only. Access to specimens and/or tissue samples complies with relevant national laws implementing the Convention on Biological Diversity and Nagoya Protocol agreement where relevant, that is, where national access laws were established and applicable at the time of collecting. Nonmonetary benefits have been shared through taxonomic training and capacity building, *e.g*., during joint field work or joint publication. Use by third parties for purposes other than noncommercial scientific research may infringe the conditions under which the genetic resources were originally accessed and should not be undertaken without obtaining consent from the corresponding author of the paper and/or obtaining permission from the original provider of the genetic material.

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AUTHOR CONTRIBUTIONS

F.D.B.S. conceived the study and wrote the first draft of the manuscript. F.D.B.S., A.C., M.O.P., B.L.S., D.N., E.J.W.M.N.V. and U.K.S. were involved with the collection of the specimens and tissue samples studied herein. F.D.B.S., A.C. and N.U.W. performed the molecular work associated with the sequencing of the partial mitochondrial genomes. F.D.B.S. conducted all molecular analyses and prepared all figures. B.L.S, D.N. and U.K.S. contributed to the improvement of all versions of the manuscript. All authors read and approved the final manuscript.

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REFERENCES

Abwe, E., Schedel, F. D. B., Snoeks, J., Manda, A. C., Schliewen, U. K. & Vreven, E. J. W. M. N. (in prep.). Hidden species richness in the Amphilius uranoscopus group (Siluriformes: Amphiliidae) of the rivers of the Kundelungu Plateau and its surroundings (Upper Lualaba, Congo basin).

Arcila, D., Ortí, G., Vari, R., Armbruster, J. W., Stiassny, M. L. J., Ko, K. D., … Betancur-R, R. (2017). Genome-wide interrogation advances resolution of recalcitrant groups in the tree of life. *Nature Ecology & Evolution*, *1*, 10.

Armbruster, J. W. (2011). Global catfish biodiversity. *American Fisheries Society Symposium*, *77*, 15–37.

Balon, E. K., & Stewart, D. J. (1983). Fish assemblages in a river with unusual gradient (Luongo, Africa - Zaire system), reflections on river zonation, and description of another new species. *Environmental Biology of Fishes*, *9*, 225–252.

Barnard, K. H. (1943). Revision of the indigenous freshwater fishes of the S.W. Cape region. *Annals of the South African Museum*, *36*, 101–262.

Betancur-R, R. (2009). Molecular phylogenetics and evolutionary history of ariid catfishes revisited: A comprehensive sampling. *BMC Evolutionary Biology*, *9*, 1–18.

Betancur-R, R., Arcila, D., Vari, R. P., Hughes, L. C., Oliveira, C., Sabaj, M. H., & Orti, G. (2019). Phylogenomic incongruence, hypothesis testing, and taxonomic sampling: The monophyly of characiform fishes. *Evolution*, *73*, 329–345.

Betancur-R, R., Wiley, E. O., Arratia, G., Acero, A., Bailly, N., Miya, M., … Ortí, G. (2017). Phylogenetic classification of bony fishes. *BMC Evolutionary Biology*, *17*, 40.

Bills, I. R., & Impson, N. D. (2013). Conservation biology of endangered freshwater fishes–linking conservation of endangered freshwater fishes with river conservation, focusing on the Cederberg. *Pretoria: Water Research Commission, No. KV 305/12*, 1–221.

Bleeker, P. (1858). De visschen van den Indischen Archipel. Beschreven en toegelicht. Siluri. *Acta Societatia Scientiarum Indo-Neèrlandicae*, *4*, 1–370.

Boulenger, G. A. (1900). On some little-known African silurid fishes of the subfamily Doradinae. *Annals and Magazine of Natural History (Series 7)*, *6*, 520–529.

Boulenger, G. A. (1901). Description of a new silurid fish of the genus Gephyroglanis, from South Africa. *Annals of the South African Museum*, *2*, 227–228.

Boulenger, G. A. (1903). On the fishes collected by Mr. G. L. bates in southern Cameroon. *Proceedings of the Zoological Society of London*, *1*, 21–29.

Chakona, A., Kadye, W. T., Bere, T., Mazungula, D. N., & Vreven, E. (2018). Evidence of hidden diversity and taxonomic conflicts in five stream fishes from the eastern Zimbabwe highlands freshwater ecoregion. *ZooKeys*, *768*, 69–95.

Chevey, P. (1931). Sur un nouveau silure géant du Bassin du Mékong Pangasianodon gigas nov. g., nov. sp. *Bulletin de la Societe Zoologique de France*, *55*, 536–542 (Date from back of volume (p. 577) as above; sometimes seen as 1930).

Covain, R., & Fisch-Muller, S. (2007). The genera of the Neotropical armored catfish subfamily Loricariinae (Siluriformes: Loricariidae): A practical key and synopsis. *Zootaxa*, *1462*, 1–40.

Cuvier, G. (1816). *Le Règne Animal distribué d'après son organisation pour servir de base à l'histoire naturelle des animaux et d'introduction à l'anatomie comparée. Les reptiles, les poissons, les mollusques et les annélides.* (Vol. 2, pp. 1–532). Paris, France: Deterville.

Cuvier, G., & Valenciennes, M. A. (1840). *Histoire Naturelle des Poissons. Tome quinzième. Suite du livre dix-septième. Siluroïdes. (Valenciennes authored volume. i-xxiv + 1–397 in Strasbourg edition.).* Paris, France: Pitois.

Day, J. J., Bills, R., & Friel, J. P. (2009). Lacustrine radiations in African *Synodontis* catfish. *Journal of Evolutionary Biology*, *22*, 805–817.

Day, J. J., Peart, C. R., Brown, K. J., Friel, J. P., Bills, R., & Moritz, T. (2013). Continental diversification of an African catfish radiation (Mochokidae: Synodontis). *Systematic Biology*, *62*, 351–365.

de Pinna, M. C. C. (1993). *Higher-level phylogeny of Siluriformes, with a new classification of the order (Teleostei, Ostariophysi)*. New York, NY: City University of New York.

de Pinna, M. C. C. (1998). Phylogenetic relationships of neotropical Siluriformes (Teleostei: Ostariophysi): Historical overview and synthesis of hypotheses. In L. R. Malabarba, R. E. Reis, R. P. Vari, Z. M. S. de Lucena, & C. A. S. de Lucena (Eds.), *Phylogeny and classification of Neotropical fishes* (pp. 279–330). Porto Alegre: Edipucrs.

Decru, E., Moelants, T., De Gelas, K., Vreven, E., Verheyen, E., & Snoeks, J. (2016). Taxonomic challenges in freshwater fishes: A mismatch between morphology and DNA barcoding in fish of the north-eastern part of The Congo basin. *Molecular Ecology Resources*, *16*, 342–352.

Diogo, R. (2004). Phylogeny, origin and biogeopraphy of catfishes: Support for a Pangean origin of 'modern teleosts' and reexamination of some Mesozoic Pangean connections between the Gondwanan and Laurasian supercontinents. *Animal Biology (former Netherlands Journal of Zoology)*, *54*, 331–351.

Diogo, R. (2005). *Adaptations, Homoplasies, constraints, and evolutionary trends: Catfish morphology, phylogeny and evolution, a case study on theoretical phylogeny and macroevolution*. Enfield, NH: Science Publishers Inc.

Diogo, R., & Bills, R. (2006). Osteology and myology of the cephalic region and pectoral girdle of the south African catfish *Austroglanis gilli*, with comments on the autapomorphies and phylogenetic relationships of the Austroglanididae (Teleostei: Siluriformes). *Animal Biology*, *56*, 39–62.

Diogo, R., & Peng, Z. (2010). State of the art of siluriform higher-level phylogeny. In T. Grande, F. J. Poyato-Ariza, & R. Diogo (Eds.), *Gonorynchiformes and ostriophysan relationships: A comprehensive review* (pp. 465–515). Enfield, NH: Science Publishers.

Ferraris, C. J., Jr. (2007). Checklist of catfishes, recent and fossil (Osteichthyes: Siluriformes), and catalogue of siluriform primary types. *Zootaxa*, *1418*, 1–628.

Fink, S. V., & Fink, W. L. (1981). Interrelationships of the ostariophysan fishes (Teleostei). *Zoological Journal of the Linnean Society*, *72*, 297–353.

Fink, S. V., & Fink, W. L. (1996). Interrelationships of ostariophysan fishes (Teleostei). In M. L. J. Stiassny, L. R. Parenti, & G. D. Johnson (Eds.), *Interrelationships of fishes* (pp. 209–249). San Diego, London, Boston, MA: Academic Press.

Fricke, R., Eschmeyer, W. N. & van der Laan, R. E. (2021). Eschmeyer's catalog of fishes: Genera, species, references. http://researcharchive.calacademy.org/research/ichthyology/catalog/fishcatmain.asp

Friel, J. P., & Vigliotta, T. R. (2006). Synodontis acanthoperca, a new species from the Ogôoué River system, Gabon with comments on spiny ornamentation and sexual dimorphism in mochokid catfishes (Siluriformes: Mochokidae). *Zootaxa*, *1125*, 45.

Friel, J. P., & Vigliotta, T. R. (2008). *Atopodontus adriaensi*, a new genus and species of African suckermouth catfish from the Ogooué and Nyanga River systems of Gabon (Siluriformes: Mochokidae). *Proceedings of the Academy of Natural Sciences of Philadelphia*, *157*, 13–23.

Friel, J. P., & Vigliotta, T. R. (2011). Three new species of African suckermouth catfishes, genus *Chiloglanis* (Siluriformes: Mochokidae), from the lower Malagarasi and Luiche rivers of western Tanzania. *Zootaxa*, *3063*, 1–21.

Hamilton, F. (1822). *An account of the fishes found in the river Ganges and its branches*. Edinburgh: Archibald Constable and Company.

Hardman, M. (2005). The phylogenetic relationships among non-diplomystid catfishes as inferred from mitochondrial cytochrome b sequences; the search for the ictalurid sister taxon (Otophysi: Siluriformes). *Molecular Phylogenetics and Evolution*, *37*, 700–720.

Howes, G. J. (1980). A new catfish from Sierra Leone. *Bulletin of the British Museum (Natural History) Zoology*, *38*, 165–170.

Huelsenbeck, J. P., & Ronquist, F. (2001). MRBAYES: Bayesian inference ofphylogenetic trees. *Bioinformatics*, *17*, 754–755.

Hughes, L. C., Ortí, G., Huang, Y., Sun, Y., Baldwin, C. C., Thompson, A. W., … Shi, Q. (2018). Comprehensive phylogeny of ray-finned fishes (Actinopterygii) based on transcriptomic and genomic data. *Proceedings of the National Academy of Sciences of the United States of America (PNAS)*, *115*, 6249–6254.

Jayaram, K. C. (1955). Taxonomic status of the Chinese catfish family Cranoglanididae Myers, 1931. *Proceedings of the Indian National Science Academy - Part B: Biological Sciences*, *21*, 256–263.

Jordan, D. S. (1923). *A classification of fishes including families and genera as far as known*. Stanford Universtiy: Stanford University Publications, University Series, Biological Sciences.

Kappas, I., Vittas, S., Pantzartzi, C. N., Drosopoulou, E., & Scouras, Z. G. (2016). A time-calibrated Mitogenome phylogeny of catfish (Teleostei: Siluriformes). *PLoS One*, *11*, e0166988.

Kearse, M., Moir, R., Wilson, A., Stones-Havas, S., Cheung, M., Sturrock, S., … Drummond, A. (2012). Geneious basic: An integrated and extendable desktop software platform for the organization and analysis of sequence data. *Bioinformatics*, *28*, 1647–1649.

Koblmüller, S., Sturmbauer, C., Verheyen, E., Meyer, A., & Salzburger, W. (2006). Mitochondrial phylogeny and phylogeography of east African squeaker catfishes (Siluriformes: Synodontis). *BMC Evolutionary Biology*, *6*, 49.

Lundberg, J. G., Sullivan, J. P., Rodiles-Hernandez, R., & Hendrickson, D. A. (2007). Discovery of African roots for the Mesoamerican Chiapas catfish, *Lacantunia enigmatica*, requires an ancient intercontinental passage. *Proceedings of the Academy of Natural Sciences of Philadelphia*, *156*, 39–53.

Ma, X., Kang, J., Chen, W., Zhou, C., & He, S. (2015). Biogeographic history and high-elevation adaptations inferred from the mitochondrial genome of Glyptosternoid fishes (Sisoridae, Siluriformes) from the southeastern Tibetan plateau. *BMC Evolutionary Biology*, *15*, 233.

Malaisse, F. (1968). *Etude ecologique de la riviere Luanza et de son bassin* (p. 473). Lubumbashi: University of Congo.

Miller, M. A., Pfeiffer, W., & Schwartz, T. (2010). Creating the CIPRES science gateway for inference of large phylogenetic trees. In *Proceedings of the gateway computing environments workshop (GCE). 14 Nov. 2010* (pp. 1–8). New Orleans, LA: Institute of Electrical and Electronics Engineers (IEEE).

Mo, T. (1991). *Anatomy, relationships and systematics of the Bagridae (Teleostei: Siluroidei) with a hypothesis of siluroid phylogeny*. Köenigstein: Koeltz Scientific Books.

Moreira, D. A., Buckup, P. A., Furtado, C., Val, A. L., Schama, R., & Parente, T. E. (2017). Reducing the information gap on Loricarioidei (Siluriformes) mitochondrial genomics. *BMC Genomics*, *18*, 345.

Moritz, T., Straube, N., & Neumann, D. (2019). The *Garra* species (Cyprinidae) of the Main Nile basin with description of three new species. *Cybium*, *43*, 311–329.

Morris, J., Ford, A. G. P., Ali, J. R., Peart, C. R., Bills, R., & Day, J. J. (2016). High levels of genetic structure and striking phenotypic variability in a sexually dimorphic suckermouth catfish from the African Highveld. *Biological Journal of the Linnean Society*, *117*, 528–546.

Nakatani, M., Miya, M., Mabuchi, K., Saitoh, K., & Nishida, M. (2011). Evolutionary history of Otophysi (Teleostei), a major clade of the modern freshwater fishes: Pangaean origin and Mesozoic radiation. *BMC Evolutionary Biology*, *11*, 177.

Nelson, J. S., Grande, T. C., & Wilson, M. V. H. (2016). *Fishes of the world*. Hoboken, NJ: John Wiley & Sons.

Neumann, D. (2010). Preservation of freshwater fishes in the field. *Abc Taxa*, *8*, 587–632.

Neumann, D., Obermaier, H., & Moritz, T. (2016). Annotated checklist for fishes of the Main Nile Basin in The Sudan and Egypt based on recent specimen records (2006-2015). *Cybium*, *40*, 287–317.

Ng, H. H. (2004). The *Microsynodontis* (Teleostei: Siluriformes: Mochokidae) of the lower Guinea region, west Central Africa, with description of eight new species. *Zootaxa*, *531*, 1–52.

Nichols, J. T., & Griscom, L. (1917). Fresh-water fishes of The Congo basin obtained by the American museum Congo expedition, 1909-1915. *Bulletin of the American Museum of Natural History*, *37*, 653–756.

Otero, O., & Gayet, M. (2001). Palaeoichthyofaunas from the lower Oligocene and Miocene of the Arabian plate: Palaeoecological and palaeobiogeographical implications. *Palaeogeography, Palaeoclimatology, Palaeoecology*, *165*, 141–169.

Peters, W. C. H. (1868). Ueber eine von dem Baron Carl von der Decken entdeckte neue Gattung von Welsen, *Chiloglanis deckenii*, und einige andere Süsswasserfische aus Ostafrika. *Monatsberichte der Königlich Preussischen Akademie der Wissenschaften zu Berlin*, *1868*, 598–602.

Pinton, A., Agnese, J. F., Paugy, D., & Otero, O. (2013). A large-scale phylogeny of Synodontis (Mochokidae, Siluriformes) reveals the influence of geological events on continental diversity during the Cenozoic. *Molecular Phylogenetics and Evolution*, *66*, 1027–1040.

Poll, M. (1959). III. Recherches sur la faune ichthyologique de la region du Stanley Pool. In *Resultats scientifiques des missions zoologiques au Stanley Pool subsidiées par le CEMUBAC (Université Libre de Bruxelles et le Musée Royal du Congo 1957–1958)* (pp. 75–174). Tervuren: Musée Royal du Congo Belgique.

Poll, M., & Gosse, J.-P. (1963). Contribution a l'etude systematique de la faune ichthyologique du Congo Central. *Annales du Musee Royal de l'Afrique Centrale Serie 8 Sciences Zoologiques*, *116*, 45–110.

Poll, M., & Stewart, D. J. (1975). Un Mochocidae et un Kneriidae nouveaux de la rivière Luongo (Zambia), affluent du bassin du Congo (Pisces). *Revue de Zoologie Africaine*, *89*, 151–158.

Posada, D. (2008). jModelTest: Phylogenetic Model Averaging. *Molecular Phylogenetics and Evolution*, *25*, 1253–1256.

Riehl, R., & Baensch, A. (1990). *Mergus Aquarien Atlas*. Band 3. Melle: Mergus.

Rivera-Rivera, C. J., & Montoya-Burgos, J. I. (2017). Trunk dental tissue evolved independently from underlying dermal bony plates but is associated with surface bones in living odontode-bearing. *Proceedings of the Royal Society B Biological Sciences*, *284*, 9.

Rivera-Rivera, C. J., & Montoya-Burgos, J. I. (2018). Back to the roots: Reducing evolutionary rate heterogeneity among sequences gives support for the early morphological hypothesis of the root of Siluriformes (Teleostei: Ostariophysi). *Molecular Phylogenetics and Evolution*, *127*, 272–279.

Rodiles-Hernandez, R., Hendrickson, D. A., Lundberg, J. G., & Humphries, J. M. (2005). *Lacantunia enigmatica* (Teleostei: Siluriformes) a new and phylogenetically puzzling freshwater fish from Mesoamerica. *Zootaxa*, *1000*, 1–24.

Ronquist, F., Teslenko, M., van der Mark, P., Ayres, D. L., Darling, A., Höhna, S., … Huelsenbeck, J. P. (2012). MrBayes 3.2: Efficient Bayesian phylogenetic inference and model choice across a large model space. *Systematic Biology*, *61*, 539–542.

Sauvage, H. E. (1879). Notice sur la faune ichthyologique de l'Ogôoué. *Bulletin de la Société Philomathique (Series 7)*, *3*, 90–103.

Schedel, F. D. B. (2020). *Phylogeny, divergence time estimates and systematics of African cichlids (Cichlidae: Pseudocrenilabrinae), with a focus on the rheophilic cichlids of east and Central Africa* (p. 255). Munich, Germany: Ludwig-Maximilians-Universität.

Schedel, F. D. B., Musilova, Z., & Schliewen, U. K. (2019). East African cichlid lineages (Teleostei: Cichlidae) might be older than their ancient host lakes: New divergence estimates for the east African cichlid radiation. *BMC Evolutionary Biology*, *19*, 25.

Schmidt, R. C., Bart, H. L., Jr., Nyingi, D. W., & Gichuki, N. N. (2014). Phylogeny of suckermouth catfishes (Mochokidae: Chiloglanis) from Kenya: The utility of growth hormone introns in species level phylogenies. *Molecular Phylogenetics and Evolution*, *79*, 415–421.

Schmidt, R. C., Bart, H. L., Jr., & Pezold, F. (2016). High levels of endemism in suckermouth catfishes (Mochokidae: Chiloglanis) from the upper Guinean forests of West Africa. *Molecular Phylogenetics and Evolution*, *100*, 199–205.

Seegers, L. (2008). *Die Welse Afrikas. Ein Handbuch für Bestimmung und Pflege*. Rodgau: Aqualog Verlag.

Silva, G. S. C., Melo, B. F., Roxo, F. F., Ochoa, L. E., Shibatta, O., Sabaj, M. H., & Oliveira, C. (2021). Phylogenomics of the bumblebee catfishes (Siluriformes: Pseudopimelodidae) using ultraconserved elements. *Journal of Zoological Systematics & Evolutionary Research*, *59*, 1662–1672.

Skelton, P. H. (1981). The description and osteology of a new species of Gephyroglanis (Siluriformes, Bagridae) from the Olifants River, south west cape, South Africa. *Annals of the Cape Provincial Museums (Natural History)*, *13*, 217–250.

Skelton, P. H. (2001). *A complete guide to the freshwater fishes of southern Africa*. Cape Town: Struik Publishers.

Skelton, P. H., Risch, L., & de Vos, L. D. G. (1984). On the generic identity of the Gephyroglanis catfishes from southern Africa (Pisces, Siluroidei, Bagridae). *Revue de Zoologie Africaine*, *98*, 337–372.

Stamatakis, A. (2014). RAxML version 8: A tool for phylogenetic analysis and post-analysis of large phylogenies. *Bioinformatics*, *30*, 1312–1313.

Stange, M., Aguirre-Fernandez, G., Cooke, R. G., Barros, T., Salzburger, W., & Sanchez-Villagra, M. R. (2016). Evolution of opercle bone shape along a macrohabitat gradient: Species identification using mtDNA and geometric morphometric analyses in neotropical sea catfishes (Ariidae). *Ecology and Evolution*, *6*, 5817–5830.

Sullivan, J. P., Lundberg, J. G., & Hardman, M. (2006). A phylogenetic analysis of the major groups of catfishes (Teleostei: Siluriformes) using rag1 and rag2 nuclear gene sequences. *Molecular Phylogenetics and Evolution*, *41*, 636–662.

Vigliotta, T. R. (2008). A phylogenetic study of the African catfish family Mochokidae (Osteichthyes, Ostariophysi, Siluriformes), with a key to genera. *Proceedings of the Academy of Natural Sciences of Philadelphia*, *157*, 73–136.

Vreven, E., & Milondo, L. (2009). Description of *Synodontis punu*, new species (Siluriformes: Mochokidae) from the lower Guinea ichthyofaunal province (Gabon and republic of Congo), Africa. *Ichthyological Exploration of Freshwaters*, *20*, 97–104.

Wang, J., Lu, B., Zan, R., Chai, J., Ma, W., Jin, W., … Chen, Z. (2016). Phylogenetic relationships of five Asian Schilbid genera including Clupisoma (Siluriformes: Schilbeidae). *PLoS One*, *11*, e0145675.

Watson, HB. (2020). Unrecognised endemic biodiversity within east African Chiloglanis catfish populations. (p. 45).

Xia, X. (2018). DAMBE7: New and improved tools for data analysis in molecular biology and evolution. *Molecular Biology and Evolution*, *35*, 1550–1552.

Xia, X., & Lemey, P. (2009). Assessing substitution saturation with DAMBE. In P. Lemey, M. Salemi, & A. Vandamme (Eds.), *The phylogenetic handbook: A practical approach to DNA and protein phylogeny* (pp. 615–630). Cambridge: Cambridge University Press.

Xia, X., Xie, Z., Salemi, M., Chen, L., & Wang, Y. (2003). An index of substitution saturation and its application. *Molecular Phylogenetics and Evolution*, *26*, 1–7.

SUPPORTING INFORMATION

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