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Cichlids of the Banc d'Arguin National Park, Mauritania: insight into the diversity of the genus *Coptodon*

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To determine the species diversity of cichlids in the Banc d'Arguin National Park (PNBA) and their phylogenetic relationships with other species in West Africa, a morphometric and meristic and molecular phylogenetic study was conducted. Both approaches not only confirm the presence of *Sarotherodon melanotheron* in PNBA but also demonstrate the presence of a second species from the genus *Coptodon*. While morphometric characteristics match the description of the Guinean tilapia *Coptodon guineensis*, phylogenetic reconstructions based on three mitochondrial and one nuclear DNA fragment demonstrate that *C. guineensis* is paraphyletic over its range. Because different lineages of *C. guineensis* are allopatric, the distribution of *C. guineensis* should be restricted to Ghana and Côte d'Ivoire. The many other lineages of this species should be considered as *C. sp. aff. guineensis*.

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Key words: Cichlidae; cryptic species; phylogeny; West Africa.

INTRODUCTION

Cichlidae is one of the most speciose families of vertebrates, found in South and Central America, Madagascar, Africa, The Middle East, India and Sri Lanka (Kocher, 2004; Salzburger & Meyer, 2004). As with many other families of fishes, cichlid taxonomy is debated because systematics have been profoundly questioned and secondarily revised

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with information from molecular phylogenetics (Chakrabarty, 2010; Dunz, 2012). Global cichlid diversity consists of major lineages with phylogenetic relationships consistent with Gondwanan break-up (Zardoya *et al.*, 1996; Streebman *et al.*, 1998; Sparks & Smith, 2004; McMahan *et al.*, 2013). All African cichlid diversity belongs to the subfamily Pseudocrenilabrinae Fowler 1934 and includes the haplotilapiine formerly referred to as 'Tilapia' (Schwarzer *et al.*, 2009; Dunz & Schlieven, 2013). The most recent comprehensive molecular phylogenetic review demonstrated that the haplotilapiine lineage is monophyletic and consists of 10 clades, with all East African cichlid tribes being recovered in a single clade (Dunz & Schlieven, 2013). Considering the molecular phylogeny and some diagnostic morphometric characteristics, Dunz & Schlieven (2013) proposed a novel generic and supra-generic classification of haplotilapiine. In this new classification, the tribes Oreochromini and Coptodonini harbour most of the genus and species diversity of West African cichlids. The extent of species diversity, however, remains to be evaluated because many new species are still being described (Dunz & Schlieven, 2010*a, b*; Dunz, 2012). Similar to the East African Great Lakes, Cameroon's crater lakes in West Africa have been a focus of much study because they host species flocks (Trewavas *et al.*, 1972; Stiassny *et al.*, 1992; Schlieven *et al.*, 1994; Schlieven & Klee, 2004; Neumann *et al.*, 2011; Martin, 2012; Lamboj, 2014). In comparison, the African cichlid diversity outside of these regions appears to be extremely poor (Teugels & Thys van den Audenaerde, 2003). Only two species, the blackchin tilapia *Sarotherodon melanotheron* Rüppell 1852 and the Guinean tilapia *Coptodon guineensis* (Günther 1862), have been described so far in near-shore African marine environments, the latter being limited to Senegalese shores according to Stiassny *et al.* (2008). Some authors, however, reported the presence of *C. guineensis* in northern Moroccan localities: the Sebket Imlily, situated at *c.* 200 km from the border with Mauritania (Qinba *et al.*, 2009), and 700 km to the North in rock pools (locally known as gueltas) along the Aabar wadi, tributary of Chbeyka wadi, Tan Tan province (south-western Morocco; Qinba *et al.*, 2012). Similarly, while only one cichlid species is known in the Banc d'Arguin National Park (PNBA) in the north of Mauritania (Sevrin-Reyssac & Richer de Forges, 1985), Imraguen (the name given to local PNBA fishermen) distinguish two cichlids based on body colouration: a black phenotype named Toumvertel kahla and a yellow phenotype named Toumvertel safra. Recent unpublished studies on cichlids targeted by fisheries at the PNBA also reported the presence of different phenotypes among the cichlids landed. All these independent observations call for further investigation to better describe the distribution range of cichlids in the Saharan area. Furthermore, because the taxonomic status of cichlids with widespread species has been controversial, such as for *C. guineensis*, the redbelly tilapia *Coptodon zillii* (Gervais 1848) and the redbreast tilapia *Coptodon rendalli* (Boulenger 1897) (for the list of synonymized species, see Eschmeyer, 2015), it is worthwhile to investigate the genetic status of these species further. In the past, genetic investigations of *S. melanotheron* demonstrated that this nominal species consisted of at least two species (and three sub-species) over its distribution from Congo to Mauritania (Falk *et al.*, 1999, 2003).

In this context, the first objective of this study was to investigate the phenotypic and genetic diversity of cichlids in the PNBA waters, by analysing their morphological and genetic variability, in order to clarify their taxonomic status. Secondly, the diversity of the tribe Coptodonini, and more specifically of some of their widespread species such as *C. guineensis*, *C. rendalli* or *C. zillii*, was evaluated by analysing several samples

of these species from remote locations over their distribution range. To recover the species diversity of this tribe and their phylogenetic relationships, a meta-analysis was performed including representative species of the different tribes and clades identified in the African cichlid phylogeny of [Schwarzer *et al.* \(2009\)](#) and [Dunz & Schliewen \(2013\)](#), and all *cytochrome oxidase I (COI)* and *control region (CR)* sequences labelled as *C. guineensis*, *C. rendalli* or *C. zillii* on GenBank.

MATERIALS AND METHODS

SAMPLING

Samples were collected at two geographic scales: one local to investigate the genetic and morphometric diversity of cichlids in the PNBA and one continental to highlight the phylogenetic relationships among the PNBA cichlid lineages and those of West African cichlids. In the PNBA, fish sampling was carried out by Imraguen fishermen using fixed gillnets at three locations: R'Gueiba (19° 25' 27.1" N; 16° 27' 56.2" W), Teichott (19° 32' 38.2" N; 16° 24' 37.6" W) and Iwik (19° 53' 08.5" N; 16° 17' 43.0" W). Each of the 393 fish collected were measured, weighed and photographed. Soft dorsal fins were clipped and stored individually in tubes of 70% ethanol. The large-scale sampling targeted some widespread species belonging to Coptodonini (*C. guineensis*, *C. rendalli* and *C. zillii*) and Oreochromini (*S. melanotheron*). Samples were collected in various countries (Algeria, Egypt, Niger, Kenya, Malawi, Mauritania, Senegal, Sierra Leone, Cameroon, Côte d'Ivoire, Ghana and Gabon; for details, see Table I). When possible, voucher specimens (Table I) have been used as in-groups; in other cases, photographs of some specimens have been uploaded to the web site <https://cichlidaebancarguinmauritania.shutterfly.com/>.

MORPHOLOGICAL DATA AND ANALYSES

The colour of each cichlid collected in the PNBA was examined immediately after landing, considering the presence of: (1) tilapian spot on the dorsal fin, (2) black stripes on flanks, (3) colour and spot pattern on the caudal fin and (4) colour of the lips and the body. Fishes stored on ice were then brought to the laboratory for morphometric analysis. A total of 21 morphometric measurements and six meristic counts were recorded from cichlids sampled in the PNBA. Morphometric measurements were taken, following Teugels & Thys van den Audenaerde (2003): standard length (L_s), head length (HL), snout length (SnL), eye diameter (ED), interorbital width (IoW), preorbital bone length (PoL), width of the fifth ceratobranchial toothplate (WTP_h), pharyngeal bone length (PhL), body depth (BD), caudal peduncle depth (CPD), caudal peduncle length (CPL), horizontal distance from front tip of snout to the articulation of first dorsal-fin ray (PdL), horizontal distance from front tip of snout to the articulation of first pectoral-fin ray (PpL), horizontal distance from front tip of snout to the articulation of first pelvic (ventral)-fin ray (PvL), pre-anal length (PaL), length of dorsal-fin base (LDFB), length of the longest dorsal-fin spine (LDFS), pectoral-fin length (PFL), pelvic (ventral)-fin length (VFL), length of the anal-fin base (AFL) and length of the third spine in the anal fin (L3SAF). Meristic counts are number of: dorsal-fin rays (NDR), dorsal-fin spines (NDS), anal-fin rays (NAR), anal-fin spines (NAS), scales along the lower lateral line (NSLL) and gill rakers on the first ceratobranchial (lower) gill arch (NGR). All measurements were recorded using a calliper and a ruler was used for L_s .

Because two phenotypes (A and B) were clearly identified from the colour pattern, morphometric and meristic differences between them were investigated through a principal component analysis (PCA) of ln-transformed morphometric data (21 measurements) using the statistical programme PAST 2.17c ([Hammer *et al.*, 2001](#)). This PCA contains 119 specimens of type A and 274 specimens of type B. In this PCA, the first principal component (PC I) integrates the most size-related variation, whereas PC II, PC III and following components are theoretically size independent. A separate PCA was performed for the meristic dataset (six counts) alone;

TABLE I. Cichlid samples used in phylogenetic analyses, including sample code, species, location, collector name and GenBank accession number. Sequences in bold are those produced in this study.

Code	Species	Locality	Collector	Voucher	S7	GenBank accession number		
						NDH2	CR	COI
J08	<i>Alcolapia alcalica</i>	No locality data available			GQ168095	GQ167781		
J02	<i>Chilochromis duponti</i>	No locality data available			GQ168090	GQ167776		
I-57	Cichlidae type A	PNBA, Mauritania	N. G. Kidé		KJ938078	KJ938133	KJ925102	KJ938191
II-17	Cichlidae type A	PNBA, Mauritania	N. G. Kidé					KJ938193
II-175	Cichlidae type A	PNBA, Mauritania	N. G. Kidé		KJ938079	KJ938134	KJ925105	KJ938194
II-176	Cichlidae type A	PNBA, Mauritania	N. G. Kidé					KJ938195
II-22	Cichlidae type A	PNBA, Mauritania	N. G. Kidé					KJ938196
II-3	Cichlidae type A	PNBA, Mauritania	N. G. Kidé					KJ938197
II-6	Cichlidae type A	PNBA, Mauritania	N. G. Kidé				KJ925106	
II-60	Cichlidae type A	PNBA, Mauritania	N. G. Kidé		KJ938080	KJ938135	KJ925103	KJ938192
II-63	Cichlidae type A	PNBA, Mauritania	N. G. Kidé		KJ938081	KJ938136	KJ925104	KJ938198
III-111	Cichlidae type A	PNBA, Mauritania	N. G. Kidé		KJ938082	KJ938137	KJ925108	KJ938199
III-227	Cichlidae type A	PNBA, Mauritania	N. G. Kidé		KJ938083	KJ938138	KJ925109	KJ938200
III-230	Cichlidae type A	PNBA, Mauritania	N. G. Kidé		KJ938084	KJ938139	KJ925107	KJ938201
III-307	Cichlidae type A	PNBA, Mauritania	N. G. Kidé			KJ938140	KJ925110	KJ938202
III-309	Cichlidae type A	PNBA, Mauritania	N. G. Kidé			KJ938141	KJ925111	KJ938203
III-310	Cichlidae type A	PNBA, Mauritania	N. G. Kidé				KJ925112	KJ938204
III-312	Cichlidae type A	PNBA, Mauritania	N. G. Kidé				KJ925113	KJ938205
III-316	Cichlidae type A	PNBA, Mauritania	N. G. Kidé				KJ925114	KJ938206
III-318	Cichlidae type A	PNBA, Mauritania	N. G. Kidé				KJ925115	KJ938207
T_3697	Cichlidae type A	PNBA, Mauritania	C. Carneiro and E. R. Correia		KJ938089	KJ938147		
I-10	Cichlidae type B	PNBA, Mauritania	N. G. Kidé			KJ938129		KJ938183
I-11	Cichlidae type B	PNBA, Mauritania	N. G. Kidé			KJ938128		

TABLE I. Continued

Code	Species	Locality	Collector	Voucher	S7	GenBank accession number		
						NDH2	CR	COI
I-130	Cichlidae type B	PNBA, Mauritania	N. G. Kidé		KJ938075	KJ938130	KJ925099	KJ938184
I-132	Cichlidae type B	PNBA, Mauritania	N. G. Kidé					KJ938185
I-19	Cichlidae type B	PNBA, Mauritania	N. G. Kidé					KJ938186
I-248	Cichlidae type B	PNBA, Mauritania	N. G. Kidé		KJ938076	KJ938131	KJ925100	KJ938187
I-249	Cichlidae type B	PNBA, Mauritania	N. G. Kidé					KJ938188
I-259	Cichlidae type B	PNBA, Mauritania	N. G. Kidé					KJ938189
I-260	Cichlidae type B	PNBA, Mauritania	N. G. Kidé		KJ938077	KJ938132	KJ925101	KJ938190
IV-223	Cichlidae type B	PNBA, Mauritania	N. G. Kidé		KJ938085	KJ938142	KJ925116	KJ938208
IV-285	Cichlidae type B	PNBA, Mauritania	N. G. Kidé		KJ938086	KJ938143	KJ925117	KJ938209
IV-286	Cichlidae type B	PNBA, Mauritania	N. G. Kidé				KJ925118	KJ938210
IV-304-R	Cichlidae type B	PNBA, Mauritania	N. G. Kidé				KJ925119	KJ938211
IV-304-T	Cichlidae type B	PNBA, Mauritania	N. G. Kidé				KJ925120	KJ938212
IV-319	Cichlidae type B	PNBA, Mauritania	N. G. Kidé				KJ925121	KJ938213
IV-58	Cichlidae type B	PNBA, Mauritania	N. G. Kidé			KJ938144	KJ925122	KJ938214
114Tjo	<i>Coelotilapia joka</i>	No locality data available		ZSM40731	JX910809	JX910895		
J33	<i>Coelotilapia joka</i>	No locality data available			GQ168117	GQ167803		
ZSM-11	<i>Coelotilapia joka</i>	No locality data available	A. Dunz		KJ938090	KJ938148	KJ925069	KJ938230
J64	<i>Coptodon louka</i>	No locality data available			GQ168136	GQ167822		
J67	<i>Coptodon</i> aff. <i>louka</i> 'Samou'	No locality data available			GQ168139	GQ167825		
J72	<i>Coptodon</i> aff. <i>zillii</i> 'Kisangani'	Kisangani, DR Congo			GQ168143	GQ167829		

TABLE I. Continued

Code	Species	Locality	Collector	Voucher	S7	GenBank accession number		
						NDH2	CR	COI
112Tba	<i>Coptodon bakossiorum</i>	Lake Bermin, Cameroon	A. Lamboj	TI102	JX910807	JX910885		
116Tbe	<i>Coptodon beminii</i>	Lake Bermin, Cameroon	A. Lamboj	TI115	JX910811	JX910888		
117Tby	<i>Coptodon bythobates</i>	Lake Bermin, Cameroon	A. Lamboj	TI117	JX910812	JX910886		
98Tcam	<i>Coptodon camerunensis</i>	Ndian River, Cameroon	I. Seidel	ZSM40013	JX910798	JX910877		
ZSM-05	<i>Coptodon camerunensis</i>	No locality data available	A. Dunz		KJ938092	KJ938150	KJ925070	KJ938224
J63	<i>Coptodon dageti</i>	No locality data available			GQ168135	GQ167821		
ZSM-02	<i>Coptodon dageti</i>	No locality data available	A. Dunz		GQ168135	GQ167821	KJ925072	KJ938221
120Tde	<i>Coptodon deckerti</i>	Lake Ejagham, Cameroon		TI142	JX910815	JX910883		
J31	<i>Coptodon discolor</i>	No locality data available			GQ168115	GQ167801		
Cg-7444	<i>Coptodon guineensis</i>	Aby, Cote d'Ivoire	J.-F. Agnèse		KJ938054	KJ938100	KJ925064	KJ938233
Cg-7445	<i>Coptodon guineensis</i>	Aby, Cote d'Ivoire	J.-F. Agnèse		KJ938055	KJ938101		KJ938234
Cg-7446	<i>Coptodon guineensis</i>	Aby, Cote d'Ivoire	J.-F. Agnèse		KJ938056	KJ938102		KJ938235
Cg-7447	<i>Coptodon guineensis</i>	Aby, Cote d'Ivoire	J.-F. Agnèse		KJ938057	KJ938103	KJ925065	

TABLE I. Continued

Code	Species	Locality	Collector	Voucher	S7	GenBank accession number		
						NDH2	CR	COI
Cg-7448	<i>Coptodon guineensis</i>	Aby, Cote d'Ivoire	J.-F. Agnèsè		KJ938058	KJ938104		KJ938236
CgD-1	<i>Coptodon guineensis</i>	Hann Bay, Dakar, Senegal	A. Pariselle		KJ938059	KJ938105	KJ925066	KJ938158
CgD-2	<i>Coptodon guineensis</i>	Hann Bay, Dakar, Senegal	A. Pariselle			KJ938106	KJ925067	KJ938159
CgD-3	<i>Coptodon guineensis</i>	Hann Bay, Dakar, Senegal	A. Pariselle			KJ925069		KJ925069
CgD-4	<i>Coptodon guineensis</i>	Hann Bay, Dakar, Senegal	A. Pariselle		KJ938060	KJ938108	KJ925068	
G59	<i>Coptodon guineensis</i>	Ivindo River, Makokou, Gabon	A. Pariselle		KJ938071	KJ938124	KJ925094	KJ938179
G60	<i>Coptodon guineensis</i>	Ivindo River, Makokou, Gabon	A. Pariselle		KJ938072	KJ938125	KJ925095	KJ938180
G92	<i>Coptodon guineensis</i>	Ivindo River, Makokou, Gabon	A. Pariselle		KJ938073	KJ938126	KJ925096	KJ938181
G93	<i>Coptodon guineensis</i>	Ivindo River, Makokou, Gabon	A. Pariselle		KJ938074	KJ938127	KJ925097	KJ938182
J84	<i>Coptodon guineensis</i>	No locality data available			GQ168151	GQ167837		
Cg-5871	<i>Coptodon guineensis</i>	Weija Lake, west of Accra, Ghana	J.-F. Agnèsè		KJ938049	KJ938095		KJ938153
Cg-5872	<i>Coptodon guineensis</i>	Weija Lake, west of Accra, Ghana	J.-F. Agnèsè		KJ938050	KJ938096		KJ938154

TABLE I. Continued

Code	Species	Locality	Collector	Voucher	S7	GenBank accession number		
						NDH2	CR	COI
Cg-5877	<i>Coptodon guineensis</i>	Weija Lake, west of Accra, Ghana	J.-F. Agnèse		KJ938051	KJ938097	KJ925062	KJ938155
Cg-5878	<i>Coptodon guineensis</i>	Weija Lake, west of Accra, Ghana	J.-F. Agnèse		KJ938052	KJ938098		KJ938156
Cg-5879	<i>Coptodon guineensis</i>	Weija Lake, west of Accra, Ghana	J.-F. Agnèse		KJ938053	KJ938099	KJ925063	KJ938157
Cz-5904	<i>Coptodon guineensis</i>	Weija Lake, west of Accra, Ghana	J.-F. Agnèse		KJ938065	KJ938113	KJ925086	KJ938167
ZSM-03	<i>Coptodon louka</i>	No locality data available	A. Dunz		GQ168136	GQ167822	KJ925074	KJ938222
J70	<i>Coptodon nyongana</i>	Dja River, Cameroon	A. Dunz		GQ168141	GQ167827		
ZSM-07	<i>Coptodon nyongana</i>	Dja River, Cameroon	A. Dunz		KJ938093	KJ938151	KJ925071	KJ938226
ZSM-08	<i>Coptodon rendalli</i>	Lake Malawi near Shire River, Malawi	A. Dunz	TI141	JX910814	JX910890	KJ925075	KJ938227
104Tre	<i>Coptodon rendalli</i>	Below Calema falls, Angola	E. Swartz	SATAB85145	JX910803	JX910889		
119Tre	<i>Coptodon rendalli</i>	Lake Malawi, Malawi	A. Spreinat	TI141	JX910814	JX910890		
Cz-6605	<i>Coptodon rendalli</i>	Victoria Lake, Kenya	J.-F. Agnèse		KJ938067	KJ938116	KJ925088	KJ938170
Cz-6606	<i>Coptodon rendalli</i>	Victoria Lake, Kenya	J.-F. Agnèse		KJ938068	KJ938117	KJ925089	KJ938171
Cz-6607	<i>Coptodon rendalli</i>	Victoria Lake, Kenya	J.-F. Agnèse		KJ938069	KJ938118		KJ938172

TABLE I. Continued

Code	Species	Locality	Collector	Voucher	S7	GenBank accession number		
						NDH2	CR	COI
Cz-6608	<i>Coptodon rendalli</i>	Victoria Lake, Kenya	J.-F. Agnès		KJ938070	KJ938119		KJ938173
Cz-6609	<i>Coptodon rendalli</i>	Victoria Lake, Kenya	J.-F. Agnès					KJ938174
121Tsn	<i>Coptodon</i> <i>snijderae</i>	Lake Bermin, Cameroon		ZSM40782	JX910816	JX910887		
ZSM-13	<i>Coptodon</i> sp. 'Cross'	Cross River at Mamfe, Cameroon	A. Dunz	TI146	JX910817	JX910884	KJ925077	KJ938231
ZSM-04	<i>Coptodon</i> sp. aff. <i>louka</i> 'Samou'	No locality data available	A. Dunz		GQ168139	GQ167825	KJ925076	KJ938223
J35	<i>Coptodon tholloni</i>	No locality data available	A. Dunz		GQ168118	GQ167804		
ZSM-09	<i>Coptodon tholloni</i>	No locality data available	A. Dunz		KJ938094	KJ938152	KJ925078	KJ938228
N1	<i>Coptodon zillii</i>	Ourgla, Algeria	A. Pariselle		KJ938087	KJ938145	KJ925125	KJ938219
N2	<i>Coptodon zillii</i>	Ourgla, Algeria	A. Pariselle		KJ938088	KJ938146	KJ925126	KJ938220
125Tzi	<i>Coptodon zillii</i>	Lake Mariout, Egypt	A. Dunz and E. Schraml	ZSM38791	JX910820	JX910880		
ZSM-06	<i>Coptodon zillii</i>	Lake Mariout, south-west of Alexandria, Egypt	A. Dunz	ZSM38791	JX910820	JX910880	KJ925079	KJ938225
Cz-7307	<i>Coptodon zillii</i>	Man, Cote d'Ivoire	J.-F. Agnès					KJ938175
Cz-7313	<i>Coptodon zillii</i>	Man, Cote d'Ivoire	J.-F. Agnès			KJ938120	KJ925090	KJ938176

TABLE I. Continued

Code	Species	Locality	Collector	Voucher	S7	GenBank accession number		
						NDH2	CR	COI
Cz-7316	<i>Coptodon zillii</i>	Man, Cote d'Ivoire	J.-F. Agnèse			KJ938121	KJ925091	KJ938177
Cz-7319	<i>Coptodon zillii</i>	Man, Cote d'Ivoire	J.-F. Agnèse			KJ938122	KJ925092	KJ938178
Cz-7322	<i>Coptodon zillii</i>	Man, Cote d'Ivoire	J.-F. Agnèse			KJ938123	KJ925093	
Cz-5462	<i>Coptodon zillii</i>	Ndjamena, Niger	J.-F. Agnèse		KJ938061		KJ925080	KJ938161
Cz-5463	<i>Coptodon zillii</i>	Ndjamena, Niger	J.-F. Agnèse		KJ938062		KJ925081	KJ938162
Cz-5464	<i>Coptodon zillii</i>	Ndjamena, Niger	J.-F. Agnèse		KJ938063		KJ925082	KJ938163
Cz-5465	<i>Coptodon zillii</i>	Ndjamena, Niger	J.-F. Agnèse				KJ925083	KJ938164
Cz-5468	<i>Coptodon zillii</i>	Ndjamena, Niger	J.-F. Agnèse		JX910790		KJ925084	KJ938165
88Tzil	<i>Coptodon zillii</i>	Orontes, Syria	N. Alwan et al.,	ZSM40036		JX910881		
Cz-5903	<i>Coptodon zillii</i>	Weija Lake, west of Accra, Ghana	J.-F. Agnèse		KJ938064	KJ938112	KJ925085	KJ938166
Cz-5905	<i>Coptodon zillii</i>	Weija Lake, west of Accra, Ghana	J.-F. Agnèse		KJ938066	KJ938114		KJ938168
Cz-5906	<i>Coptodon zillii</i>	Weija Lake, west of Accra, Ghana	J.-F. Agnèse			KJ938115	KJ925087	KJ938169
105Tzi	<i>Coptodon zillii</i>	White Nile River at Kosti, Sudan	A. Dunz	ZSM35146	JX910804	JX910878		
J03	<i>Etila nguti</i>	No locality data available			GQ168091	GQ167777		
102Get	<i>Gobiocichla ethelwynnae</i>	Cross River at Mamfe, Cameroon	J. Gottwald	PAA-443	JX910801	JX910893		

TABLE I. Continued

Code	Species	Locality	Collector	Voucher	S7	GenBank accession number		
						NDH2	CR	COI
ZSM-10	<i>Heterotilapia buttikoferi</i>	Freetown, Sierra Leone	A. Dunz		GQ168111	AF317251	KJ925098	KJ938229
J27	<i>Heterotilapia buttikoferi</i>	No locality data available			GQ168111	GQ167797		
J74	<i>Iranocichla hornuzensis</i>	No locality data available			GQ168144	GQ167830		
124Kei	<i>Konia eisenbraui</i>	Lake Barombi Mbo, Cameroon		TH150	JX910819	JX910871		
J38	<i>Oreochromis andersonii</i>	No locality data available			GQ168119	GQ167805		
J07	<i>Oreochromis niloticus</i>	No locality data available			GQ168094	GQ167780		
J10	<i>Oreochromis tanganicae</i>	No locality data available			GQ168096	GQ167782		
103Tea	<i>Pelmatotapia cabrae</i>	Lucalla River, Angola	E. Swartz, R. Bills and D. Neto	SALAB84681	JX910802	JX910874		
101Tma	<i>Pelmatotapia mariae</i>	No locality data available		ZSM39713	JX910800	JX910876		
J61	<i>Sarotherodon caudomarginatus</i>	No locality data available			GQ168133	GQ167819		
92Sgal	<i>Sarotherodon galilaeus</i>	Congo River, DR Congo	D. Neumann	ZSM37722	JX910793	JX910869		
123Slo	<i>Sarotherodon lohbergeri</i>	Lake Barombi Mbo, Cameroon		TH149	JX910818	JX910870		
J49	<i>Sarotherodon mvogoi</i>	No locality data available			GQ168125	GQ167811		
J15	<i>Sarotherodon nigripinnis</i>	No locality data available			GQ168101	GQ167787		

TABLE I. Continued

Code	Species	Locality	Collector	Voucher	S7	GenBank accession number		
						NDH2	CR	COI
I08Ssa	<i>Sarotherodon sanagaensis</i>	Sanaga River, Cameroon	A. Lamboj	ZSM38845	JX910805	JX910868		
Lgu1-E	<i>Sarotherodon melanotheron</i>	Guiers Lake, Senegal	J.-D. Durand				KJ938216	
Lgu10-E	<i>Sarotherodon melanotheron</i>	Guiers Lake, Senegal	J.-D. Durand				KJ925123	KJ938215
Lgu4-E	<i>Sarotherodon melanotheron</i>	Guiers Lake, Senegal	J.-D. Durand				KJ925124	KJ938217
Lgu7-E	<i>Sarotherodon melanotheron</i>	Guiers Lake, Senegal	J.-D. Durand					KJ938218
J18	<i>Steatocranus bleheri</i>	No locality data available			GQ168103	GQ167789		
J55	<i>Steatocranus glaber</i>	No locality data available			GQ168130	GQ167816		
J25	<i>Stomatepia mariae</i>	No locality data available			GQ168110	GQ167796		
ZSM-12	<i>Tilapia busumana</i>	Lake Bosumtwi, village Abono, Ghana	A. Dunz		KJ938091	KJ938149	KJ925073	KJ938232
J71	<i>Tilapia brevimanus</i>	No locality data available			GQ168142	GQ167828		
ZSM36123	<i>Tilapia pra</i>	Anum River, Ghana	D. Neumann, G. Baffur Entwi	ZSM36123	JX910794	JX910894		
111Tru	<i>Tilapia ruweti</i>	RD Congo	U. Schlieven	ZSM40760	JX910806	JX910826		
J30	<i>Tilapia sparrmanii</i>	No locality data available			GQ168114	GQ167800		
J51	<i>Tristramella simonis</i>	No locality data available			GQ168127	GQ167813		

COI, cytochrome oxidase I; CR, control region; PNBA, Banc d'Arguin National Park; TI and ZSM, Zoologisches Staatssammlung München; SAIAB, South African Institute for Aquatic Biodiversity.

meristics are size independent and thus PC I is the most informative component. Scores of both PCAs were combined in a single bivariate plot for most informative visualization.

MOLECULAR DATA AND ANALYSES

Phylogenetic relationships among cichlids sampled in PNBA (Mauritania), Hann Bay (Dakar, Senegal) and others belonging to Oreochromini and Coptodonini (Dunz & Schliewen, 2013) (in-group) were estimated using sequence polymorphisms of a mitochondrial (*ND2*) and a nuclear (*S7 ribosomal protein first intron*) gene. *ND2* and *S7* sequences published by Schwarzer *et al.* (2009) and Dunz & Schliewen (2013) were downloaded from GenBank and used as reference species because these contain sequences of voucher specimens stored in museums used for a revision of cichlid systematics from Africa (Dunz & Schliewen, 2013).

To investigate the phylogeographic structure and provide insight into the intraspecific genetic diversity of widespread species of *Coptodon* Gervais 1853 (*C. guineensis*, *C. rendalli* and *C. zillii*) as well as *S. melanotheron*, the *COI* portion and the mitochondrial *CR* fragment were also sequenced. These two additional mitochondrial sequences were chosen because: (1) the *COI* sequence is the marker used for DNA barcoding in the BOLD programme, so data are available on GenBank and can be used to increase the number of locations analysed for a species; (2) the *CR* is known for its high level of polymorphism and was used by Falk *et al.* (2003) to investigate the phylogeographic structure of *S. melanotheron* and *Sarotherodon nigripinnis* (Guichenot 1861) and (3) *COI* and *CR* sequences available in GenBank of species of *Coptodon* and *S. melanotheron* were added to the dataset (Table SI, Supporting Information). Only *COI* sequences of barcoding studies conducted in Africa were downloaded while most of *CR* sequences added to the present analyses belong to the study of Falk *et al.* (2003).

Genomic DNA was extracted from fin clips using DNeasy Blood & Tissue kit (QIAGEN; www.qiagen.com) following the standard protocol provided by the manufacturer. Amplifications were performed in 25 µl volumes containing 1 µl of genomic DNA, 3 mM of forward and reverse primers, 12.5 µl of FastStart PCR Master mix (Roche; https://lifesciences.roche.com/), 0.5 µl of 2% bovine serum albumin (Euromedex; www.euromedex.com) and water. Amplification of all fragments were carried out in 30–35 cycles according to the following temperature profile: 3 min at 92° C (initial denaturation), 45 s at 92° C, 45 s at 52° C (for *ND2* and *COI*) or 56° C (for *CR*), 60 s at 72° C and finally 5 min at 72° C. Specifically for the *S7* sequence amplification, a touch down PCR was adapted from the previous PCR conditions and where annealing temperature decreases 0.5° C each cycle from 62 to 58° C, totaling 35 cycles. Primers used to amplify the different gene portion were *ND2Met/ND2Trp* (Kocher *et al.*, 1995) for *ND2*, *S7RPEX1F50/S7RPEX2R50* for *S7* (Chow & Hazama, 1998), *FishF1 + FishF2/FishR1* for *COI* (Ward *et al.*, 2005) and *L-Smel/H-Smel* for *CR* (Falk *et al.*, 2003).

Portions of the nuclear intronic (*S7*) and mitochondrial *CR* sequences were aligned using Muscle (Edgar, 2004) as implemented in MEGA 5.2.2 (Tamura *et al.*, 2011) while *COI* and *ND2* gene sequences were aligned visually using the alignment editor provided by MEGA 5.2.2. Phylogenetic analyses were conducted applying the maximum likelihood (ML) approach implemented in MEGA 5.2.2. Best-fit models of sequence evolution for each dataset were estimated using the option «Best DNA model» implemented in MEGA 5.2.2 (www.megasoftware.net).

A first phylogenetic analysis was performed on *ND2* and *S7* sequences to highlight phylogenetic relationships of the samples with those analysed by Schwarzer *et al.* (2009) and Dunz & Schliewen (2013). To evaluate phylogenetic information recovered by each gene, single gene trees were built. A second phylogenetic analysis was performed on *COI* and *CR* sequences to investigate the genetic diversity of *C. guineensis*, *C. rendalli*, *C. zillii* and *S. melanotheron*. The Hasegawa Kishino Yano (HKY) model, considering non-uniformity of evolutionary rates among sites modelled using a discrete gamma distribution (+G), was used for *ND2 + S7* and *CR* datasets [Bayesian information criterion (BIC) = 19594.73 and 7965.32, respectively]; Kimura two-parameter (K2P) model, +G for *COI* (BIC = 8014.0) and invariant mutation rates among sites (+I) for *S7* (BIC = 5427.706). Node support was tested using the bootstrap procedure (Felsenstein, 1985), based on 200 pseudo-replicates. All phylogenetic trees were rooted using the out-groups *Heterochromis multidens* (Pellegrin 1900) and *Etia nguti* Schliewen & Stiassny 2003 following Dunz & Schliewen (2013).

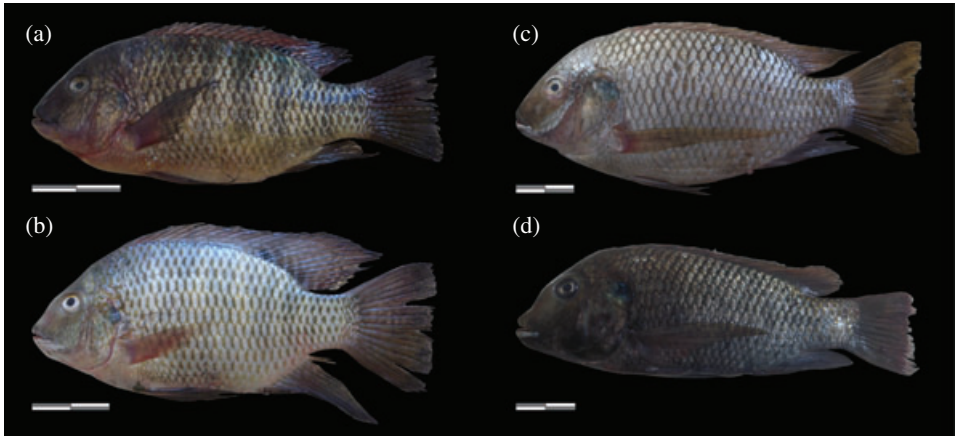


FIG. 1. Cichlidae collected in the Banc d'Arguin National Park, Mauritania: (a, b) phenotype A with colour variation and (c, d) phenotype B with its colour variation. Scale bar (white + grey) represents 2 cm.

RESULTS

PHENOTYPES, MORPHOMETRY AND MERISTICS

Different phenotypes were observed in the cichlids sampled in PNBA, based upon the general colour of body and fins. Although there was wide morphological variation, all individuals could be classified into two main phenotypes. Cichlids classified as phenotype A were characterized by: presence of a tilapia spot on dorsal fin, caudal fin spotted, greyish on its upper parts and yellowish in lower parts; the belly can present pinkish to yellowish colour while the back is brownish; large vertical bars present on sides [Fig. 1(a)]. Cichlids classified as phenotype B were characterized by: presence of a white lip (lower) and absence of spots (including the tilapia spot) on dorsal and caudal fins [Fig. 1(c)]. Large colour variation was observed within the two phenotypes, but the colour of phenotype A was usually lighter and side bands were less marked [Fig. 1(b)], while specimens of phenotype B were usually totally black [Fig. 1(d)]. Additional photographs of some specimens used in this study are available at the website: <https://cichlidaebancarguinmauritania.shutterfly.com/>.

A PCA confirms the differences observed between specimens classified as phenotype A or B (Fig. 2). Differences between the two phenotypes are mainly noticeable on the PCA I meristic axis that accounts for 81.43% of the total variance while the PCA II morphometric axis accounts for 10.75% (Fig. 2). When compared with B, specimens classified as phenotype A have shorter PFL, HL, PpL and PhL. Additionally, the NDR is higher in phenotype A *v.* B, while NGR is lower in phenotype A *v.* B (Table II).

PHYLOGENETIC RELATIONSHIPS

All sequences obtained in this study have been deposited on GenBank (Table I). Fragment size ranges between 355 and 371 for the *CR*, 469 and 492 bp for *S7*, 632 and 648 bp for *COI* and 972 bp for *ND2*. In all reconstructed phylogenetic trees, cichlids collected in PNBA belong to two divergent clades. These two clusters are composed of the same individuals presenting the same phenotype, independent of the gene

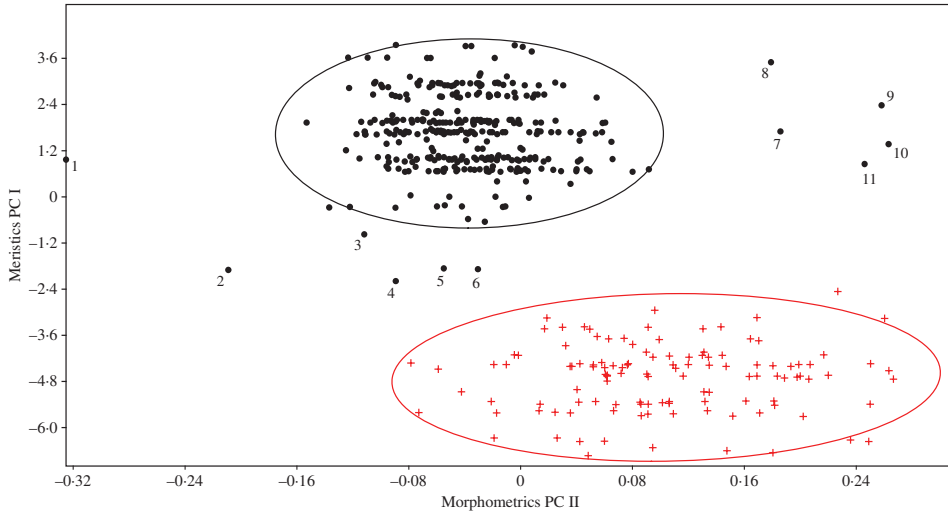


FIG. 2. Bivariate plot of principal component II (PC II; morphometrics) and PC I (meristics); 95% C.I. visualized as ellipses. +, type A ($n = 119$); ●, type B ($n = 340$). Total number of specimens in the plot ($n = 459$). Labelled specimens 1–11 refer to the type B.

sequences used for the phylogenetic reconstruction, with one exception: individual I-57 that appears in clade B in the nuclear *S7* tree [Fig. 3(a)] and in the clade A in mitochondrial trees [Figs 3(b) and 4].

The *S7* phylogenetic tree [Fig. 3(a)] presents a lower resolution than the *ND2* mitochondrial tree [Fig. 3(b)] or the multi-gene phylogenetic tree published by [Dunz & Schlieven \(2013\)](#), but the phylogenetic relationships highlighted by the *S7* tree are largely concordant, and even more highly supported than the *ND2* tree for deep divergences. Thus, in the *S7* tree [Fig. 3(a)] the *Etini* tribe is the most divergent haplotilapine followed by the *Oreochromini* tribe, as recovered in the multi-gene tree ([Dunz & Schlieven, 2013](#)). The *Oreochromini* and *Coptodonini* tribes are also monophyletic in the *S7* tree, whereas the node basal to all species of the *Coptodonini* tribe is not supported in the *ND2* tree due to the placement of the two specimens of *Coptodon tholloni* (Sauvage 1884) [CIII; Fig. 3(b)]. For recent divergences, the two *Oreochromini* clades SI and SII are not supported in the *S7* tree, unlike on the *ND2* tree [Fig. 3(a), (b)]; but the three *Coptodonini* clades (CI, CII and CIII) are well supported in the two single gene trees. Species composition of these last three clades is identical in both trees, with one exception. While morphotype A is grouped with *C. guineensis* sampled in Senegal and belongs to the CI clade in the *S7* tree, this group of sequences belongs to the CII clade in the *ND2* tree [Fig. 3(a), (b)]. Lastly, in both trees, *C. guineensis* is polyphyletic or paraphyletic.

The phylogenetic tree built from the concatenation of both *ND2* and *S7* sequences (I-57 has been removed) highlights a close relationship of individuals classified as phenotype A with *C. guineensis* collected in Senegal, and belong to a subclade including *Coptodon* sp. aff. *louka* and *Coptodon camerunensis* (Lönnberg 1903), within the clade CII of the *Coptodonini* tribe [Fig. 3(c)]. Individuals classified as phenotype B present a close relationship with *Sarotherodon nigripinnis* (Guichenot 1861), all included in the

TABLE II. Morphometric variability and meristic counts (in italics) recorded on Mauritanian cichlids presenting the phenotypes A ($n = 97$) and B ($n = 296$)

		Phenotype A		Phenotype B	
		Mean \pm s.d.		Mean \pm s.d.	
Morphometric variability	%HL	23.6 \pm 4.0		28.4 \pm 2.8	
	%SnL	11.6 \pm 2.5		12.5 \pm 1.5	
	%ED	4.8 \pm 0.6		5.2 \pm 0.6	
	%IoW	8.6 \pm 1.2		9.5 \pm 1.0	
	%PoL	7.1 \pm 1.1		8.6 \pm 1.2	
	%WTPH	6.2 \pm 0.8		6.1 \pm 0.8	
	%PhL	7.7 \pm 1.0		10.2 \pm 1.2	
	%BD	31.4 \pm 3.2		33.3 \pm 2.8	
	%CPD	14.1 \pm 1.7		14.5 \pm 1.3	
	%CPL	9.6 \pm 1.3		10.1 \pm 1.4	
	%PdL	23.0 \pm 3.1		24.8 \pm 3.2	
	%PpL	22.9 \pm 2.6		26.9 \pm 2.6	
	%PvL	28.7 \pm 3.2		31.5 \pm 3.5	
	%PaL	55.9 \pm 5.7		58.8 \pm 6.0	
	%LDFB	45.3 \pm 4.7		47.1 \pm 3.7	
	%LDFS	9.3 \pm 2.1		8.8 \pm 1.3	
	%PFL	25.1 \pm 3.6		34.7 \pm 4.0	
	%VFL	23.4 \pm 3.7		20.9 \pm 2.9	
%AFL	13.1 \pm 1.6		13.7 \pm 1.3		
%L3SAF	7.6 \pm 1.2		8.4 \pm 1.0		
		Most frequent	Minimum–maximum	Most frequent	Minimum–maximum
Meristic counts	<i>NDR</i>	14	12–15	12	11–14
	<i>NDS</i>	15	14–16	16	15–17
	<i>NAR</i>	10	9–11	10	8–11
	<i>NAS</i>	3	3	3	3
	<i>NSLL</i>	29	27–32	29	27–32
	<i>NGR</i>	18	16–20	24	20–26

%, Measure corrected by the standard length (L_S). HL, head length; SnL, snout length; ED, eye diameter; IoW, interorbital width; PoL, preorbital bone length; WTPH, width of the fifth ceratobranchial toothplate; PhL, pharyngeal bone length; BD, body depth; CPD, caudal peduncle depth; CPL, caudal peduncle length; PdL, horizontal distance from front tip of snout to the articulation of first dorsal-fin ray; PpL, horizontal distance from front tip of snout to the articulation of first pectoral-fin ray; PvL, horizontal distance from front tip of snout to the articulation of first pelvic (ventral)-fin ray; PaL, pre-anal length; LDFB, length of dorsal-fin base; LDFS, length of the longest dorsal-fin spine; PFL, pectoral-fin length; VFL, pelvic (ventral)-fin length; AFL, length of the anal-fin base; L3SAF, length of the third spine in the anal fin; NDR, dorsal-fin rays; NDS, dorsal-fin spines; NAR, anal-fin rays; NAS, anal-fin spines; NSLL, scales along the lower lateral line; NGR, gill rakers on the first ceratobranchial (lower) gill arch.

clade SI inside the Oreochromini tribe. Samples of *C. zillii* analysed in this study are all grouped with other *C. zillii* analysed by Dunz & Schliewen (2013), just as the current samples of *C. rendalli* are also grouped with *C. rendalli* analysed by Schwarzer *et al.* (2009) and Dunz & Schliewen (2013). In contrast, the current samples of *C. guineensis*, identified in the field by morphological characteristics, are included in three different

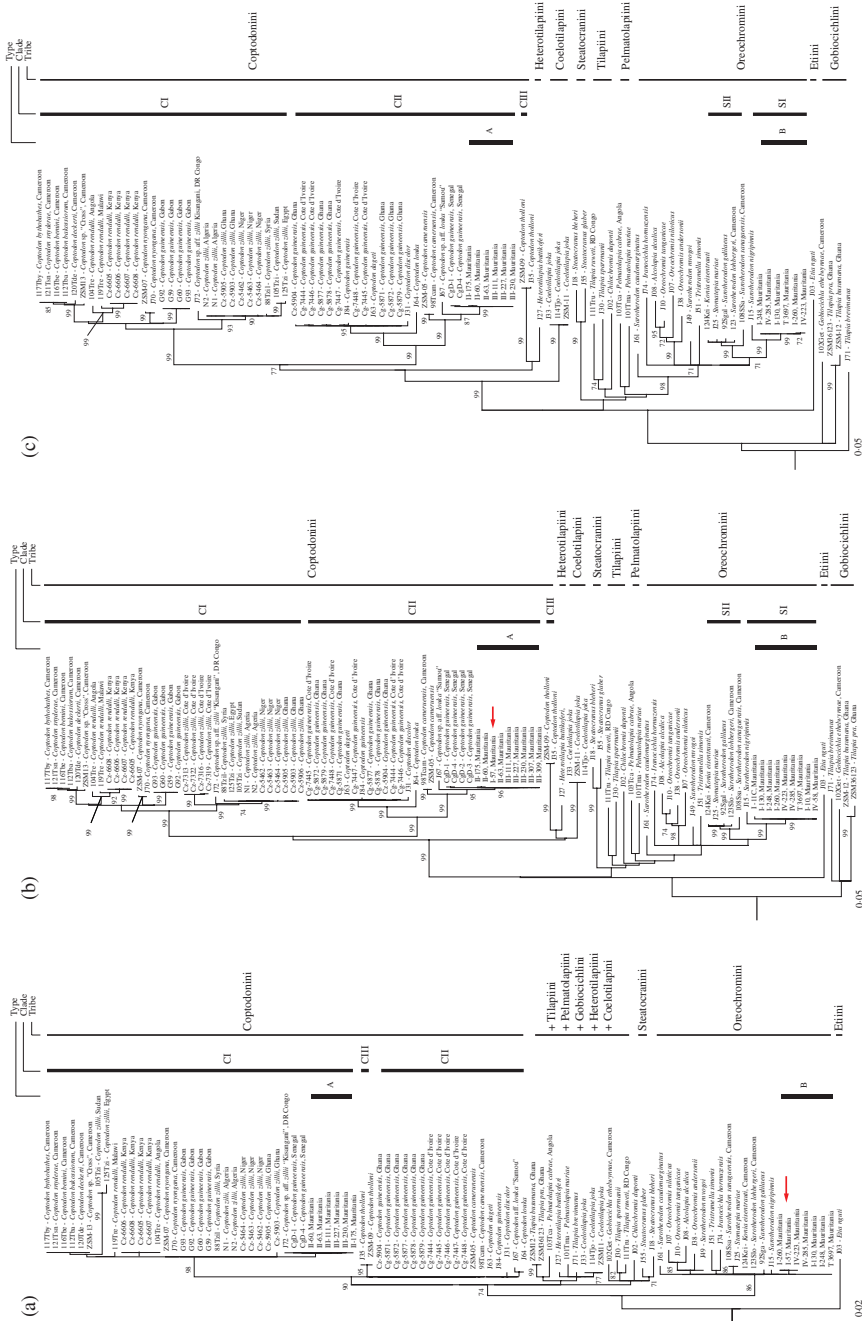


Fig. 3. Legend on next page.

subclades within two clades of the Coptodonini tribe. The samples of *C. guineensis* from Ghana are closely related to *C. guineensis* analysed by [Dunz & Schlieven \(2013\)](#), *Coptodon discolor* (Günther 1903) and *Coptodon dagei* (Thys van den Audenaerde 1971), all included in the clade CII [Fig. 3(c)]. *Coptodon guineensis* sampled in Senegal also belongs to this clade, but to a different subclade. *Coptodon guineensis* sampled in Gabon belongs to the clade CI in a subclade that includes *C. rendalli*, *C. zillii*, *C. sp. aff. zillii* ‘Kisangani’, *Coptodon nyongana* (Thys van den Audenaerde 1971), *Coptodon sp.* ‘Cross’, *Coptodon deckerti* (Thys van den Audenaerde 1967), *Coptodon bemini* (Thys van den Audenaerde 1972), *Coptodon bythobates* (Stiassny, Schlieven & Dominey 1992), *Coptodon snyderae* (Stiassny, Schlieven & Dominey 1992) and *Coptodon bakossiorum* (Stiassny, Schlieven & Dominey 1992) [Fig. 3(c)].

Phylogenetic relationships inferred from the *COI* sequence polymorphisms confirm the presence of two species in the Mauritanian cichlid samples, and that they belong to two different genera and tribes [Fig. 4(a)]. This *COI* tree confirms the monophyly of *C. zillii* that consists of two subclades: one group including all individuals sampled exclusively in Côte d’Ivoire [subclade Cz-II in Figs 4(a) and 5(b)] and the second group including all other *C. zillii* sampled over a large geographic range: from north Africa and the Levant in the north, to Niger, Ghana and Nigeria to the south [subclade Cz-I in Figs 4(a) and 5(b)]. The polyphyly of *C. guineensis* is confirmed and consists of four lineages in this *COI* tree, as sequences of *C. guineensis* from Nigeria uploaded from GenBank were grouped with *Coptodon sp.* ‘Cross’ [subclade Cg-III in Figs 4(a) and 5(a)].

Lastly, the phylogenetic relationships inferred from *CR* also support previous observations; fine intraspecific relationships are highlighted, especially in *S. melanotheron* where two subclades are retrieved. One grouped all individuals sampled in Ghana and Côte d’Ivoire [subclade Sm-II in Figs 4(b) and 5(d)] and the other one consisted of individuals sampled in Liberia, Sierra Leone, Senegal and Mauritania [subclade Sm-I in Figs 4(b) and 5(d)].

DISCUSSION

SPECIES DIVERSITY AND POLYCHROMATISM OF PNBA CICHLIDS

Both morphometric and genetic analyses clearly demonstrate the presence of two cichlid species in PNBA, Mauritania. To date, only *S. melanotheron* was recorded in

FIG. 3. Phylogenetic relationships among Mauritanian cichlids (types A and B) and other haplotilapiine species belonging to clade and tribes (with the exception of East African cichlids radiation) depicted in [Dunz & Schlieven \(2013\)](#). Maximum likelihood (ML) analyses are based on polymorphisms of (a) *S7* sequences, (b) *ND2* and (c) the concatenation of *S7* and *ND2* sequences. Branch length is proportional to the number of substitutions under (a) Kimura two-parameter (K2P) + I, (b) Hasegawa Kishino Yano (HKY) + G + I and (c) HKY + G model. Numbers on the branches are ML bootstrap values (in %, from 200 replicates), with those below 70% not shown. Scale bar: 0.1 inferred nucleotide substitution/site under (HKY + G) model. ←, point to an individual that presents different placement in phylogenetic reconstructions.

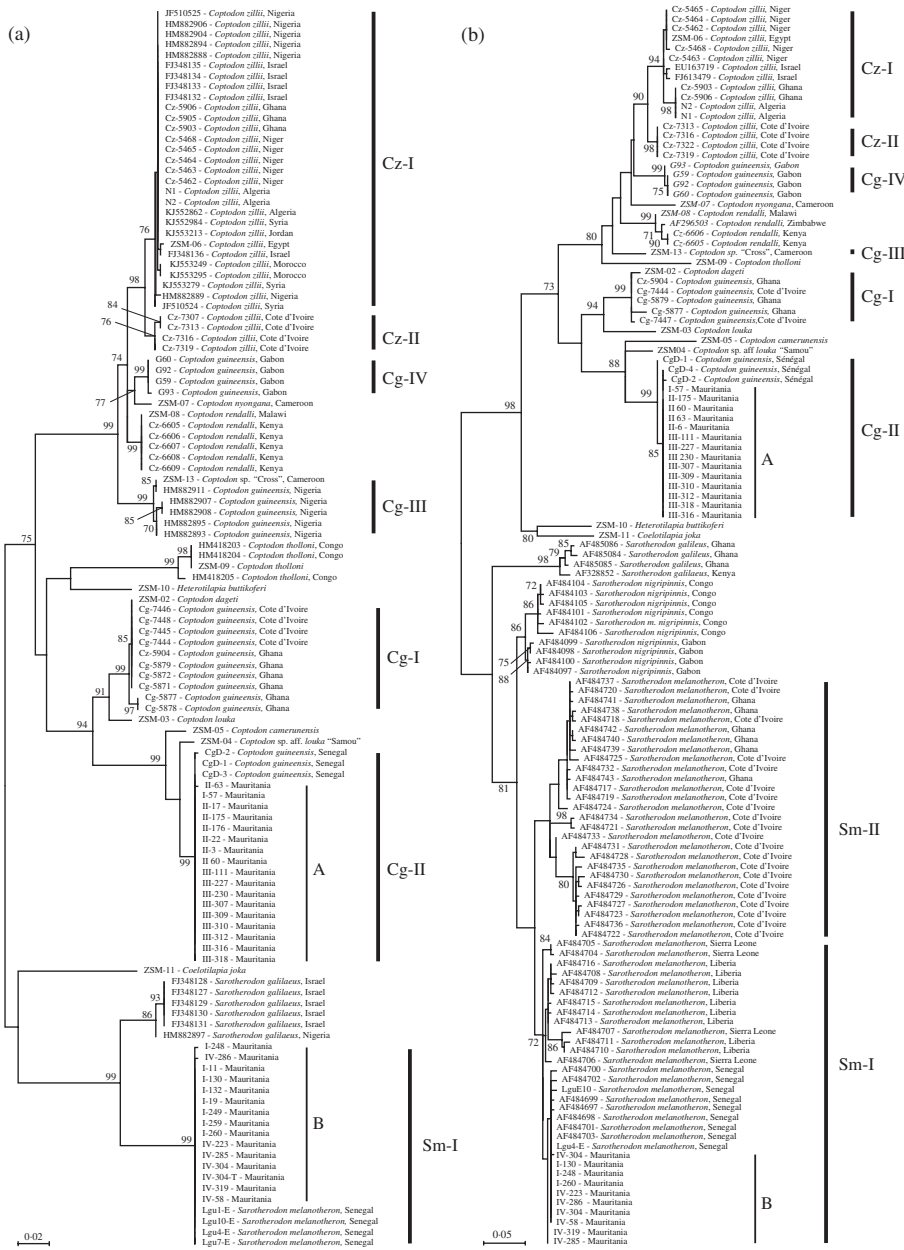


FIG. 4. Phylogenetic relationships among Mauritanian cichlids (types A and B) and other species belonging to Coptodonini, Oreochromini, Heterotilapiini and Coelotilapiini tribes as depicted in Dunz & Schlieven (2013). Maximum likelihood (ML) analyses are based on polymorphisms of (a) *cytochrome oxidase I* portion (COI) and (b) *control region* (CR). Branch length is proportional to the number of substitutions under (a) Kimura two-parameter (K2P) + G and (b) Hasegawa Kishino Yano (HKY) + G models. Numbers on the branches are ML bootstrap values (in %, from 200 replicates), with those below 70% not shown. Scale bar: 0.1 inferred nucleotide substitution/site under (K2P + G or HKY + G + I) model. (a) In the COI tree, *Coptodon guineensis* and *Coptodon zillii* subclades are highlighted (d). (b) In the CR tree, *Sarotherodon melanotheron* subclades are highlighted (d).

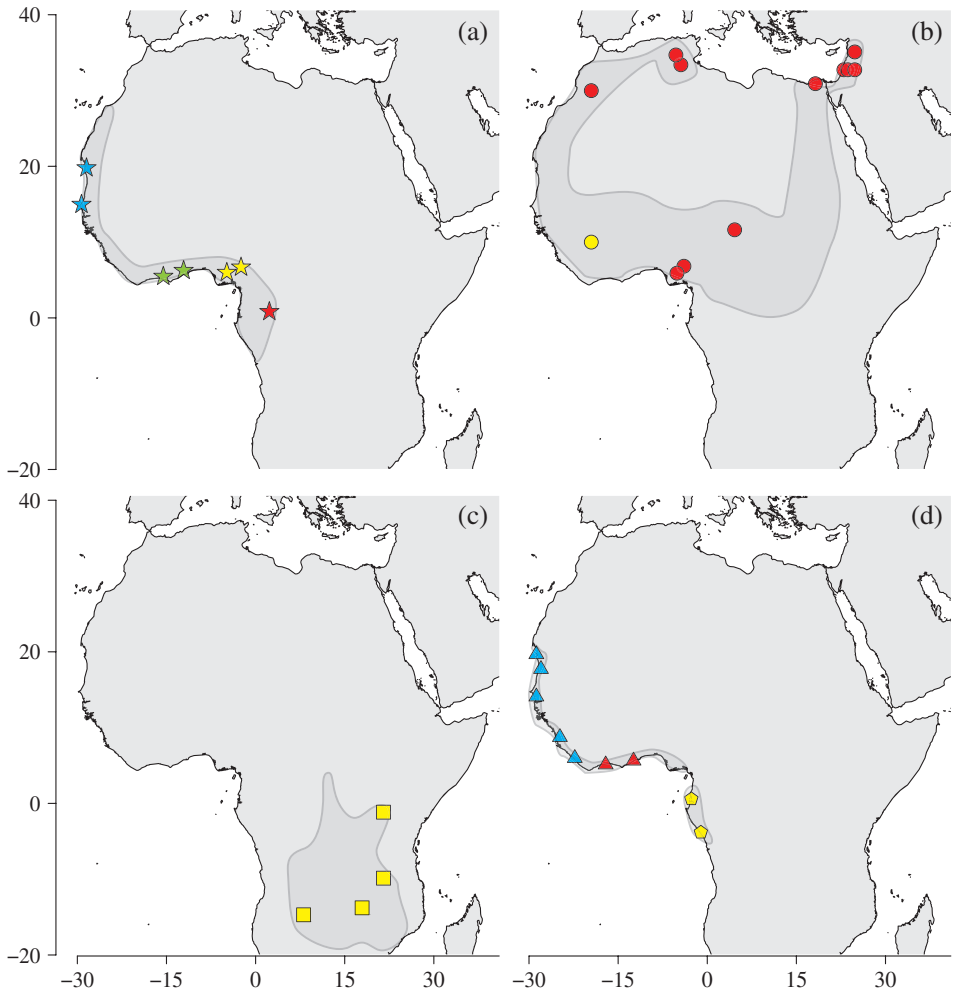


FIG. 5. Distribution of clades of (a) *Coptodon guineensis*, (b) *Coptodon zillii*, (c) *Coptodon rendalli* and (d) *Sarotherodon melanotheron sensu lato* highlighted in phylogenetic analyses based on cytochrome oxidase I (COI) and control region (CR) [Fig. 4(a), (b)]. ★, Cg-II; ☆, Cg-I; ☆, Cg-III; ★, Cg-IV; ●, Cz-I; ●, Cz-II; ■, *C. rendalli*; ▲, Sm-I; ▲, Sm-II; ●, *Sarotherodon nigripinnis*. Shaded border areas correspond to distribution range of these species or species complex.

this area, which exhibits the phenotype B, as classified by this study. All morphometric variables, meristic counts and phylogenetic relationships of phenotype B cichlids are in agreement with the description of *S. melanotheron*. This species is abundant in estuarine environments over its distribution range from Cameroun to Mauritania (Teugels & Thys van den Audenaerde, 2003). In contrast, the report of a second species is unexpected. Meristic and morphometric parameters recorded on individuals exhibiting phenotype A match with the description of a species belonging to *Coptodon*. Its presence in a marine environment and its close phylogenetic relationships with *C. guineensis* collected in Hann Bay (Dakar, Senegal), however, suggest that this species is *C. guineensis* (but see below). This species is widely distributed in West Africa,

from Angola to Senegal (Teugels & Thys van den Audenaerde, 2003). Its known northern distribution limit corresponds to the Senegal River estuary, at the border between Mauritania and Senegal. Populations attributed to this species, however, have recently been reported at more northern localities: in the Sebkhet Imlily (Morocco), situated at *c.* 200 km from the border with Mauritania in salt waters (Qninba *et al.*, 2009), and in rock pools (locally known as *gueltas*) along the Aabar wadi, tributary of Chbeyka wadi, Tan Tan province (south-western Morocco; Qninba *et al.*, 2012). Therefore, the presence of *C. guineensis* in PNBA is not completely surprising, even if previous records were absent and despite the many marked morphometric and meristic differences between *S. melanotheron* and *C. guineensis*. Together, *C. guineensis* might then be present in other isolated areas of the Atlantic Sahara (Fig. 5). Further sampling is needed to describe the distribution range of this species, which would have important implications both in terms of conservation and evolutionary biology.

There is large polychromatism within *S. melanotheron* sampled in PNBA, which can either present a silvery colour pattern or a totally black one, whereas the original species description only mentioned some black spots in adults (Teugels & Thys van den Audenaerde, 2003). Polychromatism may have a different origin in these cichlids, such as sexual dichromism or social communication (Maan & Sefc, 2013). In *S. melanotheron*, colour pattern variations are probably related to sexual maturity as black spots are often larger and darker in mature males and absent in juvenile ones (Teugels & Thys van den Audenaerde, 2003).

COPTODON GUINEENSIS: A SPECIES COMPLEX

The systematics of Cichlidae has a long history, and many successive revisions have been performed, but recent molecular phylogenies call for a new one (Nagl *et al.*, 2001; Klett & Meyer, 2002; Schwarzer *et al.*, 2009; Dunz & Schliewen, 2013). Among the most important results of these molecular phylogenetic reconstructions is the demonstration that *Tilapia sensu lato* is not a monophyletic group, but consists of several genera included in 10 lineages (Dunz & Schliewen, 2013). At the species rank, that study provided limited insights into diversity. While Dunz & Schliewen (2013) mentioned the presence in the Coptodonini tribe of 31 species distributed into three clades, they only briefly mentioned the presence of cryptic species of *C. zillii*, *C. guineensis* or *Coptodon louka* (Thys van den Audenaerde 1969), by naming some taxa as *C. sp. aff. zillii*, *C. sp. aff. guineensis* or *C. sp. aff. louka*. In this study, all phylogenetic reconstructions with all molecular markers (nuclear or mitochondrial) reveal *C. guineensis* to be paraphyletic and to consist of three to four evolutionary lineages embedded in two clades consisting of all species of *Coptodon*, with the exception of *C. tholloni* and *Coptodon congica* (Poll & Thys van den Audenaerde 1960). Consequently, *C. guineensis* is a species complex and, considering the distribution range of these lineages [Fig. 5(a)], these putative species are parapatric. Thus, the lineage representing the valid *C. guineensis* may correspond to the Cg-I lineage (Fig. 4) as its distribution ranges includes the type locality of *C. guineensis*, *i.e.* the Ghanaian Ashantee area (Bleeker in Günther, 1862). *Coptodon guineensis* is a euryhaline species with a clear affinity for marine or brackish water, as demonstrated by the origin of the current samples, which were collected in the Aby Lagoon in Côte d'Ivoire and Weija Lake in Ghana, which have salinities close to marine (Asante *et al.*,

2008). Nevertheless, it is phylogenetically extremely close to the parapatric freshwater species, *C. dageti* and *C. discolor* (Teugels & Thys van den Audenaerde, 2003). This phylogeographic pattern suggests a recent origin of *C. dageti* and *C. discolor*, which would have diverged from the ancestral *C. guineensis* when entering freshwater environments.

Other lineages of *C. guineensis* highlighted in the phylogenetic trees correspond to undescribed species or species that were invalidated after successive revisions. In Senegal, two species were described (*Tilapia affinis* Duméril 1861 and *Tilapia polycentra* Duméril 1861) that were later synonymized with *C. guineensis* (Eschmeyer, 2015). To determine which species name should be resurrected, a more complete redescription of each type species has to be performed, which is beyond the scope of this study. Among all lineages of *C. guineensis* highlighted in this study, the Mauritanian/Senegalese one (Cg-II in Fig. 4) is the closest to *C. guineensis sensu stricto* as it shares both the same mitochondrial clade [CII in Fig. 3(b)] and ecological niche (coastal environments). This species, however, has close phylogenetic relationships with freshwater species such as *C. camerunensis* and *C. sp. aff. louka* 'Samou', justifying its taxonomic status as a valid species.

The two other lineages from Nigeria and Gabon, identified morphologically as *C. guineensis*, share the same clade with *C. rendalli* and *C. zillii* [CI within Fig. 3, see also Fig. 4(a)] and are not closely related to *C. guineensis*. *Coptodon rendalli* and *C. zillii* have the widest distributions among all species of *Coptodon*; whereas *C. zillii* is known from Morocco (Qinba & Mataame, 2009) to the Democratic Republic of Congo and the Middle East (Teugels & Thys van den Audenaerde, 2003). *Coptodon rendalli* is not naturally distributed in West Africa although it is very common in aquaculture and widely distributed due to anthropogenic activities (Lazard & Rognon, 1997). The CI clade also includes species with restricted distribution ranges including species belonging to Cameroonian species flocks. With the exception of these latter species, distribution ranges of *C. zillii* and *C. rendalli* appear to be allopatric [Fig. 5(b), (c)], suggesting that vicariant events relating to landscape evolution of African rivers certainly played an important role in the divergence and evolution of *Coptodon*.

In conclusion, this study sheds new lights on *C. guineensis*, which occurs along the West African shoreline as a complex of species, each with a narrow distribution range. More phylogenetic investigations are needed to describe the species diversity inside the *C. guineensis* complex, and determine the evolutionary history underlying the diversity of *Coptodon*. If ecological speciation occurred in *Coptodon*, as suggested by phylogenetic investigations of Cameroonian crater lake cichlids, vicariance is also an important evolutionary force that needs to be evaluated by further studies.

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Supporting Information

Supporting Information may be found in the online version of this paper:

Table SI. Cichlid samples used in *COI* and *CR* phylogenetic analyses. Sample code, species, location, collector name and GenBank accession number are noted

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