The colour pattern of the caudal fin, a useful criterion for identification of two species of *Tilapia* and their hybrids

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The present work reports on some chromatic characters and their change with size in two species of Tilapia (T. zillii and T. guineensis) and their first generation hybrids. Successful reciprocal hybridizations between T. zillii and T. guineensis were performed in concrete tanks. The hybrids obtained were viable. The colour patterns of the hybrids and their parents were registered during a rearing cycle of 12 months. Between 2 and 13 cm standard length ($L_{\rm S}$), the hybrids were found to be heterogeneous and three phenotypes were observed. The type (1) phenotype had a fully yellowish caudal fin without dots, the type (2) phenotype had a bicoloured caudal fin with the upper part clear yellowish and the lower part dark yellowish and without dots and the type (3) had a similar bicoloured caudal fin, but with dots. Above 13 cm L_8 , the hybrid population was homogenous and all specimens had a bicoloured caudal fin with dots. In T. zillii, all specimens >14 cm $L_{\rm S}$ had a greyish caudal fin with dots while all T. guineensis >13 cm $L_{\rm S}$ were characterized by a bicoloured caudal fin without dots. A multivariate analysis of the morphometric and meristic characters did not allow a clear separation of all groups. The study showed that the external morphology of the hybrids was intermediate to that of the parental species. © 2006 The Authors

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INTRODUCTION

The construction of hydroelectric or agricultural dams and bridges causes modifications of the aquatic environment and the behaviour of the fish species in these water bodies (Agnèse *et al.*, 1998). In the man-made Lake Ayame (Côte d'Ivoire), which originated as a result of the construction of a hydroelectric dam on the Bia River, a large population of hybrids of *Tilapia* species was found (Pouyaud, 1995). The artificial environment of Lake Ayame in which

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the parental stocks were trapped, apparently allowed a massive hybridization (Pouvaud, 1995; Agnèse et al., 1997). These hybrids originate from crosses between *Tilapia zillii* (Gervais), widely distributed in many West African coastal basins, and Tilapia guineensis (Bleeker) naturally found in the lower courses of the rivers of the same area (Teugels & Thys van den Audenaerde, 1992). Hybrids of species of *Tilapia* represent an important component of the commercial and subsistence fisheries in some Ivorian water bodies (Gourène et al., 1999). The characterization by morphological features of these hybrids of *Tilapia* species from Lake Ayame is notoriously problematic. Conversely, identification methods based on allozymes and molecular markers can distinguish hybrids from their parents, T. zillii and T. guineensis (Pouyaud, 1995). According to Teugels & Thys van den Audenaerde (1992), the parental species can easily be identified by their pharyngeal dentition. Unfortunately, checking this character cannot easily be done in the field. Recent reports (Tchinda, 1999; Bamba, 2001) on the morphometric characterization of the hybrids of *Tilapia* species also did not allow an easy identification of the Ayame hybrids. These problems prompted a further investigation of some morphological characters of these hybrids. The development of the colour pattern of the caudal fin of T. zillii, T. guineensis and their hybrids was examined from the juvenile [2 cm standard length (L_s)] to the adult stage (20 cm L_s) in live specimens.

MATERIALS AND METHODS

This study was conducted from December 2001 to January 2003, in concrete tanks in Aboisso (5°28' N; 3°12' W) and in floating cages in Lake Ayame (5°36' N; 3°10' W). The specimens of the parental species, *T. zillii* (n = 45) and *T. guineensis* (n = 45) were sampled in Lake Ayame. Both species were raised separately and produced offspring. In addition, four *T. zillii* males and four *T. guineensis* females were crossed to obtain hybrids (F1A) and also four *T. zillii* females and four *T. guineensis* males were crossed to obtain another set of hybrids (F1B).

COLOUR PATTERN OBSERVATIONS

The colour pattern of the caudal fin was recorded during a rearing cycle of 12 months: first in concrete tanks (Aboisso) and then in floating cages (Lake Ayame). Juvenile hybrids first raised in concrete tanks and having reached an average of 10 g were then put in floating cages for further growth. During this latter 3 month period, food was distributed three times daily (0900, 1200 and 1500 hours) and for 7 days a week. Fishes were fed 30% protein pelleted commercial feed. The feeding rate was adjusted every 2 weeks after growth control. The daily ration was calculated according to the formula of Mélard (1986): $R_{max} = 0.139 M^{0.685}$, where $R_{max} = maximum$ daily rate (g of food) and M = total fish mass (g). Nine hundred specimens each of *T. zillii*, *T. guineensis*, hybrid tilapias F1A and hybrid tilapias F1B were raised during 12 months. The development of the colour pattern on the caudal fin was followed from the juvenile to the adult stage. Every 28 days, the colour characteristics were recorded for specimens from 2 and 20 cm L_S.

MORPHOMETRIC AND MERISTIC STUDY

The following morphometric characters (Fig. 1) were considered, according to the methods of Teugels & Hanssens (1994): (1) L_8 , (2) head length, (3) snout length, (4)



FIG. 1. Schematic representation of the body measurements and head measurements: 1, L_s ; 2, head length; 3, snout length; 4, eye diameter; 5, interorbital distance; 6, preorbital bone length; 7, body depth at the origin of pelvic fin; 8, caudal peduncle depth; 9, caudal peduncle length; 10, predorsal length; 11, prepectoral length; 12, prepelvic length; 13, preanal length; 14, dorsal fin base length; 15, length of the longest dorsal spine; 16, pectoral fin length; 17, pelvic fin length; 18, anal fin base length; 19, last anal spine length; 20, pectoral fin to eye distance; 21, pectoral fin to pelvic fin depth (from the base of the ventral pectoral fin ray to the base of the first pelvic fin spine); 22, dorsal fin to anal fin distance; 23, anal fin to caudal peduncle distance; 24, dorsal fin to caudal peduncle distance; 25, pectoral fin to anal fin distance; 26, pelvic fin to anal fin distance; 27, pectoral fin to pelvic fin distance (from the base of the ventral pectoral fin ray to the base of the last pelvic fin ray); 28, body depth at origin of anal fin.

eye diameter, (5) interorbital distance, (6) preorbital bone length, (7) body depth at the origin of pelvic fin, (8) caudal peduncle depth, (9) caudal peduncle length, (10) predorsal length, (11) prepectoral length, (12) prepelvic length, (13) preanal length, (14) dorsal fin base length, (15) length of the longest dorsal spine, (16) pectoral fin length, (17) pelvic fin length, (18) anal fin base length and (19) last anal spine length. Nine additional morphometric measurements, not discussed by Teugels & Hanssens (1994) were included: (20) pectoral fin to eye distance, (21) pectoral fin to pelvic fin depth (from the base of the ventral pectoral fin ray to the base of the first pelvic fin spine), (22) dorsal fin to anal fin distance, (23) anal fin to caudal peduncle distance, (24) dorsal fin to caudal peduncle distance. (25) pectoral fin to anal fin distance. (26) pelvic fin to anal fin distance, (27) pectoral fin to pelvic fin distance (from the base of the ventral pectoral fin ray to the base of the last pelvic fin ray) and (28) body depth at origin of anal fin. Meristic counts, following Teugels & Hanssens (1994) were: scales around caudal peduncle (Fig. 2), number of anal fin spines, number of soft anal fin rays, number of dorsal fin spines, number of soft dorsal fin rays, number of scales in the upper lateral line series and number of scales in the lower lateral line series (Fig. 2). Two additional meristic counts, not discussed by Teugels & Hanssens (1994) were included: number of caudal fin rays and number of scales between the upper and lower lateral lines. Morphometric and meristic characters concerning pharyngeal teeth were omitted because,

700





in this study, only external morphological characters were considered. In total, 28 measurements and nine meristic characters were considered.

STATISTICAL ANALYSIS

The morphometric and meristic characters were analysed by principal component analyses (PCA) on ln-transformed data for the measurements (Schaefer, 1991) and the raw data for the counts. Ln-transformed morphometric measurements and meristic characters were considered in two separate analyses. Factors 2 (axis 1) and 3 (axis 2) were chosen and factor 1 were ignored, to eliminate the effect of specimens size. All variables were submitted to Mann–Whitney *U*-tests to check for significant differences (P < 0.05) between species or hybrids. Statistical analysis was done using the STATISTICA 5.0 programme.

RESULTS

COLOUR PATTERN OF THE CAUDAL FIN

For all fish reared, 360 observations were made per month giving a total of 3420 observations over the 12 months. Between 2 and 14 cm L_S , some specimens of *T. zillii* had a completely yellowish or greyish caudal fin without dots [Fig. 3(a)], but more fish developed a greyish caudal fin with dots of increasing size during development. Above 14 cm L_S , all *T. zillii* had greyish caudal fins with dots on the entire caudal fin [Fig. 3(b)].

As *T. guineensis* (Fig. 4) and hybrids (Fig. 5) grew, there was a greater tendency for them to show a bicolour caudal fin (clear yellowish dorsally and dark yellowish ventrally); this trend was seen in fish growing from 2 to 13 cm L_S . Above 13 cm L_S all specimens had bicolour caudal fins. *Tilapia guineensis* did not have any dots [Fig. 4(b)] and hybrids had dots on either the upper half [Fig. 5(b)] or entirely over the caudal fin [Fig. 5(c)].

MORPHOMETRIC AND MERISTIC CHARACTER ANALYSIS

In a PCA plot of the scores of the individuals on the first axis (morphometric characters), three groups can be observed (Fig. 6). Specimens of *T. zillii* are



FIG. 3. Colouration and dots of *Tilapia zillii* caudal fin: (a) dark yellowish caudal fin without dots (6.5 cm $L_{\rm S}$) and (b) greyish with dots on the entire caudal fin (16 cm $L_{\rm S}$).

present in the positive part of this axis and *T. guineensis* in the negative part of the same axis. Specimens of the third group (hybrids F1A and F1B) are between *T. zillii* and *T. guineensis*. Last anal spine length, anal fin to caudal peduncle distance and dorsal fin to caudal peduncle distance had the highest loadings in the PCA distributions of the four populations.

The same analysis on meristic characters shows three groups as a function of axis 1 (Fig. 7). Specimens of *T. zillii* are present mostly in the negative part and specimens of *T. guineensis* mostly in the positive part of the first axis, however, there is extensive overlap between the two groups. Hybrids (F1A and F1B) are both present in the positive and in the negative part of the same axis. The number of dorsal fin spines, the number of soft dorsal fin rays and the number of soft anal fin rays had the highest loading in these distributions.

All variables were submitted to a Mann–Whitney *U*-test. This test showed that the following variables had significant differences (P < 0.05) between F1A and F1B: dorsal fin to caudal peduncle distance, anal fin to caudal peduncle distance, last anal spine length, pectoral to pelvic fin depth and number of soft anal fin rays.

DISCUSSION

The genetic identity of *T. zillii* and *T. guineensis* has been checked by complete cytochrome b sequence studies (Falk, 2004). The individuals used in this study appear to be from pure strains. The feasibility of reciprocal cross-breeding



FIG. 4. Colouration and dots of *Tilapia guineensis* caudal fin: (a) yellowish caudal fin without dots (7.5 cm $L_{\rm S}$) and (b) caudal fin bicoloured (17 cm $L_{\rm S}$) with upper half clear yellowish and lower half dark yellowish, but without dots.

between *T. zillii* and *T. guineensis* in concrete tanks is demonstrated in the present study. Hybrids obtained are viable, as demonstrated by Pouyaud (1995) in samples from Lake Ayame.

The present study of the colour pattern of the caudal fin demonstrates that, between 2 and 14 cm $L_{\rm S}$, the *T. zillii* population is heterogeneous with respect to dots on the caudal fin because it shows the progressive ontogenetic appearance of these dots on the fin; >14 cm $L_{\rm S}$, the population is homogeneous and characterized by a greyish caudal fin with dots. This appears to be the final, adult, chromatic pattern and corresponds closely to that reported in earlier studies. Thys van den Audenaerde (1964) reported the caudal fin of *T. zillii* to be brownish with yellow dots. Trewavas (1982) noted that the caudal fin of *T. zillii* is usually covered by a grey network with pale interstices. According to Pouyaud (1995), the caudal fin generally presents white dots uniformly maculated on its entire surface.

The present study shows that *T. guineensis* is also heterogeneous, between 2 and 13 cm L_S , with respect to fin colouration because it shows a progressive ontogenetic change from a yellowish to a bicolour caudal fin; >13 cm L_S , all *T. guineensis* have a bicoloured caudal fin without dots. Other descriptions of the caudal fin of *T. guineensis* vary according to presence of coloured dots



FIG. 5. Colouration and dots of hybrids (F1A and F1B) caudal fin: (a) type 2 phenotype, caudal fin bicoloured with upper half clear yellowish and lower half dark yellowish, but without dots (8 cm L_S), (b) and (c) type 3 phenotype, caudal fin bicoloured with upper half clear yellowish and lower half dark yellowish with (b) dots on the upper half (16 cm L_S) or (c) on the entire fin (16 cm L_S).

on the central part of the fin. Contrary to the present results, Thys van den Audenaerde (1964) and Trewavas (1983) found specimens to have a greyish or brownish caudal fin with some light coloured dots on the central area; Loiselle (1969) and Pouyaud (1995) stated that specimens from Lake Ayame have dots on the upper half of caudal fin while the lower half is always yellow coloured. The present work, however, agrees with the reports of Teugels & Thys van den Audenaerde (1992) and Gourène *et al.* (1995). These authors characterise

704



FIG. 6. Principal component analysis (PCA) of morphometric characters of *Tilapia zillii* (Z), *Tilapia guineensis* (G) and hybrid tilapias F1A (A) and F1B (B), following axes 1 and 2.

T. guineensis by a bicoloured caudal fin (upper half yellowish and lower half dark yellowish) without dots. During the current study on specimens raised from the juvenile to the adult stage, where all modifications in the caudal fin pattern occurring during growth were observed, no specimens of *T. guineensis* were found to have dots on the caudal fin.

In the hybrid F1A and F1B populations, several colour patterns were observed on the caudal fin. In the younger group (between 2 and 13 cm L_S), the population is heterogeneous and three phenotypes are observed. The type (1) phenotype has a fully yellowish caudal fin without dots. This phenotype was also observed in specimens of *T. zillii* (between 2 and 14 cm L_S) and *T. guineensis* (between 2 and 13 cm L_S). It is similar to one population of hybrids (*T. zillii* × *T. guineensis*) described by Pouyaud (1995); however, no data on the size of the specimens were given by this author. The type (2) phenotype has a bicoloured caudal fin (upper half clear yellow and lower half dark yellow) without dots. This phenotype has a bicoloured caudal fin (upper half clear yellow and lower half dark yellow) without dots. The type (3) phenotype has a bicoloured caudal fin. The older group (>13 cm L_S) of F1A and F1B populations is homogeneous and characterized by the type (3) phenotype



FIG. 7. Principal component analysis (PCA) of meristic characters of *Tilapia zillii* (Z), *Tilapia guineensis* (G) and hybrid tilapias F1A (A) and F1B (B), following axes 1 and 2.

of the hybrids, a bicoloured caudal fin with dots. For some specimens, the dots are few or not visible on the lower half of caudal fin. This phenotype is also reported in the study of Pouyaud (1995) for another group of hybrids of *Tilapia* species (*T. zillii* \times *T. guineensis*). Again, no data on the size of these specimens were reported.

An intermediate morphology in all hybrid specimens was also observed by Legendre *et al.* (1992) with hybrid clariid catfishes from crosses between *Heter*-*obranchus longifilis* Valenciennes and *Clarias gariepinus* (Burchell). The present study shows that final colour patterns develop through ontogeny and shows the importance of describing parental species and their hybrids over as complete an ontogenetic range as possible.

The present paper confirms the fact that the two parental species (*T. zillii* and *T. guineensis*) are morphologically similar. Phylogenetic research using allozyme data supports the hypothesis of a close relationship (Pouyaud & Agnèse, 1995; Agnèse *et al.*, 1997). More recent cytochrome *b* sequence studies, however, indicate that *T. zillii* is the sister-species to *Tilapia rendalli* Boulanger and *T. guineensis* is sister to *Tilapia Busumana* Günther (Falk, 2004).

Principal component analysis of morphometric and meristic characters distinguish between *T. zillii* and *T. guineensis* with respect to the last anal spine length, anal fin to caudal peduncle distance, dorsal fin to caudal peduncle distance, number of dorsal fin spines, number of soft dorsal fin rays and number of soft anal fin rays.

From the present work, an identification key of the parental species and their hybrids could be made, based on the colour pattern of the caudal fin.

SPECIES KEY (SPECIMENS WITH L_S >14 CM)

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