Nothobranchius oestergaardi (Cyprinodontiformes: Nothobranchiidae), a new annual killifish from Mweru Wantipa Lake drainage basin, northern Zambia

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Abstract

A new species of an annual killifish, *Nothobranchius oestergaardi*, is described based on specimens collected from an ephemeral pool in the swamps of the seasonal Mwawe River in the drainage system of the Mweru Wantipa Lake basin, northern Zambia. The new species belongs to the *Nothobranchius taeniopygus* species group and is distinguished from the other members by a diagnostic combination of male colouration and morphological characters.

Zusammenfassung

Eine neue Art saisonaler Killifische: *Nothobranchius oestergaardi*, wird auf der Grundlage von Exemplaren beschrieben, die in einem ephemeren Tümpel in den Sümpfen des saisonalen Mwawe-Flusses im Einzugsgebiet des Mweru-Wantipa-Sees im nördlichen Sambia gefunden wurden. Die neue Art gehört zur *Nothobranchius-taeniopygus*-Artengruppe und unterscheidet sich von anderen Arten dieser Gruppe durch verschiedene morphologische Merkmale und die Farbgebung des Männchens.

Résumé

Une nouvelle espèce de killie annuel, *Nothobranchius oestergaardi*, est décrite sur la base de spécimens collectés dans une mare temporaire, située dans les marécages de la rivière saisonnière Mwawe, du bassin versant du Lac Mweru Wantipa, au nord de la Zambie. Cette nouvelle espèce appartient au groupe d'espèces *Nothobranchius taeniopygus* et se distingue de ses autres membres par une combinaison diagnostique de coloration mâle et de caractères morphologiques.

Sommario

Una nuova specie di killifish annuale, *Nothobranchius oestergaardi*, è descritta sulla base di esemplari raccolti da una pozza temporanea nelle paludi del fiume stagionale Mwawe, bacino del Mweru Wantipa Lake, Zambia settentrionale. La nuova specie appartiene al gruppo di specie *Nothobranchius taeniopygus* e si distingue dagli altri membri per una combinazione diagnostica di colorazione del maschio e di caratteri morfologici.

INTRODUCTION

The killifish genus *Nothobranchius* occurs in the sub-tropical and tropical part of eastern Africa with a wide distribution from Sudan to South Africa, and from Chad to Ethiopia and the islands Zanzibar and Mafia in Tanzania (Wildekamp 2004). All known species are annual fishes, living in temporary pools and swamps formed during the rainy season (Seegers 1997; Skelton 2001).

The *Nothobranchius* species known in Zambia were previously placed in the *N. brieni* species group (Valdesalici 2010) however this group of species has now been re-assigned to the *N. taeniopygus* group, synapomorphies which are important as diagnostic characters for this group are: caudal and anal fin with a broad light sub-marginal band and dark margin in males. Four species are currently known from this country: *N. kafuensis* Wildekamp & Rosenstock from the Kafue River and upper Zambesi River basins, *N. rosenstocki* Valdesalici & Wildekamp and *N. symoensi* Wildekamp from the Luapula River basin, and *N. boklundi* Valdesalici from Luangwa River basin. In the Democratic Republic of Congo *N. brieni* Poll is known from the Lualaba River basin, *N. hassoni* Valdesalici & Wildekamp, *N. polli* Wildekamp from Lufira River basin and *N. malaissei* Wildekamp from Luapula River basin. In Tanzania *N. taeniopygus* Hilgendorf occurs and in Chad *N. rubroreticulatus* Blache & Miton is known from the Lake Chad drainage and Bahr Salamat River basin (Wildekamp 2004; Valdesalici & Wildekamp 2005; Watters 2005; Valdesalici 2010).

Table I. Morphometric data of *Nothobranchius oestergaardi*. All measurements are presented as percentages of standard length, except eye diameter and snout length are as percentages of head length; standard length in mm.

	Holotype	All males (n = 5)	Females (n = 1)
Standard length	31.8	26.4-42.7	26.0
Depth at pelvic fins	32.3	28.0-32.3	28.4
Predorsal length	57.5	57.5-62.5	66.1
Preanal length	56.2	56.2-64.4	60.7
Prepelvic length	49.6	49.6-52.3	50.0
Caudal peduncle length	21.3	17.0-21.3	18.4
Caudal peduncle depth	16.0	13.6-16.0	12.6
Dorsal fin base length	25.4	24.5-26.5	23.0
Anal fin base length	22.0	21.3-24.2	18.8
Head length	31.4	30.2-35.2	31.1
Snout length	31.0	21.5-31.0	27.1
Eye diameter	31.0	25.8-31.0	32.0

On 16 February 2010, the authors together with Jørn Boklund and Kaj Østergaard, during a fish species survey, collected *Nothobranchius* specimens from an ephemeral pool in the swamp of the seasonal Mwawe river, in the drainage system of the Mweru Wantipa Lake basin. These specimens turned out to be an undescribed species of *Nothobranchius*. This species belongs to the *N. taeniopygus* species-group but differs by male colouration and morphological characters from all other members of this complex and is herein described as *Nothobranchius oestergaardi*, new species.

MATERIALS AND METHODS

Measurements and counts were taken as described in Amiet (1987), Huber (1992) and Valdesalici (2010). Measurements were made with a digital calliper, partly under a dissecting microscope, and rounded to the nearest 0.1 mm. All measurements are presented as percentages of standard length (SL), except for eye diameter and snout length, which are given as a percentage of head length (HL). Terminology for the cephalic neuromast series follows Scheel (1968), and that for the frontal squamation as described in Hoedeman (1958). Osteological preparations (C&S) were made according to Taylor and Van Dyke (1985), but were not stained for cartilage.

Type material, additional and comparative material is deposited in the following institutions: Natural History Museum (BMNH), London; Muséum National d'Histoire Naturelle (MNHN), Paris; Royal Museum for Central Africa (MRAC), Tervuren; Museo Civico di Storia Naturale "Giacomo Doria" (MSNG), Genova; and South African Institute for Aquatic Biodiversity (SAIAB), Grahamstown.

Comparative material examined is listed in: Valdesalici (2010); data used also for comparisons in Wildekamp (1990); *Nothobranchius* aff. n*eumanni*, (Hilgendorf) MSNG 56046, 1 male, C&S 57.5 mm SL Tanzania, Malagarasi River basin (5°59'1" S, 32°48'5" E); MSNG 56045, 2 males, 36.6 & 38.1 mm SL Tanzania, Malagarasi River basin (5°57'49" S, 32°46'50" E); MRAC 2010-33-P-2-3, 2 males, 42.5 & 44.3 mm SL, Tanzania, Malagarasi River basin (5°54'54" S, 32°45'54" E); *Nothobranchius rubroreticulatus* Blache & Miton MNHN 59232, 1 female 33.9 mm SL, 2 males 31.0 & 39.0 mm SL; MNHN 59235, 2 males 20.4 & 28.1 mm SL, Chad, Lake Chad drainage (12°7' N, 15°3' E).

Nothobranchius oestergaardi n. sp.

(Figs 1-4, Table I)

Holotype: BMNH 2010.12.6.1, male, 31.8 mm SL; Zambia, Northern Province, ephemeral pool in the swamp of the seasonal Mwawe River, Mweru Wantipa Lake drainage basin, about five km west of Kalaba village, south side of the road, 8°25.300' S 29°50.474' E, collected by Giuseppe Amato, Jørn Boklund, Kaj Østergaard and Stefano Valdesalici, 16 February 2010.

Paratypes: MRAC 2010-33-P-4, 1 male, 30.4.0 mm SL; MSNG 56047A-B-C,1 male, 42.7 mm SL (preserved after seven months in captivity), 1 female, 26.0 mm SL; 1 male 29.0 mm SL, C&S; SAIAB 98224, 1 male, 26.4 mm SL; all collected with holotype.

Diagnosis: Males similar to other members of *N. taeniopygus* species-group, differing from all other species of *Nothobranchius* by caudal and anal fins having broad pale sub-marginal band and dark margin. *Nothobranchius oestergaardi* differing from members of *N. taeniopygus* species-group by following combination of characters: proximal portion of caudal fin plain red (vs. proximally irregularly spotted in *N. boklundi*, *N. hassoni*, *N. kafuensis*, *N. polli*, *N. rosenstocki*, *N. symoensi* and *N.taenipygus* or caudal fin covered with spots in *N. brieni* and *N. malaissei);* dorsal fin without coloured margin (vs. black fin margin in *N. rubroreticulatus* or broad light blue fin margin in *N. brieni*, *N. polli* and *N. symoensi)*, body without

Fig. 1. *Nothobranchius oestergaardi*, wild adult male, from the type locality, not preserved. Photo by A. Persson

Fig. 2. *Nothobranchius oestergaardi*, also a wild adult male, from the type locality, not preserved. Photo by S. Valdesalici

Fig. 3. *Nothobranchius oestergaardi*, wild adult female, from the type locality, not preserved. Photo by S. Valdesalici

distinct black spots anteriorly (vs. black spots present in *N. rosenstocki* and *N. taeniopygus);* anal fin striped (vs. anal fin irregularly striped in *N. hassoni* and *N. polli*, spots concentrated on proximal portion in *N. brieni*, *N. kafuensis*, and *N. malaissei*, or anal fin completely marbled in *N. rosenstocki* and *N. symoensi),* distal portion of anal fin cream then light blue (vs. yellow in *N. boklundi*, *N. polli,* pale yellow to yellow white in *N. taeniopygus*, light blue or orange in *N. kafuensis*, light blue in *N. rubroreticulatus* and red in *N. rosenstocki)* and with broad dark brown border (vs. narrow dark border in *N. boklundi*, *N. brieni*, *N. kafuensis*, *N. polli*, or light blue margin in *N. symoensi);* distal portion of pelvic fin plain light blue (vs. spotted in *N. boklundi*, *N. brieni*, *N. kafuensis* and *N. polli*, plain red in *N. rosenstocki*, black in *N. rubroreticulatus* and *N. taeniopygus). Nothobranchius oestergaardi* males with relatively shorter caudal peduncle compared to *N. boklundi* (17.0-21.3 vs. 19.1-26.6 % SL); shallower and longer caudal peduncle compared to *N. rosenstocki* (13.6-16.0 vs. 10.1-12.9 % SL and 17.0-21.3 vs. 12.5-14.5 % SL respectively); shallower caudal peduncle compared to *N. symoensi* (13.6-16.0 vs. 12.8 % SL); longer predorsal length compared to *N. hassoni* (57.5-62.5 vs. 52.8-56.4 % SL), *N. rosenstocki* (57.5-62.5 vs. 48.3-52.9 % SL), *N. rubroreticulatus* (57.5-62.5 vs. 53.3-57.6 % SL) and *N. symoensi* (57.5-62.5 vs. 55.1 % SL); longer prepelvic length compared to *N. brieni* (49.6-52.3 vs. 43.3-45.2 % SL), *N. taeniopygus* (49.6-52.3 vs. 42.1-48.9 % SL) and *N. symoensi* (49.6-52.3 vs. 48.2 % SL); and longer preanal length compared to *N. brieni* (56.2-64.4 vs. 52.9-54.5 % SL).

Description: See Figures 1-3 for overall appearance and Table I for morphometric data of the type series.

Robust *Nothobranchius* with deep and compressed body, maximum observed length in males 31.8 mm SL. Dorsal profile slightly concave to nearly straight on head, convex from nape to end of dorsal fin base. Ventral profile convex, slightly concave to nearly straight on caudal peduncle posterior to dorsal and anal fin. Snout slightly pointed, mouth directed upwards, lower jaw longer than upper, posterior end of rictus at same level as or slightly above centre of eye. Branchiostegal membrane projecting posteriorly from opercle.

Dorsal and anal fins located posterior to midbody, rounded, tips with short filamentous rays, dorsal fin tip reaching caudal fin. Both fins with papillate contact organs on fin rays. Dorsal fin rays 16-17; anal fin rays 16-17. Pectoral fin approximately triangular, tip reaching pelvic fin. Pelvic fin short, separated at base and tip reaching urogenital papilla. Caudal fin subtruncate.

Female smaller than male, maximum observed size 26.0 mm SL. Dorsal fin rounded. Anal fin triangular with rounded tip. Dorsal fin positioned more posteriorly (57.5-62.5 vs. 66.1% SL) compared to males. Branchiostegal membrane not projecting from opercle.

Scales cycloid, body and head entirely scaled, except for ventral surface of head. Scales in median lateral series 25-29 + 3-4 on caudal fin base. Transverse row of scales 11. Circumpeduncular scales 12. Cephalic squamation pattern variable, some specimens presenting irregular G-type. Anterior neuromast series 'open' type. Central supra-orbital

Fig. 4. *Nothobranchius oestergaardi*, BMNH 2010.12.6.1, male, holotype, 31.8 mm SL. Copyright The Natural History Museum, London.

Table II. Comparison of the morphometric values for male *Nothobranchius oestergaardi* and members of the *N. taeniopygus* species-group. Abbreviations in the table: SL = Standard length, BD = Depth at pelvic fins, PD = Predorsal length, PA = Pre-anal length, PP = Prepelvic length, CPL = Caudal peduncle length, CPD = Caudal peduncle depth, HL = Head length, SL = Snout length, ED = Eye diameter, D = number of dorsal fin rays, A = number of anal fin rays, LL = scales on lateral series. All measurements are presented as percentages of standard length, except eye diameter and snout length are as percentages of head length; standard length in mm.

Species (number of specimens)	SL	BD	PD	PA	PP	CPL	CPD	HL	SL	ED	D	A	LL
N. boklundi (6)	35.4 43.8	28.5 33.5	50.8 60.5	59.8 65.7	48.0 56.8	19.1 26.6	13.4 16.0	31.3 35.3	22.0 29.4	22.7 25.1	16 17	17 18	28 30
N. brieni (4)	33.0 47.0	30.0 32.7	55.0 58.9	52.9 54.5	43.3 45.2	16.4 19.5	13.9 14.5	32.8 33.6	26.6 27.9	24.6 28.8	16 –	16 18	28 29
N. hassoni (12)	27.0 41.6	28.6 33.0	52.8 56.4	54.8 61.8	39.7 56.5	16.0 20.0	11.7 15.5	29.7 35.3	20.2 26.9	25.7 33.0	15 17	15 17	16 30
N. kafuensis (13)	27.9 50.2	26.9 32.5	54.9 60.6	55.2 63.9	41.9 52.3	16.52 1.2	11.3 14.7	28.0 21.2	20.2 26.1	24.6 28.5	16 18	16 19	26 31
N. malaissei (7)	27.5 39	28.4 34.8	52.6 61.3	57.2 64.0	42.9 53.1	17.4 19.6	12.7 15.3	32.7 35.0	22.5 28.0	24.0 16.0	16 18	16 18	28 30
N. polli (8)	20.5 39.5	26.8 33.2	53.2 63.4	56.7 68.3	49.4 57.1	15.2 20.0	12.1 14.1	29.9 34.9	19.7 29.0	24.2 28.1	15 18	15 18	26 28
N. rosenstocki (3)	23.0 36.9	26.0 28.7	48.3 52.9	55.6 58.6	46.9 50.2	12.5 14.5	10.1 12.9	31.4 32.5	20.6 24.2	27.7 34.2	15 17	16 17	28 30
N. rubroreticulatus (3)	28.1 39.0	25.4 29.7	53.3 57.6	55.1 59.3	46.1 5.2	19.8 21.6	13.2 14.3	31.6 33.3	23.0 30.8	23.3 25.2	16 18	18 19	27 30
N. symoensi (1)	30.5	30.4	55.1	59.7	48.2	19.3	12.8	30.8	24.4	25.5	18	17	29
N. taeniopygus (42)	28.0 39.8	26.7 36.4	51.3 60.2	52.7 60.7	42.1 48.9	18.1 23.9	11.5 15.6	27.6 34.1	20.3 25.8	24.8 29.6	14 17	16 19	28 32
N. oestergaardi (5)	26.4 42.7	28.0 32.3	57.5 62.5	56.2 64.4	49.6 52.3	17.0 21.3	13.6 16.0	30.2 35.2	21.5 31.0	25.8 31.0	16 17	16 17	25 29

series separate in shallow grooves, each with two neuromasts. Posterior cephalic neuromast series curved with 2 (rarely 3) neuromasts. One neuromast on each scale of median longitudinal series. Basihyal bone sub-triangular. Six branchiostegal rays. Vomerine teeth present in small patch. Lateral process of post-temporal short. Second pharyngobranchial with 1 tooth. Interarcual cartilagine short. Single anterodorsal process of urohyal. Total vertebrae 27. Premaxilla and dentary with many irregularly distributed unicuspid, slightly curved teeth of different sizes and small number of larger teeth in outer row of upper and lower jaws.

Colour in life: Males (Fig. 1-2). Body and head scales light blue with broad dark red margin, creating reticulated pattern on body and head. Snout, frontal and dorsal portion of head red. Branchiostegal membrane whitish. Dorsal fin grey green to grey yellow with irregular rows of dark red small spots and dots; spots somewhat horizontally coalesced proximally, medially elongated on fin rays, distally denser and smaller. Anal fin with cream proximal portion with few red irregular spots, followed by red-brown band, a wider cream band becoming light blue distally and wide dark red-brown margin. Pelvic fins light blue, red-brown near base. Pectoral fins hyaline with light blue margins. Caudal fin opaque red-brown, with light blue sub-distal narrow band and dark red-brown, wider margin. Iris golden, with faint black vertical bar through centre of eye.

Female (Fig. 3). Body and head scales pale brown, with golden iridescence on scale centre. Opercular region silver to golden. Abdomen silver to golden.

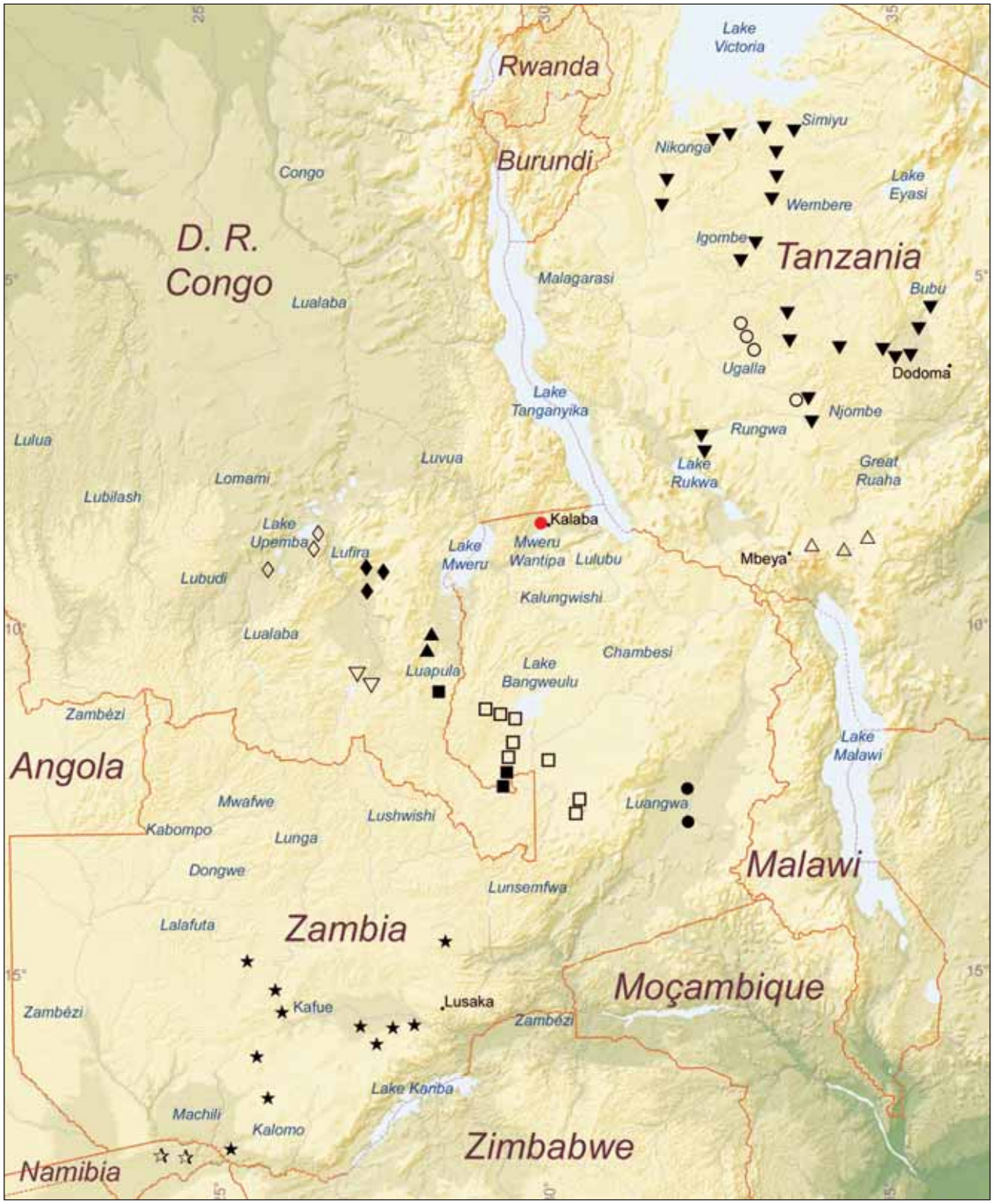

Fig. 5. Geographic distribution of *Nothobranchius oestergaardi* (red circle), *N. boklundi* (filled circle), *N. brieni* (open lozenge), *N. hassoni* (filled lozenge), *N. kafuensis* (filled star), *N. malaissei* (triangle), *N.* aff. *neumanni* "Malagarasi" (open circle), *N.* aff. *neumanni* "Mbeya" (open triangle), *N. rosenstocki* (open square), *N. symoensi* (filled square), *N.* species "Caprivi Strip" (open star) and *N. taeniopygus* (inverted triangle).

Fig. 6. The type locality of *Nothobranchius oestergaardi*, an ephemeral pool in the swamps of the seasonal river Mwawe, Mweru Wantipa Lake drainage basin, Zambia, Northern province. Photo by K. Østergaard (top). Photo by S. Valdesalici (above)

All fins hyaline. Iris golden, with faint black vertical bar through centre of eye.

Colour in alcohol: Male (Fig. 4). Body scales light brown, all scales with distinct wide dark red irregular margin. Dorsal fin light brown with dark red to brown spots. Anal fin pale white proximally with dark red to brown spots, followed by brown stripe, pale cream distal area and pale dark red to brown margin. Pelvic fins light brown with dark red spots near base. Pectoral fins hyaline. Caudal fin dark red, with distinct white sub-marginal band and wide dark brown margin. Iris grey-blue.

Female. Body light brown. Opercular and ventral area more cream-yellow. Unpaired and paired fins light brown. Iris grey-blue.

Distribution: (Fig. 5) This species is currently only known from an ephemeral pool in the swamps of the seasonal Mwawe River of the drainage system of the Mweru Wantipa Lake basin, northern Zambia.

Habitat notes: (Fig. 6) The type locality was at the time of collection a pond about 15 m in diameter and circa 1.5 m deep at its centre, extending on one side into the grass for about further 10 m and with a depth of 0.5 m. The aquatic vegetation consisted of a *Nymphea* species. The water was light clay grey and turbid, pH 6.1 (measured around 11:00 hrs), conductivity 207 µS/cm. Other fish collected at the type locality were a juvenile *Ctenopoma*, an unidentified haplochromine, *Barbus neefi* Greenwood, and *Lacustricola moeruensis* Boulenger.

Etymology: The species name is dedicated to the collector and friend Kaj Østergaard, Denmark, for his contributions over a long period of time on field investigations that have led to the discovery of many new populations of *Nothobranchius* species in numerous countries.

Key to the *Nothobranchius taeniopygus* species-group based on live colour pattern characters

1a. Black scale margins .... *Nothobranchius taeniopygus*
1b. Red scale margins..... 2
2a. Light blue sub-distal band on caudal fin 5
2b. Light blue distal margin on caudal fin 4
2c. Orange sub-distal band on caudal fin..... 3
3a. Orange or cream sub-distal band on anal fin... ..... *Nothobranchius kafuensis*
3b. Anal fin medially yellow..... ..... *Nothobranchius boklundi*
4a. Anal fin with light blue margin reduced to a light triangle in the lower corner..... ..... *Nothobranchius brieni*
4b. Anal fin with light blue margin..... ..... *Nothobranchius symoensi*
4c. Anal fin with red margin..... ..... *Nothobranchius rosenstocki*
5a. Light blue sub-distal band on anal fin..... ..... *Nothobranchius kafuensis*
5b. Cream and light blue sub-distal band on anal fin ..... *Nothobranchius oestergardi*
5c. Light blue sub-distal band on anal and dorsal fin ..... *Nothobranchius rubroreticulatus*
5d. Anal fin medially yellow..... 6
6a. Dorsal fin completely spotted .... *Nothobranchius boklundi*
6b. Dorsal fin with light blue margin..... ..... *Nothobranchius polli*
6c. Caudal fin medially yellow..... ..... *Nothobranchius hassoni*
6d. Caudal fin completely spotted..... ..... *Nothobranchius malaissei*

Discussion: *Nothobranchius oestergaardi* is a member of the *N. taeniopygus* species-group, presenting all the diagnostic features of this complex defined above. This group can be clearly separated from *N. neumanni* (Hilgendorf) *s.l.*, including the species known on western Tanzania on the Malagarasi River drainage and near Mbeya, by the unique combination of male colouration patterns and morphology. In particular, male *N. oestergaardi* can be clearly separated by the closest *Nothobranchius* aff. *neumanni* from Malagarasi River drainage, by having a uniform reticulated pattern on body and head (vs irregular pattern), the caudal fin with a broad dark red margin (vs narrow black margin); anal fin proximally spotted and cream coloured (vs. plain and grey-blue anal fin), with a sub-proximal stripe (vs. with two median irregular stripes), subdistally cream (vs. yellow subdistally) and with a broad dark red margin (vs. narrow black margin); additionally *N. oestergaardi* has a shorter predorsal length 57.5-62.5 % SL (vs. 64.2-68.8%), reduced scale number in the median lateral series 25-29 (vs. 28-34). Osteologically, the only diagnostic differences (tentative, due to the limited numbers of specimens studied) are the lateral process of the post-temporal being short (vs. longer) and a reduced number of vertebrae 27 (vs. 31). *Nothobranchius neumanni*, including the Malagarasi River drainage populations, was diagnosed in Wildekamp (1990,

2004) by its straight dorsal profile and marked, angular transition between head and body along the dorsal profile. The dorsal profile is variable in this species and also within the genus.

Members of the *N. taeniopygus* species-group present a more compressed body when compared to *N. neumanni* (Hilgendorf) *s.l. Nothobranchius rubroreticulatus* and the other members of the *N. taeniopygus* species-group are presently separated by a distance of more than 3500 km. This is a surprising result and might be explained by a paleo-link (paleo-Congo) between the Zambesi drainage basin and the Lake Chad drainage basin (Stewart 2001; Goudie 2005). Additionally, the hypothesis provided by Stankiewicz & de Wit (2006) of a Paleocene connection between the eastern Congo with paleo-Rufiji and southern Congo with the paleo-Zambezi gives connection to all the known members of the *N. teaniopygus* species-group. This results differs from the report by Shidlovskiy et al. (2010).

Further research in the area of the north-east Democratic Republic of Congo and in Central African Republic may reveal other members of the *Nothobranchius taeniopygus* species-group.

At present the *Nothobranchius* species from Caprivi Strip of Namibia (Wildekamp 2004) remain the least-known species of the *N. taeniopygus* species-group.

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REFERENCES

AMIET, J. L. 1987. *Faune du Cameroun. Fauna of Cameroon. Vol.2. Le genre* Aphyosemion *Myers (Pisces: Teleostei: Cyprinodontiformes).* Sciences Naturales, Compiègne, 262 pp.

GOUDIE A. S. 2005. The drainage of Africa since the Cretaceous. *Geomorphology* 67: 437-456.

HOEDEMAN, J. J. 1958. The frontal scalation pattern in some groups of tooth carps (Pisces, Cyprinodontiformes). *Bulletin of Aquatic Biology* 1: 23-28.

HUBER, J. H. 1992. *Review of* Rivulus. *Ecobiogeography - Relationships*. Cybium, Paris, 586 pp.

SCHEEL, J. J. 1968. *Rivulins of the Old World*. Tropical Fish Hobbyist Publication, Neptune City, 480 pp.

SEEGERS, L. 1997. *Killifishes of the World. Old World Killis II*. Verlag A.C.S. Mörfelden-Walldorf, 112 pp.

SHIDLOVSKIY, K. M., WATTERS, B. R. & WILDEKAMP, R. H. 2010. Notes on the annual killifish species *Nothobranchius rachovii* (Cyprinodontiformes; Nothobranchiidae) with the description of two new species. *Zootaxa* 2724: 37-57.

SKELTON, P. 2001. *A Complete Guide to the Freshwater Fishes of Southern Africa*. Struik Publishers, Cape Town, South Africa, 395 pp.

STANKIEWICZ, J. & DE WIT, M. J. 2006. A proposed drainage evolution model for Central Africa – Did the Congo flow east? *Journal of African Earth Sciences* 44: 75-84.

STEWART K. M. 2001. The freshwater fish of Neogene Africa (Miocene–Pleistocene): systematics and biogeography. *Fish and Fisheries* 2 : 177-230.

TAYLOR, W. R. & VAN DYKE, G. C. 1985. Revised procedures for staining and clearing small fishes and other vertebrates for bone and cartilage study. *Cybium* 9: 107-109.

VALDESALICI, S. 2010. *Nothobranchius boklundi* (Cyprinodontiformes: Nothobranchiidae): a new annual killifish with two male colour morphs from the Luangwa River basin, Zambia. *aqua, International Journal of Ichthyology* 16 (2): 51-60.

VALDESALICI, S. & WILDEKAMP, R. H. 2005. A new species of the genus *Nothobranchius* (Cyprinodontiformes: Nothobranchiidae) from Luapula River basin, Zambia. *aqua, International Journal of Ichthyology* 9 (3): 89-96.

WATTERS, B. R. 2005. *Nothobranchius rubroreticulatus* Blache and Miton, 1960. *Journal of the American Killifish Association* 38: 206-208.

WILDEKAMP, R. H. 2004. *A World of Killies. Atlas of the Oviparous Cyprinodontiform Fishes of the World. Volume 4*. The American Killifish Association, Elyria, 398 pp.