



## Three new *Troglodytella* and a new *Gorilloflasca* ciliates (Entodiniomorphida) from mountain gorillas (*Gorilla beringei beringei*) in Rwanda

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Received 24 March 2018; received in revised form 12 May 2018; accepted 14 May 2018

Available online 18 May 2018

### Abstract

Four new species, *Troglodytella gracilis*, *T. virunga*, *T. dolium*, and *Gorilloflasca longior* were described from the mountain gorillas in Rwanda. The three *Troglodytella* species share a retractable adoral ciliary zone, four non-retractable ciliary arches, two broad skeletal plates, a skeletal rod plate, numerous longitudinal cortical grooves, and four contractile vacuoles. The anterior ciliary arch is the longest of the four arches, extending transversely on the left body surface. *T. gracilis* and *T. virunga* are characterized by an elongated body, a tail flap, and a wedge-shaped macronucleus. *T. virunga* can be distinguished from *T. gracilis* by a trifurcate dorsal lobe, three right lobes, and two ventral spines. *T. dolium* has a barrel-shaped body and a rod-shaped or boomerang-shaped macronucleus. The buccal infraciliature of the three *Troglodytella* is composed of an adoral polybrachykinety, a perivestibular polybrachykinety, and paralabial kineties. *G. longior* has a long flask-shaped body, a vestibulum, a frontal lobe, a posterior cavity, an ellipsoidal or peanut-shaped macronucleus, a contractile vacuole, non-retractable adoral and vestibular ciliary zones in buccal area, a cavity ciliary zone, and two longitudinal dorsal ciliary zones. The infraciliature of *G. longior* is the same as that of *G. africana*.

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**Keywords:** Apes; *Gorilloflasca longior* n. sp.; *Troglodytella dolium* n. sp.; *Troglodytella gracilis* n. sp.; *Troglodytella virunga* n. sp.

**Abbreviations:** ACA, anterior ciliary arch; ACZ, adoral ciliary zone; AP, adoral polybrachykinety; CCZ, cavity ciliary zone; CA, cavity opening; CO, contractile vacuole; CP, cytoproct; DL, dorsal lobe; FL, frontal lobe; LCZ, left longitudinal ciliary zone; LP, left longitudinal polybrachykinety; LS, left skeletal plate; MA, macronucleus; MDA, middle dorsal ciliary arch; MI, micronucleus; PA, polybrachykinety of anterior arch; PC, posterior cavity; PCP, posterior cavity polybrachykinety; PDA, posterior dorsal ciliary arch; PK, paralabial kineties; PMD, polybrachykinety of middle dorsal arch; PPD, polybrachykinety of posterior dorsal arch; PPV, polybrachykinety of posterior ventral arch; PVA, posterior ventral ciliary arch; PVP, perivestibular polybrachykinety; RCZ, right longitudinal ciliary zone; RE, rectangles divided by grooves; RL, right lobes; RS, right skeletal plate; RP, right longitudinal polybrachykinety; SR, skeletal rod plate; TF, tail flap; VCZ, vestibular ciliary zone; VF, vestibular fiber; VK, vestibular kinety band; VL, ventral spines; VS, vestibulum.

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<https://doi.org/10.1016/j.ejop.2018.05.002>

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## Introduction

Ciliates in the order Entodiniomorpha inhabiting the intestines of primates, such as western and eastern gorillas (*Gorilla gorilla* and *Gorilla beringei*), chimpanzees (*Pan troglodytes*), bonobos (*Pan paniscus*), orangutans (*Pongo pygmaeus*), siamangs (*Hylobates syndactylus*), pygmy siamangs (*Hylobates klossi*), and guinea baboons (*Papio hamadryas*), have been found and ciliates in five genera, *Gorillofascia*, *Gorillophilus*, *Prototapirella*, *Troglodytes*, and *Troglodytella*, have been described (Ashford et al. 2000; Bonhomme et al. 1989; Brumpt and Joyeux 1912; Dollinger and Rüedi 1974; Freeman et al. 2004; Gillespie et al. 2010; Goussard et al. 1983; Hasegawa et al. 1983; Howells et al. 2011; Imai et al. 1991; Irbis et al. 2008; Ito et al. 2016, 2017; Landsoud-Soukate et al. 1995; Modrý et al. 2009; Mortelmans et al. 1971; Murray et al. 2000; Myers and Kuntz 1972; O'Donoghue et al. 1993; Pomajbíková et al. 2012; Reichenow 1917, 1920; Tokiwa et al. 2010).

We examined entodiniomorphid ciliates in the feces of wild habituated Virunga mountain gorillas (*Gorilla beringei beringei*) monitored by the Dian Fossey Gorilla Fund's Karisoke Research Center in Rwanda. We described *Prototapirella fosseyi* and *P. rwanda*, redescribed *P. gorillae* (Ito et al. 2016), and established a new genus *Gorillofascia* for *G. africana* (Ito et al. 2017). In the same samples, three new *Troglodytella* species and another new *Gorillofascia* species were discovered. Until now, three *Troglodytella* species, including *T. abressarti* Brumpt & Joyeux, 1912, *T. gorillae* Reichenow, 1917, and *T. gabonensis* Imai, Ikeda, Collet, & Bonhomme, 1991, have been reported from anthropoid primates and one *Gorillofascia* species, *G. africana* Ito, Eckardt, Stoinski, Gillespie, & Tokiwa, 2017, has been found in Virunga mountain gorillas. The infraciliature of *Gorillofascia* was described (Ito et al. 2017), while the buccal infraciliature of *Troglodytella* has never been examined. This report will focus on the description of *Troglodytella gracilis* n. sp., *T. virunga* n. sp., *T. dolium* n. sp., and *Gorillofascia longior* n. sp., and discuss the morphology of the genera *Troglodytella* and *Gorillofascia* based on the general and infraciliary morphology of these new species.

## Material and Methods

From August 2011 to August 2012, 2601 fecal samples were obtained from 120 wild habituated Virunga mountain gorilla (*Gorilla beringei beringei*) in the Volcanoes National Park, Rwanda. All gorillas are individually known and monitored daily by the field staff of the Karisoke Research Center. These fecal contents were fixed in approximately two times the volume of 10% formalin solution and transported to Emory University for microscopic examination of gastrointestinal parasites and commensals after their isolation using flotation and sedimentation techniques (Gillespie 2006). This

initial examination revealed an unexpected high diversity of ciliates. A selection of 40 preserved fecal samples collected during four different seasons with individuals of all identified ciliate types were sent to the Laboratory of the Veterinary Pathology in Japan for further examination. The infraciliary bands were stained by the pyridinated silver carbonate impregnation method, following Ito and Imai (1998). The orientation of ciliates used by Dogiel (1927) was adopted to *Troglodytella* species; the side beneath which the macronucleus lies was termed the dorsal side; the opposite one the ventral side; defining the right and left sides. The orientation of ciliates used by Ito et al. (2017) was adopted to *Gorillofascia* species; the side from which the frontal lobe arises was termed the dorsal side; the opposite one the ventral side; defining the right and left sides. Body measurements were taken from a sample of 20 fixed cells using a calibrated micrometer. Body length was determined as the distance between the anterior and posterior ends of the body. The term, polybrachykinety, refers to infraciliary bands composed of numerous, short, parallel kineties (Fernández-Galiano et al. 1985; Ito and Imai 1998).

## Results

Four new entodiniomorphid species were found in the fecal samples of the Virunga mountain gorillas in Rwanda.

Three of those species were described as members of the genus *Troglodytella* and named *T. gracilis* n. sp., *T. virunga* n. sp., and *T. dolium* n. sp. They had the morphological characteristics of *Troglodytella*: a retractable adoral ciliary zone, four non-retractable somatic ciliary arches, a long anterior ciliary arch, elongated rectangles divided by deep cortical grooves, a right broad skeletal plate between the anterior body end and anterior ciliary arch with extending posteriorly in a narrow plate, a left broad fan-shaped skeletal plate between the anterior body end and anterior ciliary arch, a skeletal rod plate along the left side of macronucleus, and a cytoproct behind the posterior ventral ciliary arch. They were detected in 21, 21, and 7 specimens of 40 fecal samples, respectively.

The fourth new species was described as a member of the genus *Gorillofascia* and named *G. longior* n. sp. It had the morphological characteristics of *Gorillofascia*: a flask-shaped body, a small round frontal lobe, a large cavity with an opening at the posterior body end, a tubular vestibulum, a non-retractable adoral ciliary zone, a vestibular ciliary zone, a cavity ciliary zone, right and left longitudinal ciliary zones on the dorsal body surface, an ellipsoidal or peanut-shaped macronucleus, a micronucleus on various sides of the macronucleus, and a contractile vacuole. It was detected in three out of 40 fecal samples.

***Troglodytella gracilis*** n. sp. (Figs. 1–8, 28–30, 36–38, Table 1)

**Diagnosis.** With the characteristics of the Genus *Troglodytella*. Body is elongated, laterally compressed with

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