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Post-emergence changes in the outer coverings of *Georissus costatus* Castelnau, 1840 (Coleoptera: Georissidae)

Изменения наружных покровов имаго *Georissus costatus* Castelnau, 1840 (Coleoptera: Georissidae) после его выхода из куколки

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КЛЮЧЕВЫЕ СЛОВА: *Neogeorissus*, группа вида *Georissus costatus*, имаго, кутикула, склеротизация, биоминерализация, синонимия.

ABSTRACT. The article discusses the morphology and properties of the outer coverings of *Georissus costatus* Castelnau, 1840 adults at different stages of their development. The study is supported by original photographs. It was found that after the adult emerges from the pupa, its outer coverings undergo a number of structural changes that affect the appearance of the beetle. 1. During sclerotisation, the cuticle develops a bright, colourful metallic sheen. 2. The surface of the cuticle becomes covered with an opaque whitish friable coating. 3. This coating forms a hard coating that partially or completely hides the coloration and sheen of the cuticle; the beetles appear dark in colour. These changes are characteristic of all species in the *G. costatus* group. Most probably, within this group, specimens with different outer coverings conditions were previously classified as separate species.

РЕЗЮМЕ. В работе обсуждаются морфология и свойства наружных покровов имаго *Georissus costatus* Castelnau, 1840 на разных стадиях их созревания. Исследование подкреплено оригинальными фотографиями. Показано, что после выхода имаго из куколки его наружные покровы претерпевают ряд структурных изменений, влияющих на внешний вид жука. 1. В ходе склеротизации кутикула приобретает яркий цветной металлический блеск. 2. Поверхность кутикулы покрывается непрозрачным белесым рыхлым слоем. 3. Из него образуется твердый слой, который частично или полностью скрывает окраску и блеск кутикулы; жуки выглядят темными. Такие изменения свойственны всем видам группы *G. costatus*. Вероятнее всего, внутри этой группы экземпляры с разным состоянием наружных покровов ранее были отнесены к самостоятельным видам.

Introduction

Georissus (*Neogeorissus*) *costatus* Castelnau, 1840 (Georissidae) is a small, inconspicuous inhabitant of the banks of small and medium-sized lowland and foothill rivers. Its life cycle, excluding migration and possibly hibernation, takes place on a strip of moist sand along the water's edge. Adults can be found on the surface and in natural cavities in the sand. They are also capable of digging small shallow pits. Pupation occurs in a pupal chamber beneath the sand surface. The species is known from the Mediterranean countries of Europe, Africa, and Asia, the Transcaucasus, the south of the European part of Russia as well as the Urals (Voronezh, Tambov, Samara, Saratov, and Orenburg Oblasts), and the south of Western Siberia (Altai Krai) [Jacobson, 1913; Fikáček, Przewożny, 2015; Litovkin, Sazhnev, 2016; Przewożny, 2022].

The original description of *G. costatus* characterizes it as black, greenish on top, and matt ("Noir, verdâtre en dessus, mat") [Castelnau, 1840: 45]. According to the original description, *Georyssus cupreus* Reiche, 1879 (= *G. costatus*) differs from it in its metallic coppery sheen ("… par son éclat cuivreux métallique …") [Reiche, 1879: 238]. Publications from different years say that *G. costatus* is matt, but it often [Ganglbauer, 1904; Steffan, 1979] or almost always [Hebauer, Klausnitzer, 1998] has a more or less pronounced metallic sheen. Sometimes, in contrast, the beetles are covered with a chalky whitish coating ("… manchmal mit kreideartigem, weissen Ueberzug …") [Reitter, 1881: 86]. Photographs of beetles with a bright metallic sheen are shown on the ZIN RAS website [https://www.zin.ru/animalia/coleoptera/rus/geocoskm.htm] and in [Zinchenko, 2014]. Photographs of

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dark-coloured beetles are shown in [Litovkin, Fikáček, 2011] and [Brojer, 2023]. The reasons for such diversity in the coloration of *G. costatus* have not yet been discussed in the literature.

In revisions of Afrotropical species of the *G. costatus* group, the presence or absence of a metallic sheen is traditionally considered as an important specific feature [Delève, 1967a, b]. Importance is also attached to the degree of tubercle and granule expression on the pronotum and elytra. At the same time, the author of the revisions assumes intraspecific variability of these features and, consequently, the possibility of synonymy among some species [Delève, 1967a, b]. Later, other species of the *G. costatus* group were described, both with and without sheen [Calamandrei, Mascagni, 1993; Fikáček, Trávníček, 2009; Fikáček *et al*., 2012]. The description of *G. chameleo* Fikáček et Trávníček, 2009 only notes that the variability of this species does not depend on geography [Fikáček, Trávníček, 2009].

In all my years of observing *G. costatus*, I have never seen live beetles with a bright metallic sheen in nature. I was familiar with such specimens from the collections of other entomologists and from the published photographs. However, the other coloration variants were represented in my collections. After observing *G. costatus* adults emerge from pupae and studying the collection material more closely, the reasons for this became clear. I found a connection between coloration and sculpture, as well as their regular change depending on the age of the adult. The preliminary results of the study are discussed in this article.

Material and methods

This study is based on personal collections and observations of *G. costatus* in the Samara and Orenburg Oblasts from 2005 to 2024 in the period from May to August. Several specimens from the Tambov and Orenburg Oblasts were obtained from colleagues. The beetles were killed by ethyl acetate vapor or in an ethanol solution. In total, more than 200 collection specimens were studied. Additionally, video and photo documentation of beetle behaviour in both their natural habitat and laboratory conditions was conducted. *Georissus costatus* pupae were obtained from first-instar larvae collected in the field and reared in laboratory conditions (Summer 2024). A total of two adults, emerging from pupae, were studied, one of which (the smaller one) appeared unhealthy (Figs 7–8). It should be noted that frequent disturbance and bright lighting during observations in the laboratory conditions may have affected the development of the adults. All the material studied is stored in my private collection.

To clean them of dirt, dry specimens were relaxed in hot, distilled water for 15 minutes, then placed in a solution of detergent (commercial dishwashing liquid) at room temperature for 1 hour. The beetles were cleaned of dirt in a drop of the same detergent solution using kolinsky brushes (sizes 0 and 00) and dissecting needles (0.3 and 0.15 mm thick) under a microscope. To remove the detergent, the beetles were kept in distilled water at room temperature for 1 hour, with periodic shaking, then mounted on paper or transparent card. In some cases, a 4% acetic acid solution was used for cleaning.

The beetles were studied using a LOMO MBS-200 stereomicroscope with magnifications up to 65×. A dual gooseneck LED illuminator with a declared colour temperature of 6500K and unknown CRI was used as the light source. Both direct hard and diffuse light were used. Photographs of collection specimens and live beetles in the laboratory were taken with a Nikon D3300 DSLR camera equipped with a LOMO 3.7×0.11 microscope objective. The collection specimens were illuminated with an LED flashlight with a colour temperature of 5700K and CRI=95+, while the live beetles were illuminated with a camera-built-in flash with a diffuser. Image capture and processing were performed using digiCamControl 2.1.2.0, Nikon Capture NX-D, Zerene Stacker 1.04, and Adobe Photoshop CS3 software.

The names given to the structures discussed below ("friable porous coating" and "hard porous coating") are based on their characteristic features. The spelling of the author's name for the taxon *G. costatus* (Castelnau, not Laporte or Laporte de Castelnau) is the same as in the work with its original description [Castelnau, 1840]. Discussion of this issue goes beyond this article.

Results

For the first hour after emerging from the pupa, a slight maroon reflection is visible on the light translucent cuticle of the adult (Fig. 3). As the cuticle darkens, it takes on an increasingly noticeable sheen. The darkening of the cuticle is uneven. During the first two to three hours, only slight darkening is visible in isolated areas, primarily along the edges of the sclerites (as in Fig. 4). Within eight to nine hours, the cuticle darkens completely, exhibiting a bright metallic maroon and green sheen on the head, pronotum, elytra, and legs (Fig. 5). The elytra are translucent, resembling smoked glass. After one day, the cuticle darkens further, the sheen fades slightly (Fig. 6), and a characteristic spotted pattern appears on the elytra (see below) (Fig. 7). The surface of the cuticle between the sculptural elements is smooth, with no pores (do not confuse these with the punctures in rows and granules) (Fig. 9). In both specimens in the experiment, the upper side of the body darkened faster than the under side. Fully mature beetles could not be obtained in the experiment, so subsequent changes were determined from collection specimens.

Specimens with fully sclerotised cuticles have a bright metallic sheen on the dorsal surface of the head, pronotum, elytra, sternum, abdomen, coxae, and femora (Fig. 13). The under side of the body is sometimes partially black. Dry (unmoistened) specimens usually have a maroon sheen with green, gold, and blue speckles. The elytra often have a faint black pattern (Fig. 13). This pattern is formed by a small transverse suture spot in the anterior third and bands that converge in a V-shape from the middle of the outer edge of the elytra to the posterior third of the suture. The surface of the cuticle between the sculptural elements is smooth (Fig. 10) and occasionally slightly rough or wrinkled. There are no pores. The tubercles on the pronotum, the ridges on the elytra, and the granules are distinct but can vary in expression. Some time after the cuticle has completely sclerotised, an additional coat forms on it, which partially or completely hides its coloration and sheen.

In videos and photographs taken in their natural habitat, the beetles appear grey and matt [e.g. https://www.zin.ru/animalia/coleoptera/rus/geocosli.htm]. When collected

in tubes, they also do not appear shiny under sunlight at 10–15× magnification. However, many specimens exhibit a slight metallic reflection when placed in alcohol for preservation. In the laboratory conditions under a microscope in direct hard light, some specimens show a greenish or reddish-copper pattern with a dull metallic sheen (Figs 1–2), while others are completely black.

Some of the collection specimens (13 specimens studied) are covered with a whitish friable porous coating (hereafter referred to as FPC) (Figs 14–15). On some specimens, the FPC turns into a continuous whitish fragile crust. The FPC covers nearly the entire body surface, excluding the ventral part of the head, tarsi, and partially the coxae and femora, as well as areas of articulation. The FPC follows the cuticle sculpture exactly, making it less expressive and smoother. The FPC seems fine-grained (hard light), has pores that correspond to puncture rows and granules, and has many additional pores dispersed evenly over the entire surface (diffused light) (Fig. 11). When dry, the FPC can easily be scraped off with a dissecting needle. It softens fairly quickly in warm water, either clean or with detergent, after which it can be completely removed from the cuticle with the tools (Figs 13–15). In some specimens, the FPC is worn away at the tips of the granules. When moistened with water, the FPC becomes grey and remains opaque. The FPC does not dissolve in organic solvents such as ethanol, isopropanol, ethyl acetate, and ortho-xylene. I do not know what the FPC looks like on live beetles. Apparently, it does not prevent movement or flight. Remnants of the FPC have been found on a brightly shiny specimen washed with water, which flew into the light and was stored in alcohol for a long time.

Most collection specimens are dark, ranging in coloration from golden-greenish with a dull metallic sheen to nearly black (Figs 16–18). The characteristic black pattern is often visible on the elytra (Fig. 18, compare it with Figs 1–2). After cleaning such specimens with a hot water and detergent solution, a hard porous coating (hereafter referred to as HPC) covering the cuticle is revealed (Figs 17–18). The HPC is very difficult to scratch with a dissecting needle. Its surface seems fine-grained and has pores, similar to the FPC from which it most likely forms (Fig. 12). The HPC is resistant to cold and hot water but softens quickly in a weak acetic acid solution, after which it can be completely removed from the cuticle with the tools.

A dull greenish metallic sheen (Fig. 17) is visible on the dorsal surface of the head, pronotum, and elytra of dry (unmoistened) beetles. This sheen can be explained

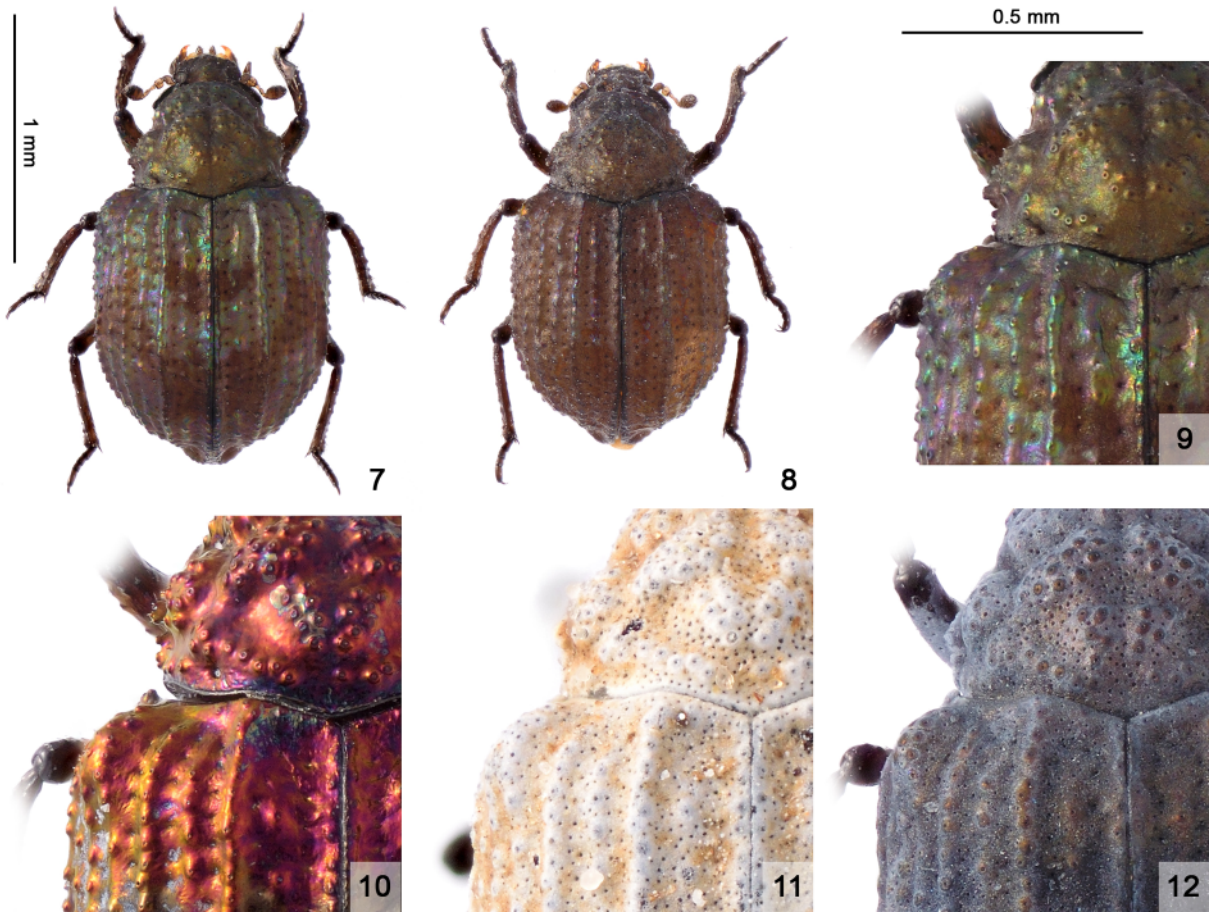
Figs. 1–6. Living individuals of *Georissus costatus*. 1–2 — beetles collected in the field with a reddish-copper (1) and greenish (2) dull metallic sheen as well as the dark pattern on the elytra; 3–6 — the adult emerging from the pupa at different stages of cuticle sclerotisation; the time after emergence is indicated in the photographs. Photographs by S.V. Litovkin.

Рис. 1–6. Живые особи *Georissus costatus*. 1–2 — собранные в природе жуки с красновато-медным (1) и зеленоватым (2) тусклым металлическим блеском и с темным рисунком на надкрыльях; 3–6 — вышедший из куколки жук на разных этапах склеротизации кутикулы; время после выхода указано на фотографиях. Фотографии С.В. Литовкина.

by a combination of two factors. First, optical effects occurring in the translucent HPC change the visible colour of the cuticle. Second, hard light reflected from the surface of the HPC creates a slight sheen effect. It is unclear whether the sheen of the cuticle itself is visible in this case. The HPC of thoroughly cleaned beetles takes on a silvery-grey reflection in hard light, which more or less masks the coloration. In diffused light, their coverings appear matt (Fig. 18) and sometimes ash-grey. A distinct maroon sheen is only visible on areas worn away to the cuticle. Moistening the coverings with water or alcohol significantly intensifies their coloration and sheen, and the silvery-grey reflection disappears. Greenish or dark blue coloration becomes evident in many dark specimens. On the ventral side of the body, the coloration is not visible, even when wet. On thoroughly cleaned beetles, a small drop of water quickly spreads over the surface of the HPC, as if impregnating it. This phenomenon can be explained by the capillary action occurring between the structural particles of the HPC.

The HPC follows the sculpture of the cuticle. The HPC fills the spaces between the tubercles of the pronotum, the elytral ridges, and the gaps between the granules on them (Fig. 12). This makes the sculpture smoother. As noted above, the HPC is often worn down to the cuticle at the tips of the granules (on the pronotum, shoulder tubercles, and elytral ridges), which makes the sculpture look even smoother.

The HPC does not prevent flight. In a Petri dish in the dark, under a point light source, some beetles attempted to fly. Colored and black specimens are usually found together. Both coloration variants were also noted among the overwintering (according to the collection date, May 10, Southern Urals) beetles. Single specimens with the HPC are brown.



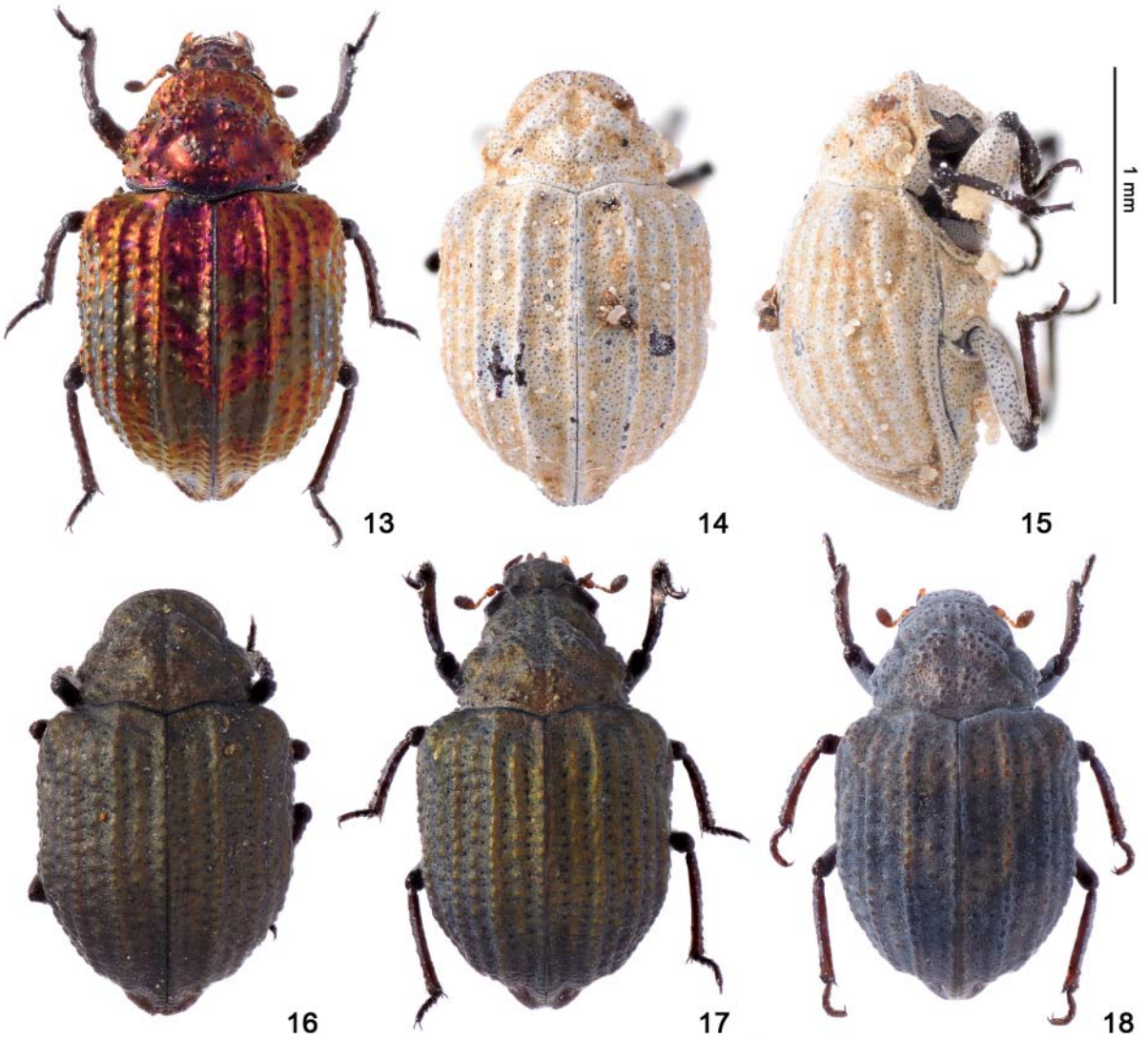
Figs. 7–12. Collection specimens of *Georissus costatus*. 7–8 — the adults reared in the laboratory conditions: one day (7) and four days (8) after emerging from the pupa; 9 — sculpture and coloration of the cuticle of the adult reared in the laboratory conditions; 10 — sculpture and coloration of the cuticle of a completely sclerotised adult collected in the field; 11 — sculpture and microsculpture of the outer coverings with the friable porous coating; 12 — sculpture and microsculpture of the outer coverings with the hard porous coating. Photographs by S.V. Litovkin.
Рис. 7–12. Коллекционные экземпляры *Georissus costatus*. 7–8 — имаго, выведенные в лабораторных условиях: через сутки (7) и через четыре дня (8) после выхода из куколки; 9 — скульптура и окраска кутикулы выведенного в лабораторных условиях имаго; 10 — скульптура и окраска кутикулы полностью склеротизованного имаго, собранного в природе; 11 — скульптура и микроскульптура покровов с рыхлым пористым слоем; 12 — скульптура и микроскульптура покровов с твердым пористым слоем. Фотографии С.В. Литовкина.

Discussion

The coloration and degree of sculptural expression in collection specimens of *G. costatus* depend on the method used to clean them of dirt. Natural coloration and sculptural variation in addition to variability are associated with gradual post-emergence changes in the outer coverings of the beetles. The details, mechanisms, and role of these changes remain to be determined. However, the observational data I have obtained allow me to draw some conclusions.

The properties of the FPC indicate that it is a natural derivative of the cuticle, rather than an artifact resulting from the fixation and/or storage of the specimen. Based on the mechanical, optical, and chemical properties of the FPC and HPC, their biomineral origin can be assumed.

The lack of field observations of beetles with a bright metallic sheen indicates that the FPC forms shortly after the adult emerges from the pupa. The small proportion (about 6.5%) of beetles covered with the FPC in the collection material indicates its relatively rapid transformation into the HPC. For most of their lives, beetles have a dark, inconspicuous coloration. It is currently unclear what determines the presence of a dull metallic sheen: age of the beetle or the structure of the HPC. The clearly



Figs. 13–18. Collection specimens of *Georissus costatus*. 13 — a specimen with a bright metallic sheen; 14–15 — the same specimen covered with the friable porous coating dorsally (14) and laterally (15); 16 — a dark-coloured specimen without preliminary cleaning, killed by ethyl acetate; 17 — a specimen cleaned with detergent, with a greenish dull metallic sheen; 18 — a specimen thoroughly cleaned with detergent, with the hard porous coating of greyish colour. Photographs by S.V. Litovkin.

Рис. 13–18. Коллекционные экземпляры *Georissus costatus*. 13 — экземпляр с ярким металлическим блеском; 14–15 — тот же экземпляр, покрытый рыхлым пористым слоем, вид сверху (14) и сбоку (15); 16 — темноокрашенный экземпляр без предварительной очистки, усыпленный этилацетатом; 17 — отмытый детергентом экземпляр с тусклым зеленоватым металлическим блеском; 18 — тщательно отмытый детергентом экземпляр с твердым пористым слоем сероватого цвета. Фотографии С.В. Литовкина.

visible whitish FPC can be used to determine the date of emergence of adults from pupae and, consequently, the time of breeding of *G. costatus*.

The whitish porous coating or crust on *G. costatus* can be mistaken for camouflage, which it is not. True camouflage made of dirt particles is not characteristic of this species. Camouflage that completely conceals the upper side of the body is known in species of the subgenus *Georissus* s.str. [Litovkin, 2018; Fikáček, 2019], and partial camouflage is known in some species of the *G.* (*N.*) *laesicollis* group [Litovkin, 2018 and unpublished data]. It is possible that the mechanism of the HPC formation and the ability to create camouflage are interrelated.

It can be argued that the described post-emergence changes in the outer coverings are characteristic of all species of the *G. costatus* group. The HPC is clearly visible in SEM photographs in the original description of *G. nemo* Fikáček, Delgado et Gentili, 2012 [Fikáček *et al*., 2012: Figs 9–13]. In a specimen of a similar species from India (private collection), a bright metallic sheen is visible on the abraded areas of the pronotum and elytra. In the descriptions of three species lacking sheen (*G. granifer* Grouvelle, 1909, *G. tuberifer* Grouvelle, 1909 and *G. subaequalis* Paulian et Legros, 1943), the pores between the tubercles of the pronotum are mentioned and illustrated [Delève, 1967b]. Specimens with a bright metallic sheen were obtained from dark and matt specimens by cleaning them in an acetic acid solution (*G. intermedius* Paulian et Legros, 1943, *G. lujai* Legros, 1943 and *G. laevigatus* Brancsik, 1914) [Delève, 1972b]. A whitish coating, similar to the FPC in *G. costatus*, has been observed by me on several specimens of *G.* (*N.*) cf. *laesicollis* Germar, 1832 from Kazakhstan. A white layer on the coverings was previously observed in *G. laesicollis* from Europe [Reitter, 1881; Mascagni, 2004]. At the same time, *G.* (s.str.) *crenulatus* (Rossi, 1794) (Russia, Kazakhstan) and *G.* (s.str.) cf. *substriatus* Heer, 1841 (Kyrgyzstan), washed of their camouflage, lack the HPC.

Based on the drawings and remarks in the descriptions, many Afrotropical species of the *G. costatus* group are practically indistinguishable in structure of the aedeagus, but differ in coloration and sculpture [Delève, 1972a, b]. Moreover, matt species have a smoothed sculpture, while brightly shiny ones have a distinct sculpture. This is well consistent with the properties of the outer coverings of *G. costatus* described above. It is obvious that different age forms and differently prepared collection specimens were described as separate species. A revision of the *G. costatus* group is necessary, as a result of which some species will most likely be synonymized.

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