# Affinity of *Aporia howarthi* Bernardi, 1961 and *A. goutellei* (Овектнüк, 1886) (Lepidoptera: Pieridae) inferred from morphology of male genitalia

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Abstract: In order to clarify the phylogenetic position of *Aporia howarthi*, we have compared the morphology of the male genitalia of four species of the genus *Aporia: crataegi* (Linnaeus, 1758), *hippia* (Bremer, 1861), *howarthi* Bernardi, 1961 and *goutellei* (Oberthür, 1886). As a result of the observation, we have found that the genitalia of *A. howarthi* are very similar to those of *A. goutellei* and that the two species share some distinctive features. Therefore, since their distribution areas are adjacent, it seems likely that *A. howarthi* speciated from *A. goutellei* which, spreading its distribution from the Hengduan Mountains to the west, lost its wing markings.

Key words: Aporia howarthi, A. goutellei, male genitalia.

Verwandtschaft von Aporia howarthi BERNARDI, 1961 und A. goutellei (ОВЕКТНÜК, 1886) (Lepidoptera: Pieridae) abgeleitet aus der Morphologie der männlichen Genitalien

Zusammenfassung: Um die verwandtschaftliche Position von Aporia howarthi zu klären, wurde die Morphologie des männlichen Genitalapparats von 4 Arten der Gattung Aporia verglichen: A. crataegi (Linnaeus, 1758), hippia (Bremer, 1861), howarthi Bernardi, 1961 und goutellei (Oberthür, 1886). Als Ergebnis stellte sich heraus, daß die Genitalien von A. howarthi denen von A. goutellei sehr ähnlich sind, insbesondere in einigen sehr spezifischen Merkmalen. Deswegen und wegen der Nachbarschaft ihrer Verbreitungsareale erscheint es uns wahrscheinlich, daß A. howarthi sich von A. goutellei abspaltete und vom Hengduan-Gebirge nach Westen ausbreitete und dabei seine Flügelmusterung weitgehend verlor.

#### Introduction

Aporia howarthi Bernardi, 1961 is a Pieridae species found only in a limited area of southeast Tibet, where it flies in the second half of May when snow is still covering the hills. Fourty years after the original description it was rediscovered by the second author.

In its original description Bernardi (1961) included *A. howarthi* in the species-group of *A. crataegi*, both for the wing markings and for the genitalia morphology. Kanoh et al. (2017) supposed a relationship with *A. goutellei* (Oberthür, 1886) on the basis of the geographical distribution and the shape of the uncus of male genitalia. However they could not reach a final conclusion, because a detailed comparison of the two species was not possible since they had available only uncus drawings from other researchers.

In the present study, the morphology of the male genitalia of the two species has been directly observed and compared, allowing a detailed analysis of their relationship. In addition they have been compared to the male genitalia of A. crataegi (Linnaeus, 1758) and A. hippia (Bremer, 1861), both belonging to the crataegi speciesgroup.

#### Material and methods

To observe the male genitalia, the abdomen of the specimen was excised, soaked, and treated in a aqueous solution of 10% potassium hydroxide at room temperature for one or two days, then washed in water and observed under a binocular stereomicroscope. The uncus was observed from the apex, and an image in the transverse plane has been drawn by focusing the microscope on the area considered to reflect the most distinctive part of the middle area.

A distribution diagram was prepared, based on specimens owned by the Research Institute of Evolutionary Biology and by the authors. Subspecies names are based on Della Bruna et al. (2013).

### Results

The specimens used for genitalia observation are shown in Figs. 1A-1D, and their male genitalia are shown in Figs. 3A-3D. The main features of the male genitalia are listed in Table 1 showing that *A. goutellei* (Oberthür, 1886) and *A. howarthi* Bernardi, 1961 are quite similar to each other and different from *A. crataegi* (Linnaeus, 1758) and *A. hippia* (Bremer, 1861).

Compared to *A. crataegi* and *A. hippia*, the common features of *A. goutellei* and *A. howarthi* are the following:

- The uncus is wide and its transverse plane is almost a circle.
- It has a flat protrusion, and the ventral side is rounded and swells to protrude to the distal side.
- On the dorsal side of the uncus there is a weak vertical linear ridge.
- The juxta is wide and heart-shaped.
- The saccus is slender and long.
- The phallus is thick.
- The valva is narrow, curved and a dent is formed inside.
- There is no clear fovea inside the valva.

The differences between *Aporia goutellei* and *A. howarthi* are small: in *A. goutellei* the uncus is wider, the uncus and the tegumen are higher and the juxta is somehow wider.

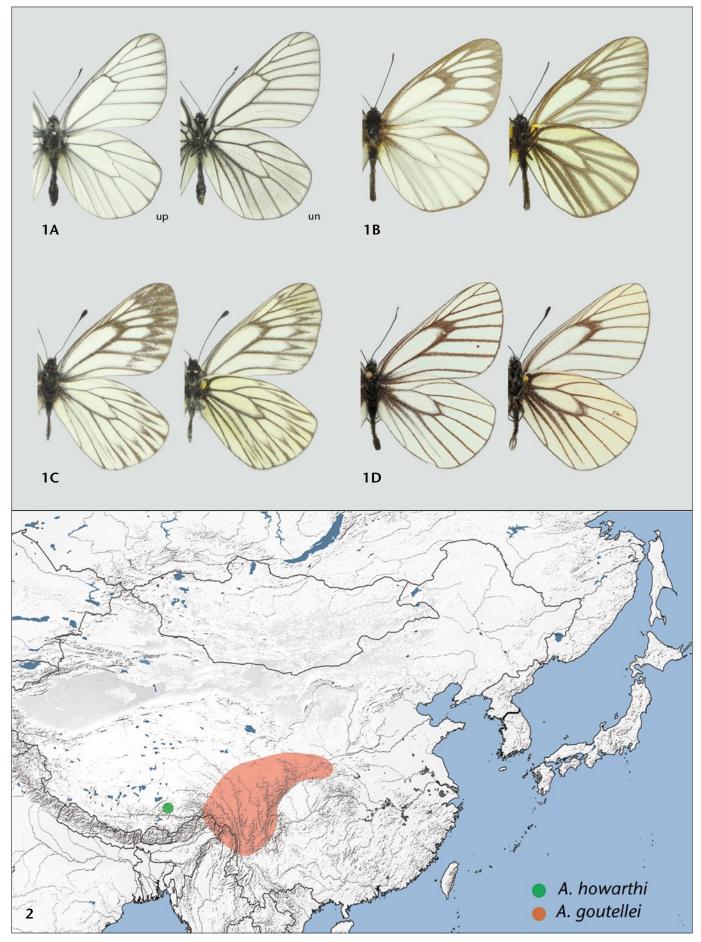
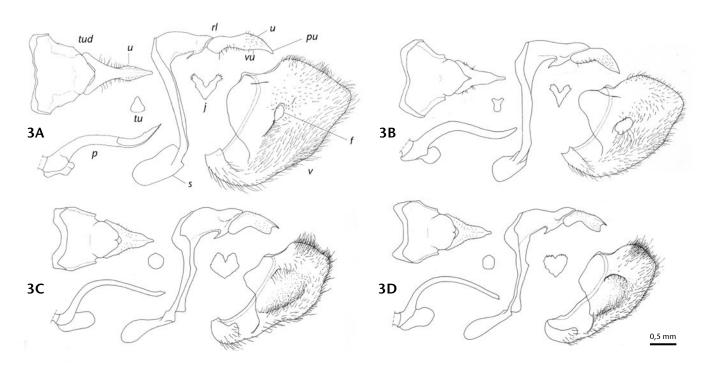


Plate: Specimens of *Aporia* and distribution map. — Fig. 1: Specimens of *Aporia crataegi, A. hippia, A. goutellei* and *A. howarthi* used for genitalia dissection. — 1A: *A. crataegi adherbal, &,* Kitami, Hokkaido, Japan, forewing length (= FWL): 37.9 mm. 1B: *A. hippia japonica, &,* Yatsugatake, Nagano, Japan, FWL: 33.7 mm. — 1C: *A. goutellei goutellei, &,* Lijiang, Yunnan, China, FWL: 29.9 mm. 1D: *A. howarthi, &,* E. of Mila pass, road Lhasa—Nyingtri, Tibet, China, FWL: 29 mm. — Fig. 2: Distibution map of *Aporia goutellei* and *A. howarthi*.



**Fig. 3:** Male genitalia of *Aporia crataegi, A. hippia, A. goutellei* and *A. howarthi.* — Abbreviations: tud = tegumen and uncus, dorsal view. rl = ring, lateral view. u = uncus. pu = protrusion of uncus. vu = ventral side of uncus. tu = transverse plane of uncus. p = phallus. j = juxta, v = right valva, inside view. s = saccus. f = fovea. — **3A:** *A. crataegi adherbal.* **3B:** *A. hippia japonica.* — **3C:** *A. goutellei goutellei.* **3D:** *A. howarthi.* 

Table 1: Characteristics of male genitalia in four species of Aporia.

Taxon	Uncus		luvto	Saccus	Phallus	Valva	
	protrusion	ventral side	Juxta	saccus	riiallus	width	fovea
Aporia crataegi adheral	cylindrical	straight	V-shaped	thick	slender	wide	clear
A. hippia japonica	cylindrical	straight	V-shaped	thick	slender	wide	clear
A. goutellei goutellei	flat	swollen	heart-shaped	slender	thick	narrow	not clear
A. howarthi	flat	swollen	heart-shaped	slender	thick	narrow	not clear

#### Discussion

Kanoh et al. (2017) showed that the genus *Aporia* can be divided into two groups by the shape of the uncus in male genitalia. One group has a long conical protrusion at uncus apex, ending with a rounded tip. The ventral side is straight. This group includes *A. crataegi*, *A. hippia*, *A. lhamo* (Овектнüк, 1893) and some other.

The second group has the uncus with a flat protrusion at apex and a rounded ventral side. This group includes *A. monbeigi* (OBERTHÜR, 1917) and *A. agathon* (GRAY, 1831) and some other.

As shown above, both *A. goutellei* and *A. howarthi* have the uncus with a protruding flat tip and a rounded ventral side, that are the characters of the second group. Furthermore, their uncus is wide and its transverse plane is close to a circle. This character, unique among the *Aporia* species (Kanoh et al. 2017), suggests that they are closely related to each other.

Bernardi (1961) included A. howarthi in the A. crataegi species-group, with A. hippia, A. bieti (Oberthür, 1884),

A. martineti (Oberthür, 1886), A. potanini Alphéraky, 1892 and A. genestieri (Oberthür, 1902), based on the wing markings and the male genitalia morphology. However, in the description of A. howarthi male genitalia, he quoted: shape of the apex of the uncus with a special form in which "the nails extend over the horseshoe", valva narrow and without fovea, and saccus long. These characters are consistent with those observed in this study, but, as clear from Table 1, they are different from A. crataegi and A. hippia, therefore A. howarthi does not belong to the A. crataegi species-group.

Aporia howarthi has no wing markings except black lining of the wing veins, same as in A. crataegi and A. hippia. However the characteristics of male genitalia suggest that the similar feature of no wing markings originated from different strains.

Kanoh et al. (2017) speculated that progression of speciation and disappearing of wing markings occurred through the geographical radiation within the genus *Aporia lhamo* and *A. monbeigi*, found in the cen-

tral portion of the Hengduan Mountains and showing complex wing markings, were thought to have retained the old original traits of the genus *Aporia*. The ancestral species progressed fading out its wing markings through spreading its distribution and made a geographical cline continued beyond species and a species, with no wing markings at all at the end of the geographical radiation.

As shown in Fig. 2, A. goutellei is widespread in China from northern Yunnan Province to Sichuan Province and to south east Tibet. A. howarthi is distributed in southeast Tibet, close to the west end of the distribution area of A. goutellei. Therefore, A. howarthi is likely to be a species originated from A. goutellei while spreading its distribution to the west, with the disappearing of wing markings.

## **Acknowledgements**

We wish to express warm thanks to Toshiaki Aoki, Shuhei Yamaguchi and Seiji Sakai.

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Received: 20. xi. 2017

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Autor(en)/Author(s): Kanoh Kazumi, Bozano Gian Cristoforo

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