

**Oleksiy Gumovsky**

**Final report of BELSPO post-doctoral project**

**"Afrotropical genera of Entedoninae (Hymenoptera: Eulophidae)" (2012-2013)**

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### **Project summary**

The aim of the project was to contribute to the taxonomic revision of a poorly studied and taxonomically difficult group of insects (parasitic wasps of the subfamily Entedoninae) from a region of high biodiversity (tropical Africa). The project facilitated the more accurate sorting of the collection of the Royal Museum for Central Africa, Tervuren, Belgium (RMCA), inventorisation of work on the types of species described by previous authors, and a comparison of these data with the data obtained from other collections. The use of the RMCA molecular genetics facilities was expected to contribute to clarification of the phylogenetic relationships of the genus-group taxa. Results are to be incorporated into the large-scale biodiversity survey "Wasps of Africa and Madagascar" (initiated by IZIKO, Museum of Cape Town, Republic of South Africa).

The RMCA houses one of the largest collections of Afrotropical insects, in particular Chalcidoidea. The generic concepts of Entedoninae are often challenging. Some Afrotropical Entedoninae, collected locally before execution of this project, represent unique derived (“weird”) forms, and their taxonomic status requires clarification. A study on the type and comparative materials housed in the RMCA collection was critical for such a clarification. Also, the stay in RMCA facilitated the identification of hitherto unsorted materials on the parasitoids (mostly eulophids) of African Gracillariidae (Lepidoptera): one of the research projects of this institution.

The anticipated outcomes from the project's execution included:

- 1) accurate sorting and estimation of the collection of Entedoninae in RMCA.
- 2) establishment of how the species described by previous authors may be incorporated into current taxonomical framework (new synonymies and combinations).
- 3) illustrations of types and other specimens stored in RMCA (with further incorporation into the Morphbank and other electronic databases).
- 4) execution of molecular work on Afrotropical entedonines using RMCA's facilities, as an aid to support or disprove the generic status of some enigmatic groups of Entedoninae.

### **Project tasks/objectives:**

Project objectives comprised the following:

- Revision of Afrotropical genera of Entedoninae,
- Revisions of most sophisticated genera and their allies.
- Phylogenetic assessment of the genera in order to find monophyletic units and evaluate their possible taxonomic range
- Identification of hitherto unsorted materials on the eulophid parasitoids of African Gracillariidae (Lepidoptera) (in cooperation with Lepidoptera experts in RMCA)

All objectives of the project were achieved, and analysis of the genetic and morphological data obtained is in progress. 14 papers are anticipated on various aspects of the project (see the list below).

### **Project activities**

The following activities were conducted during the project period (1 October 2012 to mid-November 2013) in order to achieve the above-stated objectives:

- 1) Desk-top work in the RCMA museum with collections and type materials, using visual examination by light stereomicroscopy. Unsorted specimens were identified according to the current taxonomic concepts, and the studied type specimens were checked for the presence of established generic and specific characters.

2) Digitization of the key specimens was done with the aim of generating an image library for further use in the comparison and accurate identification of specimens. Images were acquired using a Leica (color imaging) and JEOL JSM-6480LV Scanning Electronic Microscope. The use of low vacuum mode permitted the non-destructive observation of specimens which cannot be viewed at high vacuum due to their non-conductive surface. This was necessary to study type and other collection materials not suitable for dissections and coating by carbon, gold or gold-palladium, as is conventional for SEM.

3) Acquisition of new materials for the enrichment of the collection, and further molecular work. These materials were collected using sweep-netting and various traps during the field trips to Africa and received as loans or gifts from colleagues. Efforts were focused on the discrimination of representatives of Entedoninae from the mixed samples of insects, plant remnants and debris obtained.

4) Laboratory experimental studies on gregarious parasitoids of the genus *Entedon* and *Oomyzus sokolowskii* (a biocontrol agent in Africa) were continued. The isolated parasitoid larvae were observed using SEM.

5) Molecular genetic analyses: sample preparation, genomic DNA extraction, RCR and sequencing. The new sequences obtained from the freshly collected ethanol-stored specimens are expected to constitute a background library for the DNA-based identification of new and poorly known species, and also for further phylogenetic treatment of the genera. These molecular analyses are expected to be conducted in the Joint Experimental Molecular Unit (JEMU) of RMCA during the last two weeks of the visit, and data analysis thereafter.

6) Teaching and capacity-building. The stay in RMCA led to collaborations with colleagues in Europe and Africa, and therefore teaching activities were required during this visit. Dr. Gumovsky provided instructions on identification of parasitoid insects to many RMCA visitors and other students from African countries during the courses he conducted, and also during the collaborative every-day work in RMCA.

### **Duration**

06/05/2012 Project start

30/10/2013 Project end

**Field mission:** DR Congo, 14.01.2013-17.02.2013

### **Scientific missions**

- Royal Belgian Institute of Natural Sciences (RBINS, Brussels). Three visits to RBINS were arranged in order to study its dry collection of Eulophidae. The non-designated types of some Afrotropical Entedoninae are found in the wet (Congo-Brazzaville) and dry (various regions of Africa) collection of RBINS. The type and non-published types of P.L.G. Benoit were also studied.
- El Museo Nacional de Ciencias Naturales (MNCN, Madrid). The type, identified and unsorted materials at the MNCN were studied. The materials from the Mediterranean region were rather interesting, because some seem to represent the species introduced from the Afrotropical region. More than 300 photographs of the type and identified specimens were taken during the visit. Another source of interesting specimens was the unsorted alcohol-preserved materials from different regions, including the Afrotropical region (e.g. Equatorial Guinea).
- The Natural History Museum (BMNH, London). The BMNH is the repository of one of the largest collections of African insects, in particular the Chalcidoidea. As for the Afrotropical Entedoninae (the revision of the generic concepts of which is the topic of this project), the BMNH contains the primary types of fifty of the described Afrotropical species. The types, both identified and some of the unsorted materials were studied during the visit. The primary types of fifty Afrotropical entedonines were digitalized in order to facilitate the identification of the materials stored in the Royal Museum of Central Africa. Secondary types and specimens identified by other experts were also evaluated. More than 200 photographs of the type and identified specimens were taken during the visit, using the BMNH imaging facilities.

- Muséum National d'Histoire Naturelle (MNHN, Paris). MNHN houses the most complete collection of the type specimens of the species described by J. Risbec from various areas of Africa. Most of these types have never been studied, and their review and digitization was critical for the revision of African Entedoninae. The types of 29 species described by J. Risbec were studied, and some lectotypes designated and validated.
- Democratic Republic of Congo (field mission). From 14<sup>th</sup> January to 17<sup>th</sup> February 2013, O. Gumovsky participated in a field mission to DR Congo in a research group with colleagues from Ghent University and Centre de Surveillance de la Biodiversité/University of Kisangani (UNIKIS). The aim of this mission was to investigate the diversity of Entedoninae in equatorial Africa and collect fresh materials for both the RMCA collection and molecular work.

### **Lectures and seminars**

A training course entitled "Parasitoid taxonomy training", organized by *icipe*- African Insect Science for Food and Health and the Royal Museum for Central Africa (10<sup>th</sup> – 21<sup>st</sup> December 2012) was prepared (12 presentations, movies, illustrations) and discussed with Kenyan colleagues (Dr. Komi Fiaboe). The course is organized for 15 students, Ph.D. students and technicians from Togo, Kenya, Cameroon, Tanzania, Uganda and Ethiopia.

### **Main results obtained**

Main results and generalizations:

- 23 genera of Entedoninae were recorded for the Afrotropical region, 4 of them for the first time for this region, including 2 genera to be described as new to science.
- The preliminary inventories demonstrated that the species of all the hitherto known genera of Afrotropical Entedoninae (from Senegal to South Africa, from Kenya to Cameroon) are present in the natural habitats Central Africa (Congo).

- The natural tropical forest of Africa is dominated by the species of the genera represented mostly by associates of galls of Cecidomyiidae (Diptera), predominantly by the species of the genus *Omphale*.
- The phylogenetic analysis suggests that *Colpixys* is a sister group to *Entedon*, thus its generic status is retained. Three (one new) species were assigned to *Colpixys* and eleven (eight new, two from the subgenus *Cederholmia* of *Entedon*) species were assigned to *Xiphentedon* (all of Afrotropical distribution). However, *Xiphentedon*, *Cederholmia* and *Sanyangia* are regarded as junior synonyms of *Entedon* (syn. n.) because they are represented by derived species within *Entedon*, and keeping them valid renders the latter paraphyletic. Three new species groups are proposed for the species of *Entedon* having the complete lateral propodeal sulcus: the *perturbatus* (most Afrotropical, Oriental and Australasian species of *Entedon*), the *forceps* and the *kayovei* (Afrotropical species, the “*Xiphentedon*”-clade).
- Monophyly of *Entedon* + *Colpixys* is supported mainly by the possession of the lateral pronotal shoulders representing convex calli, and the reduced transepimeral sulcus. Close relationships between *Entedon* and *Colpixys* with the genera *Mestocharis*, *Achrysocharoides* and *Chrysocharis*, are supported by the possession of the peculiar semicircular plica in the upper sector of the lateral panel of the pronotum. This plica is replaced by a carina in *Pleurotropsopsis* and *Apleurotropis*, which form a sister group to the node comprising the above-mentioned taxa. The genus *Colpixys* and the groups of *Entedon*, in which the median propodeal carina is missing (*forceps* and *kayovei*), are revised.
- A preliminary analysis of the evolution of associations with beetles is presented, in which it is proposed that the diverse speciation of the *Entedon*-species was associated with the diversification of weevils, resulting from the switch of these beetles to the proliferating angiosperms in the post-Jurassic Period.

- *Afrotropopsis risbeci* Gumovsky and *Schizocharis newbyi* Kerrich are recorded as parasitoids of *Caloptilia* sp. (Gracillariidae) on leaves of *Cremaspora trifolia* (Rubiaceae) in Kenya (Arabuko Sokoke forest). *A. risbeci* was recorded for the first time from Zambia, DR Congo and Kenya. The male of *A. risbeci* is described for the first time for the genus and species. The male is similar to the males of the genus *Pleurotropopsis*.
- The genus *Omphale* is confirmed for the Afrotropical region. The two known species described for the genus, *O. gallicola* (Risbec) and *O. turgidus* Yefremova, proved to be the representatives of other genera (namely, *Sigmophora* and *Pediobopsis*, correspondingly, comb. n.). However, there are about 50 undescribed species of the genus, and moreover, they represent the majority of entedonines collected in riparian rain forests of Afrotropics.
- Molecular data suggest the existence of 6 lineages among Afrotropical Entedoninae which is in accordance with the findings of previous studies (Burks et al., 2011; Gumovsky, 2011).

## Scientific benefits

### For Oleksiy Gumovsky

This Post-doctoral stay at the MRAC allowed O. Gumovsky to achieve several tasks, which would not otherwise have been achievable.

1. He studied the invaluable types (primary and secondary) and various museum materials, all necessary for taxonomical work and his studies on African biodiversity.
2. The morphological studies conducted using SEM and transmission light microscopy, permitted comparisons of the minute morphological features of Afrotropical and European entedonines, and estimation of the degree of variety within the subfamily.
3. He collected fresh materials in Africa. This contributed to an understanding of these African fauna, and allowed for comparison between the old museum collections and local current biological materials.

4. He conducted molecular work using the excellent research facilities of the RMCA (the JEMU laboratory), which allowed re-estimation of phylogenetic relationships and taxonomic status of Afrotropical genera of Entedoninae.
5. He established cooperation in executing the following projects, curated by the researchers from RMCA and other institutions: namely “Parasitoids of Afrotropical Gracillariidae (Lepidoptera)” (Dr. Jurate DePrins, RMCA); “Parasitoids of the agromyzid leafminers in East Africa” (Dr. Komi Fiaboe, ICIPE, Kenya); “Parasitoids of West-African Psyllidae” (Prof. Joseph Lebel Tamesse, Université de Yaoundé, Cameroon), “Insect biodiversity in South of Mozambique” (Mr. Claudio Cuaranhua, Universidade Eduardo Mondlane, Centro AgroFlorestal de Machipanda-CEFLOMA, Mozambique), “Insect biodiversity in North of D.R. Congo” (Prof. A. Dudu, Mr. Jean Bakondongama, University of Kisangani, UNIKIS / Centre de Surveillance de la Biodiversité, D.R. Congo). The scientific contacts established are expected to continue as joint collaborative projects, research and expeditions.
6. Due to the temporary affiliation with RMCA, O. Gumovsky could attend some local (i.e. in Belgium) and international meetings, conferences and seminars. The information obtained from the participation in these conferences will not only be of great use for O. Gumovsky himself, but also shared with the scientific community in Ukraine.

#### For the RMCA

During his stay at the RMCA, Dr Gumovsky sorted the Entedonine collection, improving the accuracy of classifications, and added new genera, so now the RMCA collection houses the representatives of all the known genera present in the African region. Also, Dr Gumovsky took an active part in the museum's research and communication activities. He took part in the demonstration of the Scanning Electron Microscope to the visitors during the Day of Open Doors (Open Bedrijvendag, 7 October, 2012), and also prepared posters for this event. Dr Gumovsky has also actively assisted the Coordinator, Dr. Eliane De Coninck, in the training of the African visitors, especially Prof. Joseph Lebel Tamesse (Cameroon) and Mr. Claudio Cuaranhua (Mozambique).



### For Belspo and the scientific community

The work of Dr Gumovsky in RMCA resulted in the preparation of a scientific book and a series of scientific papers for peer-reviewed journals. At closing data of his post-doctoral grant, the book and one substantial paper about the phylogenetic research on the studied group have been published. The rest of the publications, as individual and in co-authorship with the RMCA and African colleagues, are in advanced stages of preparation (detailed list below). The results of the studies have been reported at two conferences (abstracts are listed below).

### **Future actions**

Writing of scientific articles containing results of the work within the project, followed by their submission to international peer-reviewed journals is anticipated. There are also some projects on collaborative teaching courses and field trips with the former ABIC visitors of RMCA.

### **Publications**

#### Published:

#### Book:

Gumovsky, A.V. 2012. *The family Eulophidae (Hymenoptera: Chalcidoidea): systematics, morphology and life histories*. Kiev, Naukova Dumka. 216 pages (BELSPO & Marie-Curie Action are acknowledged)

Paper in peer-reviewed journal

Heraty J.M., Burks R.A., Cruaud A., Gibson G.A.P., Liljeblad J., Munro J., Rasplus J.-Y., Delvare G., Jansta P., **Gumovsky A.**, Huber, J., Woolley J.B., Krogmann L., Heydon S., Polaszek A., Schmidt S., Darling D.C., Gates M.E., Mottern J., Murray E., DalMolin A., Triapitsyn S., Baur H., Pinto J.D., van Noort S., George J., Yoder M. A 2013. A phylogenetic analysis of the megadiverse Chalcidoidea (Hymenoptera). *Cladistics*, 29: 466–542. doi: 10.1111/cla.12006 (BELSPO & Marie-Curie Action are acknowledged)

Abstracts

Gumovsky A.V. (2013). Afrotropical genera of Entedoninae (Hymenoptera: Eulophidae) / *Abstracts of VIII Symposium of the Ukrainian Entomological Society*, Kiev, 26-30 August 2013. P. 211.

Ramadan, M. , Gumovsky A., Yalamar J. (2013) Parasitism of *Specularius impressithorax* (Coleoptera: Chrysomelidae: Bruchinae), an invasive species of *Erythrina* seeds on the Hawaiian Islands. *3rd International Entomophagous Insects Conference*, Orford, Quebec, Canada

Papers in preparation:

“Phylogenetic re-assessment of *Entedon* Dalman and allied genera, with description of new species from Afrotropics (Hymenoptera, Eulophidae)” (preliminary title).

“Review of Afrotropical species of the genus *Entedon* Dalman (Hymenoptera, Eulophidae)” (preliminary title).

“Review of the *cioni*-species group with a hypothesis on evolution of life history traits in the genus *Entedon* (Hymenoptera, Eulophidae)” (preliminary title).

“New records of Afrotropical Entedoninae”. (preliminary title).

“African species of the genus *Pediobopsis*”. (preliminary title).

“New host record and description of the male for *Afrotroppopsis* (Hymenoptera: Eulophidae, Entedoninae)” (preliminary title).

“A new species group of the genus *Pediobius* (Hymenoptera: Eulophidae, Entedoninae) from Africa”.

“Genera of Afrotropical Entedoninae (Hymenoptera: Eulophidae) with a review of described species“(preliminary title).

A new species and first Afrotropical records of the genus *Setelacher* (Eulophidae, Eulophinae) from Uganda.

A new species of the genus *Trisecodes* (Eulophidae, Entedoninae) from Afrotropics

A new species of *Psyllaephagus* (Encyrtidae), an endoparasitoid of *Blastopsylla occidentalis* Taylor (Psyllidae, Spondyliaspidae) in Cameroon (co-authors: J.L. Tamesse, L.Soufo, E.C. Tchanatame, V.J. Dzokou, Alex Gumovsky and Eliane De Coninck)

A description of the male and a first host record of *Afrotroppopsis risbeci* Gumovsky (Hymenoptera, Eulophidae: Entedoninae) (preliminary title, co-authors: J. De Prins, E. De Coninck)



Museum materials studied



Field work in Africa (DR Congo, January 2013)



Demonstration of the image quality of Scan Electronic Microscopy for the visitors during the Day of Open Doors (7 October, 2012)



Assisting in supervision of African visitors in the RMCA