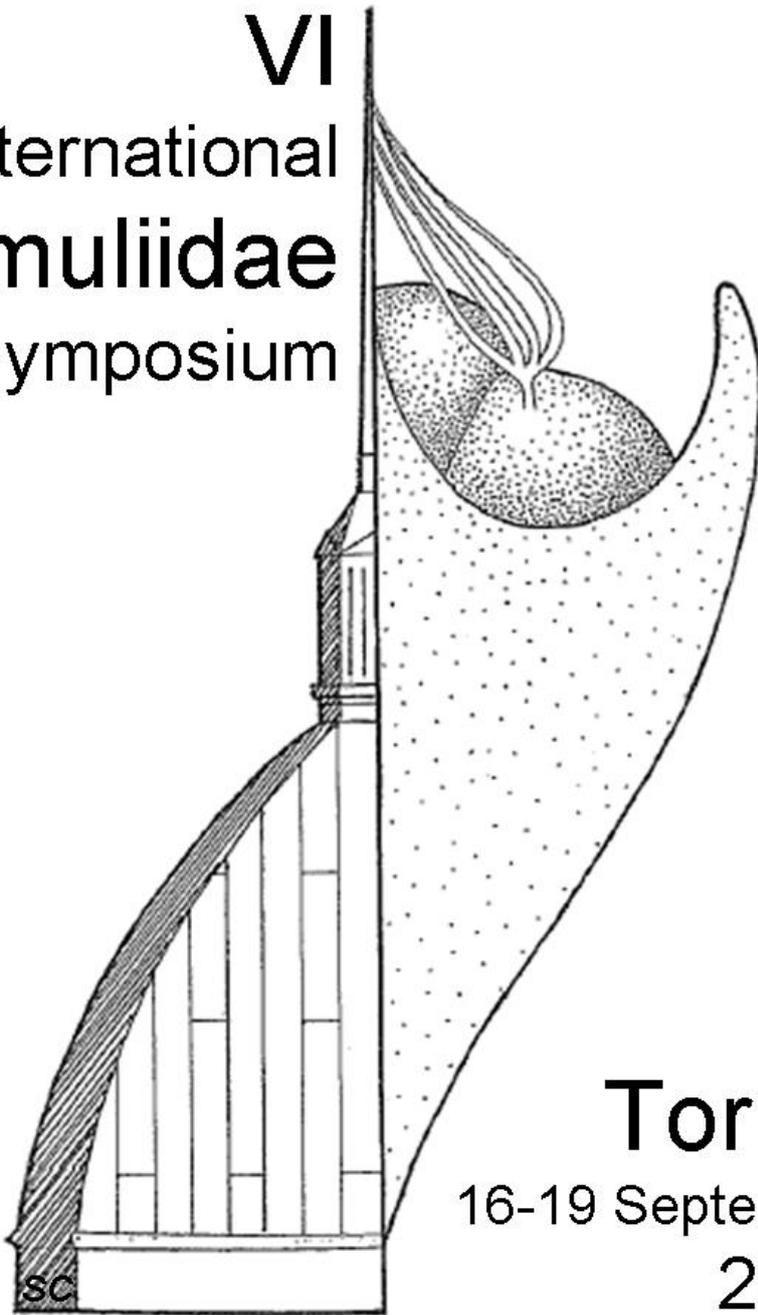


VI
International
Simuliidae
Symposium



Torino
16-19 September
2014

Abstract Book

Edited by S. Ciadamidaro and B. Maiolini

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Torino, Italy

16-19 September 2014

Host committee

Simone Ciadamidaro and Bruno Maiolini

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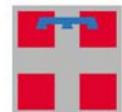
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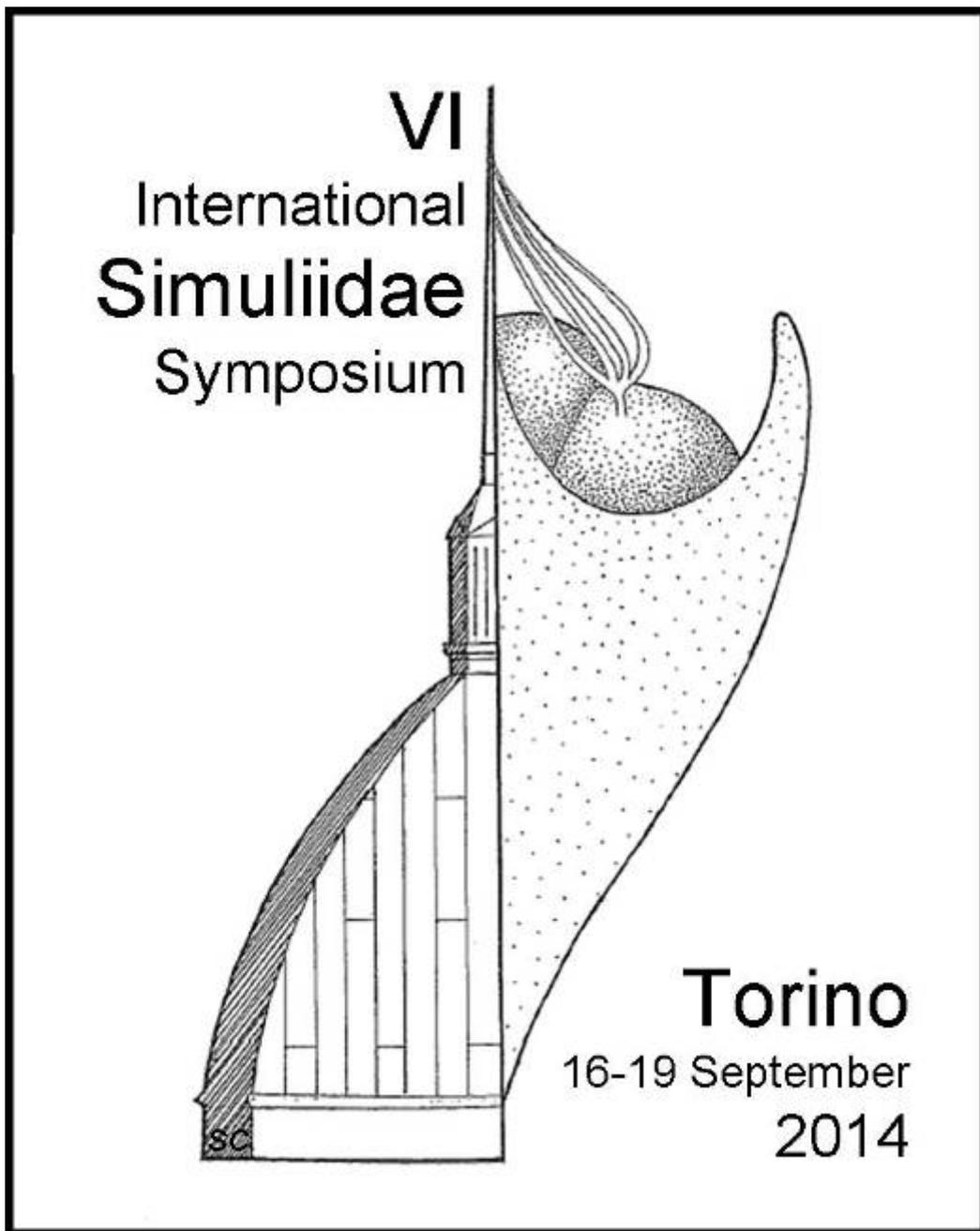
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Abstract Book

VI International Simuliidae Symposium

Torino, Italy



Dear participants to the VI International Simuliidae Symposium,

first of all, BENVENUTI A TORINO!

The organization of this sixth blackfly meeting represents for Torino, Piemonte and for Italy in general a great pleasure and an honour.

A pleasure, since it is nice to have scientists coming from all over the world and meeting in our beautiful city, that we hope you will enjoy in this late summertime. You will surely appreciate our landscapes, our monuments and our food.

An honour, since the topics we are going to discuss have great worldwide interest for human health and economy and for advances in ecology and entomology. Moreover, it is an occasion to strengthen the interest for Simuliidae in Italy, also considering their importance as bioindicators and pest control, both issues concerning blackflies, in a world contest characterized by climate change and hardly controlled river ecosystem modifications.

We thank ENEA, Università di Torino, Museo Regionale di Scienze Naturali, CISBA and the sponsor for their valuable effort in the organization of the symposium and we wish everybody a fruitful work together.

the Host Committee

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ABSTRACTS

17 SEPTEMBER 2014

LEO RIVOSECCHI, A LIFELONG PASSION FOR BLACKFLIES: THE HISTORY OF SIMULIIDAE RESEARCH IN ITALY

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On the occasion of the VI International Simuliidae Symposium, for the first time organized in Italy, Professor Leo Rivosecchi, retired researcher from the Italian National Institute of Health, was invited to present the story of his personal researches on Simuliidae through 50 years of field and laboratory activities. Starting from a short review of the birth of Simuliidae research in Italy at the beginning of the XX century, with Corti's preliminary studies, he reports on how his own interests turned to blackflies when he was still a young naturalist in the 50's, starting an intense, passionate field work to collect samples of these insects from all over Italy, depicting his first steps in blackfly systematics, his doubts, difficulties, collaborations, successes and satisfactions.

Several episodes of his professional life correspond to the advances in the knowledge on these Dipterans in Italy, leading to the publication of his "Fauna d'Italia, Simuliidae" volume, a milestone and still a reference and useful tool for anyone who wants to approach the study of blackflies in Italy and in southern Europe in general.

Leo Rivosecchi, who still collaborates with Italian and foreign researchers who refer to him in order to have suggestions and confirms in their studies, presents during this symposium his ideas on why and how Simuliidae research in Italy still needs efforts to improve the knowledges in systematic, zoogeography, and ecology of such a fascinating family of insects.

DRIFT BEHAVIOR OF BLACKFLY LARVAE IN CONTROLLED CONDITIONS

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Keywords: *Simuliidae, Drift behaviour, Artificial flumes*

Drift, i.e. the downstream transport of benthic organisms, is a primary driver in structuring benthic communities and ensuring distribution and colonization along the river continuity. Stream invertebrates may actively enter the water column to move away from unsuitable habitats, to avoid predators and competitors, or as a life stage requirement (i.e., active or behavioral drift), or being passively dislodged as a consequence of increased activity and/or changed positions on the bottom, or by the effect of the shear stress acting on the river bed which dislodges animals; during floods or high flows, (i.e., passive or catastrophic drift). While an extensive literature has explored many aspects of extrinsic and intrinsic drivers of drift, of its diurnal and seasonal timing and of the ecological meaning of this phenomenon, less specific attention has been posed specifically to drift in blackflies, though they frequently represent a major component of the drift. In one of the most relevant studies, by Adler et al. (1983) found rather homogeneous drift behaviour in five blackfly species from North America, with higher drift rates at dusk and lowest in the afternoon, when temperature reaches its daily maximum.

To fill some of these gaps, we used simulations in flumes where we manipulated the physical-chemical parameters, and assess the responses of blackflies to each alteration. We conducted three different experiments from 2008 to 2013 in a set of five artificial flumes (20 m long, section 0.30 x 0.30 m) fed by a near-to-pristine Alpine stream. Discharge in the flumes can be regulated by operating sluice gates, and temperature can be modified for short periods of time by releasing cold or warm water from a tank into the flumes.

A first experiment was conducted in 2008-2009, where we simulated the sudden and repeated changes in water temperature that affect rivers downstream of hydropower plants fed by high elevation reservoirs with hypolimnetic release. When turbines are operating and releasing water, the temperature of the receiving river reach increases of 3-4 C° in winter, and decreases of 3-6 C° in summer. These temperature changes occur suddenly, and both conditions (warming and cooling) were tested in the flumes. Blackflies reacted by behavioural drift in all cases, with some differences between decrease and increase in temperature. A second experiment was conducted in September 2013, aimed to assess the drift behaviour of blackfly larvae during repeated hydropeaking events (one five hour hydropeaking event repeated daily for five consecutive days), simulating the normal operation scheme of a storage hydropower plant. In this case, blackflies drifted at the very beginning of the event but then, differently from other taxa, resisted the higher water velocity and tended to increase in the drift after the return to baseflow.

A third experiment was conducted in October 2013, and focused on the distance travelled by blackfly larvae following a physical disturbance of the substrate. In each of the five flumes we disturbed the substrate at increasing distance from the downstream end, where the drift nets were positioned. Blackfly larvae exhibited a good capacity to re-attach to the substrate, when compared to other benthic invertebrates. Again a behavioural drift was observed when conditions returned to normality, reacting to the series of disturbances. Overall, our simulations allow providing information on the intensity and mechanisms of blackflies drift in response to discharge and temperature alterations.

BLACK FLIES (DIPTERA, SIMULIIDAE) AS ECOLOGICAL AND LANDSCAPE INDICATORS OF STREAM ECOSYSTEMS IN THE URBANIZING TERRITORY OF ROME (ITALY)

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Keywords: *Simuliidae, Biological indicators, Watercourses, Human impact, Environmental quality*

The use of Simuliidae (Diptera, Nematocera) as environmental indicators was tested in streams of central Italy, where the process of urbanization around the city of Rome is rapidly modifying territories and their use. 108 sampling sites were investigated in the spring (2008-2011) to acquire data on land use, environmental variables and black fly species assemblages in rivers. A subset of mostly unimpaired sites was sampled bi-monthly in order to study natural seasonal variations and the effects of small impacts. The comparison of species sampled and those attested in literature highlighted the absence, in the collected samples, of certain previously reported species (*Simulium reptans*, *S. (Nevermannia) venum*, *S. liriense*), possibly due to both pollution of the watercourses and changes in use of the surrounding land. Correspondence analysis enabled the identification of altitude as the main factor influencing species' distribution in the different sites considered. However, quality becomes the main factor when taking into account exclusively the sites in the plains: *S. (Odagmia) ornatum* (in particular, *S. (O.) intermedium*) and the "equinum" species group (in particular, *S. (Wilhelmia) equinum sensu strictu*) are the ones that show the greatest tolerance to chemical water pollution. Furthermore, when urbanization alters both the chemistry of the waters and the condition of the surrounding territory, *S. (O.) ornatum* s.l. is the only species still present in the streams; also this species, however, is unable to endure toxic pollution and suspended solids. The mid-high course of the Aniene river and its tributaries were sampled every two months, enabling the confirmation of the spatial-temporal black fly successions observed in the central Apennine region (area belonging to the "hirtipes" group, to the "variegatum" group and to a mixed one with more species) as already reported in literature. The results also indicate the larvae of *S. (O.) ornatum* may supplant the other species of black flies in the periods in which there is greater pressure on watercourses, and in areas where the rural use is substituted by urban landscape. In conclusion, we suggest that black flies may indeed be efficaciously adopted as environmental indicators to evaluate not only the ecological conditions of watercourses, but also of the landscape surrounding rivers.

HABITAT DEGRADATION EFFECTS ON MEXICAN BLACK FLIES (DIPTERA, SIMULIIDAE)

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Keywords: *Black flies, Mexico, Habitat degradation.*

In Mexico, nearly 90 species of black flies (Diptera: Simuliidae) has been recorded, three of them (*Simulium ochraceum* s.l., *S. metallicum* s.l., and *S. callidum*) incriminated as important vectors of human diseases such as onchocerciasis or river blindness. Recently, the elimination of onchocerciasis in Mexico has been documented. However, other medical, taxonomic, and ecologic studies involving black flies are scanty. Mexican black fly species are not currently found in localities where they were recorded for the first time over 50 years ago. This is likely because of the habitat degradation effects on black flies caused by pollution of lotic aquatic systems where black flies breed, modification of locations including draining of rivers to build roadways, construction of dams or urban expansion. Those factors have had an effect on *S. bricenoi* and *S. hinmani* (urban expansion), *Tlalcomyia revelata* (roadway construction), *S. (Hemicnetha) sp. nov.* (construction of dams), and *S. gonzalezherrejon* (river's pollution).

SPATIAL ORGANIZATION OF STREAM INSECTS OF DIFFERENT FEEDING ASSEMBLAGES IN WATERCOURSES OF WEST TUVA, WITH PARTICULAR EMPHASIS ON SIMULIIDAE

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Keywords: *Simuliidae, Stream insects, Feeding assemblages, West Tuva.*

In the watercourses of the West Tuva, the communities of stream insects, including black flies, caddisflies and stoneflies, are characterized by heterogeneity in the longitudinal profile of the rivers. High-mountain, middle-mountain, and low-mountain types of communities are well distinguished. The redistribution in the structure of domination conditioned by the gradient of biotopic characteristics in the altitude-zonal regime is observed. Feeding assemblages also undergo changes. A gradual replacement of ways to food gathering, from scraping to predation in the upper reaches to collecting in the lower reaches, is caused by changes in the food base in watercourses. The larvae of amphibiotic insects feed on algae, detritus, and small insects, selecting their food items according to a size category.

ELECTROPHYSIOLOGICAL (EAG) RESPONSES OF *SIMULIUM* SPP TO HUMAN SKIN VOLATILES

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Keywords: *Simulium*, *Blackfly*, *Skin human volatiles*, *Electroantennogram*.

Black flies are found in some parts of Spain, mainly in Catalonia and Aragon regions, but until now, in Andalucía their populations were not present in enough quantities to be a problem for inhabitants. In recent years from early summer to late October, olive farmers located in Baeza area (Andalucía, South Spain) close to Guadalquivir River, are bitten by black flies.

How *Simulium* spp (Diptera: Simuliidae) females locate and choose their host for blood meals is not yet very well understood. Females of most species of black flies feed during the day and usually they bite on the upper part of the body. Odours emitted by humans are used as olfactory cues by many mosquito species and probably by *Simulium* spp too.

Electroantennogram assay is a widely used method for the detection of behaviorally active compounds. The role of volatile semiochemicals in mediating the location and selection of human hosts by blackflies was investigated using electrophysiology (EAG). Contact solid-phase micro-extraction (SPME) technique was used to collect volatiles from human back and axillae regions. EAG antennae response was recorded from female *Simulium* spp to a broad range of human skin volatile constituents, plus two general hematophagous insect attractants such as ammonium and carbon dioxide. It was confirmed that female *Simulium* spp responded strongly to ammonia and CO₂ by EAG. Additionally in this study only 6 of the 32 tested common skin human volatiles produced consistent EAG activity. The most distinct EAG responses from these compounds were obtained to aliphatic straight-chain saturated C6-C10 aldehydes: octanal, (E)2-nonenal, nonanal and decanal. At certain concentrations the skin volatile constituents 1-octen-3-ol and 6-methyl-5-hepten-2-one elicited antennal responses. Significant differences between female and male responses were obtained.

DEVELOPING A DNA BARCODING LIBRARY FOR THE IDENTIFICATION OF BLACK FLIES (DIPTERA: SIMULIIDAE) WITH EMPHASIS ON MESOAMERICA

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Keywords: DNA barcoding, COI, Simuliidae, Medically important black flies, Mesoamerica.

Certain species of black flies are of medical importance because they transmit nematode species that cause onchocerciasis and mansonelliasis. Others can transmit *Leucocytozoon* to both domestic and wild birds, and are thought to be responsible for outbreaks of Endemic Pemphigous Foliaceous and Altamira Hemorrhagic Syndrome. Thus, correct morphological identification of black fly species is paramount for control and epidemiological studies. However, their taxonomy is extremely difficult due to their morphological homogeneity. In this study, the utility of a partial sequence of the COI gene, the DNA barcoding region, for the identification of species of black flies from Mesoamerica was assessed. For representative samples of morphologically identified species, DNA was extracted and a 656bp fragment of CO1 was PCR amplified and sequenced. In total, 32 morphospecies were analyzed, one species in the genus *Gigantodax* and 31 species in the genus *Simulium*, subgenera (*Aspathia*, *Eusimulium*, *Nolepria*, *Psaroniocompsa*, *Psilopelmia*, and *Trichodagmia*). In the Neighbour Joining analysis tree (NJ) derived from the DNA barcodes most of the specimens grouped according to species or species groups as recognized by other morphotaxonomic studies. The intraspecific genetic divergence within morphologically distinct species ranged from 0.07% to 1.65%. Higher values of genetic divergence (2.05% - 6.13%) in species complexes suggest the presence of cryptic diversity. The existence of well-defined groups within *S. callidum*, *S. quadrivittatum* and *S. samboni* revealed the possibility of cryptic species within these taxa. In addition, the suspected presence of sibling species within *S. paynei* and *S. tarsatum* is supported. DNA barcodes also showed that specimens from species that were taxonomically difficult to delimit such as *S. callidum*, *S. ochraceum* s.l., *S. pseudocallidum*, *S. trivisi*, relatives of the species complexes such as *S. metallicum* s.l. (e.g. *S. horacioi*, *S. jobbinsi*, and *S. puigi*), and *S. virgatum* s.l. (e.g. *S. paynei* and *S. tarsatum*) grouped together in the NJ analysis, suggesting the validity of their species status. DNA barcoding combined with a sound morphotaxonomic framework provided an effective approach for the delineation of medically important black flies species in Mesoamerica and for the discovery of hidden diversity in this group.

BLACKFLIES OF THE MARITIME ALPS

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Keywords: *Simuliidae*, *Maritime Alps*, *Chromosomal studies*, *Molecular studies*

Although the Maritime Alps belong to the most important biodiversity hotspots of the Mediterranean Basin, the black fly fauna of this area has not yet been thoroughly studied. Black flies were collected in the Maritime Alps from 2008 to 2013. The black fly fauna of the studied area is rich, often consisting of mountain species typical for central Europe. The most frequent and most abundant species within the analyzed samples were *Prosimulium rufipes* (Meigen, 1830), *Simulium argenteostriatum* Strobl, 1898, *S. argyreatum* Meigen, 1838 and *S. variegatum* Meigen, 1818. Morphological, cytotaxonomic and molecular study of the material revealed identity problems for most of the present species. Based on morphological characters, three species of the genus *Prosimulium* were recorded, and banding patterns of the polytene chromosomes of 71 larvae were analyzed. *Prosimulium latimucro* (Enderlein, 1925) was found at the highest elevations. Chromosomal characteristics of *P. latimucro* from the Maritime Alps differed from those of populations from the Pyrenees, the Swiss Alps, Scotland and Morocco, and probably represent a separate cytoform; conspecificity with *P. latimucro* from the type locality has to be tested. Black flies of the *P. rufipes* complex were found at all elevations from April to September. Chromosomal characteristics of 21 larvae of the *P. rufipes* complex from the Maritime Alps showed that all analyzed specimens are close to the species chromosomally characterized previously from Austria as *P. inflatum* 'aff. 3'. Individuals morphologically identified as *Prosimulium hirtipes* (Fries, 1824) were found usually not at the highest elevations and mostly not later than June, no chromosomal data are available for *P. hirtipes* from the Maritime Alps. Morphological and molecular analyses indicate either large intraspecific variability or unclear identity of several species within *Simulium* s. str. Further taxonomical study is needed.

***PROSIMULIUM HIRTIPES* IN EUROPE:
DOES *PROSIMULIUM ITALICUM* EXIST?**

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Keywords: Simuliidae, *Prosimulium hirtipes*, *Prosimulium italicum*, *Cytoforms*

Prosimulium hirtipes (Fries, 1824) – the type species of the genus *Prosimulium* – was originally described from Sweden. Later the species was reported from the majority of European countries, including all parts of the continent. The distribution extends over northern and central Russia up to China. The subspecies *P. h. italicum* Rivosecchi, 1967 was described from southern Italy, and putatively differs in the shape of the gonofurca and in small differences in the larval hypostoma. Currently, it is considered a synonym of *P. hirtipes*. No other synonymic names were identified in Europe. Larvae and pupae morphologically identified as *P. hirtipes* collected in northern Sweden, West Carpathians, Alps, Apennines and Sicily were studied. Those populations were characterized morphologically and cytotaxonomically and compared with existing data. Few morphological differences between the populations were found. Chromosomal characteristics of the population from Sicily differed from those of populations from Great Britain and central Europe, and probably represent a separate cytoform) corresponding to *P. italicum*. The chromosomal difference between Great Britain and central Europe indicates that *P. hirtipes* in Europe is probably a complex of cytoforms.

18 SEPTEMBER 2014

**THE LARGE-SCALE MAPPING OF BLACKFLY BREEDING SITES
SHOULD BE A PREREQUISITE FOR BLACKFLY CONTROL
OPERATION THAT USE AIRCRAFT TO DISPERSE LARVICIDES TO
TARGETED WATER-COURSES, BASED ON EXPERIENCES GAINED
FROM THE WHO ONCHOCERCIASIS CONTROL PROGRAMME IN
WEST AFRICA**

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In 1973 a WHO Interim Project (operating in western Burkina Faso, northern Côte d'Ivoire and western Ghana) bridged the period between the completion of a WHO Preparatory Assistance Mission to Governments (PAG) and the launching of the WHO Onchocerciasis Control Programme in West Africa (OCP) in 1974.

Early in the Interim project (IP) it became apparent that the maps available to the author were inadequate for him to plan, execute and evaluate a critical aerial spraying trial, involving the application of temephos from a helicopter, to control larval populations of *Simulium damnosum* s.l. that thrived in the Bui Gorge area of the lower Black Volta River in western Ghana (close to the Côte d'Ivoire). The author was thus obliged to digress from his other duties to conduct a low-level aerial survey of the lower Black Volta and then quickly construct a large-scale base map which would allow him to proceed with the temephos trial. A copy of the original 1973 report of that trial and of the Black Volta River will be made available for examination by Symposium participants.

During the author's employment in the OCP, he made more than 25 large-scale maps and produced illustrations for more than a dozen publications/WHO reports. Of particular value were his maps of the very large and complex rapids of the lower River Bandama (Côte d'Ivoire), where blackfly resistance to temephos was first detected, and where a very special series of maps had to be produced in order that B.t.i. could effectively control the temephos –resistance problem. Copies of those maps will be on display during the Symposium, and map "collectors" will have the opportunity to acquire digitized copies of them, in addition to discussing how they came into existence.



Massive S. damnosum s.l. breeding site, Buy area, Ghana, 1973



As part of the WHO Onchocerciasis Control Programme in West Africa (OCP), “Heliporter” aircraft made “rapid-release” applications of “Abate” (temephos EC 20%) to the R. Comoé in northern Côte d’Ivoire, to control the larvae of Simulium damnosum, s.l., which were abundant in the rapids, such as those discernible ahead of the aircraft shown in this photograph.

The width of the river at the larvicide release point was estimated to be 50 to 60 metres. Some riverine forest trees attained heights of ~40 metres. The wingspan of the aircraft was ~16 metres.

Photographed from a Bell-206 helicopter by David Baldry, VCU/CAO.

DB “takeMS” G: Comé FW release 1975. Revised May 2013.

LIVING WITH THE FLY video

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The film shows activities of the EcoHealth Project in Ghana, sponsored by the International Development Research Centre (IDRC) of Canada from 2009 to 2012. The project entitled “Eco-Health Approach to the Control of Onchocerciasis in the Volta Basin of Ghana” investigated likely effects of climate change on onchocerciasis transmission and control in the Volta Basin, using a multi-disciplinary approach involving biologists, hydrologists, climate scientists, economists and social scientists. After explanations of onchocerciasis and how it is transmitted by *Simulium damnosum* s.l., interactions between scientists and health workers with affected communities are shown. Emphasis is also placed on the biting nuisance of the flies and how this interferes with farming and other economic activities, against which simple mitigation measures such as the use of repellents and the wearing of suitable clothing can be utilised to deter flies from biting. Sequences on ivermectin distribution and its associated problems such as low coverage, non-responding cases and difficulties in filling vacant teaching and medical posts in areas affected by onchocerciasis are shown. Predictions of erratic rainfall, more storms, higher temperatures and less distinct separations of dry from wet seasons in the near future are made. How such climate changes are likely to exacerbate onchocerciasis transmission, by reducing fly development rates and increasing their populations, are explained. The use of radio, mobile telephones, workshops and drama groups to disseminate messages about the disease and its mitigation are also illustrated.

ONCHOCERCIASIS TRANSMISSION AND CLIMATE CHANGE IN WEST AFRICA

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Keywords: *Climate change, Simuliidae, Onchocerciasis, West Africa*

Climate change models predict that by 2040 temperatures in West Africa will have increased by at least 1 or 2 °C, rainfall will have become more erratic and there will be more storms and extreme weather events compared with a 1961-1990 baseline. How such changes may affect onchocerciasis transmission in West Africa was examined by developing population models for *Simulium damnosum* s.l. using three approaches: (i) regression models derived from analyses of empirical data linking blackfly numbers to rainfall and temperature in Ghana and Liberia; (ii) matrix models using field data from Ghana and Togo and (iii) ordinary differential equations, parameterised with data from studies of the bionomics of both blackflies and *Onchocerca volvulus*. The latter studies included deriving quantitative relations between temperature and development rates of eggs, larvae and pupae of *S. damnosum* s.l. and of *O. volvulus* within flies. Values for other parameters such as vector mortality rates were taken from the literature. Results from (i) suggested that blackfly numbers are likely to decrease by 13-41% with the advent of climate change but predictions from (ii) and (iii) both indicated increases in blackfly numbers and onchocerciasis transmission before 2040. Output from (iii) forecasts that fly numbers in Liberia and Ghana would peak at air temperatures of 29 and 34°C, about 3 and 7°C above current monthly averages respectively, and that transmission rates and worm loads will increase with temperature up to at least air temperatures of 33°C.

HOW TO MAKE A SIMULIID PEST

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Keywords: *Simuliidae, Pest status.*

Pest status is not a biological trait of organisms. It is the result of human interests conflicting with organismal characteristics. The factors that promote pest status are poorly understood, and the evolutionary innovations that set the scene for pest problems are generally unknown. The extrinsic factors associated with pest status of black flies, such as nutrient enrichment of larval habitats, have been more easily studied than the intrinsic, or genetic, factors. If we understand how certain species come to be pests, we can better manage existing pest problems and avoid the development of new pests. Pest status for a species of black fly is typically achieved by one of two paths: specialization to colonize the world's largest rivers or generalization to colonize a wide range of flowing waters from small streams to large rivers. Each scenario involves particular adaptations, and in both cases, the result is the same: production of enormous numbers of host-seeking female black flies. Examples of the evolution of these adaptations are highlighted with examples of species such as *Simulium venustum* in North America and *Simulium colombaschense* in Europe. Their numerous cytoforms, not all of which are pests, provide particularly compelling evidence for the evolution of traits associated with pest status.

BLACKFLIES AND HYDROPOWER: THE BUI DAM PROBLEM ON THE BLACK VOLTA RIVER, GHANA

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Keywords: *S. damnosum*, *Onchocerciasis*, *Bui dam*.

Construction of the Bui dam at 08° 16' 38"N, 02° 14' 02"W on the Black Volta river in the Brong-Ahafo Region of northern Ghana, where onchocerciasis is endemic, has been completed and its hydro-electric generating system became operational in December 2013. Although the lake created by the dam, with a maximum area of 144 km², has swamped many breeding sites of *Simulium damnosum* s.l., conditions remain ideal for breeding downstream of the dam and will be permanent. The dam has three main turbines, each with a maximum capacity of 205 m³.sec⁻¹. In addition, there is an ecological discharge turbine, with a maximum capacity of 20 m³.sec⁻¹. This allows some passage of water when the other turbines are turned off and can act as an emergency system for "black starts". At present one turbine is running continuously and a second is used only for about 5 hours from 1700 to 2200 hours to provide electricity during times of peak demand. Therefore the river level is constant for most of the day but rises to a new height every evening before subsiding again. Implications for onchocerciasis transmission and suggestions on how to control blackflies in the Black Volta river basin, with the advent of the Bui dam, will be considered and discussed.

CONTRIBUTION TO THE BLACK FLY FAUNA (DIPTERA: SIMULIIDAE) OF THE SAKHA REPUBLIC (YAKUTIA)

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Keywords: Simuliidae biodiversity, Yakutia

Republic of Sakha (Yakutia) is the largest region of Eastern Siberia (3,083,500 km²). Tundra and different types of taiga zones (north, middle and mountain types) are widely represented there. Considerable areas of Yakutia are covered by mountain landscapes. The summed length of the largest rivers (Lena, Yana, Indigirka and Kolyma) exceeds 2 million km. A dense net of watercourses provides favorable conditions for blackflies habitation.

Our own material on blackflies was collected from July to August 2011-2013 in four districts of Yakutia. Data from the collections of the Zoological institute RAS and the Institute of biological problems of Criolytozone SB RAS was also taken for the analysis. All literature sources that contributed to blackfly fauna of Yakutia were also analyzed. As a result, the fauna of Yakutia was supplemented with 4 new previously unmentioned species one of which was known only from Nearctic - *Schoenbaueria furculatum* Shewell, 1952.

According to different sources (literature, collections and our own material) there are 89 blackfly species in Yakutia. 18 species (20%) are from Holarctic and Asian-North American areas, 4 species (4,5%) – Holarctic European-Asian-Northamerican, 4 species (4,5%) – Palaearctic, European-Asian-North African, 23 species (26%) – Palaearctic European-Asian, 40 species (45%) – Palearctic Asian. Among the latter, 19 species are endemics of Russia and 10 – endemics of Yakutia.

There is only one species of the family (*Archesimulium vulgare* (Dorogostajsky, Rubzov et Vlasenko, 1935)) that was recorded from the basins of all big rivers of the territory. Distribution of blackflies of Yakutia across native zones is also irregular. Thus, only 30 species are known from the tundra zone and 29 from Northern taiga subzone, 60 from middle taiga subzone and 32 from mountain taiga subzone.

At the same time, only 6 species (*Stegopterna asema* Rubzov, 1956, *Archesimulium vulgare*, *Gnus cholodkovskii* (Rubzov, 1940), *G. decimatum* (Dorogostajsky, Rubzov et Vlasenko, 1935), *G. malyschevi* (Dorogostajsky, Rubzov et Vlasenko, 1935), *G. murmanum* Enderlein, 1935)) are found in all the mentioned zones. Six species (*Gymnopais andrei* Worobez, 1984, *G. bifistulatus* Rubzov, 1955, *G. frontatus* Yankovsky, 1982, *Sulcinephya derzhavini* (Rubzov, 1940), *Archesimulium polare* (Rubzov, 1940), *A. tumulosum* Rubtsov, 1956) were collected only from mountain regions of Yakutia. Four species were recorded in tundra: *Helodon czezanovskii* (Rubzov, 1956), *Prosimulium erythronotum* Rubzov, 1956, *P. jacuticum* Rubzov, 1973, *P. tiksii* Yankovsky, 2011.

23 species were collected only from plain taiga: *Greniera ivanovae* (Ivashchenko, 1970), *Helodon rhizomorphus* (Rubzov, 1971), *Metacnephia cuspidata* Worobez, 1987, *M. taimyrica* Patrusheva, 1976, *Wilhelmia equina* (Linnaeus, 1758), *Byssodon maculatus* (Meigen, 1804), *Cnetha chomustachi* Worobez, 1984, *C. dentatura* Worobez, 1987, *C. meigeni* (Rubzov et Carlsson, 1965), *Nevermannia angustitarse* (Lundstrom, 1911), *N. orestii* (Worobez, 1985), *Schoenbaueria brachyarthra* Rubzov, 1956, *S. dendrofilum* (Patrusheva, 1962), *S. gigantea* (Rubzov, 1940), *S. furculatum*, *S. rangiferina* (Rubzov, 1956), *S. tsharae* (Yankovsky, 1982), *Odagmia argyreata* (Meigen, 1838), *Parabyssodon transiens* (Rubzov, 1940), *Simulium lugense* Yankovsky, 1998, *S. paralongipalpe* Worobez, 1987, *S. paramorsitans* Rubzov, 1956, *S. rubtzovi* Smart, 1945, and 16 out of these were recorded only in the middle taiga zone.

SURVEY OF THE *SIMULIUM* SPECIES (DIPTERA, SIMULIIDAE) IN ALENTEJO, SOUTH OF PORTUGAL

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Key words: Simuliids, Alentejo, Portugal, Diversity.

The study of simuliids in Portugal has not been carried out during the last twenty five years. In the present, the distribution of simuliid species in temporary streams is being observed in Alentejo (South of Portugal). The preliminary results from this survey representing the collections of immature stages of simuliids from thirty different sampling places, were carried out between March and July 2014, being a contribution to acquire updated knowledge on blackfly fauna composition, as well as better understanding of the distribution preferences and relations with some associated environmental factors. Up to the moment, nevertheless the diversity of identified simuliids, two species (*Simulium* (S.) *ornatum* Meigen, 1818 (complex) and *Simulium* (*Wilhelmia*) *pseudequinum* Séguy, 1921) have been the most frequently found species. Alentejo is a remote area (with only 5% of the Portuguese population), and Alqueva (the biggest artificial lake in Europe), where the availability of water is changing in order to mitigate influence of climate change, agricultural and other human activities, that may result in new conditions for simuliid development. In the context of the growing awareness of biodiversity preservation, it is expected that the ongoing work will contribute to the knowledge of the Simuliidae in Portugal.

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OBSERVATIONS ON SIMULIIDS IN ALGARVE, SOUTH OF PORTUGAL

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Key words: *Simuliidae*, Algarve, Portugal

Algarve (southern Portugal), has a high seasonal importance for tourism, with summer being the season of peak human density, due to the good weather, its pleasant beaches and natural landscape. Furthermore, livestock production traditionally is playing an important role in the economy of Algarve region. Simuliids, commonly known as black flies (family Simuliidae) are important insects, due to their direct parasitic activity, as blood-feeders and having allergenic saliva, and by being vectors of pathogens both to humans and animals. As causing nuisance and due to biting activity, they may represent a significant threat to human and animal health, affect the tourism and sport activities, benefit of domestic animals and wildlife and cause significant losses in the economy of the region. In the present work immature stages of simuliids have been collected in the past two years (2013/2014), in nine different sampling places in the Algarve. The collected specimens were identified, to explore the relevant breeding places in the area under study. The preliminary results from the present ongoing survey, revealed the presence of *S. intermedium* Roubaud, 1906, *S. ornatum* Meigen, 1818 (complex), *S. pseudequinum* Séguy, 1921 and *Simulium velutinum* (Santos Abreu, 1922). Considering that Algarve region presents a well-defined raining season (during winter time), it might be expected that simuliids show a strong seasonal abundance outside the touristic season. However, due to the global climatic changes and human activities, the dynamics of simuliid populations in southern Portugal may be altered.

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**UPDATE OF THE BLACKFLY FAUNA OF NORTH-AFRICA (DIPTERA :
SIMULIIDAE)**

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Keywords: *Simuliidae, Blackflies, North-Africa, Algeria, Tunisia, Morocco, Fauna, Taxonomy, Chromosomes.*

The total number of species of Simuliidae currently known from North-Africa is 82, of which 42 identified species are recorded from Morocco, 27 species are listed from Algeria and 13 are cited for Tunisia.

A precise comparison is made between the representative Simuliidae fauna of each country and the latest progresses of research on North-African Blackflies are presented and commented.

NOTES ON BLACKFLY FAUNA OF THE BASENTO RIVER (BASILICATA, SOUTHERN ITALY)

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Keywords: Simuliidae, Basento, Basilicata

As reported by Leo Rivosecchi (2005), the alteration of agricultural practices and changes of landscape aspects during the last few decades caused the spreading of blackfly species of the *ornatum* species group and the remarkable suppression of populations of the *equinum* species group throughout Italy.

A short faunistic study was conducted during the period May 30th -June 5th 2013 in the Gallipoli Cognato Regional Park (Basilicata, southern Italy), in conditions of well preserved natural environment with ongoing traditional livestock production practices. Immature stages of blackflies were collected from the Basento river at two localities (Campomaggiore and Grassano) and from one confluent creek, Torrente della Rossa (Campomaggiore). Adults were collected by application of dry ice baited traps positioned close to the river banks at both localities. During the periods of swarming, adults were also captured by the sweeping net. Furthermore, in vicinity of the breeding sites, females taking the blood meal were collected from horses and dogs by entomological aspirator.

In the Basento river, where highly productive blackfly breeding sites were recorded, six blackfly species were identified: *Simulium* (*Wilhelmia*) *pseudequinum* Séguy, 1921, *S. (W.) equinum* (Linnaeus, 1758), *S. (W.) lineatum* (Meigen, 1804), *S. (Similium) intermedium* Roubaud, 1906, *S. (S.) ornatum* Meigen, 1818 (complex) and *S. (Eusimulium) velutinum* (Santos Abreu, 1922). In the Torrente della Rossa creek *S. intermedium* and *S. velutinum* were also recorded, but in low population density.

Comparing the list of identified species in this study with the last check list of the Italian blackfly fauna given by Rivosecchi in Ruffo & Stock (2005), all of the recorded species except *S. pseudequinum* represent the first records for the Basilicata region.

The three species of *Wilhelmia* subgenus, with the total portion of about 80% of the collected material in immature stages, represented the dominant blackfly taxa, with *S. pseudequinum* as the dominant species (57,86%). On the other hand, the population densities of other three recorded species was much lower (below 7,55%).

In accordance with the observations of the fauna composition in the breeding sites, *S. pseudequinum* was the dominant species collected by traps, sweeping net and from animal hosts. The biting activity of *S. pseudequinum*, *S. intermedium*, *S. ornatum* (complex) and *S. velutinum* was confirmed on horses, while *S. pseudequinum* and *S. ornatum* (complex) attacked dogs as well.

The results of the studied part of Basilicata region demonstrated that the well preserved natural environment and the permanent availability of natural hosts (especially freely grazing animals), represent excellent conditions for the production of abundant populations of mammophilic blackfly species of *Wilhelmia* subgenus.

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BIODIVERSITY OF BLACK FLIES (DIPTERA: SIMULIIDAE) IN THAILAND

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Keywords: Black fly; Biodiversity; Simuliidae; Thailand

A black fly (*Simulium nigrogilvum*) in Thailand was first reported in 1911 by Summer. More than a century later, 91 morphospecies arranged in six subgenera (*Asiosimulium*, *Daviesellum*, *Gomphostilbia*, *Nevermannia*, *Montisimulium*, *Simulium*) of the genus *Simulium* were recorded. The most diverse subgenera are *Simulium* and *Gomphostilbia* represented by 43 and 26 species, respectively. Cytological studies have been conducted in 29 species, of these, five species (*S. tani*, *S. feuerborni*, *S. angulistylum*, *S. siamense*, *S. doipuiense*) were found to be species complexes with the numbers of cytoforms ranging from two to ten. Further cryptic diversity was revealed by molecular genetic studies. Molecular divergent lineages were often found in both well examined cytogenetic species and species that has not yet been cytologically screened. Thus, integration of morphology, cytology and molecular biology is necessary for fully understanding of black fly biodiversity. Ecological studies revealed that large, fast flowing and cool streams with the presence of riparian vegetation support greater species diversity. Population genetic studies of the Thai black flies found that both historical events (i.e. Pleistocene climatic change) and ongoing processes (ongoing gene flow, geographic barrier) play significant roles in determining genetic structure and diversity.

BLACKFLY FAUNA OF MACEDONIA (NORTHERN GREECE)

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Keywords: *Simuliidae*, Macedonia, Greece

The research of the blackfly fauna composition of northern Greece which started in 2011 and 2012 in the region of Thrace has been extended to wider territory of the region of Macedonia in summer 2014.

Samplings were conducted in 33 water courses of Macedonia (creeks, streams and rivers), at altitudes from 70 m.a.s.l. to 1135 m.a.s.l. and water temperatures of 8,1 C to 28,9 C. ° Immature stages of blackflies were collected at 28 of them, at altitudes ranging between 70 m.a.s.l. and 692 m.a.s.l. and water temperatures of 13 C to 26,9 C.

The list of identified fauna within the present study consists of ten species: *Simulium* (*Simulium*) *ornatum* Meigen, 1818 (complex), *S. (S.) reptans* (Linnaeus, 1758), *S. (S.) bezzii* (Corti, 1914) (complex), *S. (S.) variegatum* Meigen, 1818, *S. (S.) argyreatum* Meigen 1838, *S. (Wilhelmia) pseudequinum* Séguy, 1921, *S. (W.) balcanicum* (Enderlein, 1924), *S. (Trichodagmia) auricoma* Meigen, 1818, *S. (Eusimulium) velutinum* (Santos Abreu, 1922) and *S. (Nevermannia) angustitarse* (Lundström, 1911). The most widespread species of the region recorded in majority of the water courses was *S. ornatum* (complex), followed by *S. bezzii* and *S. pseudequinum*. The finding of *S. argyreatum* and *S. angustitarse* in the region of Macedonia represents the first record of the presence of these two species in Greece.

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POSTERS

PAROUS RATES OF *SIMULIUM DAMNOSUM* S.L. ARE DENSITY DEPENDENT

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Keywords: *S. damnosum*, *Parous rates*

A correlation between parous rates and an index of adult numbers of *Simulium damnosum* s.l. indicates an association, but it does not prove causality nor show the direction of any causal relationship. The question of whether the adult numbers affect the parous rates or if it is vice versa is equivalent to the age old query of deciding which came first, the chicken or the egg? A method for resolving such issues based on analyses of pairs of time series was proposed in 1969 by Sir Clive W. J. Granger, who won the 2003 Nobel Prize for Economic Sciences for his research on econometrics. When Granger's method was applied to monthly numbers of adult female *Simulium damnosum* s.l. caught attempting to bite at Asubende, Ghana, and their parous rates, a significant relation ($P < 0.005$) was obtained, clearly showing that the parous rates are dependent on the adult numbers. Implications of this density dependence and results of analysing other series will be presented and discussed.

Reference

Granger, C.W.J. (1969) *Investigating causal relations by econometric models and cross-spectral methods. Econometrica* 37: 424-438.

THE SIMULIIDS (DIPTERA: SIMULIIDAE) OF NORTH AFRICA: NEW DATA ON DISTRIBUTION OF THE ALGERIAN FAUNA

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Keywords: *Algeria, Biodiversity, Simuliidae, Seybouse basin.*

Black flies constitute a large and important family (Simuliidae) of Diptera with more than 2000 species described in the world. The significance of these insects is manifested in both aquatic and terrestrial ecosystems and as economically important pests and vectors of disease agents to birds and mammals, including domestic animals and humans. Although the blackfly fauna of the Palearctic region is fairly well known, the knowledge of Algerian simuliids is not sufficient. To obtain a more accurate data of the North African simuliidae fauna, a survey of the main North Eastern Algerian hydrographic network Seybouse was conducted between 2011 and 2013 in several sampling sites located on the main river and its tributaries for which the Mediterranean character favors a significant simuliidae fauna. An annotated list of recorded species is provided. The biogeographical analysis shows that we are essentially dealing with a palearctic fauna with a West or circumediterranean distribution, Afrotropical elements are poorly represented.

SIMULIIDAE (DIPTERA, NEMATOCERA) IN THE AGRÍ VALLEY (BASILICATA, SOUTH ITALY)

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Keywords: Simuliidae, Agri valley, River biodiversity, Extractive activities

The increasing exploitation of oil reserves in the Agri river valley (Val d'Agri), in the province of Potenza, is raising serious concerns on the status of the local environment, extraordinary well preserved in that area (two National Parks and a Regional Park are present) but threatened by oil extractive operations and related human activities. In 2012-13, ENEA realized three campaigns of field activities, with water physical-chemical analyses, bio-indicator sampling and zoological studies in order to characterize the biodiversity and ecological status of the watercourses in Val d'Agri, considered to be the most susceptible environment to the ongoing pressures. The collection of Simuliidae was performed on late winter, late spring and autumn 2013, in 7 sampling sites representing almost all the course of the river: Agri0 and Agri4 in high mountain, Agri1 and Agri2 in upper Agri valley, Agri3 and Agri5 in the medium reach and Agri6 in the lower course.

A total of 9 species were identified from larvae, pupae and adults, with a considerable number of larvae too small to be identified to species. The highest number of species for site was 4, which was totalized only in spring collections (2 times: Agri1, Agri5). 2 was the highest species number per site in summer (when water scarcity was stronger) and 3 in winter. The commonest species was *Simulium (Odagmia) ornatum* s.l. in all the seasons (5 sites in spring, 4 in summer and 3 in winter) but also *S. (Nevermannia) cryophylum*, *S. (O.) variegatum* and *S. (Eusimulium) velutinum* were quite common. In spring, *S. (Wilhelmia) equinum* was collected in three sites (southern record in Italy, first in Basilicata), while only Agri6 presented this species in summer and autumn. *S. (W.) pseudoequinum* was collected only once in autumn, while *S. (S.) argyreatum* was only collected in Agri4. *Prosimulium hirtipes* was only collected in spring samplings (2 sites), while *S. (N.) angustitarse* was collected only in summer (Agri5). The unidentified larvae belonged to the groups "monticola", "vernum", "aureum" and "equinum", all of them already represented in the identified material (so that no other species may be expected in the samples).

The good diversity of species in a single river basin (compared to similar area basins in Central Italy) suggests a fair conservation of blackfly biodiversity in the Agri river, but the dominance of *S. ornatum* s.l. in most of the sites (especially Agri1 and Agri2) represents an alert on the simplification of Simuliidae assemblages in sites impaired by agriculture (Agri1) and domestic pollution (Agri2) due to the presence of increasing human productive activities in a still valuable environmental context.

ON THE DISTRIBUTION OF *SIMULIUM LUNDSTROMI* (ENDERLEIN, 1921) (DIPTERA: SIMULIIDAE) IN HUNGARY

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Keywords: *Freshwater monitoring, Water Framework Directive, Lowland blackfly species, Hungarian blackfly distribution*

Due to the monitoring of freshwaters conducted in recent years and prescribed by the Water Framework Directive (WFD), more and more distribution data are available for different blackfly species from Hungary. Lowland and hill watercourses are among the best studied areas and present more data on simuliids than from the mountain regions. In Hungary, the species *Simulium lundstromi* was collected mostly from slowly flowing, weedy lowland streams, but some occurrences from the hill zone are also reported mainly on the western part of the country. Based on the previously identified samples *S. lundstromi* can be considered as a moderately common blackfly species in Hungary.

INVESTIGATION AND MOLECULAR CHARACTERIZATION OF *SIMULIUM* SPECIES IN THE STREAMS OF ERCIYES MOUNTAIN IN TURKEY

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Key words: Black fly, Erciyes Mountain Streams, Molecular characterization, Simuliidae

This study was carried out to determine the molecular characterization of simuliid species in the small streams of Erciyes Mountain and financially supported by the Research Fund of the Erciyes University with the project number TYL-2013-4317. Between May and September 2013, simuliid larvae sampling was conducted in the research area. Morphological and chromosomal examinations of the collected larvae were performed before molecular analyses. A total of 9, 5, 2 and 1 of the examined larvae were identified as *Simulium bezzii* cytoform (Cyt) 2, *S. bezzii* Cyt 3, *S. kiritshenkoi* and *S. velutinum*, respectively. For molecular analyses of the *Simulium* species determined in the research area, the genomic DNA extractions were performed on one individual larva from each simuliid species or cytoform. PCR analyses were performed with the primer pairs that amplified mitochondrial cytochrome oxidase subunit 1 (mt-COI) and ribosomal internal transcript spacer 2 (ITS-2) gene regions. The obtained amplicons from the isolates were cloned and plasmids were obtained after the purifications. Sequence analyses were performed on the purified plasmids; unique nucleotide sequences of the isolates were determined and deposited in GenBank with the accessions KJ880275-KJ880280. Phylogenetic analyses of the mt-COI sequences from the *S. bezzii* cytoform 2 (TrERUSimbe2) and cytoform 3 (TrERUSimbe3) isolates revealed a 0.8% genetic difference from each other. In addition *S. bezzii* isolates showed 0.6±0.2% mean genetic difference with the other *S. bezzii* isolates available in GenBank within the same gene region. The data on the molecular characterization of *S. bezzii* based on ITS-2 gene region was first established with this study. ITS-2 characterization of *S. kiritshenkoi* also is the first record in the World and this isolate (TrERUSimor) showed maximum identity (99.7%) with some of the *S. trifasciatum* and *S. ornatum* isolates, which are also in the *ornatum* group, recorded from England. The *S. kiritshenkoi* (TrERUSimor) isolate was also 99.5% identical to the *S. kiritshenkoi* Sim39-kir isolate from Armenia based on the mt-COI phylogeny. *S. velutinum* TrERUSimv isolate showed maximum identity (98.9%) with the *S. aureum* Sim27-au isolate recorded from Armenia, while 4.5±0.8% mean genetic difference was determined with other *S. velutinum* isolates reported from England and Finland.

**MONITORING AND CONTROLLING THE DIFFUSION OF DIPTERA
SIMULIIDAE, IN THE SPRING WATERS AREA OF THE FRIULI
VENEZIA GIULIA REGION**

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Keywords: *Spring waters area; Bacillus thuringiensis; diffusion of Simuliidae.*

Since 1984, the presence of Diptera Simuliidae have been reported in the spring waters areas of the low Friuli plain. This presence is of particularly concern for the local population. In fact, the well oxygenated and nutrient-rich spring waters of Friuli represent an ideal environment for the completion of the entire reproductive cycle of these Diptera. Previous studies aimed to identify the possible causes and mitigations of this phenomenon (Mattassi and Rivosecchi, 1986). Two species were found responsible for the harassment, *Simulium erythrocephalum* and *Simulium paraequinum* and the massive nutrient inputs originating from agricultural sources was suggested to be the main cause of this abnormal development.

In order to solve this problem, some tests have been conducted by treating resurgence streams and irrigation ditches with an endotoxin of *Bacillus thuringiensis* serovar israelinesis based product. Such selective treatment caused the simuliids' density decrease. On the other hand, it affected only the first phase of development. Actually, other treatments are still under investigations and show a decrease in the simuliids proliferation. The last research in this area) was conducted in 2009 and revealed the presence of four species: *Simulium erythrocephalum*; *Simulium paraequinum*; *Simulium ornatum* and *Simulium lundstromi*. For each monitoring site the percentage abundance of each species and the related water physical-chemical conditions were assessed.

STUDY ON BLACK FLIES (FAMILY: SIMULIIDAE) IN SOUTH-WEST OF IRAN, 2014

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Key words: *Blackfly, Simuliidae, Iran*

Since the preliminary researches on blackflies in Iran, more than 30 years have passed and it can be argued that at this time some of the most unknown important medical insects in Iran are blackflies.

In this study we studied 50 specimens in three point of South-west of Iran, where no studies have been done on Simuliidae family, until now. We could identify 6 species between 2 stages larvae and pupae. There were *Simulium brachyantherum*, *S. variegatum*, *S. fontanum*, *Metacnephia persica*, *S. lineatum*, *S. assadovi*. The specimens were collected on Carnoy's fixative solution at -20 C°. Identification has been done using Crosskey's keys (2002).

DETAILED MAPS OF THE GEOGRAPHICAL DISTRIBUTION OF THE BLACKFLIES OF SPAIN BASED ON A LITERATURE REVIEW

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Keywords: *Simuliidae, Spain, Distribution, Ecology*

Blackfly populations in Spain have experienced a growing expansion in recent years, which has led to a significant increase in nuisances and caused serious damage in the quality of life in areas where these insects have become a real pest (Ruiz-Arrondo et al., 2014). The first paper about blackflies in Spain was published in 1888 by Antiga (1912); later publications focused on their taxonomy and distribution (Gonzalez, 1997). Although blackflies are increasingly taking an important role in the country due to its impact on public and animal health, there is still a dearth of studies published about this group to date. For this reason, the first step is to do a systematic literature review about the presence of Simuliids in our country. This paper presents the preliminary results of the bibliographical research on the ecology and distribution of a total of 53 species of 5 genera (Adler & Crosskey, 2014): *Prosimulium* Roubaud, 1906; *Urosimulium* Contini, 1963; *Greniera* Doby & David, 1959; *Metacnephia* Crosskey, 1969 and *Simulium* Latreille, 1802.

MOLECULAR TAXONOMIC ANALYSIS OF *PROSIMULIUM RACHILIENSE* DJAFAROV 1954 (DIPTERA: SIMULIIDAE) POPULATIONS IN WESTERN ANATOLIA

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Keywords: *Simuliidae, Phylogeny, Prosimulium, COI, Turkey, Blackfly*

Molecular methods, such as DNA barcoding, have been used to understand relationships between populations or between closely related species and also show utility for revealing cryptic diversity. This is also one of the best methods for confirming taxonomic identification. *Prosimulium rachiliense*, also recorded as the synonym, *pronevitschae*, is the most common species of *Prosimulium hirtipes* group in Turkey and is widely distributed in the western part of the country. Cytotaxonomical studies on its Anatolian populations revealed that it is a different cytoform from the populations in neighboring countries such as Armenia. The aim of the present study is to investigate the taxonomical status of the species in different populations by using DNA barcoding methods.

In the present study, black flies were collected at six different localities in Middle-West Turkey (Eskişehir, Kütahya and Denizli provinces) in 2013. DNA extractions were carried out and 695-bp region of the mitochondrial COI gene was amplified by polymerase chain reaction (PCR) using the universal primers.

After sequence analysis was procured, results were compared with the international data bank and 91 % - 94 % percentage overlap was found with the species *Prosimulium frontatum* Terteryan, 1956. The phylogenetic tree of six different populations as constituted with Neighbor joining method. With this study, one of the *hirtipes* group species has been explored using molecular taxonomy methods.

**EXAMINATION OF *METACNEPHIA* NEAR *LYRA* (DIPTERA:
SIMULIIDAE) WITH MOLECULAR TAXONOMY METHODS FROM
MIDDLE-WEST OF TURKEY**

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Keywords: *Molecular, Taxonomy, Simuliidae, Metacnephia, Turkey, COI*

Studies in biodiversity, systematics, community ecology and biomonitoring also depend on proper taxonomic identification. Misidentifications could cause wrong results. Using a small section of the mitochondrial gene cytochrome c oxidase unit I (COI) as a DNA barcoding for species identification could give us a more accurate result.

This study was carried out with *Metacnephia* species collected from various rivers from Eskişehir, Kütahya and Burdur provinces. The species identified as *Metacnephia* near *lyra* had its Mitochondrial COI gene amplified by polymerase chain reaction (PCR) using the universal primers from all populations.

DNA (COI gene section) sequence analysis was compared with the international data bank (NCBI) and results overlapped with *Metacnephia lyra* with 93% percent. As a result of this study taxonomical identification has been assisted with molecular taxonomic methods.

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ERRATA CORRIGE

Abstracts: changes

- Pag 8 Poster contribute
Pag 8 Manuel Elías-Gutiérrez not in the list of authors
Pag 8 **Quetzaly K. Siller-Rodríguez** added to the list of authors
Pag 26 **Peter H. Adler** added to the list of authors as second name.
Pag 40 **Peter H. Adler** added to the list of authors as Last name.
Pag 52 and 53 Two abstracts added

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BLACK FLIES AND THE COLORADO RIVER, USA – THE BULLHEAD CITY EXPERIENCE

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Keywords: Colorado river, *Simulium tribulatum*, *Bti*, Pest control

The Colorado River is the largest river in the Southwestern United States with a length of 2,334 km and a basin of 640,000 km². The river has a reported average flow of 637 m³/sec, but is known for wide ranges of flow. Today, the river has several large impoundments and provides extensive volumes of water for agriculture and nearly 40 million people, significantly reducing its flow. A river of this magnitude has probably produced large black fly populations throughout time. However, the construction of the Davis Dam near Bullhead City, Arizona, contributed to the development of a significant nuisance population.

Black flies have been reported as a nuisance in the Bullhead City, Arizona, and Laughlin, Nevada, area since the late 1960s, with the population becoming a severe nuisance problem in the early 1970s. This area is immediately downstream of Lake Mohave and the Davis Dam, which was completed in 1951, on the Colorado River. In 1974, an initial study was conducted to evaluate a wide range of factors related to the nuisance black fly populations in this area. This study identified *Simulium vittatum* [now known to be *S. tribulatum*] as the primary pest species in the area and the Colorado River as the larval habitat. Information from this study has been used as the scientific basis for a black fly suppression program in this area since 1984.

Today, the Bullhead City Pest Abatement District and Clark County, Nevada, collaborate to conduct a black fly suppression program. The program targets nuisance black fly populations in 13 miles of the Colorado River below the Davis Dam. To assure that this program is scientifically valid and is conducted in the most efficacious and cost-efficient manner, a program review was completed during 9-13 June 2014. This review will also help assure that the program continues to operate in compliance with new regulations set forth by changes to the Federal Clean Water Act and the National Pollution Discharge Elimination System that were implemented in October 2011.

Work conducted during this summer's program review confirmed that *S. tribulatum* (formerly known as 'Cytoform III-1' of *S. vittatum*) continues to be the pest species in the area. Larval surveillance revealed significant larval populations on coarse trailing grasses and rocks relatively free of algae and debris in the current. Adult monitoring detected significant adult populations early in the morning when humidity was higher and air temperatures were lower than later in the day. Larvicide applications were conducted using the Vectobac® 12AS (*Bacillus thuringiensis* subsp. *israelensis*) product. High levels of larval mortality were observed using a targeted application rate of 15 ppm. This program strategy continues to effectively manage the pest problem.

(oral presentation)

EFFICACY OF *BACILLUS THURINGIENSIS* SUBSP. *ISRAELENIS* (BTI) FOR THE CONTROL OF BLACK FLY POPULATIONS, (*SIMULIUM* SPP.) IN MANTUA PROVINCE (ITALY)

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Key words: *Black flies control*, *Simulium paraequinum*, *Simulium pseudequinum*, *Simulium erythrocephalum*, *Bacillus thuringiensis subsp. israelensis*, *VectoBac® 12AS*, *Italy*.

Black flies populations (Diptera: Simuliidae) has been reported for a long time in the basin of the river Mincio (northern Italy). This river is called Sarca before entering lake Garda, and from there it flows approximately 65 km past Mantua into the Po river. During spring and summer, there is significant tourist activity in this area. Prior to 2009, particularly in the “Sigurtà Park” (60 ha near the town of Valeggio sul Mincio, Verona province) and other theme parks, black fly outbreaks have been controlled only with broad spectrum pyrethroids insecticide treatments. Outdoor activities such as gardening and cycling have been severely discouraged due to this host-seeking behavior. In the study area they usually bite cattle and horses however bites on humans do occur. Since 2010 trials were conducted in Mantua province in northern Italy to evaluate the efficacy of the *Bacillus thuringiensis* subsp. *israelensis* strain AM65-52 Aqueous Suspension formulation (VectoBac® 12 AS) in the black flies larval control were tested concentrations of this microbial larvicide in small plain streams. Larvicidal activity was exhibited in all trials.

VectoBac® 12 AS at 2.5 ppm/min rate generated larval mortality from the treatment point to 500 meters downstream. Larvae mortality (over 80%) was observed for 3500 meters downstream of the application site when a concentration of 40 ppm/min was applied. We hope that, because Bti is an effective and environmentally friendly treatment, the use of this biocontrol method in other river basins where black fly populations still pose a danger, particularly to horses and cows, could be adopted.

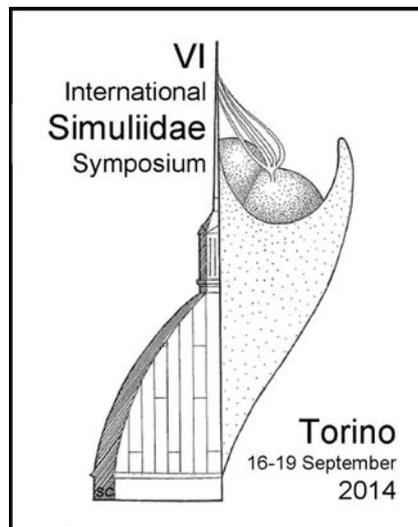
Acknowledgements

We thank Dr. Heiko Kotter (Valent BioSciences Corporation) for his useful suggestions and financial support.

(poster)

VI INTERNATIONAL SIMULIIDAE SYMPOSIUM

Torino, 16-19 September 2014



PROGRAM

Tuesday 16 September

16.00-18.00 Welcome and Registration at the University of Torino, Via Accademia Albertina 13

Wednesday 17 September

09.00-09.20 Welcome speeches

Morning session (chairing: Aleksandra Ignjatović Čupina)

09.20-10.10 **L. Rivosecchi**, S. Ciadamidaro and B. Maiolini: *Leo Rivosecchi, a lifelong passion for blackflies: the history of Simuliidae research in Italy*

10.15-10.40 **B. Maiolini** and M.C. Bruno: *Drift behaviour of blackfly larvae in controlled conditions*

10.45-11.15 Coffee break

VI International Simuliidae Symposium – Torino, 16-19 September 2014 – Abstract Book

- 11.15-11.40 **S. Ciadamidaro**, L. Mancini and L. Rivosecchi: *Blackflies (Diptera, Simuliidae) as ecological and landscape indicators of stream ecosystems in the urbanizing territory of Rome (Italy)*
- 11.45-12.10 **L.V. Petrozhitskaya** and V.I. Rodkina: *Spatial organization of stream insects of different feeding assemblages in watercourses of West Tuva, with ipcaurlar emphasis on Simuliidae*
- 12.15-12.40 **A. Ortiz**: *Electrophysiological (EAG) responses of Simulium spp to human skin volatiles*
- 12.45-13.45 Lunch break

Afternoon session (chairing: Bruno Maiolini)

- 13.45-14.10 **L.M. Hernandez-Triana**, L.G. Chaverri, M.A. Rodriguez-Perez, S. Prosser, P.D.N. Hebert, T.R. Gregory and N. Johnson: *Developing a DNA barcoding library for the identification of black flies (Diptera: Simuliidae) with emphasis on Mesoamerica*
- 14.15-14.40 **M. Kúdela**, T. Brúderová and P.H. Adler: *Blackflies of the Maritime Alps*
- 14.45-15.10 **T. Brúderová**, M. Kúdela and P.H. Adler: *Prosimulium hirtipes in Europe: does Prosimulium italicum exist?*
- 15.15-15.45 Coffee break
- 15.45-17.30 **Poster Session**

17.30 For those who wish, walk in the city centre and typical “aperitivo torinese” to have a drink together

Thursday 18 September

Morning session (chairing: Peter Adler)

- 09.00-09.30 **D. Baldry**: *The large-scale mapping of blackfly breeding sites should be a prerequisite for blackfly control operation that use aircraft to disperse larvicides to targeted water-courses, based on experiences gained from the WHO Onchocerciasis control programme in West Africa*
- 09.35-10.10 **R.A. Cheke**, M.Y. Osei-Atweneboana, C. Wiafe and M.D. Wilson: *Living with the fly (video)*

VI International Simuliidae Symposium – Torino, 16-19 September 2014 – Abstract Book

- 10.15-10.40 **R.A. Cheke**, M.G. Basanez, M. Perry, M.T. White, R. Garms, E. Obuobie, P.H.L. Lamberton, S. Young, M.Y. Osei-Atweneboana, J. Intsiful, D.A. Boakye and M.D. Wilson: *Onchocerciasis transmission and climate change in West Africa*
- 10.45-11.15 Coffee break
- 11.15-11.45 **P.H. Adler**: *How to make a dsimu pest*
lii
- 11.50-12.15 **E.W. Gray**, P.H. Adler and C. Bramley: *Black flies and the Colorado river, USA – the Bullhead city experience*
- 12.20-12.45 **R.A. Cheke**, H.M. Dobson, R.R.F. Meyer, M.Y. Osei-Atweneboana and M.D. Wilson: *Blackflies and hydropower: the Bui dam problem on the Black Volta river, Ghana*
- 12.50-13.50 Lunch break

Afternoon session (chairing: Elmer Gray)

- 13.50-14.15 **S. Aibulatov**: *Contribution to the black fly fauna (Diptera: Simuliidae) of the Sakha Republik (Yakutia)*
- 14.20-14.45 **M.J. Vila-Viçosa**, A. Ignjatović Ćupina, D. Petrić and H.C.E. Cortes: *Survey of the Simulium species (Diptera, Simuliidae) in Alentejo, South of Portugal*
- 14.50-15.15 M.J. Vila-Viçosa, A. Ignjatović Ćupina, A. Giannelli R.A.N. Ramos, F. Dantas-Torres, Y. Mutafchiev, E. Papadopoulos, L. Cardoso, D. Petrić and **H.C.E. Cortes**: *Observations on simuliids in Algarve, South of Portugal*
- 15.20-15.45 **B. Belqat** and P.H. Adler: *Update of the blackfly fauna of North-Africa (Diptera : Simuliidae)*
- 15.50-16.20 Coffee break
- 16.20-16.45 **A. Ignjatović Ćupina**, A. Giannelli, H.C.E. Cortes, R.A.N. Ramos, L. Cardoso, R.P. Lia, D. Otranto and D. Petrić: *Notes on blackfly fauna of the Basento river (Basilicata, Southern Italy)*
- 16.50-17.15 **P. Pramual**: *Biodiversity of black flies (Diptera: Simuliidae) in Thailand*
- 17.20-17.45 **A. Ignjatović Ćupina**, E. Papadopoulos, iMla.J. -Viçosa, H.C.E. Cortes and V D. Petrić: *Blackfly fauna of Macedonia (Northern Greece)*
- 20.00 Social dinner

Friday 19 September

Conference excursion

- | | |
|-------------|---|
| 09.30 | Departure from Torino by bus to visit the “ Sacra di San Michele ” |
| 12.45 | Lunch |
| 14.15 | Stop at the Metro station for those who are leaving in the afternoon |
| 15.00-18.00 | Visit to the “ Reggia di Venaria ” |
| 18.00-19.00 | Back in Torino |



Simulium argenteostriatum Strobl, 1898 ♀
(photo by S.C.)

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