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***Chrysomya albiceps* – a forensically important blow fly new for Bavaria**

(Diptera: Calliphoridae)

**Marion KOTRBA, Frank RECKEL, Jan GRUNWALD,
Michael BALKE and Sebastian SWOBODA**

Abstract

The distribution range of *Chrysomya albiceps* (WIEDEMANN, 1819) (Diptera: Calliphoridae), a hemisynanthropic and forensically important blow fly of originally tropical and subtropical distribution, has expanded considerably since the turn of the century. This paper reports the first finding of this species in Bavaria in July 2008, the second only in Germany to date, and reviews new records from the surrounding countries since the last comprehensive publication, documenting the northward progress of the species.

Zusammenfassung

Das Verbreitungsgebiet von *Chrysomya albiceps* (WIEDEMANN, 1819) (Diptera: Calliphoridae), einer hemisynanthropen, forensisch bedeutsamen Schmeißfliege von ursprünglich tropisch- subtropischer Verbreitung hat sich in den letzten Jahrzehnten deutlich ausgeweitet. Diese Studie belegt den ersten Fund der Art in Bayern im Juli 2008, den zweiten Fund in Deutschland überhaupt. Sie fasst darüber hinaus alle neueren Befunde aus den umgebenden Ländern seit der letzten umfassenden Publikation zusammen und belegt damit die Ausbreitung der Art nach Norden.

Introduction

The distribution range of *Chrysomya albiceps* (WIEDEMANN, 1819) (Diptera: Calliphoridae), a hemisynanthropic and forensically important blow fly of originally tropical and subtropical distribution, has expanded considerably since the turn of the century. For the palaeartic region the process was last summarized by GRASSBERGER et al. (2003) who depict a northward expansion from the Mediterranean region through the western and eastern parts of Europe, reaching Paris (France) and Zurich (Switzerland) in the west and Vienna (Austria) in the east (Figure 1, dotted area). More recent publications show that *C. albiceps* has continued to spread even further (POVOLNÝ 2002, AMENDT et al. 2002, VERVES 2004, SZPILA et al. 2008, DESMYTER & GOSSELIN 2009, VANIN et al. 2009, Figure 1, circles).

Detailed knowledge about the distribution range of *C. albiceps* and the history of its spread is essential, especially regarding aspects of forensic and medical entomology. Like many other representatives of the Calliphoridae, *C. albiceps* belongs to the initial colonizers of a cadaver or a dead human body. In forensic entomological casework this species can be informative in three important aspects: (1) Like other calliphorid larvae, *C. albiceps* larvae can be used for the forensic estimation of the post mortem interval by calculating the age of the oldest larval stages feeding on a corpse. (2) The second and third instar larvae of *C. albiceps* are predacious and feed on other blowfly larvae. They may seriously alter the qualitative and quantitative species composition on a corpse (GRASSBERGER et al. 2003). Heavy infestation with *C. albiceps* larvae therefore might lead to the loss of possibly older larvae of other blow fly species. This has to be taken into account when estimating the post mortem interval. (3) In regions where *C. albiceps* has not yet been recorded

its occurrence at a corpse might be used as potential evidence for a relocation of the body. In addition to its forensic relevance, *C. albiceps* can also be of medical or veterinary importance. Its larvae can cause primary and secondary cutaneous (traumatic) myiasis in human and livestock, especially after initial colonisation by *Lucilia* species (HALL & FARKAS 2000, VERVES 2004).

This paper reports the finding of *C. albiceps* in Munich, Bavaria. An updated version of the distribution range is constructed, incorporating the most recent finds. A partial *cox1* sequence is provided and compared with other published and unpublished *cox1* sequences of *C. albiceps* to test for geographic variation.

Methods and Materials

The adult specimen studied was obtained in the context of a forensic experiment (succession study) involving pig carcasses (GRUNWALD et al. 2009). It was collected by a standard fly-net sweeping technique, killed with ethyl acetate and preserved in 70% ethanol. Species identification was performed based on morphological (see results section) as well as molecular characters. One leg of the specimen was removed for DNA extraction using the Qiagen animal tissue kit (Qiagen, Hilden, Germany). Larval specimens of *Chrysomya albiceps* from Italy were obtained from STEFANO VANIN for comparative purposes. The 5' end of the mitochondrial cytochrome *c* oxidase 1 (*cox1*) gene was amplified and sequenced using primers LCO1490 / HCO2198 (FOLMER et al. 1994). PCR was conducted with Mango-Taq (Bioline) at an annealing temperature of 47°C. The detailed laboratory procedure is accessible under permalink http://zsm-entomology.de/w/index.php?title=the_beetle_D_N_A_lab&oldid=325. The sequences obtained were submitted to GenBank via EMBL (accession numbers HE 814059-61). The voucher numbers linking specimen, DNA sample and GenBank entry are MB3986 (Bavaria) and MB4309 and MB4312 (Italy: Tuscany). The vouchers were deposited in the Diptera collection at the Zoologische Staatssammlung München. The Barcoding of Life Database system at <http://www.boldsystems.org> (1. July 2011) was used for species identification based on the obtained *cox1* sequences. An alignment was created combining the new data with sequences from Belgium obtained from STIJN DESMYTER as well as data downloaded from GenBank. The closely related species *C. rufifacies* (MACQUART, 1842) was used as an outgroup. After trimming the sequences to avoid

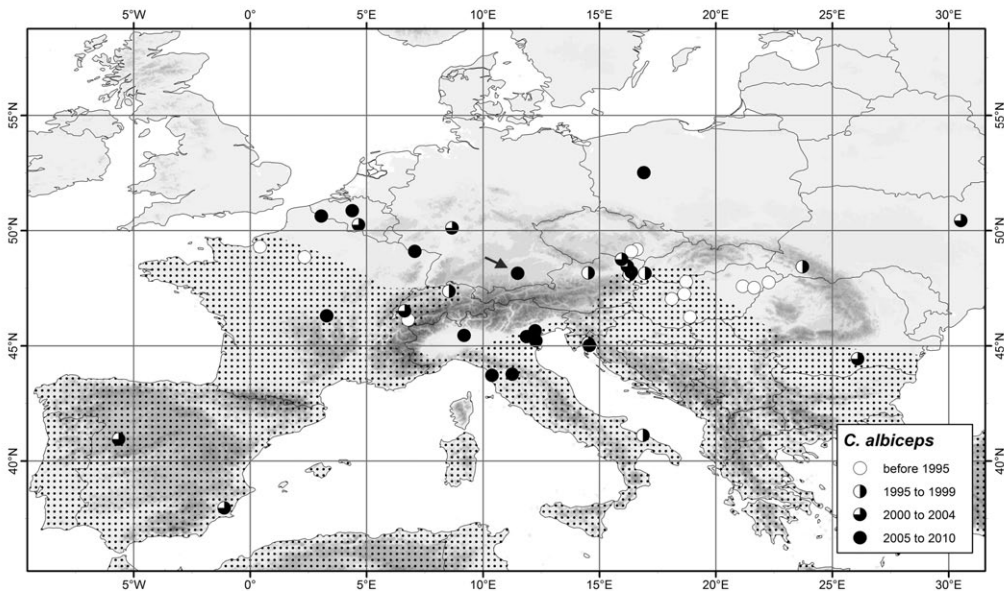


Figure 1: Distribution range of *Chrysomya albiceps*. Dotted area: distribution range from Grassberger et al. (2003); circles: individual collecting records; arrow: Munich collecting site. The filling of the circles signifies the respective time range in 5-year steps from before 1995 to 2010. The representation of individual records within the dotted distribution range is incomplete.

missing data in some of the sequences a neighbour joining tree was constructed with PAUP* (SWOFFORD 2002) software. The distribution map was generated in ESRI (R) ArcMap (TM) 9.3.1, based on topographical data obtained from the Worldclim database (<http://www.worldclim.org/>). It shows georeferenced data points. Literature data were georeferenced using Google Earth.

Results

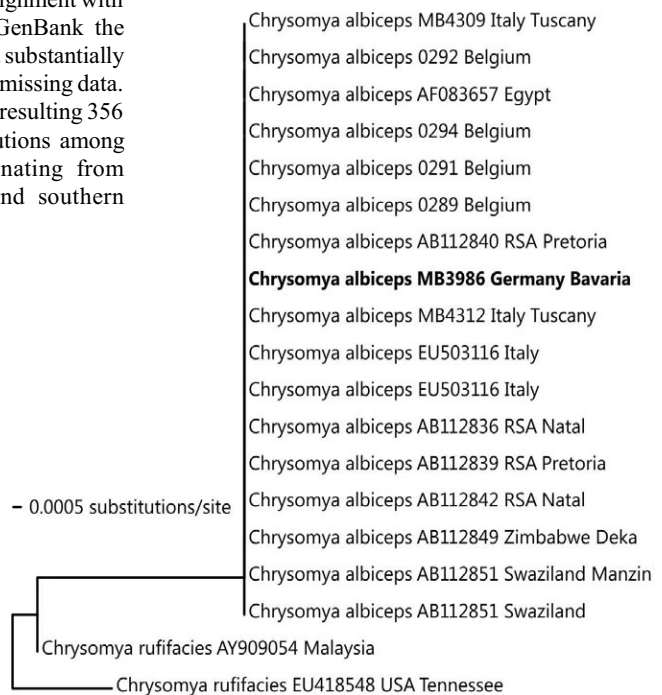
A single *Chrysomya albiceps* adult male was collected on a pig carcass in a remote area of the Botanical Garden Munich-Nymphenburg (Germany, Bavaria, Munich, Botanical Garden München-Nymphenburg “Waldspitz”, 48°09’54”N, 11°29’43”E). This is part of a large park-like area with mixed forest and ample undergrowth located in the city area of Munich. The specimen was collected in the early afternoon of July 16th 2008, a rainless summer day, at 20°C by sweeping with a fly-net above the carcass, which was 15 days old and in an advanced state of decay. No *C. albiceps* larvae were found in the larval samples collected from the carcass throughout the experiment.

Morphology: *Chrysomya albiceps* is easily identified by a range of morphological characters. The bright metallic-green adults can be distinguished from other European Chrysomyinae by the whitish-yellowish pollinosity on the face, white anterior spiracles, dark posterior edges of the abdominal tergites (SMITH 1986), and distinctive male genitalia (ROGNES 2002). Based on these characters the specimen collected in Munich was identified unambiguously. *Chrysomya rufifacies*, the only species which might easily be confused with *C. albiceps*, has not been recorded in Europe. The larvae of *C. albiceps* are characterized by numerous distinctive fleshy protuberances on their abdominal segments (“hairy maggots”) and therefore cannot be confused with any other calliphorid species occurring in Europe. It is therefore unlikely that *C. albiceps* larvae were present but not recognized in the studied larval samples. It is, however, possible that *C. albiceps* larvae were present at the carcass but not represented in the studied samples.

Cox1 sequence: A 658 base pair fragment for the 5’ end of *cox1* was obtained from the Bavarian specimen. BOLD specimen identification assigned this sequence to *Chrysomya albiceps* with specimen similarity of > 99% relative to specimens present in the database (which originated from southern Africa).

Similar fragments were obtained from two of the larval specimens from Italy. For alignment with other sequences downloaded from GenBank the obtained sequences had to be trimmed substantially at the 5’ and 3’ ends in order to avoid missing data. A neighbour joining analysis with the resulting 356 bp long alignment found no substitutions among the sequences from samples originating from Germany, Belgium, Italy, Egypt and southern Africa (Figure 2).

Figure 2: Neighbor joining tree for 5’ *cox1* sequences (356 bp) for *Chrysomya albiceps* from various regions. Specimen from Munich, Bavaria in bold.



Distribution range: The available information on the distribution of *Chrysomya albiceps* in Europe is summarized in figure 1. The dotted area illustrates the distribution range reported by GRASSBERGER et al. 2003. The circles indicate additional data from Mercier (1927), GREGOR & POVOLNÝ (1960), POVOLNÝ (2002), AMENDT et al. (2002), GRASSBERGER et al. (2004), VERVES (2004), WYSS et al. (2004), SZPILA et al. (2008), DESMYTER & GOSSELIN (2009) and VANIN et al. (2009) as well as the recent find from Munich (arrow). The filling of the circles signifies the respective time range in 5-year steps from before 1995 to 2010. The data show that the geographic range has expanded northwards since the turn of the century, now reaching Oud Heverlee (Belgium) and Frankfurt (Germany) in the west and, most recently, a location near Poznan (Poland) in the east.

Discussion

Based on morphological as well as molecular characters the specimen from Munich was unequivocally identified as *Chrysomya albiceps*. This find constitutes the first record of *C. albiceps* for Bavaria and only the second record of *C. albiceps* for Germany. The only previous find of this species in Germany was from Frankfurt (AMENDT et al. 2002). Unfortunately only a single specimen of *C. albiceps* was collected during our study. Because the available *cox1* sequences for *C. albiceps* vary considerably in length, the obtained sequences had to be trimmed substantially to avoid missing data for the alignment. The resulting comparative molecular data set did not show any substitutions and thus revealed no information regarding biogeographic relationships. It has been suggested that the rare occurrences of this species north of its original, tropical to subtropical distribution range might constitute ephemeral expansions or “pulsations” of its populations, particularly in warm and humid years (GREGOR and POVOLNÝ 1960; POVOLNÝ 2002; GOSSELIN & BRAET 2008; SZPILA et al. 2008). This hypothesis is supported by the fact that these occurrences were usually found in the later months of the year (POVOLNÝ 2002), allowing for such temporal dispersal. The new find in Munich might likewise represent a temporary range expansion, maybe even a lone traveller, from the surrounding countries. However, Munich is at approx. 48° on a similar degree of latitude as are Paris, Kolochava and other locations where the species was already found around the turn of the century. Moreover, warm years have become more common in the past years (IPCC 2007, ZORITA et al. 2008). Therefore the possible occurrence of *C. albiceps* in Bavaria has to be taken into account in scientific studies as well as in forensic casework from now on. Further studies will be necessary to resolve, whether this species has established a persistent population, leading to its regular occurrence in this region.

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Addresses of the authors:

Marion KOTRBA (Corresponding author)

SNSB, Zoologische Staatssammlung München, Münchhausenstr. 21, 81247 München, Germany, marion.kotrba@zsm.mwn.de

Frank RECKEL, Jan GRUNWALD

Bayerisches Landeskriminalamt, Maillingerstrasse 15, 81827 München, Germany, frank.reckel@polizei.bayern.de, jan.grunwald@polizei.bayern.de

Michael BALKE

SNSB, Zoologische Staatssammlung München, Münchhausenstr. 21, 81247 München, Germany, Coleoptera-ZSM@zsm.mwn.de

Sebastian SWOBODA

Westendstr. 138, 80339 München, Germany, swoboda.sebastian@alice.de

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