14th Meeting European Association for Forensic Entomology

EAFE 2017

7th-10th June 2017, Treviso, Italy

Scientific Committee

Stefano VANIN (President)

Luc BOURGUIGNON

Damien CHARABIDZE

Amoret WHITAKER

Cindy AUBERNON

Thiago DE CARVALHO MORETTI

Thierry PASQUERAULT

Jens AMENDT

Katherine BROWN

Carlo CAMPOBASSO

Organizing Committee

Stefano VANIN (President)

Valentina BUGELLI

Elisa ARENA

Sara BORTOLINI

Carlo CAMPOBASSO

Luigi MASTROGIUSEPPE

Giorgia GIORDANI

Fabiola TUCCIA

Sponsorship

CITTA' DI TREVISO
CITTA' DI VILLORBA

ACCADEMIA NAZIONALE ITALIANA DI
ENTOMOLOGIA http://www.accademiaentomologia.it/
SOCIETA' ENTOMOLOGICA ITALIANA http://www.societaentomologicaitaliana.it/
GRUPPO ITALIANO DI PATOLOGIA FORENSE http://www.gipf.it/
CONSULTA NAZIONALE GIOVANI MEDICI LEGALI
UNIVERSITARI http://www.consultagiovanimedicilegali.it/
SOCIETÀ VENEZIANA DI SCIENZE NATURALI www.svsn.it
SOCIETA' TREVIGIANA DI SCIENZE NATURALI http://www.naturalistitrevigiani.it/
ORDINE DEI GIORNALISTI DEL VENETO www.ordinegiornalisti.Veneto.it

ASCOM – CONFCOMMERCIO IMPRESE PER L'ITALIA – TREVISO
FOOD GENIUS ACADEMY www.foodgeniusacademy.com
ASSOCIAZIONE SPECIALISTI MEDICI LEGALI http://www.assomel.it/
ISTITUTO DI SCIENZE FORENSI http://www.istitutoscienzeforensi.it/

Program

7 June 2017	
18.00-19.00	Registration
19.00-	Welcome Reception

8 June 2017	**Ca' dei Carraresi**
8:30-12:30	Registration
9.15	Welcome Speech Vanin S. (President GIEF) Bourgignon L. (President EAFE) Avv. Manildo G. (Mayor of Treviso)
9.45-10.45	Cattaneo *Not only DNA, other Forensic disciplines?* (Opening lecture)
10.45-11.00	**Faris A.M., West W.R., Tomberlin J.K., Tarone A.M** An outdoor validation of laboratory based development data for *Cochliomyia macellaria* (Fabricius) (Diptera: Calliphoridae)
11.00-11.15	**Frątczak-Łagiewska K., Matuszewski S.** Sex-specific developmental models for necrophilous beetle *Creophilus maxillosus* (l.) (Staphylinidae)
11.15-11.45	Caffè and not only
11.45-12.00	**Shin S.E, Moon T.Y., Lee J.W., Hoon S.G., Ko K.S., Park S.H** Basic studies on the development of forensic entomological software in Korea
12.00-12.15	**Gemmellaro M.D, Goodson R., Sodon J., Delvillar M., Hamilton G**. Food preference of *Calliphora vicina* among human and non-human trophic substrates
12.15-12.30	**von Hoermann C., Steiger S., Ayasse M** Influence of land use on the decomposition rate of dead piglets in conjunction with the diversity of carrion insects: a large scale cadaver study
12.30-12.45	**Barnes K., Bulling M** The impact of small scale environmental variation on the relationship between recorded ambient temperature and corpse temperature; potential implications for estimating the minimum post mortem interval?
12.45-14.15	Lunch and Poster
14.30-14.45	**Brown K., Harvey E., Jetten K., Simmonds A., Smith P.** Fire flies: the effects of fire and burnt cadavers on Diptera used for forensic investigation
14.45-15.00	**Iancu L., Purcarea C.** Colonization pattern of Diptera and Coleoptera on buried remains in an urban area (Bucharest, Romania)

15.00-15.15	**Mashaly A.** Effect of clothing on insect prevalence into rabbit carcasses in three separated habitats
15.15-15.30	**Matuszewski S., Mądra-Bielewicz A.** Biotic factors affecting residency of *Necrodes littoralis* l. (Coleoptera: Silphidae) on cadavers
15.30-15.45	**Weithmann S., von Hoermann C., Steiger S., Ayasse** M.Abundance of dung-associated beetles on carrion in anthropogenic affected habitats (Coleoptera: Scarabaeoidea)
15.45-16.00	**Jang H., Shin S.E., Ko K. S., Choi B. H., Park J. H., Park S. H.** Case studies on parasitoid *Brachymeria podagrica* (Fabricius) (Hymenoptera: Chalcididae) emergenced in the rearing of necrophagous flies in Korea
16.00-16.30	Caffè and not only
16.30-16.45	**Vanin S.** 20x20 cm crime scene
16.45-17.00	**De Micco F., Cavezza A., Bugelli V., Campobasso C.P.** An Italian cold case: the sad story of the remains of a child in the suitcase
17.00-17.15	**Bugelli V., Gherardi M., Vanin S., Campobasso C.P.** Pattern of decomposition rate and insect colonization in cases of suicide by hanging
17.15-17.30	**Lo Pinto S., Giordani G., Tuccia F., Ventura F., Vanin S.** First findings of *Synthesiomyia nudiseta* (Diptera: Muscidae) in forensic cases in northern Italy
17.30-18.00	Final Discussion

9 June 2017	**Ca' dei Carraresi**
8:30-12:30	Registration
9.30-10.15	**Paciello O.** Postmortem interval estimation in animals
10.15-10.30	**Mastrogiuseppe L.** Use of insects in Forensics Veterinary
10.30-10.45	**Marchetti C., Mastrogiuseppe L., Pisani GM** Unusual entomological localizations, a wildlife experience.
10.45-11.00	**Taleb M., Tail G., Benzekkour M., Bouderbala D., Djedouani B., M.Toumi, Açıkgöz H.N.** Decomposition and arthropod colonization of wrapped rabbit carrions in north central Algeria
11.00-11.15	**Mactaggart M., Whitaker A., Hall M., Wilkinson K.** An investigation of post-feeding larval dispersal in UK blow flies
11.15-11.45	Caffè and not only
11.45-12.00	**Martín-Vega_D., Simonsen T.J., Wicklein M., Garbout A., Ahmed F., Hall M.J.R.** Metamorphic brain remodeling as an age indicator during the blow fly intra-puparial period
12.00-12.15	**Szpila K., Jafari S., Akbarzadeh K., Piwczyński M.** Taxonomy of forensically important flesh flies (Diptera: Sarcophagidae) of the middle east
12.15-12.30	**Chimeno C., Morinière J., Hausmann A., Reckel F., Grunwald J.E., Haszprunar G.** Next generation sequencing of arthropod communities on decomposing bodies – Forensic Entomology and DNA barcoding
12.30-12.45	**Weidner L.M., Nigoghosian G., Hanau C.G** Increasing the use of entomological evidence: practical food source options for crime laboratories
12.45-14.15	Lunch and Poster
14.30-15.00	**Benbow M.E., Weatherbee C.R., Pechal J.L** Maggot masses and microbiomes: the interactions of insects and microbes in the necrobiome
15.00-15.15	**Pechal J.L., Weatherbee C.R., Benbow M.E.** Community assembly of carrion and calliphorid microbiomes
15.15-15.30	**Bernhardt V., Scheid N., Holdermann T., Schäfer T., Verhoff M. A., Amendt J.** *Same same*, but different! - decoding the nutrition history of blow flies by isotope signature analysis of adult flies and their empty pupal cases
15.30-15.45	**Vanin S.** Forensic or not Forensic II
15.15-16.00	Conclusion
16.00-16.30	Caffè and not only
16.30-17.30	**EAFE Meeting**

ORAL PRESENTATIONS AND POSTERS

AN OUTDOOR VALIDATION OF LABORATORY BASED DEVELOPMENT DATA FOR *COCHLIOMYIA MACELLARIA* (FABRICIUS) (DIPTERA: CALLIPHORIDAE)

Faris A.M., West W.R., Tomberlin J.K., Tarone A.M.

Department of Entomology, Texas A&M University, College Station, TX, USA
ashmfaris@gmail.com

Knowing the age of insects collected from human remains can be informative in death investigations, and when certain assumptions are met, can be useful for estimating the postmortem interval (PMI). Such estimates often are based on species-specific development data that are temperature-dependent, highly variable, and potentially prone to an unknown degree of error. Legal and professional pressures have been placed on forensic disciplines to develop more reliable approaches in forensic sciences. One focus within forensic entomology is the approach to understanding error associated with making PMI estimates. Because of this potential error, methods employed for making such estimates must be validated to quantify the associated error. Validation studies are useful tools for determining accuracy and precision of laboratory and field data. However, few development data sets have been validated. *Cochliomyia macellaria* (Fabricius) (Diptera: Calliphoridae) is a primary colonizer of remains in the southeastern USA, and is therefore of great importance in forensic investigations. The objective of this study was to determine the accuracy of estimating the postcolonization interval when applying laboratory-based development data for *C. macellaria*. Over 12,000 *C. macellaria* immatures of a known age were allowed to develop in rearing jars outdoors. Light intensity, temperature, and humidity were recorded every ten minutes. Replicate samples of 200 individuals were collected every six hours until adult emergence was observed. Immature samples were preserved in their collected stage and stored until insect age estimates were made based on stage, larval length, and larval weight. Results show length, weight, and stage predictions with the development data set in question are generally accurate but imprecise for individuals past the second instar. Stage is an accurate predictor when all stages present in a sample are considered. Estimates made with all stages collected within a sample time point were 72% accurate. Predictions based on weight may be inaccurate due to differences in storage between the development and validation study samples. Inaccuracies due to length-based predictions may be due to differences between focal and reference populations, which could be a result of genetic or environmental differences. This validation allows forensic entomologists to quantify expected precision and accuracy derived from insect age estimates when using this development data for predictions in central Texas, USA. This study is the first validation of development data for *C. macellaria*.

SEX-SPECIFIC DEVELOPMENTAL MODELS FOR NECROPHILOUS BEETLE *CREOPHILUS MAXILLOSUS* (L.) (STAPHYLINIDAE)

Frątczak-Łagiewska K.[1,2], Matuszewski S.[1]

[1]Laboratory of Criminalistics, Adam Mickiewicz University, Poznań, Poland;
[2]Department of Animal Taxonomy and Ecology, Adam Mickiewicz University, Poznań, Poland
katarzyna.fratczak@amu.edu.pl

Differences in size between males and females, called sexual size dimorphism, are common in insects. These differences may be followed by differences in the duration of development between sexes. Accordingly, it is believed that insect sex may be used to increase the accuracy of insect age estimates in forensic entomology. The aim of this study was to test whether there are sex-specific differences in the development of *Creophilus maxillosus*. We have also created separate developmental models for males and females of *C. maxillosus* and made a validation study to test whether sex-specific models improve the accuracy of insect age estimation. The following predictions were tested: 1) males of *C. maxillosus* develop for longer than females, 2) the use of sex-specific developmental models improves the accuracy of insect age estimates and consequently the minimum PMI.

Development was studied at seven constant temperatures (15, 17.5, 20, 22.5, 25, 27.5, 30°C) and 40 insects per temperature. Beetles were kept individually in small containers and fed *ad libitum* with larval blowflies. Length and mass during larval stages and mass during the pupal stage were measured at intervals representing 10% of the stage duration. Sex for each specimen was identified after adult emergence using the shape of eighth abdominal sternite. Separate thermal summation models for males and females, as well as for the pooled sample (i.e. the general model) were calculated using the method described by Ikemoto and Takai (2000). Eight insects per temperature and sex were randomly selected to be used for the modelling purposes, rest of the specimens were used to test performance of the models in the age estimation task. Due to the large mortality at extreme temperatures, they were poorly represented in the validation sample.

As a rule, males of *C. maxillosus* developed longer than females. Differences in the duration of development between females and males were largest at 22.5 and 25°C and in the third larval and pupal stages. Males were distinctly larger (longer and heavier) than females from the beginning of third larval stage until eclosion. The sex-specific and general models for the total immature development have the same optimal temperature range, similar developmental threshold but different thermal constant *k*, which was the largest in the case of the male-specific model and the smallest in the case of the female-specific model. Despite these differences, validation study revealed just minimal and statistically insignificant differences in the accuracy of age estimates using sex-specific and general models (t test for correlated samples; $P = 0.80$).

Concluding, although there are differences in the duration of immature development between females and males of *C. maxillosus*, an increase in the accuracy of insect age estimates while using the sex-specific models is negligible. Accordingly, this study does not support the use of sex-specific developmental data for the estimation of insect age in forensic entomology.

BASIC STUDIES ON THE DEVELOPMENT OF FORENSIC ENTOMOLOGICAL SOFTWARE IN KOREA

Shin S.E[1], Moon T.Y.[2], Lee J.W.[3], Hoon S.G.[1], Ko K.S.[1], Park S.H.[1]

[1]Department of Legal Medicine, Korea University College of Medicine, Seoul, Korea; [2]Department of Biological Science, Kosin University, College of Health and Welfare, Busan, Korea; [3]Department of Statistics, Korea University, College of Political Science and Economics, Seoul, Korea
shinfbr@nate.com

The estimation of minimum postmortem interval (PMImin) using entomological evidence is known as one of the most reliable methods, even when the body is badly decayed. For the establishment of basic forensic entomological data and the development of a software for the estimation of PMI-min in Korea, a 5-year research consortium, consisting of five branches, was firstly constructed under a support from the Korean National Police Agency. The five branches cover the study on Korean necrophagous entomofauna according to natural environment and their DNA Barcoding, the construction of ecological database related to Korean necrophagous insects of forensic importance, the study on statistical models for forensic entomology and meteorology, the estimation of PMI-min based on developmental gene expression clock of necrophagous fly, and software development for entomological PMI-min estimation.

As first results of the research project, in the mid-north area of Korean peninsula, 8 orders, 20 families, 37 species were identified from decomposed piglet experiments and fly traps. 478 fly specimens were identified as 2 families, 12 species by DNA barcoding methods. In the mid and southern area, 7 orders, 35 families, 95 species were investigated from decomposed piglet experiments with coding of insect samples for fast biodiversity survey. By correlation studies on several factors related to estimation of air temperature, high correlated variables were selected and a preceding estimation algorithm was established. A genome project for *Sarcophaga peregrina*, contig assembly and annotation for 4 fly species, and the securement of growth cycle specimens were performed. A prototype homepage (http://cb0twc.axshare.com) and algorithm were constructed.

This consortium will be continued for the next four years. It is expected that investigation timeline in scene will be shortened and the capacity of investigation on cases of extremely decomposed bodies will be strengthened in Korea. This research was supported by Projects for Research and Development of Police science and Technology under Center for Research and development of Police science and Technology and Korean National Police Agency. (Grant No. PA-G000001).

FOOD PREFERENCE OF *CALLIPHORA VICINA* AMONG HUMAN AND NON-HUMAN TROPHIC SUBSTRATES

Gemmellaro M.D.[1], Goodson R.[1], Sodon J.[1], Delvillar M.[1], Hamilton G.[1]

[1] Department of Entomology, Rutgers University, New Brunswick, NJ USA
denise.gemmellaro@rutgers.edu

Adult blow flies (Diptera: Calliphoridae) are normally reared in a laboratory setting using pure water and a source of glucose, such as sugar or honey (Arias-Di-Donato and Lira 2016; Saleh et al. 2014). However, close to the moment of oviposition, it is important for them to consume a protein-based meal, whose source may or may not also represent the substrate where their eggs will be laid. From a forensic standpoint, a crime scene may present several contaminations, from garbage to food scraps; and a better understanding of the food preference of blow flies can help in the analysis of the scene; as a matter of fact, similar studies have been carried out recently focusing on other species and overall effect of a food substrate on the flies (Durdle et al. 2016). This study aims to evaluate the preference of *Calliphora vicina* (Robineau-Desvoidy) adults among potential protein-based meals.
Twenty adults of *C. vicina* (ten females and ten males) were individually color-marked and released in a clear acrylic arena; in the arena, they had access to six different food sources placed in six distinct glass petri dishes. Three of these sources were human and three were non-human. The activity of the flies inside the arena was recorded for a six-hour period using a camera (Make and model: Sony Handycam HDR-XR550V), which was placed above the arena. The number of times an individual fly visited each food source and the length of each visit were recorded. Comparisons across males and females, visits between human and non-human food substrates and the overall number and length of visits to each substrate were analyzed.

Arias-Di-Donato L and Liria J (2016) Vital Statistics of *Chrysomya megacephala* (Fabricius, 1794) (Diptera: Calliphoridae) under different diets from Venezuela. *Journal of Entomology and Zoology Studies.* 4(2): 247-251
Durdle A, Mitchell RJ, van Oorschot RA. (2016) The Food Preferences of the Blow Fly *Lucilia cuprina Offered Human Blood, Semen and Saliva, and Various Nonhuman Foods Sources. J Forensic Sci*.; 61(1): 99-103
Saleh V, Soltani A., Dabaghmanesh T., Moemenbellah-Fard M. D.. (2014) Mass Rearing and Life Table Attributes of Two Cyclorrhaphan Flies, *Lucilia sericata* Meigen (Diptera: Calliphoridae) and *Musca domestica* L. (Diptera: Muscidae) under Laboratory Conditions. *Journal of Entomology* 11(5): 291-298

INFLUENCE OF LAND USE ON THE DECOMPOSITION RATE OF DEAD PIGLETS IN CONJUNCTION WITH THE DIVERSITY OF CARRION INSECTS: A LARGE SCALE CADAVER STUDY

von Hoermann C.[1], Steiger S.[1], Ayasse M.[1]

[1] Institute of Evolutionary Ecology and Conservation Gemomics, Ulm University, Ulm, Germany
christian.hoermann-von-und@uni-ulm.de

Animal carrion is the most nutrient-rich form of dead organic matter and is considered to be an important component of recycling energy and nutrients in ecosystems. The decomposition rate of animal biomass depends on the availability of decomposers and detritivores and on biotic and abiotic factors of a certain habitat. Particularly in forest ecosystems, increasing land use intensity and land use change is a major driver of biodiversity loss. I hypothesize that anthropogenic land use intensity has an influence on the carrion insect community and consequently on the removal rate of animal biomass.

We simultaneously exposed 75 piglet cadavers on 75 forest experimental plots in three regions of Germany (DFG-supported Biodiversity Exploratories: Swabian Alb, Hainich-Dün and Schorfheide-Chorin) and collected and determined individuals of coleopteran and dipteran taxa to species level. Furthermore, we collected soil samples for isotopic analysis, dissected snout-tissue samples for measurement of microbial 16S rRNA for determination of assemblages of epinecrotic bacteria and reared fly maggots on minced meat for later species determination.

I found that not only ambient and soil temperature but also other structural habitat parameters like soil texture, soil type and management system had an influence on carcass associated carrion beetle abundance. For the forensically important calliphorid fly *Lucilia sericata* I detected that their maggots were retrieved almost exclusively in the Hainich-Dün Exploratory, which was linked to a higher humidity in the forests of this region. It became obvious that temperature and species richness of cadaver beetles (silphids and scarabaeoids) significantly lowered decomposition time whereas forest management intensity significantly increased the persistence rate of an aboveground exposed cadaver. For the first time, we could show that the *in situ* synthesis of cadaveric volatile organic compounds was linked to particular types of epinecrotic microorganisms. Concerning the input of organic carbon from decomposing mammal tissue, I found reduced ratios of organic carbon in the soil beneath the carcass in forests with higher management intensity. Overall, my findings show that the anthropogenic land use, with its impact on biotic and abiotic habitat parameters, influences the course of decomposition and the return of dead animal biomass to the ecosystem. Variations in habitat parameters are thus important factors to consider in the estimation of the post-mortem interval in criminal investigations.

THE IMPACT OF SMALL SCALE ENVIRONMENTAL VARIATION ON THE RELATIONSHIP BETWEEN RECORDED AMBIENT TEMPERATURE AND CORPSE TEMPERATURE; POTENTIAL IMPLICTIONS FOR ESTIMATING THE MINIMUM POST MORTEM INTERVAL?

Barnes K., Bulling M.

University of Derby, Derby, UK
k.barnes1@derby.ac.uk

A Forensic Entomologist can estimate the minimum post mortem interval (mPMI) using the developmental stages of initial colonisers, the blow flies. For this calculation the species of fly, age of the oldest fly and temperature of the crime scene is needed. The crime scene temperature can be calculated retrospectively using corrected data from an appropriate national weather station. The correction factor is based on correlating temperature data collected by data loggers at the location of the corpse for several days following discovery, with the temperature recorded at the weather station over the same period. Recently, work has shown that local environmental differences between the location of the corpse and that of the logger(s) can affect the relationship between the recorded ambient temperature and the temperature of the corpse, and that these differences are more apparent over summer than winter periods. This study furthers current knowledge by assessing placement of data loggers for 3 summer months (June to August) over a 3 year period (2014-2016) in relation to a model corpse.

Hourly temperature data were recorded using Tinytag data loggers in a secluded area of an urban environment (University of Derby) over three consecutive years (2014-2016). For each experiment, fresh porcine liver (100g) was placed in four modified cone traps for a six hour period (10am-4pm). Experimental sites were spaced 20 metres from each other to ensure they were independent replicates. On each of the 87 sampling days, two traps were placed approximately 1.6 metres from the ground using bird feeder poles and two were placed at ground level. Trap locations alternated each day to randomise results. One data logger (TGP-4500) recorded hourly ambient temperature and humidity throughout each experimental period (June to August) at each site. Meat temperature was recorded using a probe and data logger (TGP-4520) at trap level for the duration of each experiment (6 hours). Light levels, wind speed and rainfall were recorded three times at each site during each experiment (0, 3 and 6 hours). Data was analysed using R.

This study found that ambient temperature, amount of sunlight, time of day and location affected the temperature of samples of porcine liver, and that there were significant interactions between some of these variables. The results strongly suggest that the placement of loggers at a crime scene may affect the pattern of correlation between recorded temperature and the temperature of the corpse, and therefore will affect the estimate of the mPMI.

FIRE FLIES: THE EFFECTS OF FIRE AND BURNT CADAVERS ON DIPTERA USED FOR FORENSIC INVESTIGATION

Brown K., Harvey E., Jetten K., Simmonds A., Smith P.

The Forensic Innovation Centre, Institute of Criminal Justice Studies,
University of Portsmouth, Portsmouth, Hampshire, UK
katherine.brown@port.ac.uk

The Forensic Innovation Centre is a unique and award-winning collaboration between the University of Portsmouth and its surrounding law enforcement and emergency service partners, including Hampshire Constabulary, Hampshire Fire and Rescue and the Defence College of Policing and Guarding. This partnership has enabled unique multidisciplinary research possibilities, including student dissertations, placements and PhD research. This presentation discusses the development and current successes of our partnership, including two collaborative projects focused on the interactions between fire and insects; a markedly under-researched area often due to availability of appropriate research facilities.

In England in 2014-2015, there were 264 fire-related fatalities recorded (Home Office, 2015) and yet no published research in the UK on the effects of a) fire on the collection and interpretation of insects involved in pre-burn colonisation and b) burned cadavers on insect colonisation, succession and growth rates.

The aim of the first study was to examine the integrity of insect evidence after being exposed to an intense fire. Pupae were placed in a furnished shipping container, both at the ignition source and at opposite side of the room, exposed and protected from direct flame contact. In under three minutes, the fire reached its maximum intensity and was extinguished. The pupae were recovered and examined under stereomicroscopy for identifying features. The characteristics identified will be presented.

The second study comprised two undergraduate research projects. The students explored the effect of burning with and without accelerants, on clothed and unclothed rabbit cadavers, and monitored insect colonisation and succession throughout decomposition. In summary, clothing (in addition to burning) and heavier accelerants slowed decomposition rate and affected colonisation greater than burning alone. Burning without clothing also reduced the available food source, resulting in higher numbers of starved larvae. Primary colonisers in all cases were *Lucilia sericata* and *Calliphora vicina*, ovipositing within 1-3 days depending on material and accelerant.

The FIC model has shown that working in close collaboration with partners, particularly practitioners, enables a) rapid incorporation of knowledge and data into forensic investigative practice, and b) more focused direction of future research projects, addressing key priority areas.

Home office (2016). *Fatalities and Casualties from Fires by Nation.* Retrieved from https://www.gov.uk/government/statistical-data-sets/fire-statistics-data-tables

COLONIZATION PATTERN OF DIPTERA AND COLEOPTERA ON BURIED REMAINS IN AN URBAN AREA (BUCHAREST, ROMANIA)

Iancu L., Purcarea C.

Institute of Biology of the Romanian Academy, Bucharest, Romania
lavinia.iancu@ibiol.ro

Investigations of necrophagous insect fauna associated to buried remains are very limited to date. In these cases, the decomposition process is slower compared to exposed carcasses (Mégnin 1887; Motter 1989), the timing of insect colonization being influenced by the soil type, burial depth, precipitation rate and temperature (Amendt et al. 2010).
To extend this knowledge to the Romanian region, our experiment using buried rat (*Rattus norvegicus*) carcasses was performed in a park-type area of Bucharest during the summer period. Thirty carcasses were buried 40 cm deep, which corresponds to a mid-burial depth. The air and soil temperature and relative humidity were recorded every four hours using three button logger thermo-hygrometers, while the precipitation rate was obtained from the nearest weather station. Sampling of Diptera and Coleoptera adults and immature stages was performed daily over a 30-days period (June 2016). Specimens were preserved in 75% ethanol for taxonomic identification, and in 10 mM Tris-EDTA, pH, 8 buffer for genetic barcoding based on cytochrome oxidase I gene fragment amplification.
The air and soil temperatures during the experiment varied between 16°C and 27.3°C, and the precipitation rates alternate in the 0.51 - 5.08 mm/day range.
Necrophagous insect adults were observed on the soil surface when carcasses reached the active decay stage, however no egg clusters were noticed. Diptera adults were observed at the soil surface after 9 days' post burial, Muscidae being the first colonizers, followed by Phoridae species. Coleoptera adults belonging to Staphylinidae and Leiodidae were observed only at the end of the advanced decay stage, starting with day 26.
The insect dynamics was not affected greatly by temperature variations during this period, while the presence of all species was influenced by the precipitation regime and corresponding soil relative humidity.
Compared to the results for exposed carcass experiments in the same environment and similar climatic parameters (Iancu et al. 2016), the current data showed major differences concerning the family type, order of appearance, carcass decomposition stage and a significant delay in the insect colonization timing.
This experiment adds important data on the insect colonization pattern of buried remains, providing the first information on necrophagous insect diversity and dynamics associated to buried carcasses on the Romanian territory.

Amendt J, Goff ML, Campobasso CP, Grassberger M, 2010. Current concepts in forensic entomology. SpringerScience & Business Media, 376.
Iancu L, Sahlean T, Purcarea C, 2016. J Med Entomol. 53(1): 54-66.
Mégnin P, 1894. La Faune de cadavres: Application de l'entomologie à la médicine légale, EncyclopédieScientifique des Aide - Mémoire, Masson & Gauthier-Villars, Paris, 214.
Motter MG, 1898. J.N.Y. Entomol. Soc. 6(4): 201-233.

EFFECT OF CLOTHING ON INSECT PREVALENCE INTO RABBIT CARCASSES IN THREE SEPARATED HABITATS

Mashaly A.

Zoology department, College of Science, King Saud University Riyadh, Saudi Arabia
mmashely@ksu.edu.sa

Interest in Forensic entomology has increased at the international level, including the study of factors affecting insect colonization of carrion. Factors contributing to the decomposition process and insect succession include: habitat, season, sun exposes, outdoor or indoors, burying, clothing or burning the corpse. The purpose of this study was to determine the effect of clothing on corpse decomposition and insect prevalence on rabbit carcasses in three different habitats in the period from May to July 2015. In the agricultural habitat, carrion reached the dry stage within 12 days, whereas the carrion needed 6 days in the desert and urban habitats, in both clothed and unclothed carcasses. A total of 2925 insects included 2535 flies and 390 beetles were collected during the decay process. The prevailing species of flies belonged to families Calliphoridae and Muscidae, while families Dermestidae and Histeridae contained the prevailing species of beetles. *Lucilia sericata, Musca domestica, Dermestes maculatus,* and *Saprinus moyses* were the most abundant species occurring on both the clothed and unclothed carcasses. There were also significant differences in the insect communities evident on the clothed carcasses compared to the unclosed carcasses, and the clothed carcasses also contained the highest number of beetles and the lowest number of flies collected.

BIOTIC FACTORS AFFECTING RESIDENCY OF *NECRODES LITTORALIS* L. (COLEOPTERA: SILPHIDAE) ON CADAVERS

Matuszewski S., Mądra-Bielewicz A.

Laboratory of Criminalistics, Adam Mickiewicz University, Poznań, Poland
szymmat@amu.edu.pl

Residency of carrion insect taxa is one of the key parameters in succession-based methods for post-mortem interval estimation. Its complex causal background is poorly understood and for this reason residency of particular taxa is difficult to predict in casework. Recent studies revealed that in many carrion taxa residency is dependent on cadaver mass, season of the year and accumulating temperature. We believe that there are also biotic factors that affect residency of insects on cadavers and in this work this possibility is tested in the case of *Necrodes littoralis*. It is a carrion beetle species which regularly visits and breeds in large vertebrate cadavers including humans. Its larvae feed on cadaver soft tissues, form larval masses and may drive active decay. Because *N. littoralis* uses for breeding the same carrion resources as Calliphoridae, it is believed that these taxa are in strong competition over carrion. We hypothesize that adult *N. littoralis* visits these cadavers which are at most only partly colonized with blowflies. Therefore, the extent to which a cadaver was colonized or used by blowflies may be a good predictor of residency in adult and larval *N. littoralis*. Several predictions follow this hypothesis and in the current study they were tested by reanalyzing the results of our previous pig carcass experiments.

Analyses included data from 90 carcasses of various mass (6-64 kg), year of exposure (four years), month of exposure (April – September) and habitat type (forest and open natural habitats). Cadavers were killed and exposed according to the single protocol. Insects were sampled manually and with pitfall traps according to the similar protocol. Although in particular studies the frequency of sampling was different, until the end of active decay all cadavers were sampled at least once a day and afterwards less frequently.

It was found that adult *N. littoralis* were recorded for the longer periods and in larger numbers on cadavers which were bloated for the longer time. Because long-lasting bloating reflects low cadaver colonization by blowflies, these findings suggest that adult *N. littoralis* may in some way follow bloating as an indicator of cadaver usability for breeding purposes. Moreover, we found a positive relationship between residency of adult *N. littoralis* and 3rd larval stage of *Lucilia caesar*, the dominant blowfly species on most of our cadavers. Because both species use cadavers for breeding purposes and 3rd larval stage of blowflies is the most destructive for carrion soft tissues, this relationship may simply reflect the adaptation of adult *Necrodes* to stay on carrion as long as the blowfly 3rd instar larvae are present in order to actively reduce their population. We have also demonstrated that adult and larval *N. littoralis* were present for a longer time and in larger numbers on these cadavers which were only partly scavenged by blowflies. This finding was indicated by the strong, negative relationship between residency or abundance of adult or larval *N. littoralis* and percent of cadaver mass scavenged by blowflies.

Concluding, our results support the statement that residency of adult and larval *N. littoralis* is affected by the extent to which carrion resources were used by blowflies.

ABUNDANCE OF DUNG-ASSOCIATED BEETLES ON CARRION IN ANTHROPOGENIC AFFECTED HABITATS (COLEOPTERA: SCARABAEOIDEA)

Weithmann S., von Hoermann C., Steiger S., Ayasse M.

Institute of Evolutionary Ecology and Conservation Genomics, University of Ulm, Germany
sandra.weithmann@uni-ulm.de

In anthropogenic affected landscapes, increasing forest management is negatively influencing the biodiversity of naturally occurring organisms. Previous studies show a current loss of biodiversity due to small- and large-scale land-use intensification. Therefore, increased land-use intensity may decelerate the decomposition rate of vertebrate carcasses, slow down nutrient cycles and thus ecosystem services may get lost.
In terrestrial ecosystems, the decomposition process of vertebrate carcasses is marked by a successional colonization of various insects attracted by distinct odor bouquets of different decomposition stages. Until now, several studies investigated the role of carrion-related insect taxa (like silphids, staphylinids or histerids) during cadaver decomposition. However, dung-associated scarabaeoid beetles are frequently excluded from such studies although they are often found on carrion. Due to their previous exclusion, the aim of the current project was to examine the abundance of scarabaeoid beetles on decomposing piglet carcasses in differently managed forests. More precisely, we analyzed the influence of various habitat parameters on the decomposition rate of piglet cadavers and on the occurrence of their inhabiting scarabaeoid beetles in the biosphere reserve Schorfheide-Chorin in Germany. Schorfheide-Chorin is one out of three study regions of the "NecroPig" project, in which we exposed 75 stillborn piglet cadavers simultaneously in German forests (http://www.biodiversity-exploratories.de/1/projects/derzeit-gefoerdert/tiere/necropig/).
"NecroPig" is a large-scale project in the field of carrion ecology which examines decomposition processes in German forests taking into account the influence of land-use intensity.
In this study, a total of 9311 specimens of scarabaeoid beetles were collected, mainly represented by the geotrupids *Anoplotrupes stercorosus* (~95 %) and *Trypocopris vernalis* (~4.8 %). In respective controls, only 793 scarabaeoid specimens were retrieved. Five stages of decomposition were recognized: fresh, bloated, active decay, advanced decay and dry remains stage. The highest abundance of scarabaeoid beetles was observed at the advanced decay stage. Moreover, on species level, it turned out that exclusively *T. vernalis* occurred mainly in one specific forest stand (scots pine forest), indicating that this species is restricted to a certain forest type. Furthermore, higher ambient air temperature tended to result in faster decomposition, whereas higher land-use intensity (calculated based on the silvicultural management intensity index (SMI)) caused slower decomposition rates.
In summary, this study revealed that scarabaeoid beetles visit medium-sized cadavers not by chance but in a successional pattern and *T. vernalis* might be more sensitive to habitat alteration than other beetles due to its distribution limits.

CASE STUDIES ON PARASITOID *BRACHYMERIA PODAGRICA* (FABRICIUS) (HYMENOPTERA: CHALCIDIDAE) EMERGENCED IN THE REARING OF NECROPHAGOUS FLIES IN KOREA

Jang H.[1], Shin S.E.[1], Ko K. S.[1], Choi B. H.[2], Park J. H.[2], Park S. H.[1]

[1]Department of Legal Medicine, Korea, University College of Medicine, Seoul, Korea;
[2]Medical Examiner's Office, National Forensic Service, Wonju, Korea
kuforen@gmail.com

The unexpected emergence of parasitoid wasps in the rearing of necrophagous flies can be one of the reasons that estimation of the minimum postmortem interval (PMI-min) or the acquisition of growth information fails. Larval parasites of blowflies are well distributed over Europe and North America and have been found in Asia (Narendran, et. al., 2016). However, information about such parasitoid wasps is relatively scare. Authors report two cases of *Brachymeria podagrica*, primary solitary endoparasitoid species of family Sarcophagidae and Calliphoridae (Narendran, et. al., 2016) with a review of the literature for taxonomic keys and biological information. Four parasitoid wasps were found in the rearing of *Sarcophaga peregrina*, and *Lucilia sericata*, collected from corpses during the medicolegal investigation by South Korean authorities. The wasps were morphologically identified adults of *B. podagrica*. The body lengths were measured as 5.99 ± 0.53 mm (n=4) and the body color was shiny black. Round red hind femurs had yellow apexes and inner basal teeth, diagnostic characters of this species.

Case 1. A badly decayed corpse was found indoors in a suburb Seoul and the measured average room temperature of the scene was 28°C. 2nd instar larvae of *L. sericata* and 3rd instar larvae of *S. peregrina* were collected from the scene. Emergence of three adults *B. podagrica* from pupae of *S. peregrina* was observed after 22 days in a rearing chamber at 28°C. As *B. podagrica* requires 22 days from egg to adult in 27.2-29.4°C (R. A. Roberts, 1933), it was estimated that 3rd instar larvae of *S. peregrina* had been parasitized on the day when the corpse had been discovered.

Case 2. Pupae of Phoridae and 3rd instar larvae of *L. sericata* were collected from a badly decayed corpse during the autopsy. The corpse was discovered indoors in a suburb Seoul as the Case 1. The average room temperature was not measured. Larvae were reared at 25□ in the lab. After 22 days, an adult *B. podagrica* was emerged from a pupa of *L. sericata*. As *B. podagrica* requires 26 days from egg to adult in 24.4-26.7°C (R. A. Roberts, 1933), it was estimated that the temperature of scene had been at least 24.4-26.7°C.

From these two cases *B. podagrica,* the developmental data of parasitoid wasps may be applicable as supplemental evidences for PMI-min estimation. For more accurate and reliable estimation, the rearing of *B. podagrica* at constant temperatures should be performed.

Key words: Parasitoid, Chalcididae, *Brachymeria podagrica, Sarcophaga peregrina, Lucilia sericata*

1. T.C. Narendran, Cornelis van Achterberg, 2016, Revision of the family Chalcididae (Hymenoptera, Chalcidoidea) from Vietnam with the description of 13 new species, Zookeys, 576:1-202 doi: 10.3897/zookeys.576.8177.

2. R.A. Roberts, 1933, Biology of *Brachymeria fonscolombei* (DUFOUR), a hymenopterous parasite of blowfly larvae, Technical Bulletin No. 365, United States Department of Agriculture Washington, D. C.

20X20 cm CRIME SCENE

Vanin S.

Department of Biological Sciences, School of Applied Sciences, University of Huddersfield, UK;
GIEF, Gruppo Italiano per l'Entomologia Forense, Italy
stefano.vanin@hud.ac.uk

May a crime scene have a size of 20x20 cm?

May the investigators focus their attention to a human suspect in a so small crime scene?

May the insects kill people without biting and without any contact with the victims?

This presentation deals with a forensic case where insects caused the death of a person and two horses and they were also the cause of dramatic injuries in another person without being in contact with them.
The case was initially treated as a homicide with the search of a suspect till the unexpected results of an archaeological survey generated a new potential scenario confirmed by the entomological analysis.

AN ITALIAN COLD CASE: THE SAD STORY OF THE REMAINS OF A CHILD IN THE SUITCASE

De Micco F.[1], Cavezza A.[1], Bugelli V.[2,3,4], Campobasso C.P.[1,4]

[1] Dept. of Experimental Medicine, University "L. Vanvitelli" of Campania, Naples, Italy; [2]Dept. of Medicine and Health Sciences (DiMeS) University of Molise, Campobasso, Italy; [3]Dept of Medicine and Health Sciences, Forensic Science Section, University of Florence, Firenze, Italy;
[4]GIEF, Gruppo Italiano per l'Entomologia Forense, Italy
francesco.demicco@gmail.com

A 7-year old boy went missing on 08/11/1997 at 8:20 a.m. in Naples. Soon after the disappearance the search of missing child began. Broadcasters promptly transmitted a description of the missing child, pushing the entire community to assist in the search and safe recovery of the child. Some specifically trained search dogs were also used and they stopped repeatedly in front of the house of a neighbor. Unfortunately, every effort was in vane, and the search went on for a couple of weeks. After 10 days the neighbor, a man of 70 years old, confessed that he used to sexually abuse the child along with his sons in law. He also confessed that the perpetrators killed the child in the same day of disappearance, hitting his head and they burnt the body in a countryside for its concealment. However, no human remains were found on the death scene indicated by the killer. Eight years after the confession, the new owner of an apartment in a small town close to Naples, found a suitcase. Inside the suitcase there was a skeletonized body, still clothed with some dress remnants and personal belongings. The skeletal remains and clothes were fully covered in pupae and puparia. The autopsy findings showed no burning defects on the clothing remnants and no skeletal injuries by fire in disagreement with the confession. The autopsy showed a diastasis or springing of the coronal and sagittal sutures with no other skull fracture consistent with a relevant impact of the head that widened the suture lines. Therefore, traumatic brain injury was the leading cause of death. Based on the anthropological analysis, skeletal and dental age were consistent with a sub-adult of 7 years old. DNA analysis showed no incongruities between the missing individual and the DNA collected from parents. Unfortunately, analysis was not performed on entomological findings. The pictures taken at the autopsy show a large number of pupae and puparia morphologically resembling to those of the Calliphoridae. No Diptera adults were found and no other insect evidence was available for the estimation of the post-mortem interval (PMI). The presentation of this case will impact the scientific community on the amount of information potentially provided by a forensic entomological analysis in a cold case dealing with human remains found in enclosed sequestered environments after years. In this regard, a prompt entomological investigation could be able to provide more information regarding at least the seasonality of the insect colonization and verify the circumstances related to the death events as confessed by the perpetrators. Among these latter, an interesting question to resolve could be related to the PMI estimation and, in particular, the period of insect colonization if before or after the concealment in the suitcase. There is no evidence available to determine if the body was concealed somewhere else before the final settlement in the suitcase or sometime after that. According to some recent studies, blowflies are able to colonize resources inside closed suitcases and if the food source is not assessable the flies can lay their eggs on the surfaces of openings. In fact, first instar larvae of calliphorids are able to colonize the remains by passing through gaps between the teeth of the zipper over 0-20 mm (Bhadra P. et al, 2014).

PATTERN OF DECOMPOSITION RATE AND INSECT COLONIZATION IN CASES OF SUICIDE BY HANGING

Bugelli V.[1,2,5], Gherardi M.[3,5], Vanin S.[4,5], Campobasso C.P.[6,5]

[1]Dept. of Medicine and Health Sciences (DiMeS) University of Molise, Campobasso, Italy; [2]Dept of Medicine and Health Sciences, Forensic Science Section, University of Florence, Firenze, Italy; [3]SC Medicina Legale AUSL Valle D'Aosta; [4]Department of Chemical & Biological Sciences, School of Applied Sciences, University of Huddersfield, UK; [5]Dept. of Experimental Medicine, University "L. Vanvitelli" of Campania, Naples, Italy; [6]GIEF Gruppo Italiano per l'Entomologia Forense, Italy
vale.buge@gmail.com

Hanging is one of the most common methods of suicide worldwide. Despite this high incidence, there is a remarkably little knowledge of the patterns of cadaver colonization by insects on a hanging corpse. Hanging could be either by complete or incomplete suspension, which means toes, feet, knees or other part of legs touch the ground or other surfaces. The different type of hanging can alter body decomposition process as well as the pattern and rate of insect colonization. We describe two case studies where the hanging occurred in the same season and in the same type of habitat (woodland) with a similar post-mortem interval (PMImin) of exactly 34 days. Despite the above similarities, the bodies showed a totally different type of decomposition scored using the Total Body Score (TBS) (Megyesi's et al., 2005) and pattern of insect colonization. CASE 1: 24-year old male was found dead in the middle of July. He was found totally suspended from a branch of a big tree by a leather belt, 34 days after he disappeared. The feet were at about 1 meter above the ground in close proximity to a stone-wall. The mummification process took place especially in the upper anatomical parts (head, thorax, upper limbs). Hands and feet showed signs of epidermal maceration and skin slippage along with marbling at the lower arms and lower limbs. A TBS of 13 was assigned. An overall mean temperature of 21.5 ± 2.5°C was recorded in the 40 days before the recovery of the body. A large amount of dipteran and coleopteran larvae was found on the neck and on the body. CASE 2: 21 years old male immigrant was found at the end of September 34 days after his disappearance from the reception center. The head was disarticulated and partially skeletonized. The skull was found just approximately 2 meters away from the body. The rest of the body was pre-skeletonized, fully clothed. A TBS of 31 was assigned. A large amount of pupae and puparia were found on the ground and on the clothing remnants. Insect specimens were later identified as pupae and puparia of *Chrysomya albiceps* (Calliphoridae) and pupae of *Hermetia illucens* (Stratiomyidae). Several puparia of *C. albiceps* showed also parasitoid emergence holes. Adults of *Nasonia vitripennis* (Hymenoptera: Pteromalidae) were also found associated with the remains. A PMImin of 25-31gg was estimated according to developmental data of the species based on the mean temperature (25.1 ± 2.7°C), recorded in the 40 days before the recovery of the body. According to previous studies, different patterns of decomposition were mainly related to the diversity of arthropod community found on human remains because of the different type of hanging: the first one totally suspended, the second one in contact with the soil. According to Lynch-Aird et al. (2015) a new scoring scale for hanging bodies totally suspended should be useful in order to include the mummification of upper body parts common in these cases. In fact, desiccation of soft tissues can explain the delay in decomposition of hanging bodies as well as the decrease of internal maggot masses is mainly related to the effect of gravity causing the maggots to fall from the body.

FIRST FINDINGS OF *SYNTHESIOMYIA NUDISETA* (DIPTERA: MUSCIDAE) IN FORENSIC CASES IN NORTHERN ITALY

Lo Pinto S.[1,3], Giordani G.[2,3], Tuccia F.[2,3], Ventura F. [1], Vanin S.[2,3]

[1]Department of Legal and Forensic Medicine, University of Genova, Genova, Italy; [2]Department of Biological Sciences, School of Applied Sciences, University of Huddersfield, UK;
[3]GIEF, Gruppo Italiano per l'Entomologia Forense, Italy

In previous years, the accumulation of evidence of global warming and globalization has increased the interest in their ecological consequences; of particular interest are the distribution changes and phenology of the different insect species. This phenomenon is affecting also the entomofauna associated with carrion and cadavers that plays an essential role in the estimation of the time since death in forensic cases. The species distribution shift, in the forensic context, has been mainly observed in Diptera of different families: Calliphoridae, Stratiomyidae and Phoridae. In the past species, especially tropical, had little chance of spreading geographically, because of climatic and environmental differences.
In 2013 the presence of the tropical carrion feeding species (native of South America and East Asia), *Synthesiomyia nudiseta* (Diptera: Muscidae), was reported from forensic cases in Spain and in 2016 from Italy, where the species was collected from 5 dead bodies in the Genoa district.
The findings of *S. nudiseta* in Northern Italy are a clear example of change in the distribution of species of forensic importance. After the DNA analysis and the study of the importation data, it's possible to suggest and sustain the introduction of this species in Italy was through commercial exchanges with Asian countries instead of a variation in the species distribution area from Spain and Portugal.

ESTIMATION OF THE POSTMORTEM INTERVAL IN ANIMAL CADAVERS

Paciello O.

Department of Veterinary Medicine and animal production
University of Naples Federico II - Naples, Italy
paciello@unina.it

The estimation of the postmortem interval (PMI) is important in many human and animal death investigations. Despite many decades of research, accuracy in estimation of the time of death has not significantly improved and no single method can be reliably used to accurately estimate the time of death. The majority of the studies in the field has been focused on human bodies and, sometimes, they have been translated to animals. It has also occurred that investigations on animal cadavers served as model to better understand the physical and chemical changes that occur in humans after death. Such investigations include gross changes, microscopic changes, temperature-based methods, postmortem chemistry, molecular methods, microbial assay, ocular changes, radiography, entomology, and others. Some new studies has been performed in our Department, regarding: 1) the modifications of the lens transparency of dead dogs over the time and at low temperatures. In this first study we examined the lenses of twenty-five adult dogs at different time of death and at different storage temperature to assess variations of the optical density of the lens. In frozen dogs at -18°C for 7 days, macroscopically, we observed opacification of the lenses and increase of optical density. Instead, the removed lenses, stored at room temperature (24°-26°C) showed positivization of the optical density after 6 days post-mortem. 2) In the second study we investigated the immunohistochemical expression of two cytoskeletal proteins, desmin and dystrophin, in muscles of dead dogs over the time in order to establish if there were statistical correlation with increasing of the PMI. The histological examination showed foci of muscle disintegration characterized by ruptured fibres and a loss of cell borders after 4 days post mortem (dpm). Immunoistochemical examination showed a more rapid dystrophin degradation with complete disappearance of the immunoreactivity after 4 dpm. In contrast, desmin was detected in dog muscle for all 6 days of observation with progressive reduction of immunoreactivity cells during the time. This second study demonstrates that the muscle proteins have a time dependent degradation. Moreover, our immunohistochemical findings indicate that there is a difference in degradation among the various proteins of the muscle during storage. Although only several of the methods to estimate the PMI are currently practical for use in the workup of cases, it is expected that future research will result in improved techniques that will benefit both human and veterinary forensic investigations.

[1] Brooks J.W.Postmortem Changes in Animal Carcasses and Estimation of the Postmortem Interval, Veterinary Phatology, 53:929-40, 2016 [2] Gaballero et al. Post mortem changes produced in the muscle of sea bream (Sparus aurata) during ice storage, Acquaculture, 291:210-216, 2009 [3] Cheryl et al. The effect of post-mortem delay on immunohistochemical labelling—a short review, Comparative Clinical Pathology, 20:95-101,2011 (4) Vass et al. Decomposition chemistry of human remains: a new methodology for determining the postmortem interval. Journal of Forensic Science, 47:542–553, 2002

THE USE OF INSECTS IN VETERINARY FORENSIC FIELD INVESTIGATION

Mastrogiuseppe L.

Dipartimento di Prevenzione – Servizio Veterinario
A.S.Re.M – Campobasso, Italy
luigi.mastrogiuseppe@asrem.org

Forensic entomology is the study of insect and arthropods in criminal investigation. In the veterinary context it can be applied both living and dead animals and it can be useful in cases of cruelty, abuse cases and illegal killing of animals. Although there are many ways insects can be used in an investigation, the main application is to determine the minimum postmortem interval, or time since colonization; the time it takes for each species of fly to reach and colonize the corpse, which may or may not correlate exactly with the time of death. In fact it's important not to confuse this value with the post mortal interval (P.M.I. or time since death) as there are many environmental factors that can delay insect colonisation, such as weather conditions (first of all ambient temperature) and access to the body. It can also be used to establish whether the corpse has been moved after death, to recognize a wound, to estimate the cause of death and to associate or discard the suspect at the scene of death. Entomology applied on live animals (myiasis) is the feeding by maggots on living tissue. It not only occurs in wild animals where common wounds are untreated but occurs also primarily in domestic debilitated animals because of neglect in presence of deliberate wound cleanliness or lack of hygiene in the environment. The forensic veterinarian has to learn to properly collect and preserve insect evidence in all these cases and forward them to a forensic entomologist for correct identification and aging.

[1] Amendt J1, Richards CS, Campobasso CP, Zehner R, Hall MJ. Forensic ntomology:applications and limitations.Forensic Sci Med Pathol. 2011 Dec;7(4):379-92 [2]Brooks JW Postmortem Changes in Animal Carcasses and Estimation of the Postmortem Interval Vet Pathol. 2016 Sep;53(5):929-40 [3] Melinda D. Merck Veterinary Forensics: Animal Cruelty Investigations 1st Edition.

UNUSUAL ENTOMOLOGICAL LOCALIZATIONS; A WILDLIFE EXPERIENCE.

Marchetti C.[1] , Mastrogiuseppe L.[2], Pisani GM.[1]

[1]Department of Veterinary Medical Science, University of Parma, Parma, Italy;
[2]Servizio Veterinario -Dipartimento di Prevenzione-A.S.Re.M.,Campobasso, Italy
cristina.marchetti@nemo.unipr.it

The aim of our presentation is to communicate unusual feeding sites of maggots and beetles in wildlife fauna living in natural habitat.
Case 1- At the beginning of May, an old male roe (*Capreolus capreolus*, Linnaeus, 1758) was rescued in a field in the municipality of Noceto (Parma). The animal was ataxic and he died shortly after the hospitalization with severe cerebral myiasis. At the necropsy we diagnosed a penetrating head trauma, likely caused by the antlers of another roe during a fight, and brain invasion by maggots.
Maggots were also present not only at their preferred feeding sites (eyes, nose and mouth) but even at pharyngeal tract and esophagus.
Blowflies (Calliphoridae) and flesh flies (Sarcophagidae) cause myiases of relatively short duration by both obligate and/or facultative parasitism and infections maturing within 4– 7 days, in host body orifices or wounds (e.g. *Lucilia cuprina, Lucilia sericata, Cochliomyia hominivorax*) (Otrantoa D and Stevens J.R. 2002).
Case 2- A young female mouflon (*Ovis musimon*, Pallas, 1762) was found, at the beginning of April, in a wooded area inside of Monte Fuso's Park, an extensive wildlife park near Parma. The carcass was smeared with fecal material and showed clear signs of postmortem predation from birds (the only one predator that can enter into the area). The necropsy was performed on site and at the exam of meseraic lymph nodes all of them were enlarged and one showed internal colonization by beetles.
Case 3- At the end of October 2015 an otter (*Lutra lutra*, Linnaeus, 1758), was found in the district of Tufara (Campobasso). First we proceeded at the time of death definition. The finding of maggots into the circum anal glands and interdigital space permitted establishment of the time of colonization. The otter died for road trauma during a rainy period.
Those experiences indicate that veterinary pathologists must acquire more knowledge about forensic entomology and the estimation of colonization time is an important tool in a forensic contest.

DECOMPOSITION AND ARTHROPOD COLONIZATION OF WRAPPED RABBIT CARRIONS IN NORTH CENTRAL ALGERIA

Taleb M.[1], Tail G.[1], Benzekkour M.[1], Bouderbala D.[1], Djedouani B.[2], M.Toumi[2], Açıkgöz H.N. [3]

[1]Laboratory of Biotechnologies, Environment and Health, Faculty of Nature and Life Sciences, University of Blida 1, Blida, Algeria; [2]Entomology Laboratory, Department of Legal Medicine, National Institute for Criminalistics and Criminology, Cheraga, Algeria; [3]Forensic Biology/Forensic Entomology Laboratory, Forensic Sciences Institute, Ankara University, Ankara, Turkey
meriemtaleb1@gmail.com

Criminals often attempt to conceal evidence of murders. Several cases reporting dumped body parts or infant cadavers in plastic bags are often recorded. It is crucial to take into consideration the presence of an obstacle that may hinder or prevent the access of insects. Therefore, a field trial which represents a simulation of cases of dumped infant bodies or body parts in plastic bags was conducted using rabbit carcasses. The aim of this survey was to investigate the impact of wrapping on carcass decomposition and arthropod colonization.
The investigation was carried out during spring in the experimental station of the faculty of Nature and Life Sciences, Algeria. The coordinates of the study area are 36 ° 30 '16. 994 "North, 2 ° 52' 21.629" East; the average elevation is 188 m above sea level. Twelve live domestic rabbits (*Oryctolagus cuniculus* L.) carcasses weighing 2.55 ± 0.42 kg were used during this study. The animals were euthanized by concussion method. Six carcasses were wrapped in translucent plastic bags. The bags were not closed securely since in most cases plastic wrap was not secured tightly.
Temperature did not vary much during the survey period. All decomposition stages were observed in all carcasses, with the same durations in controls. However, durations of decomposition stages in wrapped carcasses were variable. A total of 12337 specimens, belonging to 30 families and 69 species were collected along all sampling periods. Diptera, Coleoptera and Hymenoptera orders dominated carrion communities in this study. Higher values of abundance and species richness were found in the exposed carcasses group. There was a similar composition of arthropods species between the exposed and the wrapped cadavers. We found no differences between the two groups in the sequence of occurrence of the most important carrion insects
Our observations show that the presence of plastic wrapping did not influence the accessibility of necrophagous insects. In the current study, wrapping did not delay the arrival of fly species encountered but affected the abundance of the first colonizers.

AN INVESTIGATION OF POST-FEEDING LARVAL DISPERSAL IN UK BLOW FLIES

Mactaggart M.[1,2], Whitaker A.[1,2], Hall M.[2], Wilkinson K.[1]

[1]University of Winchester, UK; [2]Natural History Museum, London, UK
m.mactaggart@unimail.winchester.ac.uk

In order to determine the minimum post-mortem interval (minPMI) in forensic investigations, it is essential to collect the oldest insect specimens associated with the body. Blow flies are the primary colonisers of a cadaver and are therefore acknowledged as the most important insects in determining minPMI. The oldest blow fly specimens are often not found on the body itself, but have dispersed to find a suitable pupariation site, and therefore may be some distance from the body on which they were feeding. To locate these dispersing larvae and puparia, it is first necessary to know where to look for them. A literature review revealed that much information is either contradictory or missing concerning the behaviour of post-feeding blow fly larvae. This has highlighted a need for a more complete study exploring the many factors that may affect larval dispersal. Perhaps most important is to determine how far the post-feeding larvae will disperse and how deep they will burrow; in order to maximise crime scene collection efforts.

Experiments were conducted to examine the dispersal behavior of UK blow flies, specifically the distance dispersed (horizontal dispersal) and the depth dispersed (vertical dispersal) prior to pupariation. Post-feeding *Calliphora vicina* larvae were introduced into different experimental apparatuses to explore the different factors: a 600 x 10 x 10 cm plastic channel was used to investigate horizontal dispersal; a 60 x 8 cm plastic pipe was used to look at vertical dispersal; and a 100 x 50 x 5 cm wooden apparatus was used to look at horizontal and vertical dispersal in conjunction with each other. The results suggest that in an experimental laboratory setting *C. vicina* can disperse up to 500 cm horizontally and up to 50 cm vertically. Further experiments will also be discussed.

METAMORPHIC BRAIN REMODELING AS AN AGE INDICATOR DURING THE BLOW FLY INTRA-PUPARIAL PERIOD

Martín-Vega D.[1], Simonsen T.J.[2], Wicklein M.[3], Garbout A. [4], Ahmed F.[4], Hall M.J.R.[1]

[1]Department of Life Sciences, Natural History Museum, London, UK; [2]Naturhistorisk Museum Aarhus, Aarhus, Denmark; [3]Department of Neuroscience, Physiology and Pharmacology, University College London, London, UK; [4]Imaging and Analysis Centre, Natural History Museum, London, UK
d.martin-vega@nhm.ac.uk

Blow flies (Diptera: Calliphoridae) are generally the first colonisers of cadavers and, therefore, exemplar forensic indicators for estimating a minimum post-mortem interval ($_{min}$PMI) if developmental data on the pertinent species are available. Within the blow fly development, the intra-puparial period, during which the developing insect lies inside an opaque puparial case that shows no significant external changes, can account for more than half of the duration of the life cycle. Hence, reliable aging methods for this developmental period are of particular importance.

Previous studies had shown the potential of non-destructive micro-computed tomography (micro-CT) for providing accurate estimations of insect age during the intra-puparial period based on both qualitative and quantitative analyses. Particular organ systems, such as the alimentary canal and the indirect flight musculature, have proved to be highly informative. In the present study, we further explore the applicability of micro-CT to forensic entomology with a focus on the morphological changes of the brain during the intra-puparial period of the blow fly *Calliphora vicina* Robineau-Desvoidy. Unlike most other larval organs, the central nervous system persists into the adult stage, but its anatomy is significantly remodeled during metamorphosis. With the use of a Nikon Metrology HMX ST 225 micro-CT scanner and a high resolution Zeiss Xradia 520 Versa 3D X-ray microscope system, we have identified informative morphological changes during the brain remodeling, which can be used as qualitative markers of age. Moreover, the analysis of tomographic data yielded quantitative measures of the volumetric changes of selected neuropils during metamorphosis. With the expected increase in its availability to medico-legal practitioners in the near future, we suggest micro-CT as a suitable tool for non-invasive internal analyses of the puparial samples collected at the forensic scene.

TAXONOMY OF FORENSICALLY IMPORTANT FLESH FLIES (DIPTERA: SARCOPHAGIDAE) OF THE MIDDLE EAST

Szpila K.[1], Jafari S.[2], Akbarzadeh K.[2], Piwczyński M.[1]

[1]Chair of Ecology and Biogeography, Faculty of Biology and Environmental Protection, Nicolaus Copernicus University, Toruń, Poland; [2]Department of Medical Entomology and Vector Control, School of Public Health, Tehran University of Medical Sciences, Tehran, Iran
szpila@umk.pl

The Middle East is a region of high diversity for flesh flies. Large parts of the region are characterised by desert or semi-desert habitats. Local conditions often facilitate intensive colonisation of vertebrate remains by flesh flies, which are competitive carrion breeders in dry habitats. The majority of forensically important flesh fly species belong to the genus *Sarcophaga*. They are grouped mostly in the subgenera *Liopygia* and *Liosarcophaga*, which include species with a strong propensity to carrion like *S. aegyptica*, *S. dux*, *S. argyrostoma*, *S. crassipalpis*, *S. ruficornis* and *S. tibialis*. Other ubiquitous species with a known preference for faeces have been reported in experimental and real cases, such as *S. africa* and *S. hirtipes*. In the subfamily Paramacronychiinae, *Wohlfahrtia* species are very common and effective as carrion breeders in true deserts. Buried carrion is utilized as a breeding substrate by species in the third subfamily of flesh flies, the Miltogramminae. Most of the above-mentioned species are taxonomically well known, but descriptions, revisions and keys are scattered in publications that are often poorly available to local scientists. Flesh flies are notoriously difficult to identify to the species level, especially females and preimaginal stages, which are usually more abundant on carrion than males. This problem can be overcome by applying molecular identification methods such as barcoding. GenBank offers barcode sequences for most of the mentioned species of *Sarcophaga* but only few of them refer to local populations from the Middle East. Sequences of species belonging to the other subfamilies are much less numerous.

To improve this situation, we: 1) performed a literature search of records of flesh flies from carrion ecology and real case studies in the Middle East; 2) prepared keys for the morphological identification of males, females and larvae; 3) created a barcode sequence library of Middle Eastern species, with special attention to species of Paramacronychiinae and Miltogramminae.

NEXT GENERATION SEQUENCING OF ARTHROPOD COMMUNITIES ON DECOMPOSING BODIES – FORENSIC ENTOMOLOGY AND DNA BARCODING

Chimeno C.[1], Morinière J.[1], Hausmann A.[1], Reckel F.[2], Grunwald J.E.[2], Haszprunar G.[1]

[1] Zoologische Staatssammlung München (SNSB-ZSM), München, Germany;
[2] Bayerisches Landeskriminalamt, Abteilung II, Sachgebiet 204, München, Germany
ca_chimeno@yahoo.com

In collaboration between the Bavarian State Criminal Police Office (BLKA) and the Bavarian State Collection of Zoology (SNSB-ZSM), the application of Next Generation Sequencing (NGS) was tested with the goal of identifying arthropod communities within bulk samples collected on carcasses. A major challenge of forensic entomology is warranting rapid, exact, and reliable species identification as immature life stages of many arthropods share similar features, making their morphology-based distinction difficult or impossible. Often, collected larvae need to be reared under special conditions to the imago, which offers better differential features. This time-consuming method is not beneficial if fast identification is of crucial interest. In this case, DNA barcoding provides a promising alternative using molecular markers for species identification. Within the framework of the German Barcode of Life (GBOL) project, the SNSB-ZSM aims to obtain genetic fingerprints (DNA barcodes) of all German animals. Approximately 20,000 animal species have already been successfully barcoded and provided for the international Barcode of Life Database (BOLD).
Here, we present the results of a three-phase project in which we established (i) a reference library containing approximately 100 species which play an important role in forensic entomology to back up the NGS-based analyses of (ii) 30 arthropod bulk samples collected from dead human bodies which were provided by the local morgue in Munich and (iii) over 450 arthropod communities which were collected from decomposing pigs over a period of three months (July – September 2016) with the goal of identifying species as being relevant for forensic investigations. The last phase consisted of analysing the succession of arthropod species present on the decomposing pigs.
Our results indicate that DNA barcoding is a promising tool for forensic entomologists, because it facilitates accurate and fast species identification of all developmental stages in the absence of specialized taxonomists. The use of Next Generation Sequencing on mixed bulk samples containing hundreds of individuals is especially interesting when facing large aggregations of eggs and larvae. Although it was not the focus of our study, and we did not replicate the experiment with other pigs, preliminary results show differences in the arthropod communities due to the presence or absence of clothes.

INCREASING THE USE OF ENTOMOLOGICAL EVIDENCE: PRACTICAL FOOD SOURCE OPTIONS FOR CRIME LABORATORIES

Weidner L.M.[1], Nigoghosian G.[1], Hanau C.G.[1]

[1] Purdue University, West Lafayette, Indiana, 47907, USA
lweidner@purdue.edu

Forensic entomology is a well-established tool for evaluating death, or abuse, of a person or companion animal. Insect evidence provides valuable information as related to time of colonization (TOC) and movement of remains from one location to another. Blow flies are commonly found on human remains throughout most stages of decomposition and consequently when entomological evidence is collected these tend to be the most abundant taxa. However, very few crime laboratories across the United States have established collection and rearing protocols for these forensically important insects. Some of the main challenges for using blow flies as evidence are likely due to the fact that proper collection techniques are not always widely disseminated, and it can be difficult to have access to an appropriate food source. Further, the majority of crime laboratories are not equipped with a forensic entomologist on staff. Thus, when crime scene investigators or pathologists collect these insects, they are often mishandled (e.g., placed into containers with no air holes, no food, or a food source that is not sustainable for their development). This study analyzed alternative food sources for blow flies that are easily accessible and cost efficient, including; tuna, wet cat food, and beef liver (control). Two species were used, *Phormia regina* (Meigen) and *Cochliomyia macellaria* Fabricius. Survivorship and development time were analyzed for each species across each food source. It was found that *P. regina* had significant differences in survivorship based on food source, while food source had no effect on *C. macellaria.* Significant differences were also found in development times across food source within each species. This experiment will provide an overview of possible food alternatives that could be used as a sustainable food option in laboratories when immediate assistance from a forensic entomologist cannot be obtained.

MAGGOT MASSES AND MICROBIOMES: THE INTERACTIONS OF INSECTS AND MICROBES IN THE NECROBIOME

Benbow M.E.[1,2], Weatherbee C.R.[1], Pechal J.L.[1]

[1] Department of Entomology, Michigan State University, East Lansing, MI, USA;
[2] Department of Osteopathic Medical Specialties, Michigan State University, East Lansing, MI, USA
benbow@msu.edu

Research into the potential use of postmortem microbiomes of the necrobiome (i.e, community of species associated with carrion) has grown over the last several years. The premise for this research is based on experiments that have shown microbial communities go through community succession that may be important for forensics (Pechal et al., 2014a; Metcalf et al., 2016). However, despite the demonstrated potential of using this next generation biological tool in forensics, few studies have attempted to collect, analyze and interpret the microbial communities associated with insects and other arthropods that colonize decomposed remains. The influence of necrophagous insects on postmortem microbial communities has rarely been studied, even though evidence suggests that they play a significant role in postmortem microbial succession dynamics of carrion (Pechal et al., 2014b). In this presentation we address an important question about microbiomes in decomposition: How do necrophagous fly larval masses influence the microbiome succession of a decomposing carcass? To answer this question, we used swine carcasses (N=6) to characterize microbial communities every 12h using high-throughput 16S rRNA sequencing from three carcass locations: skin; internal microbiome of surface decontaminated individual larvae (maggots); and the maggot masses (external communities of larvae). Co-occurring blow fly species were collected from masses: *Phormia regina*, *Lucilia coeruleiviridis*, and *Cochliomyia macellaria*. The most predominate species at the beginning of decomposition was *C. macellaria*, but was replaced by *P. regina* during later decomposition, with *L. coeruleiviridis* intermediate throughout decomposition. This succession suggests that blow fly species may be associated with microbial temporal shifts. Proteobacteria and Firmicutes inversely changed in relative abundance over decomposition. The relative abundances of microbial families changed during decomposition and across sampling locations, suggesting significant interactions between the environment, microbes, and insect larvae. After maggot masses had formed, the individual larval internal microbiomes and maggot mass microbiome communities converged with the skin communities, indicating that the larvae and masses were likely incorporating microbes from the carcass. Further, the Xanthomonadaceae (often environmental taxa associated with flies) were very low in relative abundance on carcass skin prior to maggot mass formation, but then significantly increased with larval presence; indicating the blow flies may inoculate carcasses with their own, exogenous microbes. These reciprocal interactions of insects and microbes during decomposition offer new insight and potential challenges into using microbiomes during death investigation.

Pechal *et al.* 2014a. The potential use of bacterial community succession in forensics as described by high throughput metagenomic sequencing. *Int J Legal Med* **128**, 193-205. Metcalf, *et al.* 2016. Microbial community assembly and metabolic function during mammalian corpse decomposition. *Science* **351**, 158-162. Pechal *et al.* 2014b. Delayed insect access alters carrion decomposition and necrophagous insect community assembly. *Ecosphere* **5**, art45.

COMMUNITY ASSEMBLY OF CARRION AND CALLIPHORID MICROBIOMES

Pechal J.L.[1], Weatherbee C.R.[1], Benbow M.E.[1,2,3]

[1] Department of Entomology, Michigan State University, East Lansing, MI, USA; [2] Department of Osteopathic Medical Specialties, Michigan State University, East Lansing, MI, USA; [3] Ecology, Evolutionary Biology, and Behavior Program, Michigan State University, East Lansing, MI, USA
pechalje@msu.edu

Communities consist of a collective group of interacting species or genes that fluctuate in space and time. The resulting dynamic interactions among species can have tremendous impact on host or ecosystem structure and function. A microscopic group of organisms, including but not limited to bacteria, archaea, protists, fungi, and viruses (or the microbiome), has garnered an intensification of research interests due to its known functional role in controlling, among many other examples, host physiology, behavior, nutrition, and pathogenesis. However, the complexities of the microbiome constituents during vertebrate decomposition have only recently begun to be holistically explored. Developing a better understanding of the basic ecological interactions of micro- and macro-organisms associated with carrion during decomposition and their community assembly has potential for increased use in forensics.

Replicate swine carcasses (*Sus scrofa* L., *ca*. 19 kg) were placed in a grassy field surrounded by deciduous forest in the Pre-Wisconsinan Drift Plains ecosystem (West Lafayette, IN, USA). Carcasses were similarly oriented on two East-to-West transects that were no less than 50 m apart; all carcasses were covered with anti-scavenging cages. Blow fly (Diptera: Calliphoridae) adult and larval communities were sampled every 12 h until the carcasses had skeletonized. Adult blow flies were passively collected using bait and glue traps, while larval specimens were collected by hand during mass formations. The carcass epinecrotic microbial communities were concurrently collected every 12 h via swabbing from three anatomic areas (mouth, skin, rectum) during decomposition. Targeted amplicon based (16s rRNA – bacteria and archaea) high-throughput next generation sequencing was used to characterize: the carcass microbiomes to assess postmortem succession; and the internal blow fly microbiomes of larvae and adults to assess ontogenetic variation. Carcass decomposition from fresh to skeletal remains took eight days at a mean ambient temperature of 23.0 ± 0.8^{o}C. The three prominent blow fly taxa collected were *Phormia regina* (Meigen), *Cochliomyia macellaria* (Fabricius), and *Lucilia coeruleiviridis* (Macquart), while the two predominate bacteria phyla present were Firmicutes and Proteobacteria across all samples (carcass and calliphorid adults and larvae). Overall, there was evidence of succession for all taxa sampled as decomposition progressed.

Our results demonstrate the community assembly patterns of the predominant micro- and macro-organisms of the necrobiome during vertebrate decomposition. Continued research of these dynamic necrobiome communities by using novel genomic tools is warranted in order to achieve a better understanding of how species interact throughout decomposition. Resulting information from microbe-insect interactions could span across basic and applied disciplines in the future for this emerging field, such as estimating the postmortem interval for forensic practitioners.

SAME SAME, BUT DIFFERENT! - DECODING THE NUTRITION HISTORY OF BLOW FLIES BY ISOTOPE SIGNATURE ANALYSIS OF ADULT FLIES AND THEIR EMPTY PUPAL CASES

Bernhardt V.[1], Scheid N.[2], Holdermann T.[2], Schäfer T.[2], Verhoff M. A.[1], Amendt J.[1]

[1]Institute of Legal Medicine, Goethe-University, Frankfurt/Main, Germany;
[2]Forensic Science Institute, Central Analytics II, Federal Criminal Police Office, Wiesbaden, Germany
bernhardt@med.uni-frankfurt.de

Since only the feeding larval stages of forensically important flies can be assigned to a particular corpse with certainty, the association of empty puparia or adult flies and a decomposing human cadaver at a crime scene may be called into doubt; as such specimens could result from another food source and not from the corpse in question. This is especially valid for adult flies, as these could potentially have arrived at the scene only minutes before the investigators did. In light of the fact that recent techniques in forensic entomology target age estimation of adult flies and weathering of empty puparia as new tools for the analysis of the minimum time since death, there is need of a way to guarantee that specimens actually developed on the corpse in question.

We investigated stable carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$) isotopes in human muscle tissues (n = 10) and in the muscle tissue of 13 different animal species (n = 3-5 each) by means of isotope-ratio mass spectrometry. These isotopes were also investigated for adult specimens of the forensically important blow fly *Lucilia sericata* that had been reared on the above mentioned tissues and, additionally, for the empty puparial cases.

Adult flies grown on human tissue revealed an average $\delta^{13}C$ value of -23.05‰ (± 0.55‰) and an average $\delta^{15}N$ value of 11.38‰ (± 0.95‰). This displays an average $\delta^{13}C$ depletion of -0.39‰ and an average $\delta^{15}N$ enrichment of 2.64‰ in adult flies compared to the human tissue itself. In empty puparia, we saw an average $\delta^{13}C$ value of -23.35‰ (± 0.63‰) and an average $\delta^{15}N$ value of 6.49‰ (± 0.81‰). Compared to human muscle tissue, we found an average $\delta^{13}C$ depletion of -0.69‰ and an average $\delta^{15}N$ depletion of - 2.25‰ in empty puparia.

The $\delta^{13}C$ and $\delta^{15}N$ ratios of the human muscle tissues were in accordance with literature values for Germany. During metamorphosis, adult flies become enriched with ^{15}N, as can be observed at each change of trophic level. Since chemical bonds including ^{14}N break down more rapidly than chemical bonds including ^{15}N, excretion products -such as the chitin in empty puparia- are ^{15}N-depleted.

All of the analyzed animal species showed a $\delta^{13}C$ and $\delta^{15}N$ ratio pattern that can be clearly distinguished from the human $\delta^{13}C$ and $\delta^{15}N$ ratio pattern.

FORENSIC OR NOT FORENSIC II

Vanin S.

[2]Department of Biological Sciences, School of Applied Sciences, University of Huddersfield, UK;
[3]GIEF, Gruppo Italiano per l'Entomologia Forense, Italy
stefano.vanin@hud.ac.uk

Forensic biology is defined as the application of Biology to legal investigation and it covers a wide spectrum of disciplines from genetics to zoology, of organisms from algae to vertebrates and of cases from homicide to protect organisms' trade.
More specifically Forensic Entomology deals with the use of insects and other arthropods in medico legal investigations. If we consider as well urban and store products entomology, forensic entomology deals with the application of the knowledge about insects to legal investigations.
In all these definitions, and in others reported in the wide literature about this discipline there are three common words that make the difference between Forensic entomology and the other branches of Entomology: the magic words are "application", "legal" and "investigations/cases". We can apply our knowledge, analyses, studies to a legal investigation and we are doing Forensic Entomology.
However Forensic Entomology has developed in the last year an important core of "forensic" applicable studies and several researchers are exploring the new frontiers of the discipline with fascinating and promising results.
Despite the high scientific level of the produced works, the lack of knowledge in the forensic and legal language is evident and for this reason some publications reflect the spectacular CSI scenarios.
"Determination of the PMI", "solving a crime" and other sentences are only two examples.
This presentation will discuss and underline some language and conceptual errors found on the nowadays literature related with entomological studies.

QUANTITIATIVE PTERIDINE FLUORESCENCE AND N-PENTACOSANE ANALYSIS: POSSIBLE AGE-GRADING TECHNIQUES FOR THE ADULT STAGES OF THE BLOW FLY *CALLIPHORA VICINA* (DIPTERA: CALLIPHORIDAE)

Bernhardt V., Kinast R., Verhoff M.A., Zehner R., Toennes S.W., Amendt J.

Institute of Legal Medicine, Goethe-University, Frankfurt/Main, Germany
bernhardt@med.uni-frankfurt.de

In forensic entomology, the estimation of the minimal postmortem interval (PMI_{min}) is restricted to the juvenile stages of necrophagous insects and only works until the end of metamorphosis and emergence of the adult stages. Since many adult flies remain at the scene after hatching, especially at indoor crime scenes, being able to estimate their age would extend the calculable PMI_{min} by several weeks. Recently, several promising age-dependent morphological and physiological characteristics of adult flies have been investigated in medical and forensic entomology; however, the results are still preliminary and restricted to a few species only.

We examined adults of the forensically important blow fly *Calliphora vicina* and investigated the fluorescence levels of pteridine (a natural pigment) in the eyes as well as the amount of n-pentacosane (a hydrocarbon that helps prevent desiccation) on the legs by means of fluorescence analysis, respectively, by means of gas chromatography–mass spectrometry. From Day 1 to Day 20 (n-pentacosane), respectively, to Day 25 (pteridine) post-emergence, flies were kept at up to three different temperature regimes (20°C, 25°C, and fluctuating temperatures in the context of a field study) and at 12:12 L:D.

The fluorescence level of pteridine increased linearly with increasing age (females: R^2 = 0.777; males: R^2 = 0.802). Neither head weight nor temperature had an effect on pteridine fluorescence, but the difference between sexes was significant ($p < 0.001$). The n-pentacosane amounts of the legs increased linearly (R^2 = 0.949); no significant differences between sexes or leg weight could be detected.While the variation of the single characteristics seems too big, combining several aging methods to obtain more precise results seems promising. Here it is advantageous that different body parts of the same specimen can be used to analyze different characteristics like cuticular hydrocarbons (legs), pteridine (head/eyes), and maybe additionally the gonotrophic stage of the female abdomen.

A FORENSIC ENTOMOLOGICAL CASE OF NEGLECT OF AN ELDERLY MAN IN CALABRIA, SOUTHERN ITALY

Bonacci T.[1], Vercillo V.[2], Benecke M.[3]

[1] DiBEST Department, University of Calabria, 87036 Rende (CS), Italy;
[2] Azienda Sanitaria Provinciale, sezione di Medicina legale di Cosenza (CS), Italy;
[3] International Forensic Research & Consulting, Cologne, Germany
teresa.bonacci@unical.it

Some insect species are valuable as forensic indicators in cases of abuse and neglect. They deposit their eggs in and around clothing and skin and feed at open wounds, ulcers, natural openings, etc. (Zumpt, 1965, Sherman and Hall, 2000). In particular, immature stages of Calliphorids may give information on how long a person was neglected (Lord, 1990; Baumjohann et al., 2011) but also Muscids, Sarcophagids and other species may prove cases of neglect (Benecke et al., 2001, 2004). Here, we present a case of neglect of an 80-year-old incontinent, elderly man with a psychiatric illness in the urban area of Acri (Cosenza), Calabria, Italy. The colonization with Diptera larvae before his death (myiasis sensu strictu) shed a light on his suffering and to prove the neglect before death. In the apartment, garbage, feces and urine were scattered around. The window was closed. Cause of death was hypertrophic cardiomyopathy and chronic active hepatitis. The temperature in Acri city before the discovery of the corpse was 23.9 ± 1.35°C. On the corpse and near anal genital area, few mature larvae of *Musca domestica* Linnaeus, 1758 and *Fannia scalaris* (Fabricius, 1794) were found; also, dead adults of *M. domestica* were collected from the room (July 9, 2016). Both species are attracted to feces and urine. Also, 2nd and 3rd instar larvae of *Lucilia sericata* were found (but no dead Calliphorids). On the floor, dead adult *Musca domestica* L. and *Fannia scalaris* and many active and empty puparia of both species were found spread on the floor near the corpse and near the closed window of the room where was found the corpse. Some larvae were put in hot water first, then stored in 90% ethanol; some specimens were reared to adult stage. External examination of the corpse also revealed multiple irregular, brown injuries (1–4 mm) on the skin, typical of ant action (Bonacci and Vercillo, 2015). However, no Formicidae were found on the corpse and inside the apartment. Since the larvae of *Lucilia sericata* take 3.5 (25°C) to 4.5 days (22°C) to reach end of L3 (data from Austria; Grassberger & Reiter, 2001), and 10 (25°C) to 13 (20°C) days to reach the end of postfeeding state (data from Pakistan; El-Kady et al., 1999), we discuss the possibility that the family of the man neglected him over the course of at least one week.

Baumjohann K. Schiwy-Bochat KH., Rothschild M A. 2011. Maggots reveal a case of antemortal insect infestation. Int J Legal Med, DOI 10.1007/s00414-011-0571-8.
Benecke M., Lessig R. 2001. Chil neglect and forensic entomology. Forensic Science International, 120: 155-159.
Benecke M. Josephi E., Zweihoff R. 2004. Neglect of the elderly: forensic entomology cases and considerations. Forensic Science International, 146S: S195-S199.
Bonacci T., Vercillo V. 2015. Outdoor post-mortem bite injuries by Tapinoma nigerrimum (Hymenoptera, Formicidae) on a human corpse: case report. Journal of Forensic and Legal Medicine, 33: 5-8.
El-Kady EM., Kheirallah AM., Kayed AN., Dekinesh SI., Ahmed ZA. 1999. The development and growth of the house fly *Musca domestica vicina* and the blowfly *Lucilia sericata*.Pakistan Journal of Biological Sciences, 2(2): 498-502.
Grassberger M., Reiter C. 2001. Effect of temperature on Lucilia sericata (Diptera: Calliphoridae) development with special reference to isomegalen- and isomorphen-diagram. Forensic Science International, 120: 32-36.
Lord WD. 1990. Case histories of the use of insects in investigations, In Catts PE., Haskell NH. (eds.). Entomology & Death, A procedural guide, Joyce's Print Shop, Clemson, pp, 9-37.
Sherman RA., Hall, MJT. 2000. Medicinal maggots: an ancient remedy for some contemporary afflictions. Annu Rev. Entomol, 45: 55-81.
Zumpt F. 1965. Myasis in man and animals in the Old World: a textbook for Physicians, Veterinarians ans Zoologists. Butterworth, London, England, K. 267 pp.

ESTIMATING OF THE POSTMORTEM INTERVAL: MEDICAL LEGAL AND ENTOMOLOGICAL APPROACH

Defilippo F.[1], Guerra E.[2], Gaudio R.M.[2], Dottori M.[1], Bonilauri P.[1]

[1]Istituto Zooprofilattico Lombardia e Emilia Romagna, Reggio Emilia, Italy; [2] Unit of Legal Medicine – Department of Medical Sciences, University of Ferrara, Ferrara, Italy
francesco.defilippo@izsler.it

This study discusses 8 cases of cadavers that had been transferred for forensic entomology investigations to the Department of Forensic Medicine, Faculty of Medicine, Ferrara University, northern Italy, from 2013 to 2016. Variable death scenes were determined, including outdoor and indoor environments. The fly specimens found on the corpses obtained were the most commonly of the blow fly of family Calliphoridae, and consisted of *Chrysomya albiceps*, *Lucilia sericata*, *Lucilia caesar*, *Calliphora vicina*, *Calliphora loewi* and *Protophormia terraenovae*. The cadavers were examined and analyzed to evaluate the reliability of the entomological method in estimation of time elapsed since death, in relation to medicolegal approaches. The time elapsed since death was estimated collecting sufficient immature insect forms from the corpse according to the method initially described by Reiter (1984). The postmortem intervals recorded by the Medical Officer at the time of autopsy, were based on the "postmortem changes" in the body. The entomological approach for the postmortem interval (DENT) was compared to methods based on "postmortem changes" (DML) by t-test, linear correlation (Pearson's r) and Spearman's rank coefficient of Correlation, $p<0.01$ was chosen as the level of statistical significance (p-value).

The DML estimate in 8 considered cases range between 4 to 20.5 days (average 10.5), the DENT between 3.5 to 18 days (average 8.6). The mean difference between methods was 1.9 days less for DENT, but difference was not statistically significant (test -0.63 $p=0.54$). Considering the intervals of the two estimations, DML and DENT obtain comparable precision when the death was dated in less than 7 days, while DENT give more precise estimation (less interval range) in the other cases. In particular the entomological dating gives the maximum error of 2 days, when the death occurred after 18 days, while the coroner obtain a mean error in time elapsed since death, of 4 days. A Pearson's product-moment correlation was run to assess the relationship between DENT (x) and DML (y). There was a strong positive correlation between DENT and DML, $r(8) = 0.992$, $p < 0.01$, with $R2 = 98.3\%$. Results also confirmed by Spearman's rank coefficient of Correlation, 0.956.

The statistical evaluation indicates that an estimate of the postmortem interval by an entomological approach do not differ from methods based on "postmortem changes" with a strong positive correlation between the two methods. In average the DENT were two days less than DML and produce a smaller interval.

In conclusion, in our study, in absence of eye witness in the police report (gold standard), is not possible to establish which method between entomological and "postmortem changes" in the body, is more accurate and confirms the importance of combining the two approaches in dating the postmortem interval.

Reiter C (1984) ZumWachstumsverhalten der Maden der blauen Schmeiβfliege Calliphora vicina. Z Rechtsmed 91:295–308

STORED-PRODUCT FORENSIC ENTOMOLOGY: STUDY OF DEVELOPMENT OF *Plodia interpunctella* (LEPIDOPTERA: PYRALIDAE) AND USE OF DATA IN DETERMINING TIME INTERVALS OF FOOD INFESTATION

Defilippo F., Pinna M., Calzolari M., Dottori M., Bonilauri P.

Istituto Zooprofilattico Lombardia e Emilia Romagna, Reggio Emilia, Italy
francesco.defilippo@izsler.it

Forensic entomology is the intersection between insect science and the legal system. Although most recent attention has focused on the subspecialty known as "medico-criminal entomology," where insect development and succession on a corpse are used to estimate how long the decedent has been dead, there are additional areas where entomology is of legal interest. Stored-product entomology includes the study of a variety of important insects with legal implications. The typical case involves infestations during the harvesting and storage of crops and subsequent invasion by an insect pest, the infestation of food sold by retailers to the public or the invasion of foodstuffs in the domestic kitchen by insect pests, which may result in prosecution and substantial fines. In such situations the size/developmental stage of insects, together with ambient conditions may be used to estimate the age of insects and therefore minimum time since infestation. Time frames provided may be used to determine whether infestations are the fault of the producer, the retailer, or are the result of long domestic storage or even deliberate contamination during the shelf life of the product. This study discusses the effect of two different larval diets (D1=standard diet, D2=chocolate) and temperature on developmental rate of *Plodia interpunctella*. The study was conducted in a small climatic room with 60% ± 5% relative humidity and a photoperiod of 14:1 0 h (L: D), using temperatures of 20, 23, 25 and 27°C ± 0.5°C. We report the lower developmental thresholds estimated from the linear regression of developmental rates for both diets tested, the ADD (Accumulated Degree Days) for the period egg-eclosion, and the larval growth curves plotted for each of the constant temperature regimes. The lower thresholds for total immature development were estimated as 15.6 and 17°C for D1 and D2 respectively. These data allowed the calculation of the ADD. Different degree-days for each diets were observed (230°C for D1 and 346°C for D2). The growth rates obtained by analysis of the growth curves of the larvae to the stage pupal were significantly different ($p < 0.01$) between the 2 growth substrates and temperatures tested. Since *P.interpunctella* infests products stored in an environment with a roughly constant temperature, the use of the growth curve and rates could provide a quick and precise minimal estimate for the infestation time. Furthermore precise values of developmental minima and degree-day estimates for different larval substrates could be of great value for forensic entomological casework.

EFFECT OF ANTIFREEZE (ETHYLENE GLYCOL) ON THE LIFE CYCLE OF OF THREE SPECIES OF BLOWFLIES (DIPTERA: CALLIPHORIDAE)

Essarras A.[a], Pazzi M.[b], Magni P.A.[a]

[a] School of Veterinary & Life Sciences, Medical & Molecular Sciences Cluster, Murdoch University, Perth, Western Australia; [b] Department of Chemistry, University of Turin, Torino, Italy
p.magni@murdoch.edu.au

Entomotoxicology is a forensic discipline amidst extensive development; it involves the analysis of the effects of toxicological substances on forensically significant insects. Furthermore, this branch of forensic entomology considers the use of insects as an alternative matrix to detect drugs or other toxic substances in decomposing bodies, or in the absence of tissues or biological fluids.

Generally, the main focus of forensic entomology is about the estimation of the minimum time since death (minPMI) of a deceased human or animal using the succession status of carrion insects and/or the identification of the insects' species and age. However, in numerous studies it has been demonstrated that the estimation of such time frames may be severely compromised by the presence of drugs and toxins in the insects' food substrate. While the detection of drugs, metals, pesticides and alcohol has been reported in entomotoxicological studies, no studies regarding the effects of the common car antifreeze – ethylene glycol – on blowflies is available in the present literature.

Ethylene glycol is extremely easy to purchase, it is odorless and sweet. It may be consumed accidentally or purposefully in an attempt to cause death. Several cases report fatalities of humans (especially homeless people in cold cities) and pets. Toxicity and death may occur even after drinking a small amount of ethylene glycol, as the lethal dose for both humans and cats is 1-1.5 ml/Kg (ethylene glycol concentration 95%).

The present study describes for the first time the effects of ethylene glycol on the morphology (length of larvae and pupae), survival, developmental rate and F1 viability of three blowfly species (Diptera: Calliphoridae): *Lucilia sericata* (Meigen)*, L. cuprina* (Wiedemann) and *Calliphora dubia* (Macquart).

Blowflies were reared on substrates (beef liver) spiked with 3 different concentrations of ethylene glycol that could cause death in either a human or a cat: $1/2LD_{50}$, LD_{50}, $2LD_{50}$.

The results show a pattern between the concentration of ethylene glycol and the developmental time, the survival and the morphology of the three blowfly species considered in this study. At present, F1 viability is under analysis.

EFFECTS OF PARACETAMOL ON THE DEVELOPMENT OF *MEGASELIA SCALARIS* (DIPTERA: PHORIDAE)

King S. J. [1], Lo Faro A. F. [2], Wyville S.[1], Vanin S.[1,3]

[1]School of Applied Sciences, University of Huddersfield, UK; [2] Dipartimento di Scienze Biomediche-Chirurgiche ed Odontoiatriche, sezione Tossicologia Forense, Universita' degli Studi di Milano, Italy;
[3]GIEF Gruppo Italiano per l'Entomologia Forense, Italy
stefano.vanin@hud.ac.uk

In Forensic Entomology the estimation of the age of the insects is used for the estimation of the minimum post mortem interval (mPMI). Insect development rate is mainly temperature dependent despite other parameters that can affect the development. Several studies demonstrated that drugs and other chemicals can affect the growth of larvae, feeding on the dead body, leading to incorrect mPMI estimations. *Megaselia scalaris* is a forensically important fly especially in indoor cases, where it can be the first colonizer of a body. Paracetamol is an everyday drug used globally and was tested until now only on blowflies (Calliphoridae). No data are available for the Phoridae. Flies were reared on pet food homogenized with 100mg/g, 40mg/g, 20mg/g, 10mg/g and 0mg/g (control) of paracetamol. Pupal width and length were measured before extraction and HPLC Results showed that paracetamol had a negative effect on *M. scalaris* pupae size, with a reduced length and width at increased amount of paracetamol. In addition experiments demonstrated a general developmental delay in the larvae-pupae stage for the paracetamol treated groups when compared with the control. HPLC analyses were able to detect the presence of the drugs on the pupae; further analyses are requested to identify the minimum detectable dose.

OVERVIEW OF FORENSICALLY IMPORTANT SEPSIDAE (DIPTERA) OF THE CZECH REPUBLIC

Klimesova V., Oleksakova T., Sulakova H.

Institute of Criminalistics Prague (ICP), Prague, Czech Republic
tereza.oleksakova@pcr.cz

This research is a combination of search, retrieval and an empirical part based on observing two experiments which were conducted between 2011 and 2012 and between 2012 and 2013. In both experiments model carcasses of pigs (*Sus scrofa* f. *domestica* Linnaeus, 1758) were used. Pig carcasses were a substitution for human corpses and imitate real crime scenes. Samples were collected from dead pigs and from nearby vegetation. The main goal of the experimental part was to observe and annotate which species of Sepsidae family were present at particular periods of time and to elaborate on possible relationships between their presence and phases of cadaver decomposition.

Several methods were used to collect insects in the experimental area: insect nets, entomological tweezers, exhausters, pitfall traps and pyramidal traps (Slaff et al. 1984, Barták and Roháček 2011). Identification of species was based on identification keys from various monographies.

Specimens are deposited in the collections of the Institute of Criminalistics Prague (ICP). The most suitable method for obtaining a sufficient number of representative samples (data) was determined as a combination of pyramid traps, pitfall traps and entomological tweezers.

During trials, altogether 15,195 adult specimens of family Sepsidae were collected which belonged to 15 species. The most abundant species was *Nemopoda nitidula* (Fallén, 1820) (spring = 45.54%, summer = 36.46%) and which larvae were collected from both carcasses. The experiment leads to the broadening of the available knowledge about Sepsidae family and helped to verify applied data collecting methods.

Barták M. & Roháček J. (2011): Records of interesting flies (Diptera) attracted to meat baited pyramidal trap on sapping stump of European walnut (*Juglans regia*) in Central Bohemia (Czech Republic). Časopis slezskeho zemskeho muzea, 60/3: 223-233

Slaff, M., Haefner J. D., Parsons R. E. and Wilson F. (1984): A modified pyramidal emergence trap for collecting mosquitoes. Mosquito News. 44: 197-199

THE FAMILY MUSCIDAE (DIPTERA) AND THEIR PREFERENCES OF DECOMPOSITION STAGE OF CARCASSES IN THE CZECH REPUBLIC

Klimesova V., Oleksakova T., Sulakova H.

Institute of Criminalistics Prague (ICP), Prague, Czech Republic
vanda.klimesova@pcr.cz

After death, the body takes a natural course of events and reactions leading to decomposition of the body. The volatile molecules called apeneumones escaping from the decomposing body attract insects and decomposition of a dead body through invertebrates' activity is called a succession of saprophagous organisms on the cadaver. This process brings about changes on the body, which are called successional waves. Each of these waves has its own characteristic representatives of the order insect.
In years 2011 to 2015, three field experiments were performed in the capital city of Prague to study decomposition and insect colonization of large cadavers in conditions of the Central Europe. Experiments in turns followed decomposition in outdoor environments with the beginning in spring, summer and winter. As the test objects, a cadaver of domestic pig (*Sus scrofa* f. *domestica* Linnaeus, 1758) weighing 50 kg to 65 kg was used for each test. Our paper presents results of family Muscidae, which was collected during all three studies, with focusing on its using in forensic practice. Altogether 29,237 specimens of the muscids were collected, which belonged to 51 species. It was 16.6% (n = 307) of the total number of Muscidae family which are recorded in the Czech Republic. In all experiments, the species Hydrotaea ignava (Harris, 1780) was dominant (spring = 75%, summer = 81%, winter = 41%), which is a typical representative of necrophagous fauna on animal cadavers and human corpses in outdoor habitats during second and/or third successional stages (active decay phase) in the Czech Republic.

Joseph I, Mathew D G, Sathyan P & Vargheese G (2011): J Forensic Dent Sci. 3/2: 89-91
Eliášová H & Šuláková H (2012): Forenzní biologie. [Forensic biology] In: Štefan J. & Hladík J. (eds.) Soudní lékařství a jeho moderní trendy. Grada, Praha: 281-325

INTERSPECIFIC AGGREGATION OF NECROPHAGOUS DIPTERA LARVAE

Komo L., Charabidze D.

EA 7367 - UTML – Forensic Taphonomy Unit, Univ Lille, 59000 Lille, France
larissa.komo@univ-lille2.fr

Every organism (hence also every fly) is interested in its fitness. This mainly includes reproducing and making sure that their offspring survive, develop successfully up to an adult stage and finally also reproduce. According to the Allee principle, there should exist a positive association between individual fitness and the population size. Therefore, the benefits for aggregated maggots feeding efficiently on carrion should increase with their number until reaching a threshold. This, in turn, may imply that maggots from different species live together in a relationship in which every maggot benefits from the activity of other maggots. However, up to now, no experiment has proven this hypothesis.

The present study analysed the interspecific aggregation and segregation within maggots of *Lucilia sericata*, *Calliphora vicina* and *C. vomitoria*. On an agar-liver medium were two spots determined and each framed by an upended petri dish with 50 maggots in there eating on the medium. After 3 hours, 100 new maggots were placed on the medium in one line and the same distance to both aggregation spots. After 20 more hours, the individuals were counted under both petri dishes, in close proximity to the petri dishes and in surroundings. The petri dish which contained the largest amount of maggots was designated as the 'winner spot' while the opposite petri dish was the 'loser spot'.

Our experiments demonstrated that later added maggots moved to the pre-existing aggregations. Thereby, they had one favourite aggregation and thus created a clear winner spot which also attracted the maggots from the loser spot. Between 62 and 100 % of all larvae were located within or in close proximity to the winner spot at the end of the experiments (mean 84 %, n = 16). Thereby, the winner spot exhibited an average of 60 maggots of those which pertain not to the species that were placed at this spot before. In contrast, the loser spot included an average of 4 maggots of those which pertain not to the species that were placed there before. The later added maggots did not choose an aggregation according to their species. The winner spot alternated also between sides inside the set-up regardless of the species.

These results confirm the existence of collective choice, shared aggregation mechanism and the minor importance of the species within necrophagous larvae of the family Calliphoridae. The benefits of aggregation in a homogeneous environment likely outweigh the differences of species. These benefits probably include the accelerated larval growth due to exo-digestion (in prep.). Naturally, a fast development is beneficial for maggots to reduce the risk of being eaten up from predators. Further research will analyse the effect of food characteristics on the aggregation intensity.

FORENSIC ENTOMOLOGY IN FRENCH CRIMINAL CASES: TRENDS OF IMMATURE STAGES ASSOCIATIONS

Lefebvre F., Dourel L., Rothlisberger F., Pasquerault T., Vincent B., Cervantes L., Aimar S.

Department of Forensic Fauna Flora (3F) – IRCGN (France);
Forensic Sciences Institute of the French Gendarmerie
dfff.dcih.ircgn@gendarmerie.interieur.gouv.fr

Created in 1992, the Forensic Fauna Flora Unit (ex Entomology) of the Forensic Science Institute of the French Gendarmerie has provided mainly forensic entomology expertise for the French metropolitan area and Corsica Island. Only 4% come from French overseas territories (mainly French West Indies).
The department receives judiciary requests mainly from rural areas (71%). The diversity of the corpse's situation and environmental conditions must be integrated in the PMI estimations. From these cases, some information appears concerning geographical, environmental parameters and some different trends regarding the species identified.
Development stages of species at the moment of the discovery of a corpse are studied. The aim is to show a pattern of necrophagous species developmental trends. About 1300 entomology cases are analysed in this way.
The presence of 10 taxa is mainly used: *Calliphora* sp., *Lucilia* sp., *Phormia* sp., *Protophormia* sp., *Chrysomya* sp., Sarcophagidae, *Muscina* sp., *Ophyra* sp. Fanniidae and Piophilidae.
The methodology compares the taxa 2 by 2 when they are simultaneously sampled with active immature stages (larvae or pupae).
It is observed:

- Trend of equal development stages between necrophagous' families Calliphoridae and Sarcophagidae.
- High trend of equal development stages between *Calliphora* sp., *Lucilia* sp. and *Protophormia* sp.
- *Chrysomya* sp. and *Phormia* sp. tend to be older than *Calliphora* sp. and *Lucilia* sp.
- *Ophyra* sp.
- Fanniidae and Piophilidae tend to be younger than *Chrysomya* sp. and *Phormia* sp.

In post mortem interval estimation, many parameters can affect the accuracy of the result. The faunal succession is one of them and its study need to be integrated in a wide model of FE analysis linking samples data and statistical approach.
To be continued…

SPECIES IDENTIFICATION OF AFRICAN BLOWFLIES (DIPTERA: CALLIPHORIDAE) OF FORENSIC IMPORTANCE

Lutz L. [a], Williams K. A.[b,c], Villet M. H.[c], Ekanem M.[d], Szpila K.[e]

[a]Institute of Forensic Medicine, Goethe-University Frankfurt, Frankfurt am Main, GE; [b]Entomology Department, Durban Natural Science Museum, Durban, ZA; [c]Southern African Forensic Entomology Research Laboratory, Department of Zoology and Entomology, Rhodes University, Grahamstown, ZA [d] Department of Zoology, University of Uyo, Uyo, NG; [e]Chair of Ecology and Biogeography, Faculty of Biology and Environmental Protection, Nicolaus Copernicus University, Toruń, PL
lena@die-lutzens.de

One of the most important taxa in forensic entomology are necrophagous blowflies (Diptera: Calliphoridae), as they usually colonise a body minutes after death. They are also relevant to problems in human and animal health. However, finding relevant information about these flies requires their correct identification. Unfortunately, the blowfly faunas of most African countries (except Namibia and the Republic of South Africa) remain barely studied and identification keys, if available, do not cover the fauna of the whole continent and are often poorly illustrated. This shortage of reliable, accessible identification tools for blowflies of forensic importance poses a serious obstacle to the development of forensic entomology in these countries. To address this, a high quality key to the adults of all species of forensically relevant blowflies of Africa has been developed, based on high quality entomological material and modern techniques of image-stacking stereomicroscopy. In contrast to previous keys, which are mainly works with drawings; the diagnostic characters and full habitus pictures of the species are illustrated with photographs in the present key. In total, 16 blowfly species of the genera *Calliphora, Chrysomya, Lucilia* and *Hemipyrellia* are keyed and information of their geographical distribution in Africa is given. Problematic diagnostic characters, such as the darkened anterior wing margin of some *Chrysomya* species or characters used in previous keys for the distinction between *L. cuprina* and *L. sericata* and their possible hybrids, are discussed. Due to the illustration of characters via high quality photographs, this new guide can be easily applied even by investigators inexperienced in the taxonomy of blowflies and will be available through a highly accessible online platform.

BLOWFLY PUPARIA IN A HERMETIC CONTAINER: SURVIVAL UNDER DECREASING OXYGEN CONDITIONS

Mądra-Bielewicz A.[1], Frątczak-Łagiewska K.[1,2], Matuszewski S.[1]

[1]Laboratory of Criminalistics, Adam Mickiewicz University, Poznań, Poland; [2]Department of Animal Taxonomy and Ecology, Adam Mickiewicz University, Poznań, Poland
szymmat@amu.edu.pl

Despite widely accepted standards for sampling and preservation of insect evidence, unrepresentative samples or improperly preserved evidence are encountered frequently in forensic investigations. Incorrect preservation may affect the scope of laboratory procedures to be performed using insect evidence. Moreover, in some instances it may elicit extra questions, which are irrelevant in cases with properly preserved insect samples. This experiment was provoked by a recent case in which fly puparia were preserved in an airtight glass jar with no preservative inside. Upon inspection of the jar it was discovered that during the 4-month storage, that had elapsed prior the sample arrival in the laboratory, some insects had emerged in the jar. Because the oldest puparia sampled on a crime scene were the ones from which adults had eclosed in the jar, the minimum PMI was based on the age of these specimens. However, due to the lack of scientific data about survival of fly intra-puparial forms in hermetic containers, the age estimate was less accurate than it could have been if these data had been available. Consequently, as a by-product of the case it was decided to make a basic study to answer the following questions provoked by the circumstances of the case. How long may intra-puparial forms survive inside a hermetic container? Does the number of puparia in the container affect their survival? Does the age of insect inside the puparium affect its ability to survive inside the container under conditions of decreasing oxygen? Are forensically important species equally sensitive to hypoxia?

Puparia of *Lucilia sericata* and *Calliphora vomitoria* (Linnaeus, 1758) were chosen for the study. Experiments were performed in insect incubators at 22.5 °C for a photoperiod (h) of 12:12 (L:D). Glass jars (twist type with a metal lid) were used as containers. Puparia of both species were placed in each container (ratio 1:1). The first experiment tested the effects of blow fly species, number of puparia, container volume, damp paper substrate, and container conditions on the survival of intra-puparial forms. Three trials were made with 16 containers in each. The second experiment tested the effects of the age of intra-puparial forms and their number on the survival rate in hermetic containers. Three trials were made with 30 containers in each.

The study demonstrated that the survival of blowfly intra-puparial forms inside airtight containers is dependent on container volume, number of puparia inside, and their age. The survival in both species was found to increase with an increase in the volume of air per 1 mg of puparium per day of the development in a hermetic container. Below 0.05 ml of air, no insect survived, and above 0.2 ml of air per 1 mg of puparium per day, the survival reached its maximum. These results suggest that blowflies reveal a single, general pattern of survival under decreasing oxygen conditions and that this pattern is a product of number of developing insects, their age and the initial amount of available air. Implications for forensic entomology are discussed

EARLY COLONISATION OF INDOOR CARCASSES BY BLOW FLIES (DIPTERA: CALLIPHORIDAE): AN EXPERIMENTAL STUDY FROM CENTRAL SPAIN

Martín-Vega D.[1,2], Martín Nieto C.[1], Cifrián B.[1], Baz A.[1], Díaz-Aranda L.M.[1]

[1]Departamento de Ciencias de la Vida, Universidad de Alcalá, Alcalá de Henares, Spain
[2]Department of Life Sciences, Natural History Museum, London, UK
daniel.martinve@uah.es; d.martin-vega@nhm.ac.uk

Due to their ubiquity and synanthropy, blow flies (Diptera: Calliphoridae) are generally the first colonisers of cadavers and, therefore, frequently used to estimate a minimum post-mortem interval ($_{min}$PMI). Whereas in outdoor situations blow flies are expected to locate and colonise exposed cadavers within hours or even minutes after death, it is usually assumed that the colonisation of a cadaver indoors might be delayed for an uncertain period of time. This uncertainty severely limits the informativity of $_{min}$PMI estimates based on entomological evidence. Moreover, these limitations are emphasised by the lack of experimental data on insect colonisation of indoor carrion and by the fact that most of the forensic cases involving entomological evidence have been reported to occur indoors. In this study we investigate the early colonisation of pig carcasses placed indoors in a building located in the centre of an urban environment in central Spain. Three carcasses were placed in three equal rooms with a window half opened during summer 2013, autumn 2013, winter 2014, spring 2014 and summer 2014. The species composition and their contribution to the carrion colonisation differed among seasons. *Calliphora vicina* Robineau-Desvoidy was the sole coloniser of carcasses in winter and colonised the carcasses within the first 24–48 hours in every season, although *Lucilia sericata* (Meigen) was the first coloniser in most summer carcasses. On the other hand, *Calliphora vomitoria* (L.) and *Chrysomya albiceps* (Wiedemann) colonised the carcasses significantly later in spring and in spring and summer, respectively, with a delay of several days. In autumn, however, there were no significant differences in the colonisation times by *C. vicina*, *L. sericata* and *Ch. albiceps*. *C. vicina* and *L. sericata* showed a clear preference for ovipositing in the natural orifices of the carcasses, whereas *Ch. albiceps* oviposited more frequently on trunk and legs. We hope that the current study will contribute to fill the existing gap in experimental data on insect colonisation of carrion indoors, particularly in the Mediterranean region.

EXPOSING PROBLEMS TEACHING STUDENTS MORPHOLOGICAL SPECIES IDENTIFICATION

Nigoghosian G.[1], Weidner L.[1], Nunes L.[2], Stamper T.[1]

[1]Department of Entomology, Purdue University, West Lafayette, IN 47907, USA; [2]Center for Instructional Excellence, Purdue University, West Lafayette, IN 47907, USA
lweidner@purdue.edu

When dealing with physical remains, morphological assessment for species is a traditional approach to entomological specimen identification. A dichotomous key guides the user through taxa determination for a specimen by providing a series of dual-choice nodes that center around morphological differences. Each nodal choice leads to either a new set of dichotomous choices or a taxa decision. We evaluated student's ability to utilize a dichotomous key down to species for a limited set of taxa, by reviewing their nodal decisions along with their confidence level using a Likert scale (1-5). Along with individual decision recording, students conducted a post-decision group comparison, following a think-pair-share active learning model. If student answers were not the same, they re-evaluated their specimen until a mutual evidence-based decision was reached. We analysed student identification success as well as the correlation between confidence and accuracy. Students displayed high decision confidence but low accuracy. Although no significant difference was found in accuracy between male and females, we observed a higher initial accuracy from students enrolled in STEAM majors when compared to non-STEAM majors. From these data we aim to improve student training in the use of dichotomous keys for species identification, with a continued approach that can be then used to provide guidelines for how forensic scientists should approach dichotomous key training.

THE STUDY AND APPLICATION OF UNDERWATER DECOMPOSITION FROM AN ENTOMOLOGICAL PERSPECTIVE FOR THE PURPOSE OF POST-MORTEM INTERVAL ESTIMATION

Ody H., Smith P., Brown K.

The Forensic Innovation Centre, Institute of Criminal Justice Studies,
University of Portsmouth, Portsmouth, Hampshire, UK
helen.ody@port.ac.uk

Although the decomposition of human remains in water is known to differ from that on land, very little research has been conducted into how these differences extend to forensic entomology. A small number of studies into aquatic decomposition and forensic entomology have been conducted worldwide, however very little of this research has been conducted in the UK. A full understanding of taphonomic processes in aquatic environments is important for accurate minimum post-mortem interval estimation, especially in areas with easy access to water.

Following a pilot study in which rabbit carcasses were decomposed in lidded plastic boxes in fresh stream and sea water, a field study is underway in South East England to compare decomposition and insect succession on piglet carcasses in a freshwater pond (Wickham, UK) and in sea water in an area of Langstone Harbour (Portsmouth, UK). Insect specimens have been collected from piglet carcasses across two sampling periods (June - August 2016, November 2016 – present) and observations on the decomposition state of the carcasses have been recorded. Water samples have also been taken in order to investigate possible changes in microfauna taking place throughout the decomposition period. Insect samples have also been taken from two other piglet carcasses which have been decomposed on land in a wooded area (Wickham, UK), across the same periods in order to provide an indication of which species are being attracted to non-submerged carrion in the area. This can also be compared to other studies describing insect succession on land in Portsmouth.

A preliminary report on species colonisation patterns for each habitat will be presented along with some observations and recommendations for methods for undertaking aquatic forensic entomology research on a small scale. Ultimately the field research will be presented alongside data from questionnaires which are currently being distributed to members of relevant professions.

THE EFFECT OF SUITCASE CONCEALMENT ON THE INSECT COLONIZATION: A PILOT STUDY IN WESTERN AUSTRALIA

Petersen C., Georgy J., Magni P.A.

School of Veterinary & Life Sciences, Medical & Molecular Sciences Cluster,
Murdoch University, Perth, Western Australia
p.magni@murdoch.edu.au

Decomposition is a complex and continuous process that involves the breakdown of soft tissues post mortem. This is often mediated by the action of necrophagous macro- and micro-fauna, especially insects (e.g. Diptera, Coleoptera). Depending on the geographic region and the author, the number and the extent of decomposition stages has varied from one to as many as nine. However, the decomposition process is often broadly categorised into five stages (fresh, bloated, active decay, advanced decay, and dry/skeleton). Generally, each stage is characterised by specific changes in tissue morphology and insect activity. Although researchers frequently use decomposition stages, it is important to understand that in reality, the process of decay doesn't occur in discrete stages (as it is a continuous process).

Even though the basic sequence of decay is generally the same for all carcasses, the rate at which decomposition occurs varies between bodies as a result of many interrelated variables. Both intrinsic (variables concerning the corpse itself, e.g. body size, condition of body, health of the individual, cause of death, and presence of clothing) and extrinsic factors (variables in the external environment, e.g. temperature, humidity, precipitation, sun/shade, accessibility to insects) may directly influence the rate of decomposition, with a number of these factors having been previously investigated.

The present pilot study investigated suitcases as a barrier that may affect the access of bodies to insects, and therefore modify or delay the typical decomposition process.

The field of experiment has been held at Murdoch University Vet Farm, over a period of six weeks (April-May 2017). A total of six pig carcasses (*Sus scrofa* L., approx. 7-11 kg) and 6 identical suitcases were used over the course of the study. Five pigs were singularly placed in suitcases, one pig was caged and exposed and one suitcase was left empty. Experimental pigs were placed approximately 20 meters apart from each other. Temperature data loggers were used to record the temperature and humidity within the suitcases, as well as the ambient temperature/humidity of the study field. The exposed pig and suitcases were photographed and sampled daily. Insect samples collected were reared in laboratory until the adult instar. The first suitcase was opened after 15 days from the beginning of the experiment, while the others every 7 days until the end of the study.

Results regarding the decomposition process inside/outside the suitcases as well as the insect species colonizing the carcasses will be presented. At present the study is currently ongoing.

SCANNING ELECTRON MICROSCOPY INVESTIGATIONS ON ANTENNAL SENSILLA IN *HERMETIA ILLUCENS* (DIPTERA: STRATIOMYIDAE)

Pezzi M.[1,2], Leis M.[1,2], Chicca M.[1], Falabella P.[3], Salvia R.[3], Scala A.[3], Nardiello M.[3], Scieuzo C.[3], Grossi G.[3], Farina D.[3], Whitmore D.[4]

[1]Department of Life Sciences and Biotechnology, University of Ferrara, Ferrara, Italy; [2]Laboratory TekneHub, Technopole of University of Ferrara, Ferrara, Italy; [3]Department of Sciences, University of Basilicata, Potenza, Italy; [4]Department of Life Sciences, Natural History Museum, London, United Kingdom
patrizia.falabella@unibas.it

The black soldier fly, *Hermetia illucens* (L.) (Diptera: Stratiomyidae), a slender, wasp-like dipteran of forensic relevance, is often used to estimate *postmortem* interval. Investigations by scanning electron microscopy (SEM) were conducted on adults of both sexes of *H. illucens*, examining, in detail, the sensilla and microtrichia of the three regions of the antenna (scape, pedicel and flagellum). Three different types of sensilla and four types of microtrichia were found on the antennal structures. Chaetic sensilla and hair-like microtrichia were detected on the scape and pedicel. Different sensorial structures were detected on the flagellomeres according to their position: two types of pit sensilla on flagellomeres 1 to 6, and trichoid sensilla in an oval depression on flagellomeres 4 to 6. Lanceolate microtrichia were present on flagellomeres 1 to 6, scimitar-like microtrichia on flagellomeres 7 and 8, and finely tapered microtrichia in the depression of the eighth flagellomere. The pit sensilla were of two types: a complex sensillum with an arbor-like structure and a grooved one, and a simpler sensillum with digitiform structures. Sex-related differences were detected in the length of chaetic sensilla of the pedicel: three different lengths were found in males and two in females. These ultrastructural studies represent the basis for further investigations on the electrophysiology and molecular biology of these sensorial structures in *H. illucens*, probably involved in the ability of this species in finding corpses on which to develop.

A MODIFIED TRAP TO MONITOR AND CONTROL POPULATIONS OF MEDICALLY IMPORTANT BLOW FLIES IN THAILAND

Sanit S., Samerjai C., Klong-klaew T., Sontigun N., Limsopatham K., Suwannayod S., Sukontason K., Sukontason K.L.

Department of Parasitology, Faculty of Medicine, Chiang Mai University, Chiang Mai, Thailand
sang.ngeab@gmail.com

Necrophagous blow flies are medically-important in Thailand. They are commonly found around human dwellings (e.g., house, market and garbage) and can act as vectors for various pathogens. Therefore strategies to control the fly population are necessary. In Thailand fly traps have been used for control fly populations and the monitoring of flies. The first aim of this study was to improve the commonly used, modified WHO fly trap, invented by Prof. Kom Sukontason (fig.1). Compared to previous traps, the new, improved trap (fig.2) has many advantages because it is small, easy to regulate and effective to collect flies. The modified trap consists of two parts: 1) The trap base is made from acrylic plastic size 22 × 22 × 9 cm (collecting area 20 × 20 cm) that is controlled by an electronic board and 2) The collecting bag is made from paper size 20 × 20 × 22 cm. The efficiency of the modified fly-trap was proved by comparing the samples with the ones achieved by using sticky traps and the sweeping net method. The bait used for all collection methods was 1-day tainted beef mixed with offal. Fly samples were taken once every 2 weeks for 6 months (Apr-Sep 2015) with a 1-hr collection period between 1 p.m.-4 p.m. The sticky traps were put nearby the bait. For the fly sampling with a net, flies were collected every ten minutes for 1 hr. The collection with the modified fly-trap started when the collecting bag was open to allow flies entering the trap for 10 minutes. After that, the collecting bag was closed for 30 sec and the cycle started again. During the study a total of 11,466 flies were captured, containing *Chrysomya megacephala* (59.62%), *Chrysomya rufifacies* (38.56%), *Hemipyrellia ligurriens* (1.03%), *Lucillia cuprina* (0.59%), *Ceylonomyia nigripes* (0.17%), *Chrysomya villenuvi* (0.02%) and *Chrysomya chani* (0.01%). The results indicated that the sticky trap (2396 flies; 21%) was the least efficient at collecting blow flies and the performance of the modified fly-trap (4791 flies; 42%) was similar to the sweeping nets (4279 flies; 37%) (P>0.05) when catching blow flies. Therefore, the modified fly-trap is an effortless method which is not just useful for fly population control but also for monitoring approaches.

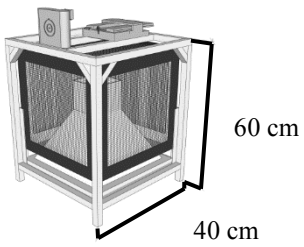

Fig. 1 Prototype trap invented by Prof. Kom Sukontason

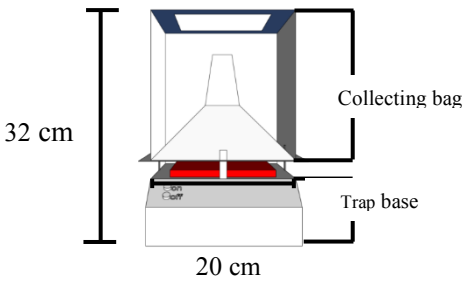

Fig. 2 Modified fly-trap

INVESTIGATIONS ON MOLECULAR MECHANISMS AND MORPHOLOGY OF STRUCTURES INVOLVED IN CHEMORECEPTION IN *HERMETIA ILLUCENS* (DIPTERA: STRATIOMYIDAE)

Scala A.[1], Pezzi M.[2,3], Salvia R.[1], Leis M.[2,3], Chicca M.[2], Whitmore D.[4], Vogel H.[5], Nardiello M.[1], Scieuzo C.[1], Grossi G.[1], Farina D.[1], Falabella P.[1]

[1] Department of Sciences, University of Basilicata, Potenza, Italy; [2]Department of Life Sciences and Biotechnology, University of Ferrara, Ferrara, Italy; [3]Laboratory TekneHub, Technopole of University of Ferrara, Ferrara, Italy; [4]Department of Life Sciences, Natural History Museum, London, United Kingdom; [5]Department of Entomology, Max Planck Institute for Chemical Ecology, Jena, Germany
patrizia.falabella@unibas.it

Insects use chemical perception to interact with other insects and with the environment. The perception of the volatile substances, linked to the processes of feeding, coupling and escape from predators, is related to molecules belonging to the families of Odorant Binding Proteins (OBPs), Olfactory Receptors (ORs), Ionotropic Receptors (IRs), Chemosensory Proteins (CSPs).The black soldier-fly *Hermetia illucens* (Diptera: Stratiomyidae), during the larval stage, is a generalist detritivore which is often present on corpses in later stages of decomposition and may be useful in forensic entomology for estimation of *postmortem* interval. *Hermetia illucens* has a highly sensitive olfactory system, specialized in perceiving molecules linked to the decomposition of animal and vegetal materials. Investigations on molecular mechanisms involved in feeding source recognition by OBPs, through the perception of volatile attractants, and the study of the structures involved in olfactory perception may contribute to the description of ethological details of this species. An identification of the protein profile has been conducted based on the construction *de novo* and annotation of *H. illucens* larval transcriptome, using specific bioinformatics tools (Blast2GO) and searching for transcripts encoding for proteins involved in processes mediating olfactory perception. The OBPs and other molecules involved in olfactory perception mechanisms have been identified through specific software. A parallel investigation by scanning electron microscopy has been conducted on *H. illucens* larvae to identify structures possibly related to the olfactory perception. The preliminary data obtained by the two experimental approaches will be useful to increase knowledge about the role of this species in forensic entomology.

A PRELIMINARY STUDY ON THE CARRION INSECT COMMUNITIES AND THEIR SUCCESSION IN LUANDA, ANGOLA

Sebastião M.[1], Prado e Castro C.[2*]

[1]Central Laboratory of Criminalistics, Criminal Investigation Service, Interior Ministry. Luanda, Angol;
[2]Centre for Functional Ecology, Department of Life Sciences, University of Coimbra. Coimbra, Portugal
catarinapcastro@gmail.com

The study of arthropod fauna involved in the decomposition process of animal matter is crucial for forensic purposes. This knowledge is especially needed in countries with high levels of criminality and where the arthropod fauna are still under-recorded and not well known, despite its interest. This is the case of many African countries, including Angola.
In this work, we present data from the first survey on sarcosaprophagous Diptera and Coleoptera in Angola.
The study was conducted in Luanda over a 17-day period, from December 2011 to January 2012. Pig carcasses were used as animal models and collections of the entomofauna, both adults and immatures (for rearing) were performed daily.
A total of 881 specimens were collected during the study, belonging to 18 species. Three of them were not known previously from Angola. *Chrysomya albiceps*, *C. megacephala*, *C. putoria*, *C. marginalis* (Calliphoridae) and *Liosarcophaga emmrichiana* (Sarcophagidae) were the Diptera observed to breed on the carcasses. *Dermestes maculatus* (Dermestidae), *Saprinus splendens* (Histeridae) and *Necrobia rufipes* (Cleridae) were the most represented Coleoptera. Because of the hot temperatures during the experimental period (~27ºC) decomposition was extremely fast. *Chrysomya* spp. were dominant on the carcasses, especially *Chrysomya albiceps*. Periods of insect activity on carcasses are presented and comparisons are made with other carrion communities reported from the Afrotropical region.

TO STUDY THE INSECT SUCCESSION AND DIVERSITY ON DECOMPOSED CALF CARCASS

Singh S., Bala M.

Department of Zoology & Environmental Sciences Punjabi University Patiala, Punjab
Sabi_ghotra88@yahoo.com

Forensic entomology is the application and study of insects and other arthropod biology to criminal matters. It involves the interaction of arthropods (mainly insects) with legal activity. Insect succession is an ordered invasion pattern of the insects on the carcass. These insects arrive on the carcass for their requirements like food, shelter and oviposition. Biological studies for example taxonomic study, ecological study, ethological study, reproductive and developmental study of these insects encompass the forensic investigation. To check the forensically important insect fauna, an experiment was conducted on a calf weighing about 25 kg, in month of February. The temperature in month of February was 22±1°C. The carcass was left to decompose. It took 26 days for complete decomposition. The process of complete decomposition was divided into Fresh, Bloated, Active decay, Dry decay and Skeletal stages. The larvae and adults of the insects were collected from all decomposition stages and brought to laboratory for the identification. The identification of these insects was done with the help of a taxonomic key. A total 150 adults belonging 3 orders (Diptera, Coleoptera and Hymenoptera), 9 families and 18 species were collected during complete decomposition. In the Diptera 3 families Calliphoridae, Sarcophagidae, Muscidae were reported with 7 species i.e. *C. megacephala, C. rififacies, L. cuprina, L. sericata, Sarcophaga sp., Musca domestica, Musca sorbens.* In Coleoptera 5 families Histeridae, Staphylinidae, Dermestidae, Tenebrionidae, Cleridae were reported with 8 species i.e. *Dermestes maculates, Tenebrio molitor, Blapstinus sp., Typhaea stercorea, Philonthus longicornis, Necrobia sp., Pterostichus melanarius, Hybosoridea sp.* In Hymenoptera only one family Formicidae was reported with 3 species i.e. *Camponotus compressus, Pheidole indica, Anochetus graeffei*. There were total eighteen species *C. megacephala, C. rififacies, L. cuprina, L. sericata, Sarcophaga sp., Musca domestica, Musca sorbens, Dermestes maculates, Tenebrio molitor, Blapstinus sp., Typhaea stercorea, Philonthus longicornis, Necrobia sp., Pterostichus melanarius, Hybosoridea sp. Camponotus compressus, Pheidole indica, Anochetus graeffei* observed during the decomposition process of the carcass at different decomposition stages.

FORENSIC ENTOMOLOGY IN THE CZECH REPUBLIC

Sulakova H., Klimesova V., Oleksakova T.

Institute of Criminalistics Prague (ICP), Prague, Czech Republic
hana.sulakova@pcr.cz

Forensic entomology has had a forty-year tradition in the Czech Republic. The first entomological case was the murder of a young girl in Prague, which has never been resolved, because her murderer is unknown to this day. In the 70s of the 20th century, the next entomological cases occurred, which especially dealt with violent crimes. Among the first pioneers, who applied this field in forensic practice, were mainly university specialists in insects and private entomologists.

During the 1980s and 1990s, two or three cases per year were processed; mostly in the Institute of Criminalistics in Prague, but some civilian specialists, dipterologists, from universities participated in some of them. Greater development of forensic entomology occurred at the beginning of the 21st century, when the Institute of Criminalistics in Prague engaged a zoologist who should deepen the use of special biology in forensic practice. Through the zoologist, the Institute of Criminalistic in Prague became a member of the European Association for Forensic Entomology (EAFE).

In the Czech Republic, entomological traces at the crime scene are usually collected by a trained criminalistics technician, who specializes in searching and collecting all evidence at the crime scene. Each forensic technician has to complete six months of basic training, where he or she learns to provide all the evidence, including biological traces. Then forensic technicians have, as a part of their lifelong training, to pass a three-day entomological course. The course solves both the theoretical and practical issues of collecting necrophagous insects at the crime scene. For each course, animal cadavers of varying degrees of degradation (from completely fresh to skeletal) are prepared. Technicians work in pairs, as at a real crime scene. At the Institute of Criminalistics in Prague in 2010, the first version of an entomological sampling kit was made out, inspired by the French and Belgian sampling kits.

Forensic entomology is currently used in criminal cases and civil-law disputes. The Institute of Criminalistics in Prague also cooperates with archaeologists because the knowledge and principles of forensic entomology can be applied to description of the circumstances of death or funeral rituals in the same way.

Šálková T, Bezděk A, Březinová H, Farkašová K, Houfková P, Chvojka O, John J, Kmošek J, Koník P, Kovačiková L, Michálek J, Msallamová Š, Novák J, Pavelka J, Šuláková H, Bešta T, Myšková E, Weiter L & Zronek P (2015) Bioarchaeological recontruction of the funerál rite – case study based on organic material from the Hallstatt Period tumulus at the site Zahrádka (South Bohemia, Czech Rebublic). Památky archeologické CVI: 95-135Šuláková H & Barták M (2013) Forensically important Calliphoridae (Diptera) associated with animal and human decompositon in the Czech Republic: preliminary results. Čas. Slez. Muz. Opava (A), 62: 255-266.

RECORDS AND CHECKLIST OF CARRION COLEOPTERA IN ALGERIA

Taleb M.[1], Tail G.[1], Djedouani B.[2], Toumi M.[2], Açıkgöz H.N.[3]

[1]Laboratory of Biotechnologies, Environment and Health, Faculty of Nature and Life Sciences, University of Blida 1, Blida, Algeria; [2]Entomology Laboratory, Department of Legal Medicine, National Institute for Criminalistics and Criminology, Cheraga, Algeria; [3]Forensic Biology/Forensic Entomology Laboratory, Forensic Sciences Institute, Ankara University, Ankara, Turkey
meriemtaleb1@gmail.com

Knowledge of the carrion-breeding species present in a location during a particular time is important and necessary to apply forensic techniques. Algeria is situated in North Africa, bordered by the Mediterranean Sea in the north. Data regarding the occurrence of carrion Coleoptera are not available in Algeria, therefore, the aim of this work was to identify and inventory the most important carrion Coleoptera species in Algeria to improve fundamental knowledge regarding the distribution of these species.

The specimens were collected between 2011 and 2015 on human corpses, animal carcasses and baited traps and were morphologically identified.

Dermestidae, Staphylinidae, Silphidae and Histeridae were the most abundant families. *Dermestes frischii* (Kugelann, 1792) (Coleoptera, Dermestidae) was the most frequemt species followed by *philonthus* sp. (Stephens, 1829) (Coleoptera, Staphylinidae), *Creophilus maxillosus* (Linnaeus, 1758) (Coleoptera, Staphylinidae), *Saprinus semistriatus* (Scriba, 1790) (Coleoptera, Histeridae) and *Thanatophilus sinuatus* (Fabricius, 1775) (Coleoptera, Silphidae).

LOCOMOTOR ACTIVITY OF *DERMESTES LARDARIUS* (COLEOPTERA: DERMESTIDAE) AND *NECROBIA RUFIPES* (COLEOPTERA: CLERIDAE) IN LONG AND SHORT PHOTOPERIODS

Wyville S.[1], Mensah V.[1], Vanin S.[1,2]

[1]FLEA, School of Applied Sciences, University of Huddersfield, UK;
[2]GIEF, Gruppo Italiano per l'Entomologia Forense.
stefano.vanin@hud.ac.uk

The circadian clock mechanism is one whose importance is often overlooked despite it being the regulator of physiological and metabolic mechanisms in all the organisms, including insects. It synchronizes behaviour to the 24 hour cycle of the earth. The knowledge of this cycle is vital to the field of forensic entomology, as it determines the behaviour of insects with regards to locomotor activity, foraging and feeding, mating and emergence. The understanding of how insects behave on a day to day basis allows a better estimation of minimum Post Morten Interval (mPMI) to be obtained.
It has been reported that *Dermestes* and *Necrobia* species are nocturnal and therefore less active (if at all) during daylight hours. Verification of this and monitoring levels of activity spread throughout a 24 hour period could have implications on the accuracy of estimating mPMI using the currently accepted behavioural patterns.
Within this study the locomotor behaviour has been obtained using LD 12:12 conditions to form a baseline for our observations, followed by LD 16:08 and LD 08:16 in order to describe how these beetles respond to a longer and shorter photoperiod. Data was obtained in controlled conditions of 25°C and using Trikinetics technology often used with *Drosophila* species. This allows for mechanical data collection rather than relying on observational data.
Both species showed both diurnal and nocturnal locomotor activity with a clear anticipation in *Dermeste lardarius* of the dark-light transition, while *Necrobia rufipes* specimens lacked this anticipation. Both species showed a clear rhythmicity when, after a three days entrainment in LD, were exposed to a continuous dark condition (DD). This rhythmicity is maintained for several days independently to the length of the light phase during the entrainment, however the rhythmicity is less robust in *Dermeste lardarius* when exposed to a continuous light period (LL).
Our results underline that in these beetle species, as already demonstrated for other insects of forensic interest (eg. *Calliphora vomitoria, Lucilia sericata* and *Megaselia scalaris*), the locomotor activity is under clock regulation, however light stimuli play an important role in the amount of activity, with a greater movement during the light phase.