International Journal of Legal Medicine (2019) 133:1267–1278
https://doi.org/10.1007/s00414-018-1967-5

ORIGINAL ARTICLE

Environmental factors influencing flight activity of forensically important female blow flies in Central Europe

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Received: 5 September 2018 / Accepted: 7 November 2018 / Published online: 27 November 2018

Abstract

In forensic entomology, evaluation of a possible delay between a person's death and insect colonization is crucial. We monitored the seasonal flight activities of the most abundant blow flies in an urban habitat in Frankfurt/Germany based on 152 sampling days between April and October 2017. Thirty-six thousand female specimens of 12 necrophagous taxa were sampled as a possible groundwork for establishing a prediction tool for the activity of certain forensically relevant taxa. The most abundant taxon was *Lucilia sericata* ($n = 19{,}544$), followed by *Lucilia caesar* ($n = 8025$), *Calliphora vicina* ($n = 5224$), and *Lucilia ampullacea* ($n = 1834$). Up to six environmental parameters were statistically significant predictors of fly presence, leading to unique patterns of seasonal and daily activity for all four species. In detail, our analysis proved that *L. sericata* is a sun-loving, high-summer species that dominates the warmer months and is mostly influenced by mean day temperature. In contrast, *L. caesar* seems to be a shade-loving species that dominates in autumn resp. late-season and is mainly influenced by mean day temperature and wind speed. The activity of *L. ampullacea* was highly related to mean day temperature and relative humidity. In contrast to all other species, *C. vicina* behaved differently, particularly due to its occurrence throughout the entire sampling interval and the higher tolerance limits for the measured abiotic parameters, especially temperature. The present study is groundwork for establishing a prediction tool for the flight and oviposition activity of forensically relevant taxa.

Keywords Female blowflies · Forensic entomology · Abiotic parameters · Monitoring

Introduction

A major task in forensic entomology is the estimation of time between insect colonization of a human body and its discovery [1]. Blow flies are usually the first insects at a cadaver, often arriving minutes to hours after death [2–4]. This makes it one of the most important taxa in forensic entomology. Age determination of their offspring [5, 6], developing on a cadaver, can give evidence for the minimum post-mortem interval (PMI_{min}), which corresponds to the time since the first insect colonization. It is important not to mistake that period with the time since death because there can be variable time gaps of less than an hour up to several days (or even longer) between death and first colonization by insects [6]. In recent years, several attempts have been made to focus attention on this problem by identifying reasons for, and possible periods of, such a delayed arrival of insects. The variety of names that exists for this phenomenon, e.g., pre-colonization phase (pre-CP) [7], pre-colonization interval (pre-CI) [8], or pre-appearance interval (PAI) [9] illustrates the difficulty of coping with this task. It is known that colonization timings of blow flies are not constant because crime-specific circumstances, like the accessibility of the body for insects [10–12], or ecological and climatic parameters affect their arrival times [7, 13–17]. Abiotic variables such as temperature, precipitation, relative humidity, barometric pressure, light intensity, wind speed, and seasonality have an effect upon flight activity and colonization time. These variables seem to have a stronger impact than biotic factors such as inter- and intraspecific competition, given that unfavorable climate parameters can completely prevent flight activity [7]. Temperature has been reported as the most important environmental factor for the activity of blow flies, and the overall

Electronic supplementary material The online version of this article (https://doi.org/10.1007/s00414-018-1967-5) contains supplementary material, which is available to authorized users.

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positive influence of temperature on blow fly flight activity was postulated in several studies [7, 13, 18–21]. Next to temperature, rain seems to be the most important other weather factor affecting insect activity [22]. The inhibitory effect of rain was observed in field studies [7] where it was seen to cause oviposition delays of 1–3 days [23]. Not only rain itself, but also its duration seems to be of great importance, as blow flies can become active again during rain breaks [3]. Strong and persistent rainfalls, however, continuously inhibit their activity [24, 25]. Relative humidity is another negative factor for the activity of blow flies [7, 20, 26, 27]. Changes in barometric pressure can announce changes in weather conditions, and blow flies might be able to detect such fluctuations with external baroreceptors located on their antennae [28, 29]. Edwards [30] observed increased activity for the blow fly *Calliphora vicina* with altering barometric pressure, and other studies have revealed that decreasing barometric pressure can act as a positive predictor for blow fly activity [7, 30].

An increase in light intensity can be correlated with an increase in the flight activity of blow flies [31, 32]. In contrast, decreasing and low light intensities, (or complete darkness e.g. at night) inhibit or totally halt colonization of carcasses by blow flies [15, 33, 34]. An example for the inhibitory effect of low light levels on flight activity is the phenomena that *Chrysomya megacephala* is more likely to walk than to fly to carrion under such conditions [35]. The affinity for certain light intensities can reflect the habitat preferences of different fly species [7, 36] and should be considered not only on a species, but also on a gender level, because the abilities and preferences of females are most important when it comes to the detection and colonization of a body. Wind speed is central for the flight activity of insects [31]. So far, wind speeds above 29 km h^{-1} have been reported as the maximum value for flight activity [7, 31, 37], while wind speeds below 10 km h^{-1} seem to be optimal for oviposition [38]. Moreover, the absence of wind seems to have an inhibiting effect on the colonization of carrion by blow flies [7]. Seasonality has a big impact on the activity and abundance of blow flies due to the high correlation of season with abiotic factors. Nevertheless, a possible interrelation of season and flight activity has to be investigated separately for each of the relevant necrophagous blow fly species because each taxa may show an own set of seasonal adaptations that underlie their specific seasonal abundance and activity.

In summary, a large number of single or interacting abiotic factors influence the flight activity and oviposition of forensically important blow flies. Despite some past efforts in statistically modeling such impact on the activity of selected blow flies [7, 17, 21, 39], more knowledge is essential to understand the presence or absence, respectively, the immediate or delayed arrival, of blow fly species on cadavers. There are no studies in Europe that identify and also quantify the impact of abiotic factors, such as temperature or precipitation, as relevant for the activity of blow flies from a forensic point of view (but see Rose and Wall 2011 [17]). The present study monitored a blow fly population by means of bait traps at five sampling localities in an urban habitat in Frankfurt/Germany during the main activity period of blow flies. Based on 152 sampling days from April to October 2017, the seasonal activity and distribution of the most abundant forensically important blow fly taxa were analyzed. The overall goal was to understand their flight frequency and abundance in terms of climate variables and season.

Material and methods

Sampling parameters

Original Red Top ® Flycatchers (3 l, Ashmoat Ltd., Suffolk) were modified by cutting off the lower part of the plastic bag (Supplementary Fig. 1). Every sampling day, a baited urine beaker was taped to the lower part of the trap. Mashed chicken liver (60 g), which had been allowed to decompose for 1 day at room temperature, was used as bait and as an olfactory stimulus. The urine beaker was covered with a layer of insect gauze to prevent direct oviposition on the bait. The traps were placed approximately 1.5–2 m above ground to minimize access for crawling insects, such as ants, stabilized with two ropes, and fastened to the ground with tent pegs. Every 24 h, at 7 a.m., the traps were replaced with new baited traps to ensure that the olfactory and visual stimuli had almost the same quality for each sampling day. Sampling was conducted at five sites (Table 1) in an urban habitat in Frankfurt/Germany (50° 6′ 39.32” N 8° 40’ 55.656” E), and the traps were randomly assigned to the different locations each day to exclude the possibility of a trap bias. The sites were all within a radius of approximately 2 km and at least 150 m apart from each other, with the widest distance being 430 m. All traps were in an urban habitat with a high density of streets and houses in the close distance (Supplementary Fig. 2). The different sites were quite similarly structured to make the data comparable. Monitoring was conducted from the 12 April to the 20 October 2017. The definitions of the National Weather Service (Deutscher Wetterdienst, DWD) were applied to determine the seasons of insect activity. On the basis of these definitions, there were 3 months per season. Samples were taken on 152 sampling occasions representing 760 samples (152 sampling occasions × 5 traps) spread over the spring, summer, and autumn seasons. The traps were placed on 52 randomly selected days in summer, while in spring and autumn the traps were sampled on 50 randomly selected days. The collected blow flies (Diptera: Calliphoridae) were morphologically identified to species level using Keys by Rognes [40] and Szpila [41] before being counted and sorted by sex.

Table 1 List of the five sampling locations

Location	Latitude (+ 50°)	Longitude (+ 8°)	Urban habitat
1	5′ 37.73” N	39′ 56.73″ E	Garden of the Institute of Legal Medicine
2	5′ 43.55” N	40′ 10.29″ E	Small park behind an apartment building
3	5′ 51.09” N	40′ 3.12″ E	A grass strip with a tree on a backstreet parking lot
4	5′ 33.40” N	39′ 57.26″ E	Between some trees on site of a tennis court next to a railway track
5	5′ 30.53” N	39′ 37.32″ E	Green area in front of a residential home of the University Hospital

Environmental parameter

The following environmental parameters were recorded for each sampling day: mean day temperature (°C), maximum and minimum day temperature (°C), precipitation (mm), wind speed (km h^{-1}), barometric pressure (h Pa), relative humidity (%), and number of hours of sunshine for each day (h). These data were collected from three local weather stations because none of the weather stations measured all of the required parameters. The weather station "Frankfurt-Höchst" was 8.8 km westwards of the sampling sites; "Offenbach-Wetterpark" was 8.52 km east of the sampling sites; and "Frankfurt-Westend" was 2.42 km north of the sampling sites. For the calculation of the parameters, hourly values during daylight were used. A "day" was defined as being from 7 a.m. until sunset. The sunset times differed depending on the respective season and were obtained from the DWD. Degree-day accumulation (DDA) over 11 different temperature thresholds (0–10 °C) was calculated using minimum and maximum daily temperatures starting on 12 April. A specific rain-index was calculated for the abiotic factor "rain," which indicates the amount of rain and also takes into account the duration of rain during daylight hours.

$$rain{-}index = (rain\ amount\ (mm) \times hours\ of\ rain\ on\ a\ day) \times \left(\frac{h\ rain\ on\ a\ day}{h\ daylight}\right)$$

Statistical analysis

The activity data of the four most abundant blow flies (i.e., *Calliphora vicina*, *Lucilia ampullacea*, *Lucilia caesar*, and *Lucilia sericata*) were analyzed due to their importance in forensic case work as the colonizing specimens. This data covered 152 sampling days with five traps, for a total of 760 samples. First, we computed a canonical correlation analysis using the *CCorA* function from the package *vegan* [42] to examine the relationships between the environmental variables and the abundance of the four species identified above. Then, to identify abiotic parameters affecting the presence and abundance of each species, a series of generalized additive models (GAMs) was performed. The dependent variable was daily abundance of female specimens of each of the four species fitted by using negative binominal distribution. For the independent variables a set of abiotic factors namely, mean day temperature, hours of sunshine, barometric pressure, relative humidity, wind speed, precipitation and the duration of rain on a day, were used at the start for each model due to their ecological importance for a flight activity of blow flies [7]. Prior to the modeling, the lower threshold temperatures for each species were determined with a series of GAMs including DDA for the given lower temperature thresholds and the random effect of site. Based on this, accumulated degree days (ADD) above 0 °C were used for *C. vicina* in the further models and ADD above 10 °C for all other species. For all models, the random effect of sampling sites was accounted for. The models were performed for each species separately, starting with the before-mentioned set of abiotic variables and compared using the Akaike information criterion (AIC) to identify the best model. Final models were those with the lowest AIC value, selected with a threshold value of 2. Abiotic variables that did not improve the AIC value of the model were removed, resulting in different sets of important abiotic parameters for each species. All analyses were conducted using RStudio version 3.4.3 [43].

Results

During the 152 sampling occasions, a total of 50,592 specimens were sampled; 96.24% of these were necrophagous blow flies, represented by 12 species. *Lucilia sericata* was by far the most abundant blow fly, followed by *L. caesar*, *C. vicina*, and *L. ampullacea* (Table 2). These species represented 96.4% of all sampled blow flies and were therefore used for further analysis of the dependence of their flight activity on abiotic variables. For all four species, a clearly female-biased sex ratio was recorded, for which *L. sericata* and *C. vicina* were quite similar with a 2:1 female to male ratio. The trap catches of *L. ampullacea* and *L. caesar* were substantially dominated by females, resulting in sex ratios of 4.8: 1 and 7.6:1.

Table 2 List of the four most abundant blow flies sampled from April to October 2017. Information of their number and sex ratio is included

Species	Sex	Number	Sex ratio (female: male)
Calliphora vicina Robineau-Desvoidy, 1830	Female	5.312	2.1:1
	Male	2.546	
Lucilia ampullacea Villeneuve, 1922	Female	1.834	4.8:1
	Male	381	
Lucilia caesar (Linnaeus, 1758)	Female	8.025	7.6:1
	Male	1.049	
Lucilia sericata (Meigen, 1826)	Female	19.557	2.3:1
	Male	8.236	

Flight activity and influence of environmental parameters

Calliphora vicina was the most frequent species and was sampled in 95% ($n = 145$) of all sampling occasions from spring to autumn. *Lucilia ampullacea* was sampled in 79% ($n = 120$) of all days but only commenced its flight activity on 10 May. *Lucilia sericata* was by far the most abundant species, but was only sampled in 77% ($n = 117$) of all sampling occasions. *Lucilia caesar* was the species least caught, in 75% ($n = 114$) of all sampling occasions. To assess the relationship between the environmental parameters and the abundance of these four blow fly species, a canonical correlation analysis was performed. On the first horizontal axis, the analysis shows that all species were positively influenced by daytime temperature and sunshine hours, and negatively affected by the amount and duration of precipitation in a day, as well as by relative humidity (Fig. 1). On the second vertical axis, relative humidity and ADD were shown to have a positive effect on *L. ampullacea* and *L. caesar*, and a negative effect on *C. vicina* (Fig. 1). To quantify the influence of abiotic factors on the frequency of this species on a single species level, GAMs were performed.

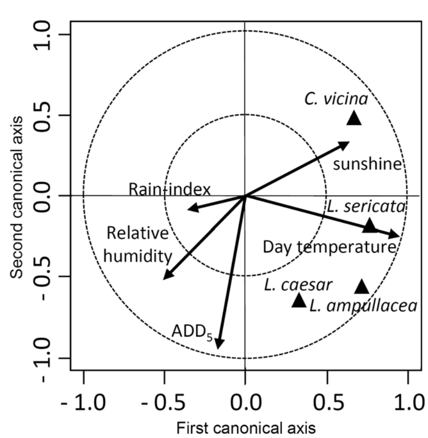

Fig. 1 Canonical correlation analysis between environmental parameters (*arrows*) and species abundance (Pillai's trace = 0.97, $p < 0.001$, canonical correlations = 0.80 (axis 1) and 0.48 (axis 2), redundancy $r^2 = 0.37$)

Calliphora vicina

The GAM fitted to the abundance data of *C. vicina* explains 47.7% of the variability in species abundance and is presented in Fig. 2(a–f) and Table 3. Figure 2a illustrates the effect of ADD above 0 °C on the abundance of female *C. vicina*. Overall, this species was active from the beginning of the sampling with the highest abundance in the end of spring, strongly decreased flight activity throughout the warmer months and increased again in autumn. Four environmental factors influenced the frequency of *C. vicina* significantly (Table 3). Mean day temperature had a strong positive influence on the activity of *C. vicina* up to temperatures of 20 °C (Fig. 2b). Barometric pressure had a positive effect (Fig. 2d) and relative humidity a largely negative effect (Fig. 2e). For the hours of sunshine intermediate values seems to be associated with the highest specimens counts (Fig. 2c). Overall, the abundance of this species differed between the five sampling sites (Fig. 2f).

Lucilia sericata

The GAM, fitted to the activity data of *L. sericata*, explains 70.5% of the variability in species abundance and is presented in Fig. 3(a–e) and Table 3. *Lucilia sericata* was primarily active in summer, and almost absent in spring and autumn (Fig. 3a). Mean day temperature was the most important abiotic parameter that significantly influenced positive the activity of this species (Table 3), whose flight activity increased when the mean day temperature exceeded 15 °C (Fig. 3b). Two additional abiotic parameters significantly affected the frequency of this species (Table 3). Barometric pressure influenced the flight activity of *L. sericata* positively (Fig. 3c), whereas relative humidity had a negative effect (Fig. 3d). The hours of sunshine and the interaction between the amount and duration of rain improved the overall model but did not influence the species significantly (Table 3). As with *C. vicina*, the abundance of *L. sericata* differed between the sites, but on marginally (Fig. 3e).

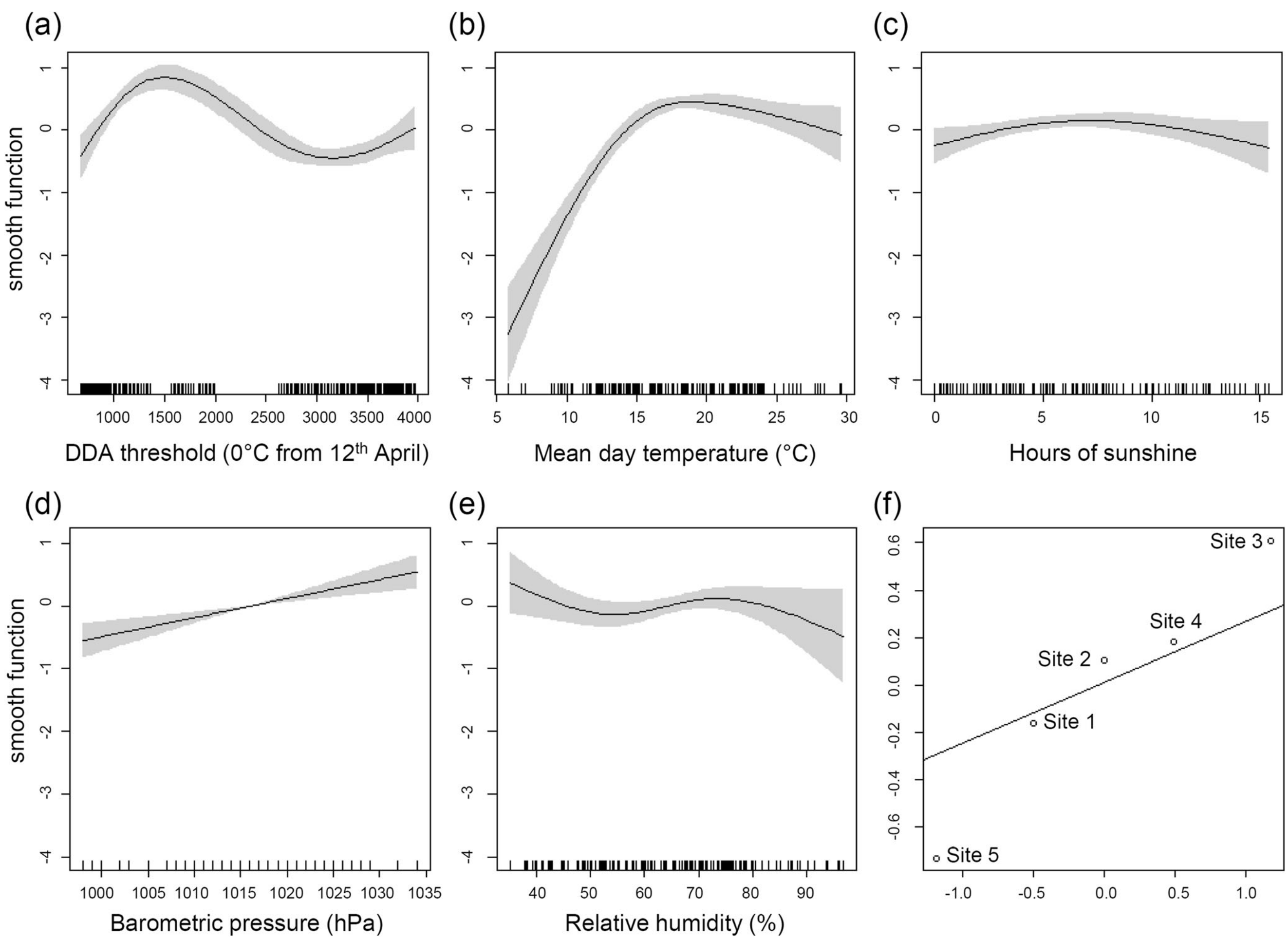

Fig. 2 **a–f** Variables affecting the abundance of female *Calliphora vicina* in Frankfurt am Main from April to October 2017. The figure presents the value of the smooth parameter function associated with each significant independent variable. Shaded areas represent the standard error of the mean associated with the given smooth function

Lucilia caesar

The GAM fitted to the activity data of *L. caesar* explains 66% of the variability in species abundance and is presented in Fig. 4(a–d) and Table 3. The activity of *L. caesar* was characterized by reduced activity in summer and a high frequency in autumn (Fig. 4a) which is also illustrated by the overall positive effect of accumulated degree days on the abundance of this species in the canonical correlation analysis (Fig. 1). Two abiotic parameters affected the abundance

Table 3 Approximate significance of smooth terms in the additive models of the daily abundance of the four most abundant blow fly species

Environmental parameter	X^2 value of parameters included in the models [*edf* value]			
	Calliphora vicina	*Lucilia sericata*	*Lucilia caesar*	*Lucilia ampullacea*
$ADD_{0\|10}$	106.536 [3.806] ***	134.471 [3.893] ***	236.055 [3.906] ***	20.101 [3.519] ***
Mean day temperature	107.081 [2.898] ***	179.366 [2.637] ***	143.700 [3.409] ***	114.175 [2.394] ***
Hours of sunshine	8.875 [2.337] *	6.201 [2.318] n.s	6.450 [2.119] n.s	–
Barometric pressure	15.876 [1.004] ***	21.326 [2.876] ***	–	5.939 [2.228] n.s
Relative humidity	10.032 [2.753] *	15.176 [2.708] **	5.882 [2.382] n.s	18.765 [1.001] ***
Wind speed	3.494 [1.236] n.s	–	18.700 [1.000] ***	4.818 [1.350] n.s
Precipitation × hours of rain	19.598 [5.235] **	5.693 [3.871] n.s	–	11.984 [3.001] **
Sites	144.737 [3.898] ***	140.516 [3.886] ***	221.601 [3.935] ***	207.159 [3.994] ***

p value *** < 0.001; ** < 0.01, * < 0.05; *n.s* not significant; − not included in the model

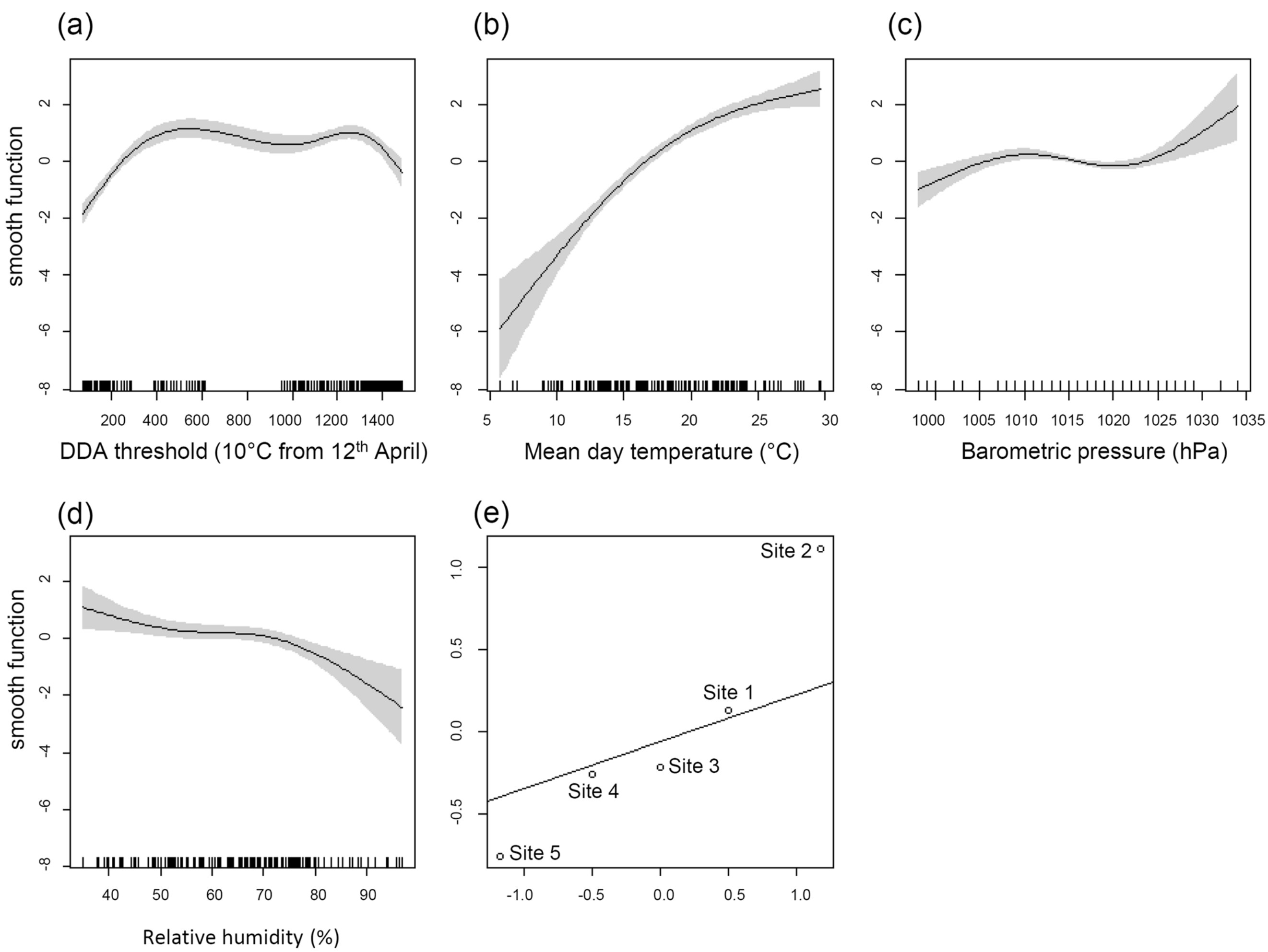

Fig. 3 **a–e** Variables affecting the abundance of female *Lucilia sericata* in Frankfurt am Main from April to October 2017. The figure presents the value of the smooth parameter function associated with each significant independent variable. Shaded areas represent the standard error of the mean associated with the given smooth function

significantly (Table 3). Mean day temperature had a strong positive effect (Fig. 4b) whereas wind speed a strong negative (Fig. 4c). Relative humidity and the hours of sunshine improved the model but did not significantly influence species abundance (Table 3). The trap catches differed strongly between the sites (Fig. 4d).

Lucilia ampullacea

The GAM fitted to the activity data of *L. ampullacea* explains 52.3% of the variability in species abundance and is presented in Fig. 5(a–d) and Table 3. *Lucilia ampullacea* was like *L. sericata* primarily active in summer when mean day temperature exceeded 15 °C (Fig. 5a) but still was only slightly influenced by ADD_{10}. Three abiotic variables significantly affected the abundance with mean day temperature (Fig. 5b) and relative humidity (Fig. 5c) being positive predictors for a flight activity and the interaction of precipitation and the duration of rain on a day being a negative predictor. Wind speed and barometric pressure were included in the model but did not influence the frequency of this species. Likewise, to all other species, specimen counts differed between the sampling sites (Fig. 5d).

Discussion

This study is the first attempt in Central Europe to understand the flight activity of forensically important female blow flies and the dependence of their flight activity on abiotic variables. The effectiveness of our methodology in monitoring cadaver-associated fauna and in sampling sufficient data to analyze the dependency of activity on environmental parameters is supported by the fact that necrophagous blow flies comprised over 96% of all the flies collected in the samples. The observed female-biased sex ratio in our trap catches is not surprising and well documented in other studies [44, 45]. Females are more likely to be attracted to carrion baited traps because they searching for oviposition substrate or a source of protein uptake for a successful egg development [44]. In

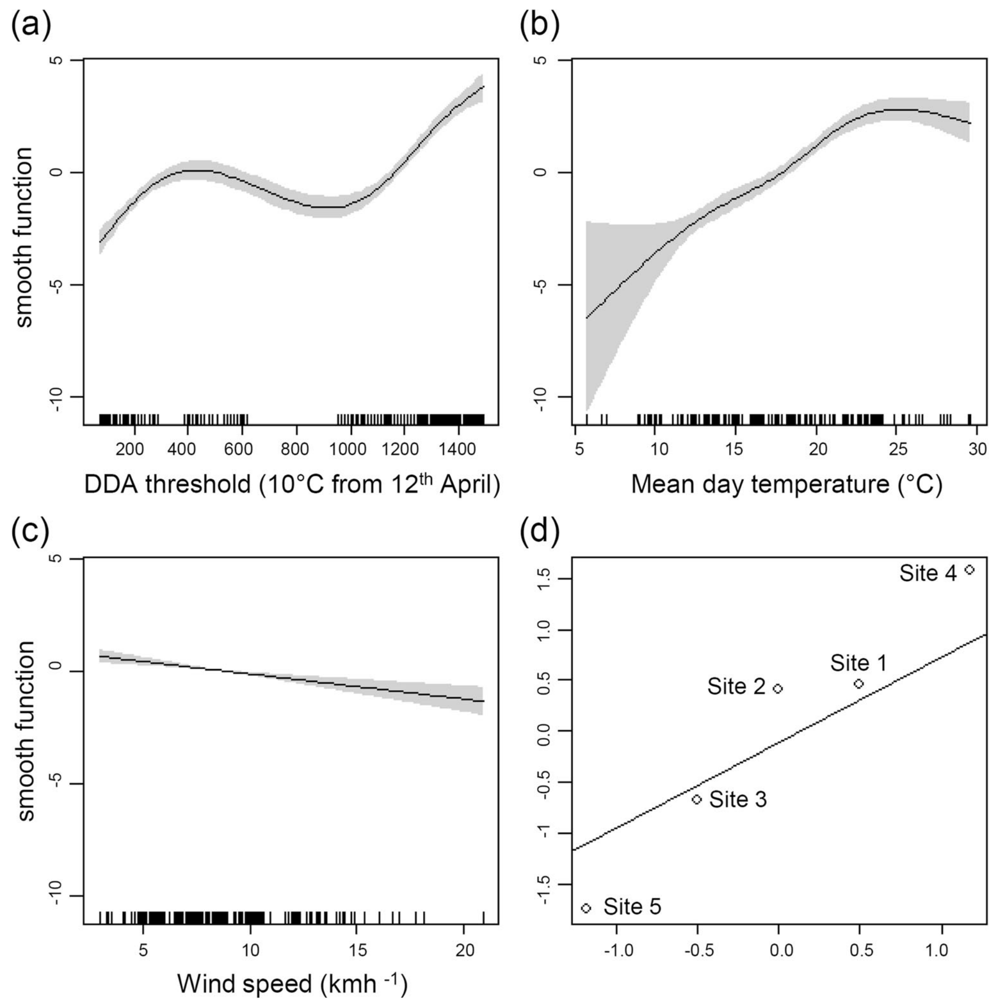

Fig. 4 **a–d** Variables affecting the abundance of female *Lucilia caesar* in Frankfurt am Main from April to October 2017. The figure presents the value of the smooth parameter function associated with each significant independent variable. Shaded areas represent the standard error of the mean associated with the given smooth function

contrast to other studies, the most common species were analyzed individually in this study and only female specimens were used to obtain information on species- and sex-specific behavior towards the measured abiotic variables. The use of GAMs allowed the identification of climate factors significantly related to the flight activity of the analyzed species. For all four abundant species, *C. vicina*, *L. ampullacea*, *L. caesar*, *and L. sericata*, mean day temperature was the most important abiotic variable that was significantly related to flight activity. Mean day temperature positively affected all species, i.e. flight activity also increased with an increase in temperature. These findings are congruent with many other studies [7, 13, 18–21], which identify temperature as the most important environmental parameter for the activity of blow flies. Especially, the minimum temperatures at which blow flies are still active are strongly determined by the energy demand of such an activity, and species-specific lower temperature thresholds act as important climatic factors determining insect flight [19]. Despite the overall positive effect of mean temperature for all species, the aforementioned species-specific differences were also found. For *L. sericata*, a well-known, sun-loving [18, 46], high-summer species in Central Europe [47], temperature was by far the most important factor. This finding may explain the dominance of this species especially during summer in our study. Even though the first appearance of *L. sericata* in this study was recorded in April, when the mean day temperature was 15 °C, a strong increase in activity and the emergence of new specimens was only noted when the mean day temperature exceeded 20 °C. These findings are also supported by records of species abundance on human bodies in the area of Frankfurt/Germany in 2015 to 2016 [48]. The authors of this study identified *L. sericata* as the most dominant species, and the most abundant one on human cadavers, during the warm summer season [48], which fits well with our observation and the high influence of temperature on the flight activity of this species. Our analysis also showed that the upper temperature threshold for activity of this species exceeded a maximum mean day temperature of 30 °C (Fig. 3b) and, as a consequence, temperatures of up to 40 °C may not limit flight activity in *L. sericata*. The other *Lucilia* species, *L. ampullacea* and *L. caesar*, were also positively influenced by temperature, but commenced their flight activity only on days with temperatures higher than 12 °C. In contrast, *C. vicina* was also positively affected by

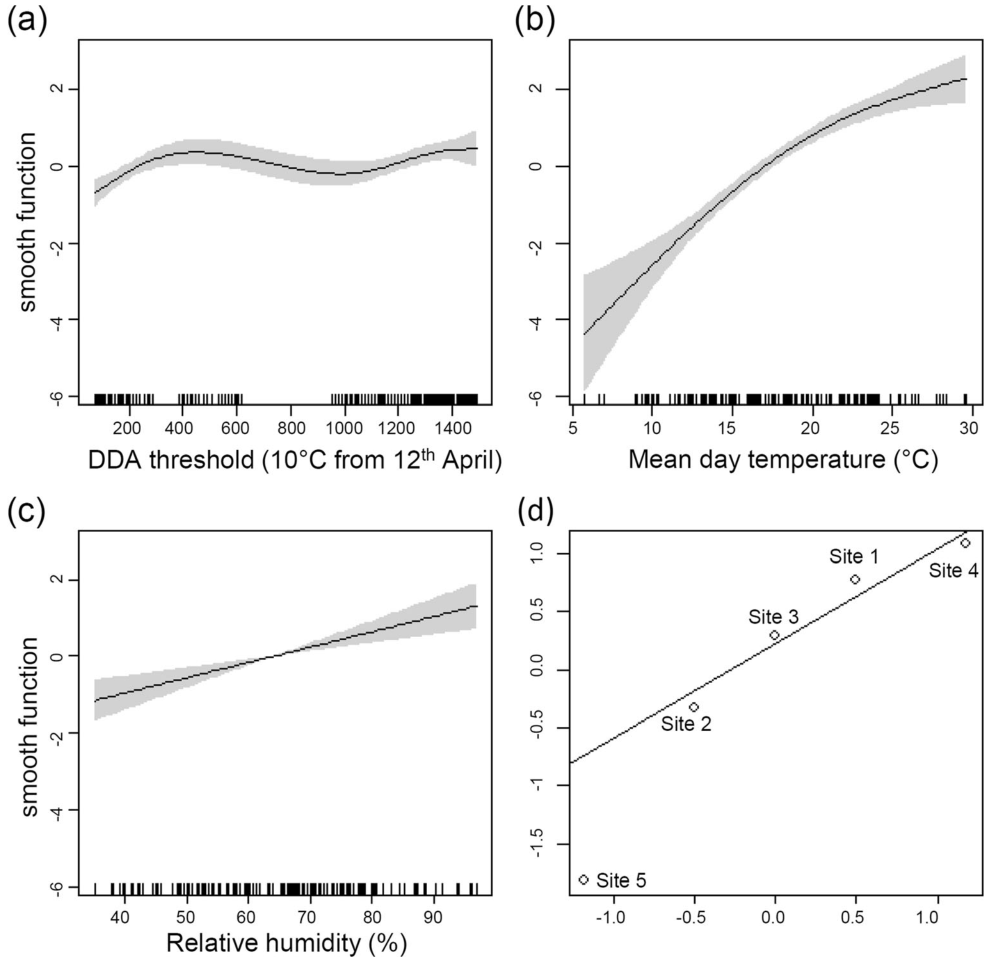

Fig. 5 **a–d** Variables affecting the abundance of female *Lucilia ampullacea* in Frankfurt am Main from April to October 2017. The figure presents the value of the smooth parameter function associated with each significant independent variable. Shaded areas represent the standard error of the mean associated with the given smooth function

increasing temperature, but with a much higher tolerance limit for cold temperatures than the *Lucilia* species. *Calliphora vicina* was the only species to appear throughout the entire sampling period from March until October, and was already found to be active in the same location during the end of January (unpublished data). *Calliphora vicina* possesses the advantage of a higher metabolic rate and, therefore, better storage of heat, especially on cold days [49]. In summary, *Lucilia* species occur over a shorter period of the year and are almost exclusively limited to the warmer months. The higher developmental temperature thresholds of the *Lucilia* species in comparison to *C. vicina* may be a cause for the later seasonal occurrence of adults in the year and may be a reason for the greater dependence of their flight activity on ambient temperature.

Next to temperature, many studies identify rain as the most important factor for insect life. Although some studies reported an inhibitory effect of rain [7], they also observed that flies resumed their activity during rain breaks [3]. In the present study, "rain" was analyzed by merging the duration and the amount of rain during a day. The results of the canonical correlation indicated that all species were negatively influenced by rain. However, a significant negative effect could only be observed for *C. vicina* and *L. ampullacea* in the GAMs. For these species, the probability of flight activity is low on days with constant and high precipitation. Our study is one of the first studies to analyze a combined variable for the factor rain. The overall negative influence of that rain-index on flight activity indicates that the hours of rain on a day, i.e. its duration, seems to have a greater influence on flight activity than the amount of precipitation in general (e.g., measured in mm per m^2).

Changes in barometric pressure can forecast changes in weather conditions, i.e. a drop in barometric pressure usually forecasts precipitation, whereas a rise usually precedes sunny and dry weather [50]. So far, not much research has been done to analyze the effects of changes in air pressure on the flight activity of female blow flies, and, in the research that has been done to date, the observed effects were not significant [7]. However, it has been discovered that both an increase and decrease in atmospheric pressure can affect insects directly by influencing their rate of development, the time of their ecdysis or emergence, their feeding, oviposition, general locomotion, and flight activity [51]. Our results showed that barometric pressure had a significant effect (Table 3) on flight activity for at least three species. For *L. sericata* and *C. vicina*, it was actually one of the most important environmental factors, after temperature. With a rise in barometric pressure, the activity of both species increased likewise. For *L. ampullacea*, this environmental parameter had a marginal positive but nervertheless, significant effect.

Blow flies like to oviposit in moist places because these offer the perfect conditions for their offspring [2, 6, 52, 53]. A higher relative humidity could, thus, be postulated to act as an indicator for humid or moist oviposition sites. In our study, relative humidity was, indeed, found to be a significant factor for three of the most abundant species. However, for *C. vicina* and *L. sericata*, we found a negative relation, especially for values > 80%, i.e. with increasing relative humidity the flight activity of these species decreased. These results are consistent with those from other studies that found a negative relation between flight activity of blow flies and relative humidity [7, 20, 26, 27]. A high-relative humidity, mirrored in a drop in barometric pressure, is thus an indicator for rain and can be used by these flies as a signal to search for shelter or to avoid oviposition [7]. *Lucilia ampullacea* was, actually, the only species that was positively influenced by relative humidity. For this species, the result fits in with the aforementioned preference for a higher relative humidity that affords better conditions for successful development of juvenile stages [36]. In addition, because the species is strongly negatively affected by heavy rainfall, *L. ampullacea* may increase its activity before rain, as its flight activity is completely prevented during rain.

Models could be improved for only three species by including the hours of sunshine as an explanatory variable, and only *C. vicina* was significantly influenced by this factor. Reasons for this could be the different microclimates at the different sampling sites (direct sun exposure vs. shade), which likely did not match the data from the local weather station at all times. Such possible differences are a permanent challenge in forensic entomology when it comes to the reconstruction of crime scene temperatures [54–56]. This might be even more complicated as flies are “moving targets” and reconstructing a climatic parameter like, e.g., temperature for a sessile/immobile situation like a maggot mass on a body is different than getting an idea about the climate situation of a certain area which impacts the flight activity of its insects. However, for *C. vicina*, sunshine has a big impact on days with a low mean day temperature. On such days, for example, in winter, *C. vicina* heats up its body in sun spots on walls (personal observation). This behavior enables the adults to fly even when the temperature is close to or below the lower activity threshold of 2.5–4 °C [49]. The preference of *L. sericata* for sunshine can be linked to the habitat preference of this species. Many authors describe this species as a sun-loving, high-summer species that can withstand high temperatures and breeds mainly in open habitats where sun exposure is high [57, 58]. Therefore, the preference for high temperatures and sunshine in this species is a result of the seasonal phenology and association with open habitats. Such seasonal phenology of *L. sericata* might differ across different regions in Europe due to adaptations to other temperature profiles especially in the summer month. An example is given in the study of Díaz-Aranda et al. [59] where *L. sericata* was the dominating species in spring and autumn. Habitat preference may also be the explanation for the negative influence of sunshine on the activity of *L. caesar*. *Lucilia caesar* is a typical woodland species [58, 60] that prefers shady habitats [46, 61, 62] but still can be found both in open habitats and in forest areas [57].

Wind speed is central among the various weather factors that influence the flight activity of insects [31]. Especially for blow flies, which depend on olfactory clues to detect relevant food sources, wind serves as an important carrier of information, even over long distances [35, 63]. Therefore, it would not be surprising if this parameter were to have a larger impact on the flight activity of female blow flies. Nevertheless, in our study, only one species was significantly influenced by wind speed. For *L. caesar*, wind speed was a negative predictor of flight activity, and a decline in flight activity was observed with an increase in wind speed [7, 38]. The strong influence of this factor on *L. caesar* might be related to its usual occurrence in forests. In forests, particularly in deciduous ones, the closed canopy provides protection towards rain and wind. Without the protection of the forest, *L. caesar* may, therefore, be more sensitive to rain and wind than other species.

In summary, all species revealed species-specific adaptations to the environmental parameters monitored in our study. *L. sericata* is a typical summer species that needs high temperatures and sunshine. In contrast, *L. caesar* seems to be a late season, woodland, shade-loving species that is influenced by parameters of inclement weather conditions. *Lucilia ampullacea* is also a summer species that is mostly influenced by temperature and relative humidity. To date, *L. ampullacea* is an underrepresented species in the forensic entomology literature [48]. However, in the last few years, it has been described as the second most abundant species colonizing human remains in Frankfurt [48] and was sampled as the third most abundant blow fly species in Catope-watertraps in the area of Frankfurt/Germany [64]. In this regard, new studies that are focusing on the developmental patterns, as well as the activity and abundance patterns, of *L. ampullacea* seem to be worthwhile.

The species with the highest tolerance limit for the measured environmental parameters, and with the lowest overall influence of these parameters on flight activity, was *C. vicina*. Less than 50% of the variability in the abundance of this species could be explained by the measured abiotic variables. Here, one reason could be the monitoring time that was restricted to late spring till the beginning of autumn and therefore did not cover the entire activity of this species. Especially, in summer we could observed a strong decrease in the flight activity of this species which can be explained by the low tolerance of *C. vicina* to mean day temperatures >25 °C. Definitely, *C. vicina* is a thermophobic species and more abundant under colder temperature conditions. Thus, high temperatures in summer can inhibit flight activity in this species.

Despite the different flight activity patterns noted among the species, our study could confirm the general assumption that temperature is a relevant factor shaping the distribution and the flight activity of blow flies. Temperature was, in fact, the most important parameter in all four of the abundant species we looked at in this study. Seasonality, strongly linked with specific temperature profiles, seems to define the abundance of the most forensically important blow fly species. The specific temperature profiles can vary from region to region in Europe and must be taken into account in forensic case work. *Lucilia sericata* is a good example of such geographic patterns. In our study, it was clearly associated with high temperatures and was the dominant species in summer, while in southern Europe phenology may change to an almost year-around occurrence in Italy [65] or to dominance in spring and autumn in Spain [59]. It might be a challenge of the nearest future to proof possible new modifications in behavior of the analyzed species due to climate change and related heat periods.

In conclusion, this study, which collected new information on the most important blow fly species for forensic case work, is one of the first in Europe to look at and compare data for the influence of abiotic factors on the flight activity of blow flies on an individual species level. Knowing the activity patterns of forensically important flies and understanding the main factors driving their flight activity is the very first step in modeling colonization probabilities in a forensic context. A next analysis could focus on the ovarian cycle of forensically important female blow flies and on factors that could have a significant influence on their eggload or oviposition behavior, as not every female fly on the wing with access to a dead body will necessarily guarantee an oviposition event. Low temperatures, which might still be suitable for flight activity, could conceivably inhibit oviposition. Generally, temperatures below 12 °C and above 30 °C have been reported as unfavorable for the colonization of corpses by most blow fly species [36, 66]. Exceptions do, however, exist [67, 68], so this general statement might prove to be quite variable at a species level. Ody et al. [69] recently established lower temperature thresholds for oviposition of 16 °C and 17.5 °C for *Calliphora vomitoria* and *Lucilia sericata*, respectively, in their study, while *Calliphora vicina* continued to lay eggs at 10 °C. These data were, however, obtained in the laboratory. The determination of oviposition temperature thresholds in the field would, thus, definitely improve the evaluation of a possible delay in the colonization of a dead body by blow flies. A year-round monitoring, i.e., not just focusing on the main windows of fly activity, could help to understand these thresholds and their impact not just on flight activity, but also on oviposition events, especially for species such as *C. vicina* which can be active over all seasons.

On the basis of the flight activity parameters provided by our study for the relevant blow fly species, a targeted oviposition study is now possible on suitable (= flight-active) days. Special “oviposition traps” could help in this endeavor. The design of appropriate traps is, however, an ambitious task. Synthetic bait, to ensure a constant and uniform attraction over time, would be an important component of such traps to avoid biased results due to changes in the attractiveness of the bait. By using such a universal attractor, a sound analysis of the data—which is crucial for statistically modeling the impact of these effects—could be achieved.

Understanding the flight activity and oviposition patterns of individual blow fly species definitely leads to a better understanding and interpretation of possible delays in the colonization of a dead body in forensic contexts. Ultimately, there will, however, always be very case- and site-specific random effects that cannot always be taken into account. An open or a closed window, for example, or a wrapped body, or body-specific factors, such as medication or intoxication, can impact prompt colonization of a body by blow flies. The ability to evaluate the magnitude of such impacts depends not only on the experience of the forensic expert, but also on the quality of the data that she or he can refer to in the report.

Acknowledgements The authors would like to thank D. Ricciardi and V. Bernhardt for their assistance during fieldwork, an anonymous colleague for helping with the statistical analysis, and the two anonymous reviewers for their careful reading and commenting of the manuscript.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

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