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#### Article

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**A biological and procedural review of forensically significant  
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**Abstract**

The analyses of the insect species found on decomposing remains may provide useful information for the estimation of the minimum time elapsed since death and other parameters, such as causes and circumstances of death. The majority of research has focused on the early colonising species, typically blowflies, while research concerning late colonising insects is currently sparse. Dermestid beetles of the genus *Dermestes* L. (Coleoptera: Dermestidae) are one of the predominant insect species associated with decomposing remains during dry decay and skeletal stages of decomposition. In some dry environments *Dermestes species* are likely to be the only necrophagous insects feeding on the decomposing remains. Furthermore, *Dermestes species* (immature and adults), their remains (cast skins and fecal material) and their artifacts (pupal chambers) are frequently found associated with ancient remains (e.g. mummies, fossils). *Dermestes species* have a worldwide distribution and are considered important in decomposition processes, forensic investigations and economically as a known pest of stored products. Despite their recognised forensic importance, there is limited data documenting the ecology, biology and the growth rates of the forensically relevant species.

The aim of this review is to provide a comprehensive synopsis on the available literature concerning *Dermestes* species associated with forensic cases. In particular, aspects of colonisation behaviour, growth rates for forensic taxa and potential best practice guidelines for forensic casework encompassing late colonising *Dermestes species* are discussed.

**Keywords**

*Dermestes* spp., ecology, development, decomposition, forensic entomology

## Introduction

Forensic entomology is involved with insects and other arthropods present at crime scenes (Magni et al. 2008, Byrd and Castner 2010a). When immature insects are found on decomposing remains, the correct sampling, measuring and subsequent interpretation can provide useful information such as the minimum post-mortem interval (minPMI) and the post-mortem movement of the remains, detection of toxicological substances and/or human DNA from the crop and gut of larvae (Di Luise et al. 2008). One of the most important estimates of minPMI is based on the age of immature insects inhabiting decomposing remains and knowledge of initial colonisation timeframes for the identified species (Byrd and Castner 2010a). It has been determined that insects colonize remains in a predictable manner and that the development of these colonising offspring is strongly correlated with climatic conditions such as temperature (Smith 1986). As such, the observed assemblage of species present on the remains, along with associated thermal history, can be used to determine the time elapsed since death (Byrd and Castner 2010a). To date, most research has been conducted on decomposition processes in terrestrial environments and in different climatic situations (Haglund and Sorg 1997, Byrd and Castner 2010a). The majority of research has focused on the life history and behaviour of early colonising species, typically blowflies (Diptera: Calliphoridae), and the decomposition stages associated with these species (Smith 1986). There is currently a paucity of research concerning late colonising insects, yet expertise in forensic entomology is often required in cases where human and animal remains are in a late stage of decay (Archer et al. 2005). As a consequence, data relevant to minPMI determination for late stages of decomposition (skeletal, mummified and dry) are scant and less accurate (Magni et al. 2008, Haskell and Williams 2009). Numerous cases of skeletonized or mummified bodies are found in houses, weeks, months or even years after death (Hönigschnabl et al. 2002, Archer et al. 2005, Magni et al. 2008, Nilsson and Logdberg 2008, Campobasso et al. 2009, Charabidze et al. 2013). As well, insects and/or their remnants may also be found in ancient tombs associated with archaeological human remains. This type of insect material is used to define the peri- (= around) and post-mortem events

50 (“archaeoentomology”) or funerary practice (“funerary archaeoentomology”) in paleo-forensic  
51 contexts (Huchet 1996, 2014).

52 During late decay and skeletal stages of decomposition, beetles are the predominant insect species  
53 associated with remains and typically the dominant species are from the family Dermestidae  
54 (Coleoptera) (Smith 1986). When decomposition takes place in dry environments (e.g. desert)  
55 dermestid beetles are likely to be the only insects present on the remains (Bellussi 1933).  
56 Furthermore, insects belonging to the family Dermestidae are frequently found associated with  
57 mummies (Lesne 1930) and marks attributed to such beetles found on fossils have been used for  
58 paleoecology and paleontology studies.

59 Dermestids undergo complete metamorphosis (egg, larva, pupa, adult). Moulting larvae produce  
60 cast skins (exuviae) and when fully developed, larvae bury into a variety of materials to pupate,  
61 forming “pupal chambers” (Bruesch 2011). The total developmental time from egg to adult is  
62 highly variable depending on the species, environmental temperatures and humidity, food source  
63 and population size (see details in the subsequent paragraphs). As a consequence of human  
64 civilisation and the habit of curing meats with salt for preservation, it is now known that the  
65 development time and the survival of different species of dermestids may also be affected by the  
66 salt content in the food source (Osuji 1975a). Such knowledge, although derived in a non-forensic  
67 context, can be useful when interpreting a minPMI where decomposing remains, for instance, have  
68 spent time in salt water (Magni et al. 2015).

69 Dermestids are a widely distributed family of beetles that are found across Europe, the Americas,  
70 Canada, Africa and Asia. Such an extensive distribution is likely due to global trading (Bruesch  
71 2011). Worldwide there at least 1000 species of dermestid beetle, but the global fauna is still poorly  
72 known and this has been compounded by the many nomenclatural changes and the existing  
73 synonyms (Háva 2003). However, the majority of the forensically relevant species of dermestids are  
74 from the genus *Dermestes* L.. They are primarily considered and treated as pests of stored products  
75 (Bruesch 2011), but they are also important in the decomposition process of humans and other  
76 animals (Smith 1986).

The value of biological knowledge relating to insects of forensic importance cannot be overstated and yet no single resource exists that centralises such data for use in forensic case work. The wide scope of research disciplines across which potentially relevant data has been generated inhibits location and accessibility of data for forensic application. Additionally, while a plethora of research and case studies report the presence of Dermestidae colonising decomposing remains (Charabidze et al. 2013), there is limited data documenting the necessary development rates of relevant *Dermestes* species to determine a minPMI. Where reference data does exist, it is often difficult to locate due to the lack of peer reviewed publication and/or confidentiality issues surrounding case work.

This review aims to address the issue of access to reference data pertinent to minPMI estimation for late stage decomposition by providing a comprehensive reference guide to the available research on *Dermestes* species. In particular, aspects of colonisation behaviour and growth rate for forensic taxa are discussed and best practice guidelines for forensic casework encompassing late colonising *Dermestes* species are outlined. Gaps in the current knowledge base, relevant to forensic investigation, requiring further research are also identified.

### **Dermestes species at the death scene**

Beetles of *Dermestes* species are regularly found at crime scenes, especially in city apartments and houses where environmental conditions are dry and warm. Investigation of indoor death scenes, involving late stage decomposition, regularly involves the discovery of such beetles feeding directly upon decomposing remains (Schroeder et al. 2002). Social isolation of elderly people is a problem in many big cities around the world (Kulshrestha and Satpathy 2001, Hönigschnabl et al. 2002, Archer et al. 2005, Nilsson and Logdberg 2008, Campobasso et al. 2009, Williams 2009) and every year skeletonized and naturally mummified bodies are found in houses and apartments (Hönigschnabl et al. 2002, Magni et al. 2008, Charabidze et al. 2013). As such, the likelihood of encountering adults, larvae and/or by-products of *Dermestes* species at a crime scene is high, which exemplifies the potential of this group as useful indicators of minPMI in such cases.

Numerous studies have demonstrated the significance of the presence of *Dermestes species* in biomass removal, showing a reduction of up to 50% of carcass biomass by insect activity in xerophytic and mesophytic habitats (Lord and Burger 1984, Early and Goff 1986, Hewadikaram and Goff 1991). In habitats where dermestids are absent (e.g. rain forest), biomass reduction for the same period was only 10% (Tullis and Goff 1987, Richards and Goff 1997). In contrast to outdoor crime scenes, indoor death scenes are associated with a lower number of species involved in the process of decomposition (e.g. restricted access and/or insecticides/insect traps are present) and a slower progression in insect succession (Magni et al. 2008). The speed and extent of biomass removal for indoor decomposition is highly dependent on the environmental situation (e.g. cleanliness and accessibility).

Natural mummification takes place when the remains lose fluids to the environment via evaporation (Haglund and Sorg 1997). Experiments performed in different natural environments suggest that extremes of heat or cold and appropriate air currents can facilitate this process (Haglund and Sorg 1997). Sometimes domestic dwellings can be a suitable environment for mummification due to the presence of carpets, sheets, blankets and other coverings which facilitate the absorption of putrefactive fluids (Campobasso et al. 2009). The identification of the time since death of mummified remains is extremely complex both from medical and entomological perspectives. The time needed for mummification varies drastically based on the carcass type (e.g. size, amount of fat tissue), the location and the external environment. As such, the succession of insects associated with remains during late stage decomposition is not easily predictable without greater understanding of the species-specific influence of relevant biotic and abiotic factors (Archer et al. 2005, Campobasso et al. 2009).

As discussed, there is certainly evidence indicating the importance and advantages of further developing our understanding of dermestid biology in the context of their role in decomposition and forensic investigation. Additionally, adult dermestids, cast skins and faecal material persist in the environment for considerable periods of time (Byrd and Castner 2010b). Such remnants have been used for toxicological analyses in forensic cases where human decomposed tissue is highly

degraded or absent entirely (Miller et al. 1994, Wolff et al. 2004). It has also been proposed that host DNA may be identified from this insect material (Manhoff et al. 1991). As well, dermestids and other invertebrate necrofauna are used in an archaeological funerary context to obtain information on the immediate environment of the site or the grave at the time of the burial, (Huchet et al. 2013b) the post-mortem stages and even on the duration and process of the mummy's embalming (Huchet 2010). In paleontology and "paleo-forensics" adult species of Dermestes have been found perfectly preserved (Huchet et al. 2013b), as have larvae and pupal chambers (Martin and West 1995, Laudet and Antoine 2004, Huchet et al. 2013a, Huchet 2014) (Fig. 5). As such, adults, larvae and remnants of Dermestes species are frequently associated with death scenes and offer a wealth of potential information to forensic investigators where relevant biological data is available. Unfortunately, the most beneficial aspect of dermestid evidence associated with decomposing remains, which could be the indication of minPMI, is hampered by a paucity of such data.

#### **Dermestes species** as indicators of minPMI

The Dermestidae family represents one of the most economically important insect groups in the world (Crowson 1967). Although they are viewed primarily as a pest of stored products (Hinton 1945) they have also been used as a means of removing hide and tissue from animal specimens in the case of museum collections (Munro 1966, Halls and Russel 1993, Offele et al. 2007). More importantly in regard to forensic investigations, dermestid beetles of the genus Dermestes are included in the list of the most common necrophagous insect species visiting, rather than utilising the remains (Smith 1986).

Considering the ability of Dermestes species to locate a deceased animal for resource exploitation and the late stage of decomposition at which they typically arrive, they can be useful in determining the minPMI (Smith 1986). Dermestes species, however, are not the most predictable of colonisers with colonising time frames strongly influenced by the death scene and associated environmental conditions. Dermestes species are most frequently associated with advanced decomposition,



arriving and colonizing remains when only skin and bone remain approximately 3-6 months following death (Bornemissza 1957, Reed 1958). This concurs with the study of Mégnin who first documented *Dermestes species* on exposed remains in a temperate climate during the third wave of decomposition, when the fats were rancid (after 3-6 months) (Mégnin 1894). Mégnin also indicated that *other species of the family Dermestidae* were most prevalent during the 7<sup>th</sup> wave, when the remains were completely dry (after 1-3 years) (Mégnin 1894). More recent studies, however, have reported somewhat contrary dermestid arrival times on remains as early as 3-11 days, although larvae were not collected until later (during the dry stage, days 12-66+ and the remains stage, days 25+) (Early and Goff 1986, Hewadikaram and Goff 1991, Richards and Goff 1997). In one case, where conditions were extremely dry (deserts habitat) *Dermestes species* were found on dog carcasses as early as 24 hours after exposure (Bellussi 1933). Tomberlin (2009) supports this, stating that dermestid beetles are typically found on remains throughout the decay process, however many species will be present on dried remains. Dermestids may visit decomposing remains during all decomposition stages but demonstrate a preference for dry remains in respect to abundance, oviposition and feeding activity. Arrival prior to preferred conditions may simply be an adaptive response related to competitive advantage, like many opportunistic decomposers that feed on a variety of resources (VanLaeroven 2010).

While influenced by environmental conditions, these broad colonisation timeframes may also be related to differences in the colonisation behaviour of a variety of species as, in most cases, the dermestid beetles collected in association with forensic research are not identified beyond taxonomic family (Archer et al. 2005, Campobasso et al. 2009). This oversight by practitioners of not identifying the actual species can lead to some of the literature based conclusions that dermestid beetles typically only consume dry remains. However, certain adult species such as *Dermestes maculatus* DeGeer prefer moist muscle tissue and ligamentous remains. In addition, *D. maculatus* can occasionally act as a predator of blowfly larvae and/or consume dead insects (Braak 1987). A summer experiment on the decomposition pattern of shaded and exposed pigs reported *D. frischii* (Kugelan) and *D. undulatus* (Brahm) attending pig remains on day 23 and 24 respectively, but only

on the exposed pigs (Shean et al. 1993). No dermestid species were found on shaded pigs for the total duration of the experiment (Shean et al. 1993). Thus, the arrival time and succession of dermestids is not necessarily tied to the decomposition stage of the remains but may depend on species-specific preferences for environmental conditions (VanLaeroven 2010). Environmental conditions, as well as *priority effects* and *exclusion mechanisms*, can also affect the decompositional pattern and these factors may determine whether the decomposing remains will be colonized by dermestids (Bellussi 1933, Charabidze et al. 2013).

*Priority effects* occur when a species that is already present either inhibits or facilitates other species that subsequently arrive at the resource. Priority effects have been demonstrated for necrophagous insects on carrion (Hanski 1987). For instance, the utilisation of a carrion resource by blowflies potentially facilitates future colonization by dermestid beetles (Schoenly and Reid 1987). However, for some dermestid species, such as *D. maculatus*, colonisation of decomposing remains occurred prior to blow fly colonisation in the case of woodland (Braak 1987) and desert (Bellussi 1933) environments.

*Exclusion mechanisms* affirm one or several mechanisms (e.g. repulsion, competition, predation) linked to the presence of one species and decreasing the probability that additional species would subsequently colonize the remains. One review work of forensic case records provided an indicator of exclusion mechanisms acting on *Dermestes species* colonisation patterns. Charabidze et al. (2013) conducted an analysis of forensic cases occurring in France over a 20 year period. *Dermestes species* were only observed in 81 of the 1093 cases included in the analysis (Charabidze et al. 2013). As acknowledged by Charabidze et al. (2013) sampling bias in regard to the different personnel involved in the cases reviewed and their training levels could account for the low number of reported observations. Interestingly, however, in 78% of these 81 cases only a single *Dermestes* species was observed suggesting the possible influence of an as yet unidentified exclusion mechanism (Charabidze et al. 2013). It was also noted that the species distribution was clearly more balanced in indoor cases than in outdoor death scenes (Charabidze et al. 2013). Accordingly, Charabidze et al. (2013) suggests that experiments under controlled conditions are required to

determine the potential mechanisms driving the colonisation patterns observed. Future studies are required to investigate whether certain dermestid species are competitively excluded by other necrophagous insects (such as blowflies) or if these species are simply poor dispersers and are unable to reach the carrion resource until later in the community assembly.

Upon review, the unpredictability often reported for the colonization timeframes of forensically relevant Dermestidae appears largely a consequence of inadequate research. In the absence of comprehensive biological and ecological data the development of an accurate predictive model for the estimation of minPMI is unfeasible. Investigation of the colonising factors, specific to relevant species, would greatly enhance our current understanding of Dermestidae succession and add considerable value to the group as an additional PMI estimation tool. Few controlled studies on the resource location preferences of *Dermestes species* exist (Table 1), and further work is needed to document *Dermestes species* arrival and oviposition timeframes under death scene conditions. Here we provide a comprehensive reference guide (Table 1) to the relevant literature as an aid to forensic case work involving the collection of Dermestidae and their by-products, and as a basis for determination of the direction of future research requirements.

**Species of Forensic Relevance**

Within the family Dermestidae, the species most frequently observed in association with decomposing remains are those within the genus *Dermestes*. Adult beetles of this genus are recognized by their oval shape and dark colour (black or dark brown) with a number of light coloured spots situated around the margin (Munro 1966). Often covered with scales which form patterns useful in their identification they typically vary in length between 3 to 12 mm (Munro 1966). Members of this genus are commonly referred to as hide, skin, larder, leather, tallow, incinerator, bacon and carpet beetles which reflects their dietary choices (Bruesch 2011). *Dermestes* species are common and many are cosmopolitan, but only 14 have been reported in association with both human and animal remains (Table 1). In the majority of these cases only a single *Dermestes* species has been reported in association with remains (Charabidze et al. 2013).

The biology of the different *Dermestes* species is very similar (Munro 1966). Male beetles excrete a pheromone to attract females and within a very short time many adult beetles may arrive on a corpse and a large number of eggs will be laid continuously over a few months (Levinson et al. 1978, Levinson et al. 1981, Jacobs and Renner 1988, Conquest 1999). *Mature* larvae are *generally* brownish in colour, 11-13 mm in length and are covered with strong bristle-like hairs of different sizes (setae), the shorter ones being borne in tufts. *Futhermore*, according to Hinton (1945), these hairs can be moved or vibrated when larvae are threatened. *Larvae of Dermestes species* are characterized by two curved spines (urogomphi) which are visible on the last body segment (Bruesch 2011). The number, position and length of urogomphi are used in species identification (Bruesch 2011). A complete description and dichotomous key of the superfamily Dermestoidea and the family Dermestidae are provided by Hinton (1945), Crowson (1967), Hinton and Corbet (1975) and Veer et al. (1996). Crowson (1967) also traces the phylogeny of Dermestoidea based on the morphological features of the adult and larva.

The most commonly reported and widely distributed species of *Dermestes* is *D. maculatus*. This one species has been the focus of *many cases due to its* potential as an indicator of time since death. *Unlike* the majority of the 14 species of forensic interest, a reasonable amount of data are available detailing the reproductive behaviour and development of *D. maculatus*.

### ***Dermestes maculatus*** (DeGeer)

*Dermestes maculatus* females lay eggs that are 2 mm long and creamy in color. Eggs are laid singly or in batches of 2-20 eggs and hatch in 2-20 days (Bruesch 2011). A single female can produce between 198 and 845 eggs in her lifetime (Grady 1928, Kreyenberg 1928). A complete study on *D. maculatus*' oviposition and longevity at different temperature and humidity ranges reported the approximate developmental periods for *D. maculatus*' eggs as 7 days at 20 °C, 4 days at 24 °C, 3 days at 28 °C and 2 days at 32 °C and that humidity has little or no effect on developmental timeframes (Scoggin and Tauber 1949).

Larval development is also temperature driven and reports of developmental timeframes indicate that the larva undergo a first moult two days after hatching at 28-30 °C followed by 5 moults at intervals of 5 days (Hinton 1945). In adverse conditions, however, the number of moults can increase (Grady 1928). Smit's (1931) study of *D. maculatus* development failed to define the environment in which larvae were located during development but reports that the larval period can range from a minimum of 35 days in summer to a maximum of 238 days during the rest of the year. The larvae cease to feed 4 days before pupation and then wander in search of shelter in which to pupate (Smit 1931). The larvae can delay pupation by more than 20 days if a suitable pupation site cannot be found, but this period can compromise their body mass and their survival (Archer and Elgar 1998).

At average temperatures and humidity the life cycle of *D. maculatus* requires 60 to 70 days to progress to completion (Walker 1944). The optimum temperature for the development of *D. maculatus* colonies at constant conditions in a laboratory is 25–30 °C (Raspi and Antonelli 1996, Richardson and Goff 2001), which results in an average life cycle duration of 35.1–43.9 days. Howe (1965) describes *D. maculatus* as a species that needs high temperature (lower limit of temperature required to survive 20 °C; optimal range of temperatures 30-35 °C) and moderate relative humidity (lower limit of r.h. 30 %). At 15 °C no individuals completed development to the adult stage although some individuals remained alive at this temperature for up to nine months (Howe 1965). Kulshrestha and Satpathy (2001) report *D. maculatus* on human remains at an ambient temperature of 16.5 °C and 71% average humidity. A small number of adults were present, but no larvae or pupae were observed (Kulshrestha and Satpathy 2001). *D. maculatus* appears to only be able to reach full development if the temperature remains above 18 °C (Raspi and Antonelli 1996). Under such conditions this species takes 96 days after oviposition to reach the adult stage (Raspi and Antonelli 1996). Hinton (1945) showed that temperatures of 28–30 °C resulted in *D. maculatus* completing their life cycle in 22 days. At lower temperatures life cycles of 40–50 days were reported (Hinton 1945). At 29 °C the average length of the various stages is: egg - 3 days, larva - 30 days, pupa - 7 days and adult before oviposition - 5 days (Russell 1947).

292 Some authors have studied the life history of *D. maculatus* on mulberry silkworm cocoons (Paul et  
293 al. 1962, Rajashekhargouda and Devaiah 1985 ). They reported that eggs hatched within 2-6 (mean  
294 2.4) days, the larval period was 37-69 (mean 65.5) days and the pupal period 617 (mean 13.3) days.  
295 These data must be considered with caution when used for the evaluation of a minPMI as different  
296 diets can affect insect development (da Silva Ribeiro and Von Zuben 2010). In general, the total  
297 time required to complete development from egg to adult was inversely related to temperature and  
298 ranged from a mean of 89.7 days at 20 °C to a mean of 36.4 days at 35 °C (Richardson and Goff  
299 2001). The quickest larval development occurs in 23.4 days at 33 °C and 70% r.h. (Howe 1953).  
300 The fastest pupal development of 4.4 days takes place at 37 °C and 70% r.h. (Howe 1953).  
301 A study on *D. maculatus*' larval and pupal development at different moisture levels on various  
302 media identified that when moisture levels are low (10-15%) larval mortality is also low.  
303 Additionally, the number of the larval instars as well as the duration of larval develop<sup>ment</sup>  
304 decreases and larger adults emerge. Inversley, high moisture levels (46%) caused high mortality and  
305 a skewed sex ratio with fewer female adults emerging (Scoggin and Tauber 1951). Under  
306 favourable conditions there may be 6 generations per year (Mallis 2011).  
307 Bellemare and Brunelle (1950) reported an interaction effect between temperature and development  
308 for *D. maculatus* reared under different constant temperatures and relative humidities (25, 28, 31,  
309 34 °C and 0, 20, 50, 70 and 100 % r.h.). Complete larval development occurred only at 70 and  
310 100% r.h. and the duration of the larval period ranged from a minimum of 2.4 days (31-34 °C and  
311 100 % r.h.) to a maximum of 5.8 days (25 °C and 70% r.h.). In contrast, only temperature affected  
312 the duration of the pupal period (from 8.5-8.6 days at 25 °C and any r.h. to 5 days at 34 °C and any  
313 r.h.) (Bellemare and Brunelle 1950). Toye (1970) reported similar developmental times for *D.*  
314 *maculatus* reared under a constant temperature of 25 ± 1 °C over 2 ranges of humidity (10–60 % and  
315 50–100 %). *D. maculatus* showed a preference for a relative humidity of 50-60% (Toye 1970). The  
316 behaviour of *D. maculatus* infesting dried fish in Nigeria under different combinations of humidity  
317 and temperature were also observed (Toye 1970). It was also noted that during the morning when  
318 the fish carcass temperatures were 24-26 °C the beetles feed on the carcass' surface, but as soon as

the ambient temperature increases to 29-47 °C *D. maculatus* move inside the carcass, where the internal temperature is lower (29-42 °C, 40-70 % r.h.). Where *D. maculatus* were raised on fish with a high lipid content as a food source, a shorter length of larval stage was recorded (Osuji 1975a).

The length of the larval period can also be affected by the size of the larval cohort (Rakowski and Cymborowski 1982). Metamorphosis time is affected by larval density as well as by chemical compounds liberated in the faeces by both adults and larvae (Rakowski and Cymborowski 1982). Therefore, as with blowflies, dermestid population size as well as temperature should be taken into consideration when estimating the age of the larvae present on the remains.

It is also important to note that many food stuffs are cured using salt and this information may be useful in relation to bodies that have been submerged following drowning in salt water or following a tsunamis\_ (Magni et al. 2015). Salt affects both development and survival of *D. maculatus*; in experiments at 30 °C, larval development took 37 days on fish with 3.5 % salt content compared with 21.5 days on unsalted fish, and mortality reached 100% when the salt content was increased to 9.2 % by brining for 1.5 h (Osuji 1975a, Osuji 1975b, Ezenwaji and Obayi 2004, Zakka et al. 2013).

A few studies have assessed the cues used to locate and colonise decomposing remains, but the role of visual, olfactory and tactile cues in attracting male and female dermestids to remains is largely unknown. vonHoermann *et al.* (2011) recently demonstrated that freshly emerged male *D. maculatus* are attracted to the EAD-active compound benzyl butyrate released in high levels following bloat during the decomposition process but were unable to demonstrate a similar consistent preference by females (von Hoermann et al. 2011). Additional olfactory cues such as male released pheromones and prey derived odour cues were not assessed but are possible sources of attraction and identification cues for resource location by *Dermestes* beetles. Understanding the cues used by *Dermestes* species to locate resources in patchy environments is an essential component required for establishing succession timeframes and ultimately developing the group as reliable indicators for minPMI.



Given the prevalence of *D. maculatus* in forensic investigations more research is needed to quantify and measure the impact of cohort density, food source, temperature and humidity and odour cues on the species' behaviour and development. In particular, experimentally determined lower developmental thresholds and thermal constants for species development are needed to allow application of current mathematical models to determine the age of an individual.

### ***Dermestes ater* (DeGeer)**

The species *D. ater* has been reported infesting bodies both in Europe and Asia (Kumara et al. 2009, Charabidze et al. 2013). The female is capable of laying up to 400 eggs over a two month period. The biology of *D. ater* has been studied by Kumar et al. (1998) on dried mulberry silkworm pupae. Eggs hatched within 3-6 days (average 4.5 days), the larval period lasted 27-28 days, and the pupal period 7-8 days at room temperature (temperature not reported) (Kumar et al. 1998). The life-cycle takes about 6 weeks at 27-28 °C on fishmeal with drinking water (Roth and Willis 1950). The absence of drinking water retarded larval development. *D. ater* is also adversely affected by the presence of salt in their food source (Osuji 1975a).

### ***Dermestes frischii* (Kugelann)**

The dermestid *D. frischii* has been reported infesting bodies both in France and Spain (Arnaldos et al. 2004, Charabidze et al. 2013) and it is occasionally associated with *D. maculatus* in sampling decomposing remains (Paul et al. 1962). The quickest larval development of this species occurs in 23.4 days at 33 °C and 70% r.h., while the fastest pupal development takes place at 37 °C and 70% r.h. in 4.4 days (Kreyenberg 1928). Howe (1965) describes *D. frischii* as a species that shows a lower limit of survival at temperatures of 22 °C and an optimal survival rate at ranges of temperature between 31-34 °C. Furthermore *D. frischii* needs a high rate of humidity (lower limit of r.h. 50) to develop successfully (Howe 1953, Howe 1965).

By contrast to *D. maculatus* and *D. ater*, *D. frischii* is relatively tolerant of salt. At 30 °C and 75% r.h., the total development period of 34 days on unsalted fishmeal increased only to 42 and 53 days



when the salt contents were 14 % and 25 %, respectively, though a salt content of 60% prevented development (Amos 1968). However, in these experiments, the presence of salt even at 14 % had a considerable effect on larval mortality and on egg-laying (Amos 1968).

***Dermestes lardarius* (L.)**

Cases involving *D. lardarius* are reported in France and Germany (Benecke 2010, Charabidze et al. 2013). Eggs are generally 2 mm in length and the female lays eggs over a 2-3 months period (Hickin 1964). The total number of eggs laid varies from 200 to 800 (Hickin 1964) but females have been observed to lay as few as 102-174 eggs (Kreyenberg 1928). Eggs are laid from June through August and the incubation period lasts approximately 12 days (Mallis 2011). At 17 °C eggs hatch in 9 days, but at high temperatures (25-28 °C), this is reduced to 2.5 days (Hickin 1964). *D. lardarius* breed optimally at 25 °C and 80% r.h. (Coombs 1978). The larvae moult up to six times and tend to avoid light. Larvae eat constantly until the last moult when they begin to wander in search of a suitable place to pupate. The pupal stage extends from 3 days to a week or longer, depending on the environmental conditions and a generation may be completed in 40-50 days under suitable conditions (Mallis 2011). The optimum temperature for the development of this species is from 18 to 20 °C (Kreyenberg 1928). In general there is usually one generation a year, but in some situations up to 5 a year have been observed (Hinton 1945). Under optimal conditions, male *D. lardarius* completes 4 instars, whilst the female completes 5 instars (Gennard 2012).

**Feeding Artefacts**

Adults and larvae of *Dermestes species* have strong mouthparts which make it possible for them to consume hard materials. Experiments have demonstrated that larder beetles can penetrate lead with ease and tin with some difficulty, but they are unable to perforate zinc or aluminium (Bauer and Vollenbruck 1930). Dermestidae, together with Mallophaga and Tineidae (Lepidoptera), include the only species of higher organisms able to digest keratin (Caldeira et al. 2007). Adults and larvae require the same types of food, such as skins, fur, woollens, leather, feathers, bones and dry animal

matters. The genus name *Dermestes* as well as the family name Dermestidae is derived from Greek and means “to devour a skin”, a habit that is typical of this genus (Bruesch 2011). However, they can also infest cheese, mushrooms, pet food, dry fish, bacon, ham and occasionally bird and rodent nests (they are apparently attracted by the animal remains), vegetable products (chocolate, copra and cocoa beans) and waste materials burnt in incinerators (even where obsolete incinerator shafts are unlikely to have been removed and could remain a source of infestation) (Munro 1966, Smith 1986, Gerozisis and Hadlington 1995, Byrd and Castner 2010a, Mallis 2011). Larval infestation of *D. lardarius* (L.) have been associated with the presence of dead clusters of flies and dead face flies (Mallis 2011). They are particularly common in dead insect accumulations found in the pan beneath electrocuting insect light traps (Mallis 2011).

The feeding behaviour of *Dermestes* beetles can extend the decomposition process. Generally, the skin of the carcass tends to remain intact (Byrd and Castner 2010a) but is sometimes littered with many holes that can be both symmetrical, uniform and rounded or irregular in form and size (Byrd and Castner 2010a) (Fig. 1, 2). The mature larva has the habit of boring into various hard substances in order to pupate and may cause damage to the remains that can be mistaken for prior injury (e.g. gunshot wounds) (Byrd and Castner 2010a). Larvae usually form shallow tunnels (pupation chambers), sometimes up to 30 cm deep and then use the final larval skin as a plug (Hickin 1964). Brimblecombe (1938) observed severe damage by *D. maculatus* to a mill in which the larvae climbed some 7.3 m to 11 m. However, if they are unable to bore a tunnel the larval skin remains attached to protect the pupa from predaceous insects (Hickin 1964). Care should be taken that these artefacts are not misinterpreted as gunshot wounds, lacerations or possible abrasions.

Pupal chambers created by beetles of *Dermestes species* also been observed on human bones from the Middle Bronze Age (Huchet et al. 2013a) and in fossils from the late Pliocene and middle-late Pleistocene (Martin and West 1995). The pupal chambers were described using CT scans, 3D imaging techniques and SEM photographs and such traces contributed to the understanding of funerary practices (Huchet et al. 2013a), paleoecology and paleoclimatology (Martin and West 1995, Laudet and Antoine 2004). Pathologists and anthropologists examining more recent remains

are generally not familiar with such artefacts and, without the expertise of an entomologist, can misinterpret or ignore the information such remnants may provide.

**Dermestid Frass**

One unique attribute of dermestid larvae are their faecal residues, usually referred to as “frass”. Frass is a term given to insect excrements, or faeces, especially when they are dry in nature. Dermestid beetles excrete a light brown, stringy and powdery material which in large amounts can resemble sawdust, and when an abundant supply of food is present the faecal pellets are excreted in a bead-like chain (Hickin 1964, Byrd and Castner 2010a) (Fig 3, 4). The digestive track of dermestid beetles is lined with a peritrophic membrane, which functions to protect against abrasion as food passes through the digestive system (Bolognesi et al. 2008). Dermestid frass is essentially faecal material wrapped in a peritrophic membrane, which has a distinct appearance resembling pencil shavings (Tomberlin 2009).

The gross anatomy of the *D. maculatus* larval midgut has been described (Rahman et al. 1993) while a detailed histological and ultrastructural analysis of the digestive system including the identification and distribution of key digestive enzymes of *D. maculatus* has also been documented (Caldeira et al. 2007). The gut of larvae is composed of a short foregut, a large midgut, and a large hindgut (Caldeira et al. 2007). The food ingested by insects usually passes through the foregut and is then enclosed by the peritrophic membrane in the midgut. In *Dermestes species*, the food is digested at first by enzymes that penetrate into the endoperitrophic space (inside the peritrophic membrane), then by enzymes acting on diffused material in the ectoperitrophic space (between the peritrophic membrane and the midgut epithelium), and finally at the midgut cell surface (Caldeira et al. 2007).

The peritrophic membrane is a film that surrounds the food bolus in most insects. It is formed by a network of chitin and proteins (Caldeira et al. 2007). Since the insect midgut epithelium lacks a mucus coating, the peritrophic membrane is considered to be the analogous to that of the mucus that lubricates the mucosa, protecting against food abrasion and microorganisms (Caldeira et al. 2007,

Bolognesi et al. 2008). However, the peritrophic membrane also has specific functions depending on the fact that it compartmentalizes the midgut lumen into an endoperitrophic space (inside peritrophic membrane) and an ectoperitrophic space (space between peritrophic membrane and midgut epithelium) (Bolognesi et al. 2008). This functions to (1) prevent non-specific food binding onto the cell surface; (2) restrict oligomer hydrolases to the ectoperitrophic space in; and (3) prevent enzyme excretion by allowing enzyme recycling (Caldeira et al. 2007).

In respect to forensic investigations, frass is commonly present where human remains have reached an advanced state of decomposition and/or become mummified. Frass will often be present long after the beetle larvae have fed on the remains and completed development (Tomberlin 2009). As such, the occurrence of frass at a crime scene may provide additional information in the calculation of time since death because it is generally indicative of an extended minPMI (Byrd and Castner 2010a). Currently, most pathologists or medical examiners have limited knowledge about the occurrence or nature of dermestid frass and what it indicates when found at a death investigation. Where frass is documented in the literature, the information provided is limited to presence and absence observations. Generally this is accompanied by broad time frames of when frass occurs on human remains, which can range between 1 month and 10 years after death (Byrd and Castner 2010b). In a recent case in northern Italy, dermestid frass was observed on mummified remains concealed in an apartment for 18 years (P.A.M., unpublished data).

Given the potential for frass to persist long after insect life cycles are completed in association with remains further emphasis should be placed on its identification and collection from crime scenes. Following a death event, when frass is evident, a complete entomological assessment should be considered by a qualified forensic entomologist before attempting a minPMI determination (Voigt 1965, Wolf et al. 2006, Byrd and Castner 2010a). The presence of dermestid frass can only be viewed as an additional aid when estimating the time since death due to the inexact time frames that the literature documents. Nonetheless, forensic entomologists will continue to research this biological artefact as well as additional methods to quantify the minPMI in cases where many months, or even years, have elapsed.

**Recommended Collection Procedures**

When dermestids are located at a crime scene or on decomposing remains care should be taken to collect both alive and dead specimens. Sometime this distinction is not easy because adults prefer a dark environment showing a negative response to light (negative phototaxis) and will, when touched, readily “play dead” (thanatosis) (Gennard 2012). Appropriate safety procedures should be applied during collection as the minute barbed hairs (hastisetæ) and the slender elongate hairs (spicisetæ) of dermestid larvae have urticating proprieties and apparently can cause enteric problems. In addition, insect emanations such as scales, antennae, faeces and saliva are suspected as being source of sensitizing antigens that can produce allergic conditions (Patton 1931, Cuesta-Herranz et al. 1997, Goddard 2003). For any hypersensitive individual attending to the crime scene this can mean rhinitis, urticarial, ectzema and asthma (Goddard 2003). The symptoms experienced after ingesting dermestid larvae have been attributed to mechanical action of the hastisetæ and spicisetæ resulting in tissue damage or irritation in the alimentary tract. Clinical symptoms include diarrhoea, abdominal pain and perianal itch (Jupp 1956). Moreover, since beetles of the genus *Dermestes* feed on decomposing remains and hides, the possibility they may spread the bacilli or spores of anthrax has been raised (anthrax bacilli have been recovered from the faeces of a dermestid) (Bruesch 2011).

Care should also be used in sampling dermestid frass because they are fragile and can crumble very easily (Byrd and Castner 2010a). Lastly, and most importantly from a forensic perspective, care should be taken when collecting living dermestids as the adults have cannibalistic and predaceous habits consuming eggs, larvae and pupae and older larvae may eat exposed pupae.

Dermestids should be preserved in 80 % ethanol when collected for morphological analyses (Amendt et al. 2007). Numerous difficulties can arise when utilising traditional morphology methods for species identification, and as such DNA techniques are becoming more commonplace for this purpose (Magni et al. 2012). In such cases dermestids should be preserved in 100 % ethanol

(Magni et al. 2008). Dermestids and their remains can be also used for entomotoxicology analyses. Entomotoxicology studies the potential uses of insects for detecting drugs or other toxic substances that may otherwise not be measurable in decomposing tissues. Necrophagous insects, feeding on the decomposing remains, accumulate toxins present in their food substrate. These insects can sometimes provide a more reliable and sensitive result than from highly decomposed remains (Magni et al. 2014), and for such an analysis should be preserved at approximately -6 °C (Magni et al. 2008).

### Conclusion

Despite the common occurrence of dermestids dermestids and especially *Dermestes* species on decomposing remains, basic biological and behavioural data pertinent to forensic investigations are lacking or of limited application. Relatively few studies of decomposition and insect succession of remains have identified immature and adult specimens of dermestids beyond their taxonomic family, and consequently there are few succession records beyond reporting the presence of adults at a scene. Where species attending remains are identified, records of the timeframes of beetle arrival, oviposition and development are extremely limited and geographically specific. Here we have presented all the known literature relevant to forensic case work and identified areas for future research aimed at improving the information that may be provided by the family Dermestidae as an aspect of forensic evidence.

As discussed, the unpredictability of colonization timeframes often reported for forensically relevant Dermestidae is used to discount their potential as indicators of minPMI but this is largely a consequence of inadequate research. Research is urgently needed to further develop our understanding of the factors driving species-specific resource location by dermestids along with adequate documentation of species-specific arrival and oviposition timeframes on decomposing remains across geographic locations. Additionally, basic life history parameters, particularly lower developmental threshold and thermal parameters for forensically relevant species are needed. Such data is needed for identification and incorporation of the relevant factors affecting development

time into predictive models for larval aging. Finally, dermestid artefacts have considerable potential to provide forensic investigators with additional crime scene information. Unfortunately, such artefacts are frequently missed and, ignored or of limited value without further development of analysis approaches. Additionally, familiarity with dermestid artefacts, their collection and value as forms of evidence should be included in training packages for crime scene officers, pathologists and other law enforcement personnel involved in processing decomposing remains for forensic investigation.

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<b>DERMESTIDS</b>	<b>COUNTRY</b>	<b>SUCCESSION ON ANIMALS, PRESENCE IN FOOD AND OTHER MATERIALS</b>	<b>CASE STUDY (HUMAN REMAINS, MEDICAL CASE, ARCHAEOENTOMOLOGY)</b>	<b>DESCRIPTION AND ECOLOGY (FOOD PREFERENCE, PREDATORY HABITS, CANNIBALISM)</b>	<b>DEVELOPMENT (LIFE CYCLE, OPTIMUM AND LAB REARING)</b>	<b>OTHER</b>
Dermestids in general/ <i>Dermestes</i> sp.	USA, Canada, Africa, South America	(Hickin 1964, Wolff et al. 2001, Anderson 2010, Byrd and Castner 2010a, Bruesch 2011, Saunders 2011)	(Goddard 2003, Archer et al. 2005, Arnaldos et al. 2005, Anderson 2010, Byrd and Castner 2010a, Bruesch 2011)	(Munro 1966, Crowson 1967, Hinton and Corbet 1975, Braak 1987, VanLaereven 2010)	(Byrd and Castner 2010b, Byrd and Tomberlin 2010)	
Dermestes frass			(Miller et al. 1994, Archer et al. 2005, Wolf et al. 2006, Campobasso et al. 2009, Byrd and Castner 2010b)	(Byrd and Castner 2010b)		Toxicology (Miller et al. 1994, Byrd and Castner 2010b)



<i>Dermestes ater</i> (DeGeer) Synonymous with <i>Dermestes cadaverinus</i> Fabricius	France, USA, Argentina, Australia, Malaysia	(Fuller 1934, Hinton and Corbet 1975, Early and Goff 1986, Goff 1991, Avila and Goff 1998, Davis and Goff 2000, Shalaby et al. 2000, Oliva 2001, Centeno et al. 2002, Voss et al. 2008, Kelly et al. 2009, Kumara et al. 2009, Voss et al. 2009, Byrd and Castner 2010b, Bruesch 2011)	(Kumara et al. 2009, Charabidze et al. 2013)	(Munro 1966, Woodroffe and Coombs 1979, Menezes et al. 2005, Byrd and Castner 2010b, Bruesch 2011)	(Roth and Willis 1950, Coombs 1981)	Toxicology (Byrd and Castner 2010b)
<i>Dermestes bicolor</i> Fabricius	France		(Charabidze et al. 2013)			
<i>Dermestes caninus</i> (Germar)	USA except for the Pacific Northwest	(Reed 1958, Payne 1965, Payne and King 1972, Watson and Carlton 2003, Watson and Carlton 2005, Byrd and Castner 2010b)		(Byrd and Castner 2010b)	(Byrd and Castner 2010b)	

<i>Dermestes carnivorou r</i> Fabricius	Mexico		(Huchet et al. 2013b, Muñiz Vélez 2001)			
<i>Dermestes frischii</i> (Kugelann)	Canada, USA, Europe, Asia, Africa	(Early and Goff 1986, Anderson and VanLaerho ven 1996, Avila and Goff 1998, Shalaby et al. 2000, Centeno et al. 2002, Arnaldos et al. 2005, Matuszews ki et al. 2008, Özdemir and Sert 2009, Matuszews ki et al. 2010, Bruesch 2011, Prado e Castro 2011)	(Arnaldos et al. 2004, Charabidze et al. 2013)	(Munro 1966, Woodroffe and Coombs 1979, Bruesch 2011)	(Howe 1953, Howe 1965, Amos 1968, Bruesch 2011)	
<i>Dermestes haemorroid alis</i> Küster	France		(Charabidze et al. 2013)	(Woodroff e and Coombs 1979)		
<i>Dermestes intermedius</i> (Kalik)	Turkey	(Özdemir and Sert 2009)				

<i>Dermestes lardarius</i> (L.)	France, Vienna, Germany	(Grassberger and Frank 2004, Tomberlin and Talley 2010, Bruesch 2011, Saunders 2011)	(Benecke 2010, Charabidze et al. 2013)	(Kreyenbergr 1928, Herrick 1936, Hinton 1945, Hickin 1964, Munro 1966, Woodroffe and Coombs 1979, Jacob and Fleming 1980a, 1984, Fleming and Jacob 1986, Bruesch 2011, Weier 2011)	(Coombs 1978, Jacob and Fleming 1980a, Jacob and Fleming 1980b, Jacob and Fleming 1984, Byrd and Castner 2010b, Bruesch 2011)	
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<i>Dermestes maculatus</i> (DeGeer) Synonymus with <i>Dermestes vulpinus</i> Fabricius	France, Spain, Italy, USA, Argentina, Australia, Vienna, South Africa, India, Colombia, Canada, China	(Kreyenberg 1928, Fuller 1934, Early and Goff 1986, Goff 1991, Avila and Goff 1998, Davis and Goff 2000, Shalaby et al. 2000, Richardson and Goff 2001, Centeno et al. 2002, Grassberger and Frank 2004, Arnaldos et al. 2005, Sharanowski et al. 2008, Voss et al. 2008, Wang et al. 2008, Kelly et al. 2009, Segura et al. 2009, Voss et al. 2009, Tomberlin and Talley 2010, Bruesch 2011, Saunders 2011)	(Miller et al. 1994, Kulshrestha and Satpathy 2001, Schroeder et al. 2002, Arnaldos et al. 2004, Turchetto and Vanin 2004, Charabidze et al. 2013)	(Kreyenberg 1928, Hickin 1964, Munro 1966, Woodroffe and Coombs 1979, Braak 1987, Archer and Elgar 1998, 1999, Byrd and Castner 2010b, VanLaerven 2010, Bruesch 2011, Weier 2011)	(Smit 1931, Walker 1944, Russell 1947, Scoggin and Tauber 1949, Scoggin and Tauber 1951, Howe 1965, Archer and Elgar 1998, Richardson and Goff 2001, Bruesch 2011)	Antibacterial Activity (Barnes et al. 2010)  Toxicology (Miller et al. 1994)
<i>Dermestes marmoratus</i> (Say)	USA	(Payne 1965, Payne and King 1972, De Jong and Hoback 2006)		(Bruesch 2011)		

<i>Dermestes murinus</i> (L.)	Poland	(Matuszewski et al. 2008, 2010)				
<i>Dermestes olivieri</i> (Lepesme)	Turkey	(Özdemir and Sert 2009)				
<i>Dermestes peruvianus</i> (Castelnau)	France, Argentina, South America, Sweden	(Oliva 2001, Bruesch 2011, Saunders 2011)	(Charabidze et al. 2013)	(Woodroffe and Coombs 1979, Bruesch 2011)		
<i>Dermestes undulatus</i> (Brahm)	France, Turkey, USA	(Shean et al. 1993, Özdemir and Sert 2009)	(Charabidze et al. 2013)			

908 Fig. 1. Mummified corpse found in a city apartment in Turin, North of Italy. Irregular holes are  
909 visible over the surface of the remaining skin. Active insects and their remains (puparia, dermestid  
910 frass, cast skins) are also visible on the body.

911





912 Fig. 2 Mummified corpse found in a city apartment in Turin, North of Italy. Particular of the head  
913 where active insects and their remains are present.



914 Fig. 3. Mummified corpse found in a city apartment in Turin, North of Italy. Particular of  
915 dermestid frass associated with the feet.  
916





917 Fig. 4 Mummified corpse found in a city apartment in Turin, North of Italy. Particular of dermestid  
918 frass associated with the lateral view of the right leg.

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Fig. 5. Larval exuviae of *Dermestes* species (a) collected on a mummified corpse (b) from a female found in a Coptic grave (V-VI century A.D.) during an archaeological excavation in Antinopolis (Sheikh 'Ibada), Egypt.

