MEDICAL AND VETERINARY ENTOMOLOGY

MYIASIS AND FORENSIC ENTOMOLOGY Assoc. Prof. Marija lvković marija.ivkovic@biol.pmf.hr

- Myiasis Invasion of living tissue of vertebrates with fly larvae
- Families of flies implicated in Myiasis Calliphoridae (Blowfly), Oestridae (Botfly), Sarcophagidae (Fleshfly),









- Myiasis Invasion of living tissue of vertebrates with fly larvae
 - Most often characterized with a place of entry:
 - Gastrointestinal myases
 - Urogenital myases
 - Ocular or Ophthalmic myases
 - Auricular myiasis
 - Nasopharyngeal myases
 - Cutaneous myases (with an opening they are called Traumatic myases)







- Myiasis Invasion of living tissue of vertebrates with fly larvae
- RANDOM When eggs or larvae of a fly are eaten, most often only digestive disorders – finding larvae in children's feces - most often they are not "real" myiasis
- FACULTATIVE Includes larvae that can (and often do) exploit living tissue, but may also complete their development on corpses and carrion or decaying organic matter (most commonly mild infections) – Most cutaneous myiasis of humans by Calliphoridae and Sarcophagidae where larvae develop in open wounds and ulcers
- OBLIGATORY Includes larvae that ALWAYS require living tissue, i.e. a living host, to complete their development (often strong infections and very invasive)



- Myiasis Invasion of living tissue of vertebrates with fly larvae
- FAKULTATIVE







- Myiasis Invasion of living tissue of vertebrates with fly larvae
- Maggot therapy controlled therapy that uses sterile fly larvae that feed exclusively on necrotic tissue and secrete allantoin (promotes wound healing)
- Used in Mayan culture, Aboriginal people, Northern Myanmar people,...
- 3 species are used: Lucilia sericata, Lucilia illustris and Phormia regina





- Myiasis Invasion of living tissue of vertebrates with fly larvae
- CALLIPHORIDAE "Screwworms" genera Cochliomyia i Chrysomya obligate miases
- Pregnant females attracted to fresh wounds, organic secretions or exposed mucosa where eggs are laid
- Larvae feed on healthy tissue of domestic animals and humans





- Myiasis Invasion of living tissue of vertebrates with fly larvae
- Ivermectin treatment in destructive rhino-orbital myiasis



The release of sterile males is used to reduce/exterminate these flies in the southern US and Mexico, but it remains a major problem in Central and South America.





Cochliomyia hominivorax

MYIASIS

- Myiasis Invasion of living tissue of vertebrates with fly larvae
- OESTRIDAE (Botflies) 4 subfamilies (Cuterebrinae (New World) – cutaneous, Hypodermatinae (Old World) – cutaneous, Oestrinae (Old World, origin Africa) – nasal, Gasterophilinae (Old World, Africa) – gastric







- Myiasis Invasion of living tissue of vertebrates with fly larvae
- OESTRIDAE Torsalo (*Dermatobia hominis*)
- Common in Central and South America, most often a pest on cattle
- Eggs are laid on an "intermediary" (mosquitoes, various types of flies), larvae hatch when the intermediary feeds on vertebrates - PHORESY
- The larva enters the skin and forms a small blister on the skin, within which it grows (Furuncular myiasis)





- Myiasis Invasion of living tissue of vertebrates with fly larvae
- OESTRIDAE Torsalo (Dermatobia hominis)





- Myiasis Invasion of living tissue of vertebrates with fly larvae
- OESTRIDAE *Cuterebra* spp.
- Most often on rodents, cats and dogs









Scholl i sur. 2019

- Myiasis Invasion of living tissue of vertebrates with fly larvae
- OESTRIDAE Oestrus ovis
- Nasal botfly of sheep













- Myiasis Invasion of living tissue of vertebrates with fly larvae TREATMENT AND PREVENTION
- The treatment is different it depends on the place of myiasis, the number of larvae, ...
 - Manual/Surgical removal of larvae
 - Surgical removal of the entire tissue affected by the larvae
 - Antibiotics are given to prevent secondary infections (although these are rare)
 - Ivermectin
 - Purgatives in the case of intestinal myiasis
- Prevention
 - Insect repellents
 - Maintaining hygiene
 - Detailed attention to children, elderly and comatose patients
 - Wearing clothes
 - Prophylaxis only in cattle (Ivermectin or something similar)

- Class Insecta Order Diptera Superfamily Hippoboscoidea
- The superfamily contains the following families: Hippoboscidae, Streblidae, Nycteribiidae and Glossinidae
- With the exception of the Glossinidae family, the other families do not feed on humans and do not transmit diseases to humans
- Most often, they are ectoparasites on bats, domestic animals and birds



- Class Insecta Order Diptera Superfamily Hippoboscoidea
- The superfamily contains the families: Hippoboscidae, Streblidae, Nycteribiidae and Glossinidae - all feed on blood
- Most often, they are ectoparasites on bats, domestic animals and birds
- All members of this superfamily have a "uterus" and "mammary glands" and only one larva that leaves the female in the 3rd stage (the pupa - because it resembles a pupa)
- Most often, they are dorsoventrally flattened, and sometimes representatives of the Nycterbiidae are laterally, so they resemble fleas





- Class Insecta Order Diptera Superfamily Hippoboscoidea
- The families Streblidae and Nycteribiidae are exclusively ectoparasites on bats
- Most adults lose their wings or have very reduced wings, but retain their halteres
- Nycteribiidae often look like spiders, so they are also called "spider-like bat fly"







Nycteribiidae

Reeves & Lloyd 2019

HIPPOBOSCOIDEA



- Class Insecta Order Diptera Superfamily Hippoboscoidea
- Family Hippoboscidae they are primarily ectoparasites on birds and mammals - morphological adaptations of the legs to feathers or hair
- In some species, after settling on the host, the wings are torn off at the base (*Lipoptena*, *Neolipoptena*) and the histolysis of the flight muscles occurs



HIPPOBOSCOIDEA

- Class Insecta Order Diptera Family Hippoboscidae
 - Melophagus ovinus a wingless ectoparasite that lives its entire life on sheep
 - Present worldwide except in the tropics
 - The pupae stick to the sheep's wool
 - It can cause dermatitis and bites can be painful, depending on the person and location







HIPPOBOSCOIDEA





- Class Insecta Order Diptera Family Hippoboscidae
 - Hippobosca equina ectoparasite of the Equidae family Europe, Asia, North Africa







- Class Insecta Order Diptera Family Hippoboscidae
 - Lipoptena cervi primarily a parasite on deer in Europe, Siberia and North China
 - Mass emergences of adults appear in early autumn (Russia) then people may be bitten and dermatitis may appear (?) - severe itching
 - It can transmit the bacterium *Bartonella schoenbuchensis* a papule on the skin that can be present for up to a year





- Class Insecta Order Diptera Family Hippoboscidae
 - They can transmit some pathogens to animals, such as West Nile Virus, vectors of the facultative protist *Haemoproteus* that cause avian malaria, the filarial worm *Acanthocheilonema dracunculoides* to dogs
 - They transmit to sheep *Trypanosoma melophagium* the vector is *Melophagus ovinus*
 - And some other diseases caused by *Trypanosoma* in other mammals and birds





 Application of the science of insects and other groups of Arthropoda in legal research



- 3 main areas:
 - Entomology of stored food







Urban entomology









FORENSIC ENTOMOLOGY

Medically significant Arthropode

- 3 main areas:
- Entomology of stored food
- Urban entomology
- Medico-legal entomology (Forensic entomology)
- Insects and other arthropods involved in murders, rapes, suicides, physical abuse, neglect, suspicious death, etc...

• HOW?

- Determination of post-mortem interval (PMI)
- Evidence that the body was moved, transported or hidden
- Evidence of neglect
- Evidence of drug use
- Evidence of rape





- Determination of post-mortem interval (PMI)
- The time that has passed since the moment of death
- If you know how long it takes necrophagous insects found on the body to develop to a certain life stage under certain conditions (temperature, humidity, presence of light, etc.), then you can give an estimate of the minimum time that has passed since the moment of death
- This time from the time of death is generally considered to be equal to or longer than the post-infestation interval (PII) unless... there is no evidence of myiasis



Krinsky 2019

Medically significant Arthropoda

- Determination of post-mortem interval (PMI)
- To accurately determine the post-mortem interval, you need:
- The collected material should be accurately identified
- The successional position of the collected species should be known
- Know the development time of each stage in the life cycle of the collected species (how many days under certain conditions)
- Be aware of factors (physico-chemical) that affect growth and decomposition
- Flies (especially larval stages) are the most important organisms involved in determining the post-mortem interval

Why accurate identification is important?





FORENSIC ENTOMOLOGY



- **Determination of post-mortem interval (PMI)**
- **Different types of insects:**
- They live in different parts of the world
- The development of individual life stages is different
- They visit corpses at different times of decomposition
- The collected material should be accurately identified
- The successional position of the collected species should be known
- Know the development time of each stage in the life cycle of the collected species (how many days under certain conditions)
- Be aware of factors (physico-chemical) that affect growth and decomposition





- Determination of post-mortem interval (PMI)
- The collected material should be accurately identified
- Morphological features
- 1st larval stage anterior vents absent, posterior vents with 2 slits
- 2nd larval stage anterior vents present, posterior vents with 2 slits
- 3rd larval stage anterior vents present, posterior vents with 3 slits





- Determination of post-mortem interval (PMI)
- The collected material should be accurately identified
- Larvae of closely related species are very difficult to identify to species what can we do?
- Grow to adults because adults are easier to identify
- DNA typing If equipment is available, the DNA of the sample can be compared to known species of confirmed identification (database)





FORENSIC ENTOMOLOGY

- Determination of post-mortem interval (PMI)
- The successional position of the collected species should be known



Fig. 4 Arthropod succession on dog carcases in the USA (Tennessee). After Kauppala (*in* Nuorteva, 1977) from data presented by Reed (1958).





- Determination of post-mortem interval (PMI)
- Succession of insects



Dan 1

- Determination of post-mortem interval (PMI)
- Succession of insects



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DAY 18

- Determination of post-mortem interval (PMI)
- Succession of insects



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- Succession of insects





Time

- **Determination of post-mortem interval (PMI)** •
- **Succession of insects**

Calliphoridae (Zujare)]	
Muscidae (Muhe)			
Sarcophagidae (Mesaruše)			
Razl	ičite porodice s	kupine Acalyptratae	
	Piophilidae (Skočne muhe)	
Staphylinidae (Kusokril	lci)		
Silphidae (Strvina	ari)		
		Dermestidae (Kožo	niedi)



- **Determination of post-mortem interval (Plvi)**
- **Succession of insects Community on carcasses**
- Necrophagous species Mostly Diptera and Coleoptera only groups that significantly contribute to the decomposition of corpses/carrion
- Predators/parasites of necrophagous species (necrophilic species) various types of dipterans and beetles, wasps, ants,...
- **Omnivorous species different species of wasps, beetles and ants**
- Adventitious species springtails (Collembola) and spiders





- **Determination of post-mortem interval (Plvi)**
- Succession of insects Necrophagous Diptera
- Calliphoridae (Blowflies) among the first visitors to the carcass











Phormia regina



- **Determination of post-mortem interval (Plvi)**
- Succession of insects Necrophagous Diptera
- Calliphoridae (Blowflies) among the first visitors to the carcass

Lucilia sericata









- Determination of post-mortem interval (Plvi)
- Succession of insects Necrophagous Diptera
- Sarcophagidae (Fleshflies) They often come after blowflies on the carcass, but can outrun them in crowded situations
- There is no egg stage, this family of Diptera is larviparous







- **Determination of post-mortem interval (PMI)**
- Succession of insects Necrophagous Diptera
- Muscidae (True flies) more often attracted to excretions and urine than to carcasses
- **Common on bodies in closed spaces**



Hydrotaea (Ophyra) leucostoma



Musca domestica



- Determination of post-mortem interval (PMI)
- Succession of insects Necrophagous Diptera
- Different families of the Acalyptratae group often come late to the carcass



- Determination of post-mortem interval (PMI)
- Succession of insects Necrophagous Coleoptera
- Staphylinidae (Rove beetle) Adults and larvae of some species are important predators of fly larvae and eggs
- Often the first beetles to come to the carcass



Creophiulus maxillosius

- Determination of post-mortem interval (PMI)
- Succession of insects Necrophagous Coleoptera
- Silphidae (Carrion beetle) necrophages and/or predators
- Different species prefer different levels of carcass decomposition



- Determination of post-mortem interval (PMI)
- Succession of insects Necrophagous Coleoptera
- Dermestidae (Skin beetle) Adults and larvae are "scavengers" and some species feed on decaying/dry skin and tissue
- They are often used to clean old bones and carcasses





Dermestes maculatus



- Determination of post-mortem interval (PMI)
- To know the development time of each stage in the life cycle of the collected species (how many days under certain conditions)
 - The life cycle of 11 species of blowflies and fleshflies grown at 27°C and 50% relative humidity

Species	Egg (hrs)	1 st instar (hrs)	2 nd instar (hrs)	3 rd instar (hrs)	Prepupa (hrs)	Pupa (days)	Total dev time (hrs)
Sarcophaga cooleyi		24	18	48	96	9	402
Sarcophaga shermani		22	16	48	104	8	382
Sarcophaga bullata		26	18	54	112	12	498
Phormia regina	16	18	11	36	84	6	309
Protophormia terranovae	15	17	11	34	80	6	301
Lucilia (= Phaenicia) sericata	18	20	12	40	90	7	348
Eucalliphora lilaea	22	22	14	36	92	6	330
Cynomyopsis cadaverina	19	20	16	72	96	9	439
Calliphora vomitoria	26	24	48	60	360	14	854
Calliphora vicina	24	24	20	48	128	11	508
Calliphora terranovae	25	28	22	44	144	12	551

- Determination of post-mortem interval (PMI)
- To know the development time of each stage in the life cycle of the collected species (how many days under certain conditions)
 - ADH (Accumulated degree hour) Time required to reach a certain developmental stage at a certain temperature (Standard technique used to estimate the rate of insect development during a time period with temperature compensation (number of hours x °C)
 - If ADH and temperature in degrees Celsius are known - the age of the larvae can be determined

Prosječno minimalno trajanje razvojnih stadija i ADH za vrstu Lucilia sericata

	22 Degrees C			
	Hours	ADH at End of Stadium		
Egg	23	506		
1st Instar	27	1100		
2nd Instar	22	1584		
Feeding 3rd Instar	22	2068		
Postfeeding 3rd Instar	108	4444		
Pupa	143	7590		
total	345 (14.4 days)	7590		





FORENSIC ENTOMOLOGY

- Determination of post-mortem interval (PMI)
- To know the development time of each stage in the life cycle of the collected species (how many days under certain conditions)
- IT IS ONLY VALID IF THE RELATIONSHIP BETWEEN SPECIES DEVELOPMENT TIME AND TEMPERATURE IS LINEAR!!!!

	22 Degrees C		29 Degrees C			
	Hours	ADH at End of Stadium	Hours (predicted)	Hours (observed)	ADH (observed)	
Egg	23	506	17,5	18	522	
1st Instar	27	1100	20,4	16	986	
2nd Instar	22	1584	16,7	16	1450	
Feeding 3rd Instar	22	2068	16,7	22	2088	
Postfeeding 3rd Instar	108	4444	81,9	94	4814	
Pupa	143	7590	108,5	130	8584	
total	345 (14.4 days)	7590	261.7 (10.0 days)	296 (12.3 days)	8584	



- Determination of post-mortem interval (PMI)
- To know the development time of each stage in the life cycle of the collected species (how many days under certain conditions)
- IT IS ONLY VALID IF THE RELATIONSHIP BETWEEN SPECIES DEVELOPMENT TIME AND TEMPERATURE IS LINEAR!!!!

22 Degrees C

29 Degrees C

- The predicted development time at 29°C (extrapolation based on ADH measured at 22°C) was 10.8 days to complete development. The actual value was 12.3 days.
- Most cultivation experiments are carried out at the same temperature, but in natural conditions it fluctuates much more.
- Fluctuations affect some species significantly more than others and different models are designed to incorporate this.
- Although, the mean temperature values during 24 h are usually acceptable if the fluctuations remain within a narrow range.

i upa	140	1000	100,0	100	0004	
total	345 (14.4 days)	7590	261.7 (10.0 days)	296 (12.3 days)	8584	



- Determination of post-mortem interval (PMI)
- Be aware of factors (physico-chemical) that affect growth and decomposition
- Factors affecting growth:
- Temperature The most important factor affecting growth rate, in cold temperatures the growth rate will be significantly reduced and diapause may be induced
- Food Availability Larvae that are starved will develop at the same rate as others, but will be significantly smaller
- Medicines, Drugs and Chemicals Cocaine and Ecstasy will speed up the growth rate while an insecticide like Malathion will slow down the growth

rate





- Determination of post-mortem interval (PMI)
- Be aware of factors (physico-chemical) that affect growth and decomposition
- Factors influencing the colonization of insects:
- Location of the body/carcass Geographical location, indoor or outdoor, hay or sun, forest or field,
- Temperature Many species are inactive below a certain temperature and will not lay eggs
- Dressing or body wrapping Can delay colonization or exclude some species, can also affect local carcass conditions (T°, humidity, etc....)
- Burial or drowning Even a few cm of soil or water will significantly prevent or affect insect colonization of the carcass
- Body movement Changing habitat after death will have an impact on the insect fauna (eg. in the trunk of a car 2 days before it was dumped in the forest)
- Medicines, drugs and chemicals Consumption of chemicals (eg. insecticides) can affect or even delay the colonization of the carcass

- Forensic entomology
- Determination of the post-mortem interval
- Evidence that the body was moved Larvae and pupae may be found where the body was before it was removed, also the lack of early colonizers on the exposed body may mean that the carcass was hidden in an environment where insects could not get to before it was removed and was exposed to air
- Evidence of abuse and neglect Some species of flies (most commonly Calliphoridae) will inhabit the body while it is still alive (myiasis), the larvae will feed on excrement, fresh or decaying tissue
- Evidence of "drugs" Drug testing on highly decomposed tissue usually gives poor results. Feeding on tissue, the larvae accumulate various substances in their tissues and enable the detection of even very small amounts of drugs
- Identification of the rapist DNA extracted from a blood meal from a pubic louse of a rape victim enabled confirmation of the suspect

FORENSIC ENTOMOLOGY

COLLECTING INSECTS FOR FORENSIC INVESTIGATIONS



• Forensic entomology – History

- The earliest crime, for which insects are known to have offered evidence, took place in China between 907 and 960 - flies landed on part of the head of a murdered man, thereby showing where the fatal blow had been delivered
- The first described crime where insects (flies) exposed the killer was described by Sun Tzu (The Art of War) in 1247 in China a Chinese farmer was killed with a sickle, during the investigation all farmers had to put their sickles on the ground (70 to 80 sickles), in a short time flies came to just one sickle and thereby incriminated the owner of the sickle for murder

THE ART OF WAR



SUN TZU

