COLLECTION AND PRESERVATION OF INSECTS
Objective

After class, students are able to describe...
- methods for collecting and preserving of insects
Collecting

1. Habitats and niches (breeding sites)
2. Timing and seasonality (diurnal, nocturnal/hot, rainy or dry season)
3. Biology and behaviour (life cycle/animal or human-biting insects)
4. Permit requirements (e.g. National Park)

Upton and Mantle (2010)
Breeding habitat

*Anopheles* spp. (larval stage)

*Simulium* spp. (larval stage)

Black flies (แมลงชีวิตดำ)
Timing and seasonality

Black fly collection (adult female) during 7-9 a.m.
Biology

Life cycle

Mosquitoes

Adult

Egg

Larva (Nymph & naiad)

Pupa

Dragonfly Nymph

Dragonfly Emerging from Nymph Stage

Adult Dragonfly
Behaviour

Anthropophilic
(human-biting mosquitoes)

Zoophilic
(animal-biting mosquitoes)

Original photograph from Dr. Petchaboon Poolphol,
Office of Disease Prevention and Control No. 10th,
Department of Disease Control, Ministry of Public Health,
Ubon Ratchathani, Thailand
Collecting Methods and Equipment

1. Land insects

**Arial net:** light nets for capture of insects in flight
(flies, mosquitoes, butterflies, dragonflies, wasps, bees)

**Sweep net:** heavy, canvas bag with heavy mounting ring for net, large numbers of insects found in grass and plant foliage
(small bugs, beetles, spiders)
- Aspirator: small insects (mosquitoes, biting midges, black flies)
Glass vial aspirator

Mouth aspirator with HEPA filter
- Sifting soil and leaf litter: spring tail, beetles
- **Berlese (Tullgren) funnel (extraction device):** mites
▪ Trap: adult insects

Flight-interception traps: malaise trap and window trap

Malaise trap (e.g. black flies)
Window trap: heavy bodied insects (e.g. beetles)

Upton and Mantle (2010)
Sticky trap: bugs, bees, flies
Bait trap: blow flies

Klong-klaew et al. (2017)
Sontigun et al. (2018)

Sanit et al. (2018)
Baited pitfall trap: ground-dwelling insects (e.g. ground beetles)

https://en.wikipedia.org/wiki/Pitfall_trap
Light trap and sheet: night-flying insects

Upton and Mantle (2010)
Light traps (with CO₂): mosquitoes, biting midges, sand flies

Miniature CDC Light Trap with UV Light

CDC mini light trap with incandescent light

https://www.bioquipinc.com
2. Aquatic insects

- Hands, forceps: larvae/pupae of black flies
- Dip net: larvae of mosquitoes, mayflies, naiad of dragonfly
- White (plastic) tray: larvae of mosquitoes

- Pipette/sucking tube: larvae of mosquitoes
Killing Methods

- Freezer
  - small moths, butterflies (kill immediately prior to pinning and spreading)

- Killing bottle/tube: adult stage of most insects
  - Size of the bottle depend on the type of insect
  - Wide-mounted and made of glass
    (polypropylene or polythene jars can be used)
  - Clean bottle and avoiding from direct sunlight
  - Awareness of delicate specimen
  - Labeled POISON and keep out of reach of children
  - Handled with great care and read the chemical MSDS
Killing bottle

Cork

Absorbent paper

Cotton wool/sawdust

Plaster & killing agent (e.g. ethyl acetate)
Use of liquid for killing and fixing

- 70-95% ethyl alcohol
- Hood’s solution (70-80% ethyl alcohol, glycerin)

1. Kahle’s solution
   - 95% ethyl alcohol
   - distilled water
   - formaldehyde
   - glacial acetic acid

2. Pampel’s solution
   - 95% ethyl alcohol
   - distilled water
   - 40% formalin
   - glacial acetic acid

3. Alcoholic Bouin’s solution
   - 80% ethyl alcohol
   - formaldehyde
   - glacial acetic acid
   - picric acid

Millar et al. (2000)
Larvae/nymph (naiad)

- 70-80°C hot water (e.g. mosquito larvae)
- 70-95% ethyl alcohol
- KAAD mixture
  - 95% ethyl alcohol
  - kerosene
  - glacial acetic acid
  - dioxane
- XA mixture
  - 95% ethyl alcohol
  - xylene
Preservation

- Temporary storage
  - Envelope: butterflies, dragonflies, stone flies

- Permanent storage
  - Dry preservation
  - Wet preservation
  - Microscope slide
- Dry preservation
  - Relaxing methods: dishes, boxes, Barber’s relaxing fluid

Barber’s fluid
- 95% ethanol
- Distilled water
- Ethyl acetate
- Benzene

Millar et al. (2000)
- Pinning insects
  - entomological pins
  - forceps

Pin positioning for 8 orders
Wing arrangement for pinning insects

**BUTTERFLIES & MOTHS**
Hind margin of the front wing at right angles to the body. Rear wing arranged so only a small notch between front and rear wings.

**DRAGONFLIES, WASPS, LACEWINGS**
Front margin of the rear wings at right angles to the body. Front wings pushed forward to clear edge of rear wing. In wasps front and rear wings may remain hooked together.

**GRASSHOPPERS, COCKROACHES, STICK INSECTS, MANTIDS**
Set only the wings on left side. Front margin of rear wing at right angles to the body. Front wing pushed forward to clear edge of rear wing.

**FLIES**
Set the wings slightly forward of right angles to the body.

**BEETLES AND BUGS**
Do not spread the wings. Pin through the front half of right wing so pin emerges between mid- and hind legs.

Mounting beetle on a card platform

Millar et al. (2000)
Pinning adults in Simuliidae

Hernández (2008)
Spreading board

Insect braced on mounting board
Drying

- Room temperature
- Drying box: 100 Watt lamp
- Incubator: 40-50°C
Preserving box

- Creosote
- P-dichlorobenzene
Wet preservation

- Following fixation, the most commonly used preservative is ethyl alcohol in various concentrations.
- Aphids and scale insects be preserved in lactic-alcohol, a mixture of 2 parts 95% ethyl alcohol + 1 part 75% lactic acid.
- 80-95% ethyl alcohol for molecular studies
- Isopropanol can be applied for preserving
- Stored with a data label in a separate glass vial
- Sealed vial to prevent evaporation

Upton and Mantle (2010)
Preserving jar

Storing small tubes containing specimens
Microscope slide
- Hoyer’s media, Euparol, Canada balsam
Labelling

- Pinning insects: white cards
- Insects preserved in fluid: white paper & pencil
- Mounting slide
Mounting slide of mosquito larva

THAILAND
Chumphon. 06.V.2018
Ban Kon Thae
Coll: Name
HBT: water jar
Slide#F-8204

Culex quinquefasciatus
Det. Name
Pinning mosquito adult

THAILAND:
A2 BCLp12(1)-21 ♀
Lampang Province,
Ko Kha District,
Ban Don Thum.

Human-baited traps
23.XI.2013
Coll: Choochote et al.

Anopheles (Anopheles) saeungae
Det. Taai & Harbach 2014
Storage Cabinet

Original photograph from Dr. Wichai Srisuka
Entomology Section, Queen Sirikit Botanic Garden
Chiang Mai, Thailand
Preferred methods of preserving insects and arachnids

- **Alderflies**: set, both wings spread
- **Antlions and Lacewings**: set, both wings spread
- **Ants**: preserved wet; card platforms
- **Aphids**: preserved wet; microscope slides
- **Armoured scale insects**: preserved dry (or wet); microscope slides
- **Bees**: larger than 8 mm: pinned
  - smaller than 8 mm: glued to pin
- **Bedbugs**: preserved wet; microscope slides
- **Beetles**: larger than 8 mm: pinned
  - smaller than 8 mm: card platforms
- **Booklice**: preserved wet; microscope slides
- **Bristletails**: preserved wet; microscope slides
- **Bugs**: larger than 8 mm: pinned
  - smaller than 8 mm: card points
- **Butterflies**: set, both wings spread

Millar et al. (2000)
Caddisflies: pinned
Cockroaches: pinned
Crickets: pinned
Dragonflies and Damselflies: set, both wings spread
Earwigs: pinned; card platforms
Eggs: preserved wet
Fishmoths: preserved wet
Fleas: preserved wet; microscope slides
Flies: larger than 8 mm: pinned
                      smaller than 8 mm: minutent pins
Grasshoppers and Locusts: pinned, one wing spread
Harvestmen: preserved wet
Larvae: preserved wet
Lice: preserved wet; microscope slides
Mayflies: preserved wet
Mealybugs: preserved wet; microscope slides
Mites: microscope slides; preserved wet
Moths: larger than 8 mm: set, both wings spread
                 smaller than 8 mm: minutent pins
Nymphs: preserved wet
Parasitic wasps: card platforms; card points; microscope slides
Praying mantids: pinned
Pseudoscorpions: preserved wet
Schizomida: preserved wet
Scorpionflies: set, both wings spread
Scorpions: preserved wet
Soft scale insects: preserved wet; microscope slides
Spiders: preserved wet
Stick insects: pinned
Stoneflies: preserved wet
Sun-spiders: preserved wet
Termites: preserved wet
Thrips: preserved wet; microscope slides
Ticks: preserved wet
Wasps: larger than 8 mm: pinned
                      smaller than 8 mm: glued to pin
Whip-spiders: preserved wet
Additional Readings

