Biomedical Parasitology

Arthropod control

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Background

- Most novel chemical compounds are developed for agricultural pest control, not for ectoparasites
- Insecticides lead to resistance (it is an arms race!)
- Understanding chemical spectrum of activity, mode of action, resistance mechanisms, is essential for effective chemical control

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History

- 1867: first scientifically-made insecticide developed (arsenical Paris green)
- 1874: DDT (dichlorodiphenyltrichloroethane) synthesized
- 1939: Muller discovered insecticidal properties of DDT
- 1943: DDT used to control louse-borne typhus in Naples
- 1948: Muller awarded the Nobel Prize

- 1950s: Organophosphates
- 1960s: Carbamates
- 1970s: Pyrethroids
- 1980s: Insect growth regulators

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Major groups of insecticides

- Insecticide (acaricide, arthropodicide)

- Organochlorines (e.g. DDT)
- Organophosphates (OP; e.g. malathion)
- Carbamates (e.g. carbaryl)
- Pyrethroids (e.g. permethrin)
- Insect growth regulators (IGRs; e.g. buprofezin)
- Bacterial toxins (*Bacillus* toxins)

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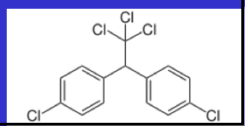
Organochlorines

- Containing chlorine
- Target nerve system: sodium channels, GABA receptors (gamma-aminobutyric acid)
- DDT: opens sodium channels in neurons, leading to continuous, uncontrolled muscle movement & eventually death
- Simple structure, cheap
- Persistent in environment, wildlife & humans
- Reduced use since 1970s

DDT



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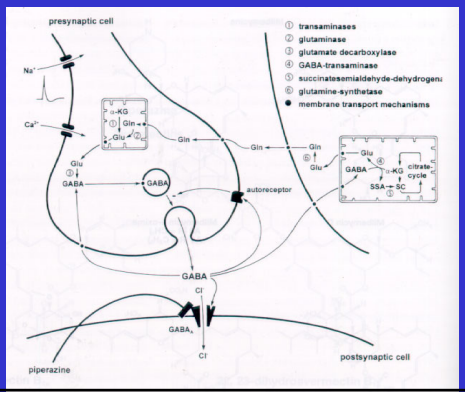
Interfere with GABA neurotransmitters



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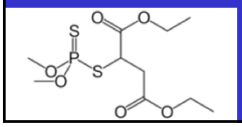
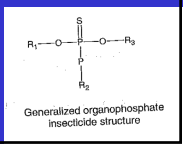
Organophosphates

- Containing phosphorus
- Bind acetylcholinesterase (AChE) at nerve junction
- Once bound, AChE cannot remove acetylcholine (ACh)
- Ach continues stimulating muscle movement, leading to paralysis & death
- Less stable than organochlorines but does not accumulate in animal body (less toxic to humans)



malathion



Generalized organophosphate insecticide structure

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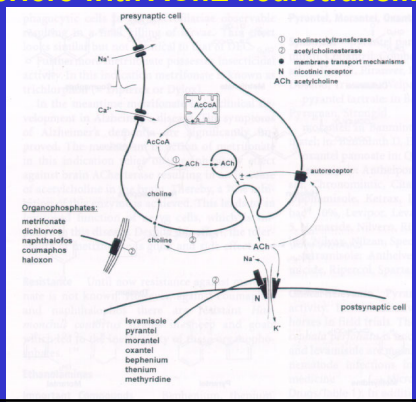
Interfere with AChE neurotransmitters



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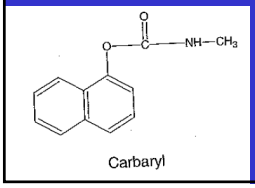
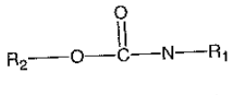
Carbamates

- Derived from carbamic acid (NH_2COOH)
- Same mode of action as OP - target AChE
- Unstable (breaks down in environment in weeks/months)
- Low toxicity to humans and livestock



Carbaryl



Generalized carbamate structure

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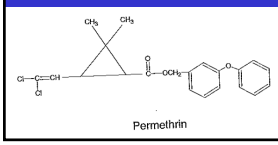
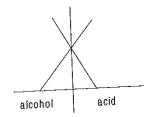
Pyrethroids

- Developed from pyrethrum flowers
- Same mode of action as DDT (target sodium channels)
- Unstable (safe to humans and livestock)
- Most successful commercially



Permethrin



Generalized pyrethroid structure

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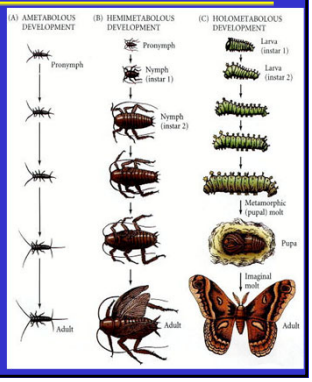
Insect growth regulators

- Target hormonal systems that control molting & metamorphosis
- Low toxicity to mammals
- Highly species specific
- Take long time to kill
- High cost



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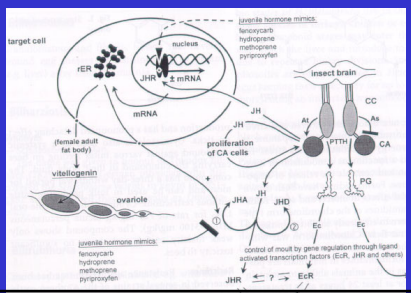
Arthropodicidal drugs

1980's	- insect growth regulator (cyromazine)	disrupt cuticle
	- " " (benzoylphenylureas)	inhibit cuticle
	- " " (juvenoids)	mimic juvenile h



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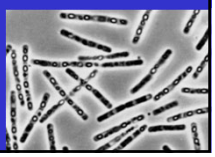
Bacillus toxins

- Enzymes (Cry toxins) secreted by *Bacillus thuringiensis*
- Bind to receptors on surface of midgut epithelial cells, break cells & kill insects
- Active against moths, flies, wasps, beetles, nematodes
- Non-toxic to vertebrates & other arthropods
- Environmentally safe; used since 1938
  - Genetically modified plants contain Bt-toxin gene (corn, maize, cotton, peanut, potato, tobacco, etc)



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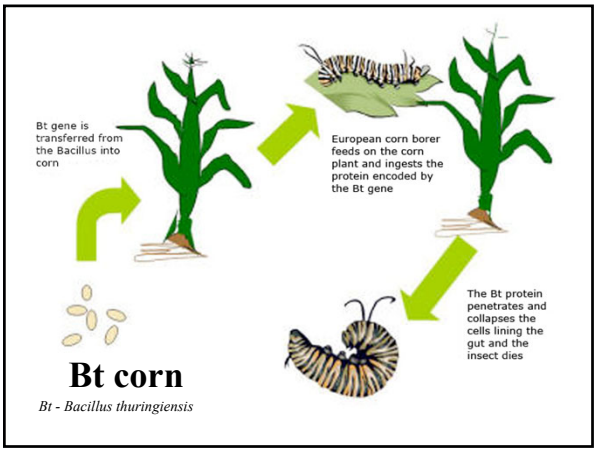

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Bt cotton

A 12-day-old cotton bollworm larva raised on a diet with no Bt. Source: USDA

A 12-day-old cotton bollworm larva raised on a diet containing Bt proteins. Source: USDA

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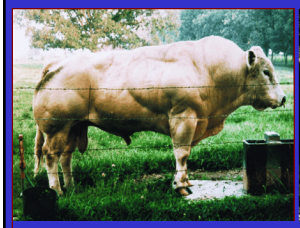
Bt sheep/cattle?



control flystrike, grubs, lice, etc?

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Treatment applications

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Formulations

- Dry formulations
  - Dusts
  - Granules
  - Wettable powders
  - Soluble powders
  - Baits
- Liquid formulations
  - Emulsifiable concentrates
  - Solutions
  - Flowables
  - Aerosols

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PROBLEMS

Emergence of drug resistance due to:
- under-dosing (sublethal doses)
- poor compliance (treatment not completed)

Resistance found against:
- antimalarials (chloroquine)
- anticoccidials (ionophores, sulfonamides)
- anthelmintics (white/clear drenches)
- insecticides (DDT, organophosphates)

Need to understand mode of action of drug

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Resistance

- WHO definition: "the development of an ability in a strain of some organism to tolerate doses of a toxicant that would prove lethal to a majority of individuals in a normal population of the same species."
- Insecticides kill susceptible individuals but select the resistant individuals
- Resistance is inheritable and has a genetic basis
- Mutations - phenotype changes - detoxification ↑ - sensitivity ↓ - survivability ↑ - more offspring

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Detection of resistance

Dose-response bioassay

- Treat parasite samples with insecticides in a range of doses (at least 5)
- Measure mortality at each dose
- Plot dose against mortality - sigmoid curve
- Plot log dose against probit mortality - straight line
- Establish dose required to kill a given % of treated individuals
- Measurement of resistance
  - LD50 - log dose that kills 50% of the population
- Resistance level can be compared between populations
- Discriminating concentration: all susceptible killed but no resistant ones killed

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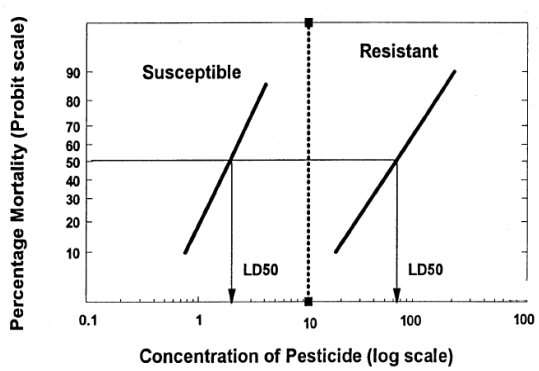
Dose-response bioassay



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Possible mechanisms of resistance

DRUG LEVEL

- **Exclusion**
  - **decreased drug import**
  - **increased drug export**
- **Sequestration**
  - **drug-binding molecule**
  - **drug compartmentalization**
- **Metabolism**
  - **pro-drug not activated**
  - **increased drug inactivation**

TARGET LEVEL

- **Modified**
  - **decreased affinity**
- **Amplified**
  - **increased sequestration**
  - **increased threshold**
- **Missing**
  - **target bypass**
- **Repaired**
  - **increased damage repair**
- **Protected**
  - **protected by substrate**

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Mechanisms of resistance

- Changes in exoskeleton: reduce or slow down insecticide penetration
- Changes in metabolic enzymes:
  - Monooxygenases, esterases, glutathione S-transferases (GST)
  - Detoxification ability ↑ or more enzymes produced
  - Genetic basis: point mutations or gene duplication

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Mechanisms of resistance

- Target sites insensitive
  - Acetylcholinesterase (AChE)
  - Sodium channel proteins
  - GABA receptors
  - Genetic basis: point mutations
- Behavior changes - reduce contact with insecticide

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Management strategies

- Aim to reduce the frequency of resistant allele (R); increase the frequency of susceptible allele (s)
- When using single insecticides
  - Use a dose high enough to kill at least all heterozygotes

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Management strategies

- When using multiple insecticides
  - Using insecticides with different modes of action
  - Rotation in time
  - Delay resistance development to each insecticide
- In combination with non-chemical control
  - Environment/animal management
  - Biological control
  - Genetic control
  - Vaccination

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Summary

- 6 groups of insecticides: organochlorines, organophosphates, carbamates, pyrethroids, IGRs, *Bacillus* toxins
- Different formulations for different parasites and condition
- Chemical use leads to resistance
- Resistance detection: dose-mortality plot, LD50, discrimination concentration
- Mechanisms of resistance: exoskeleton, metabolic enzymes, target sites, behavior
- Management strategies: aim to reduce the frequency of resistant allele; slow down resistance development

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