

Implementation of Biological Control of Major Insect Pests and Weeds in Northern Plains

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Western SARE-Professional Development Program
Montana State University-Western Triangle Agricultural Research Center

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**Using Microbes
to Manage Insect Pests
Where are We?
What Does The Future Hold?**


Stefan Jaronski
USDA Agricultural Service,
Northern Plains Agricultural Research Lab
Sidney MT USA



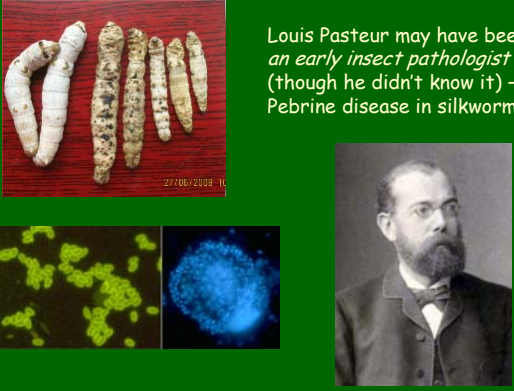
Microbes attacking insects is not a new idea

- Earliest reports of insect disease:
- 700 BC: China
- 322 BC- Aristotle
 - "Historia Animalium"
- 29-32 BC- Virgil
 - "Georgica"

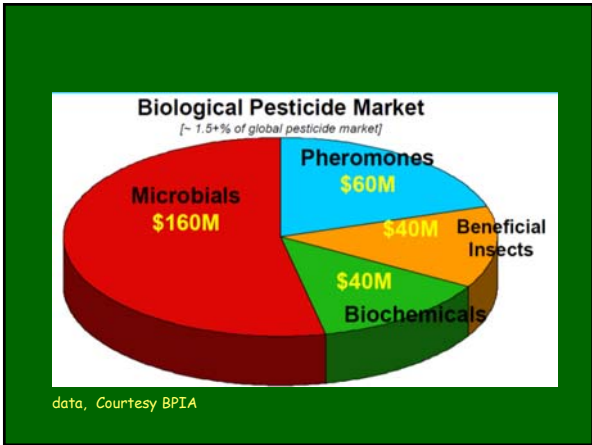


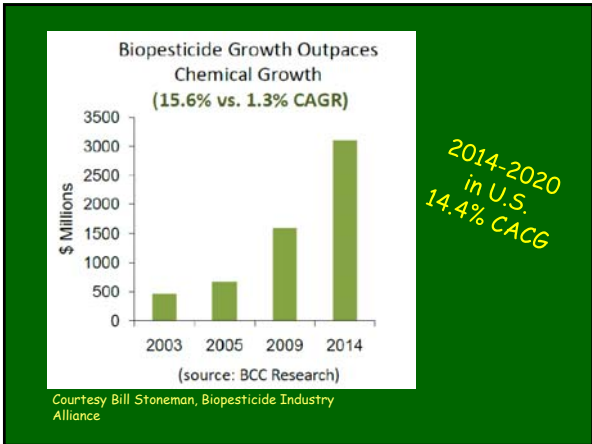


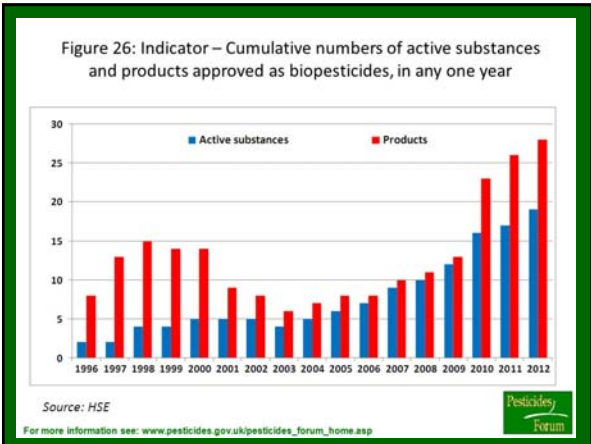
Earliest Illustration of Fungi Killing Insects c.1600

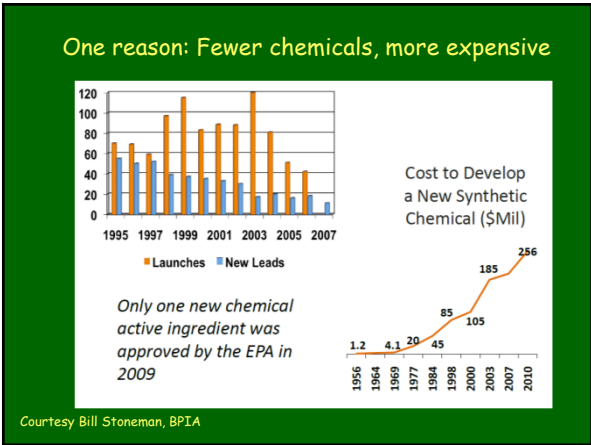


Louis Pasteur may have been an early insect pathologist (though he didn't know it) — Pebrine disease in silkworms









- Current US Microbial Insecticides**
- | | |
|---------------------------------|---------------------------|
| Granulosis Virus | Chromobacterium subtsugae |
| NucleoPolyhedrosis Virus | Paenibacillus lentimorbis |
| Bacillus popilliae | Pasteuria penetrans |
| Bacillus thuringiensis kurstaki | Paranosema locustae |
| B thur. aizawai | Lagenidium giganteum |
| B thur. israelensis | Beauveria bassiana |
| Bacillus sphaericus | Metarhizium anisopliae |
| | Isaria fumosorosea |

**U.S. Companies
'into' microbials**

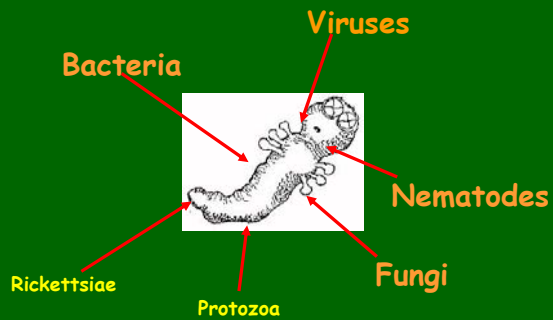
Agraquest	Marrone
Agrivir	BioInnovations
Anatis	M&R Durango
Bioprotection	Novozymes
Arvesta	Biologicals
Becker	Planet Natural
Microbials	Reuter
Biotepp	Troy Biosciences
Certis	US Forest Service
JABB	Valent Biosciences
LAM Intl	
Bayer	BASF
Syngenta	Monsanto

AgBiochem
 Arysta Lifescience NA
 AZ Cotton Research & Protection Council
 Bayer Crop Science
 Bioworks
 Circle One Global Inc.
 Growth Products Ltd
 Jet Harvest Systems
 Montana Microbial Products
 Myco-Forests Corp
 Mycologic Inc.
 Natural Industries Inc.
 Nufarm
 NW Agricultural Products
 Premier Horticulture Inc
 Prophya
 Sylvan Bioproducts
 Verdera Oy
 OmniLytics

but first, a primer...

[Insect Pathology 101]

Insects have their fill of diseases ...



Viruses:
 "Baculoviruses"
 "Nucleopolyhedrosis Viruses"
 "Granulosis Viruses"

Virus particles

Virus-killed Caterpillar

Viruses in gut wall

Viruses:

How the Baculovirus Works

The Baculovirus is sprayed onto the foliage

The caterpillar consumes the Baculovirus

The protein encapsulating the Baculovirus DNA dissolves and the DNA enters the stomach cells

Baculovirus DNA is replicated by stomach cells until the stomach cells rupture. The caterpillar stops feeding

The caterpillar dies within days

Baculovirus DNA is spread throughout the caterpillar causing a general systemic infection

Each virus specific to particular moth species

Some virus products

Gemstar

CYD-X


Madex 3

SPOD-X LC

USDA does not endorse any commercial products. These are provided as examples only.

The Bacteria

Bacillus thuringiensis; *Bacillus sphaericus*

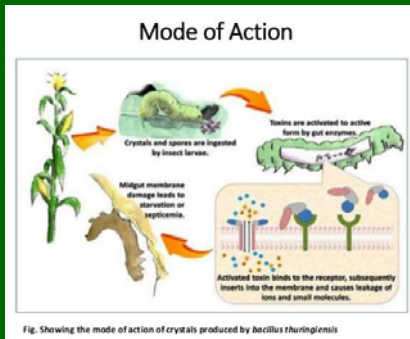


XenTari
DIPeL
Gnatrol
Vectobac
Vectolex
DIPeL

Novodor
Teknar
Novodor
MVP
Javelin
Cutlass
JavelinWG


The 'Bts'

- moths
- beetles
- mosquitoes
- blackflies
- gnats

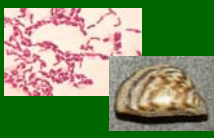


Other bacteria are true pathogens


Paenibacillus popilliae



Bacillus lentimorbus

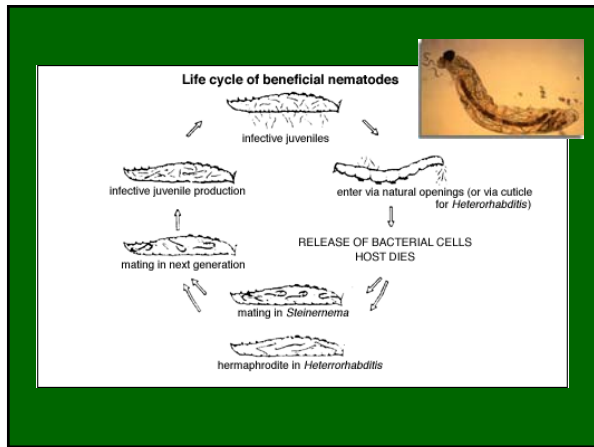


Pasteuria penetrans



Chromobacterium substugae **GRANDEVO**





- ### Some Common Nematode Products:
- Nemasys, Nemasys H (Microbio)
 - Scanmask, Ecomask, Heteromask (Biologic Co.)
 - Grubstake Hb, Grubstake-Hm, GnatNot (Integrated BioControl Systems, Inc.)
 - Entonem, Larvanem, Scia-Rid (Koppert B.V.)

Nematodes actively seek out their targets

Types of Nematode Behavior:


- **Cruisers**  *Heterorhabditis*
Steinernema riobravis
- **Stalkers**  *Steinernema*
carpocapsae

Nematodes are Biologicals

Need refrigerated storage and have definite shelf life
Are affected by soil conditions (moisture, heat, porosity)
Can be affected by greenhouse chemicals.

So how are we using these microbials?

As **inundative** (albeit biological) **catastrophic density independent** mortality factors
i.e. like chemicals ...



Those danged microbes don't work well enough!

- Too weak (need too much, so too expensive)
- Don't last long enough
- (sometimes) Too complicated to think about

How to make them work better, cheaper?

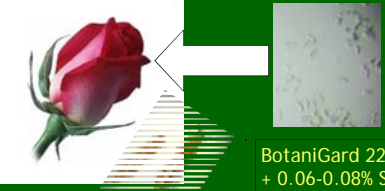
The 'traditional' approaches

- Deliver them more efficiently
- Make them more persistent, work longer
- Make them more virulent



How to make microbes better, cheaper?

Make application more efficient thru formulations



BotaniGard 22WP
+ 0.06-0.08% Silwet L77®

Spores penetrated in substantial numbers into 5-6th petals of unopened flower

Control much better than 22WP alone

How to make microbes better, cheaper?

Creative, 'traditional' approaches

Make application more efficient thru *novel* formulations

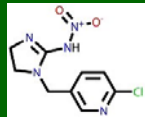
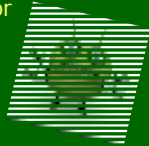
Carnauba wax carrier e.g. Entostat®;
or Candelilla wax powder



How to make microbes better, cheaper?

Creative, 'traditional' approaches

Combine chemical stressors with
fungi
→ to stress insects' immunity
→ alter behavior



How to make them work better, cheaper?

Make (get) a 'better' microbe

How?

Traditional

- Screening for the 'best' isolates
 - Let Nature provide
- Classical mutation selection - tradeoffs

Novel

- Transgenic approaches: virulence factors, enhanced detox mechanisms
 - BUT, regulatory, societal challenges



Let's think outside the box

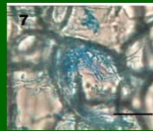


- **Bring the insect to the microbe**
 - Fatty acids attractive to grasshoppers
- **Use the insect to vector the microbe to its kin**
 - Japanese beetle
 - Pine bark beetle
 - Adult click beetle
- **Use another insect to transfer the microbe to where it's needed**
 - bees vector *Beauveria* to flowers
- **Take advantage of target insect's behavior to increase transfer efficiency:**
 - spraying bark over which Gypsy Moth larvae must crawl;
 - Spray mosquito resting habitat (resting boxes in urban area)
 - Put the spores where (cherry fruit fly) larvae fall to pupate



Outside the box

The insect pathogenic fungi, Beauveria, Metarhizium, Isaria as plant endophytes



Gomez-Vidal et al., 2006
Microb 37 (2006) 624-632

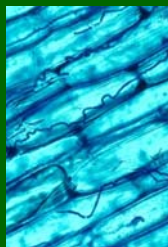
Beauveria are endophytic in maize, cocoa, date palm, coffee, grapes, tomato, banana, sorghum, medicinal poppy, jute, broad bean, cassava, cotton, strawberries, wheat ...

Metarhizium are endophytic in rape, beans, switchgrass, yew, rice; more famously associated with root systems.

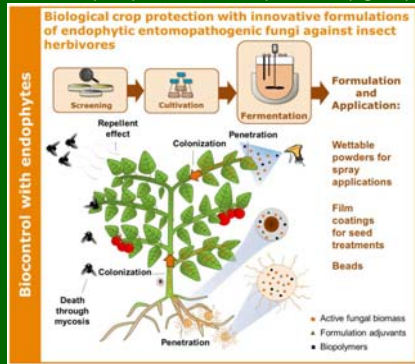
Beauveria, Metarhizium, Isaria as endophytes

Can be artificially introduced in at least some plants

- Foliar application
- Seed treatment
- Root dip



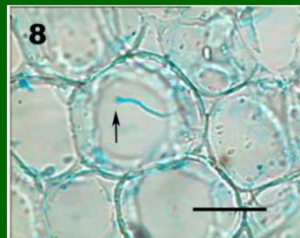
Visualization of the concept by one university-industry group



Courtesy Anant Patel, Bielefeld University

How do these endophytic fungi act on an insect pest?


- Direct infection of insects
- Indirect effect - secreted metabolites



from Quesada-Morago et al. 2006

But wait! There's more!

And more...



Pure culture of *Metarhizium anisopliae* LHL07 reprograms soybean to higher growth and mitigates salt stress

Abdul Latif Khan · Muhammad Hanuman · Sumera Afzal Khan · Saraj-Muhammad · Zahra Khara Shiverani · Muhammad Hanuman · Shady ur Rehman · Jang-Gook Kim · In-Jung Lee

Metarhizium anisopliae LHL07-inoculated soybean plants displayed significantly


- higher shoot length,
- shoot fresh and dry biomass,
- chlorophyll contents,
- transpiration rate,
- photosynthetic rate and leaf area,

under salt-induced salt stress as compared to non-inoculated control plants.

Phytohormone Effects?

Metarhizium LHL07 elevated proline and reduced superoxide dismutase and malondialdehyde, reduced abscisic acid and elevated jasmonic acid levels

Induction of Systemic Resistance *without* endophytism does occur




Physiological and Molecular Plant Pathology

Volume 61, Issue 1, November 2002, Pages 209-216

Regular Article

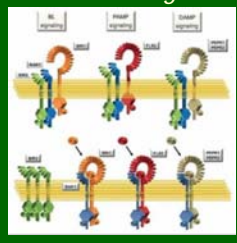
Characterisation of systemic resistance in sugar beet elicited by a non-pathogenic, phyllosphere-colonizing *Bacillus mycoloides*, biological control agent

R.L. Bargabus, N.K. Zidack, J.E. Sherwood, B.J. Jacobsen



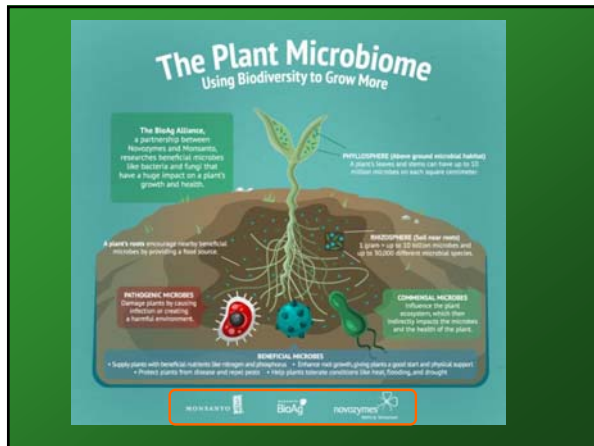

Induction of Systemic Resistance

Plants can recognize molecular patterns



PAMPs
MAMPs
DAMPs
HAMPs

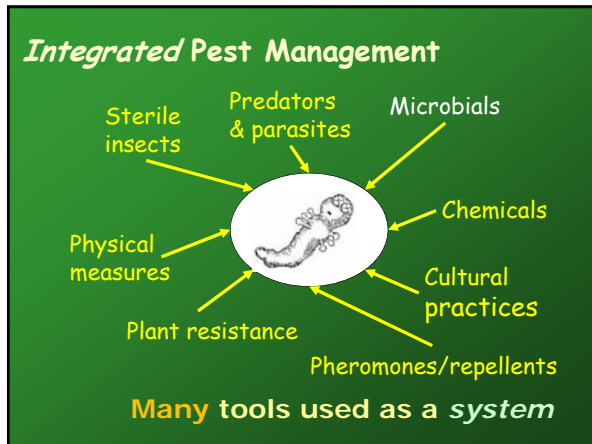
Microbes on plant's cuticle are a molecular pattern ...



But before we get too excited
at these sci-fi prospects,
Let's not forget

There is no such thing as a magic bullet

(if you think there is, I have a bridge over the
Yellowstone River to sell you).

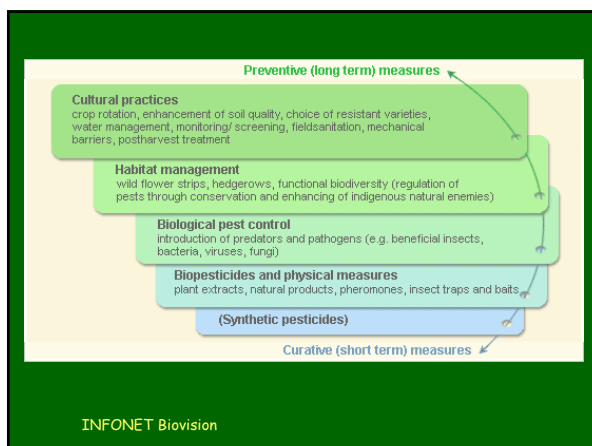


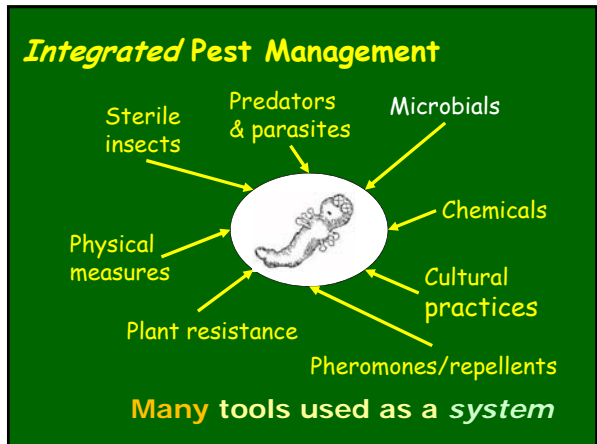
Integrated Pest Management

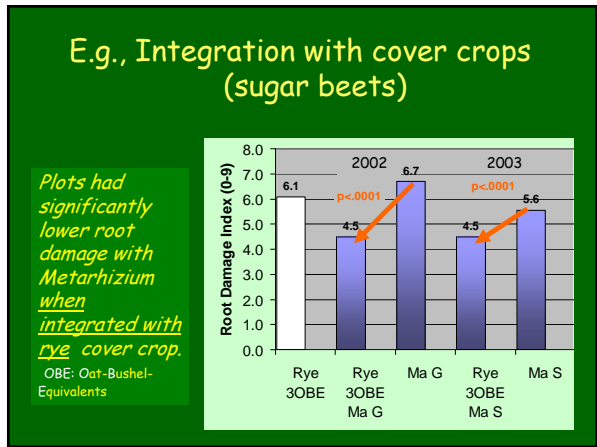
(U.S. National IPM Network).

“Integrated Pest Management (IPM) is a

- sustainable approach
- to managing pests
- by combining biological, cultural, physical and chemical tools in a way that
- minimizes economic, health, and environmental risks”.







Another example:

Strawberry IPM system — UC Dept of Agriculture & Natural Resources Coop Extension

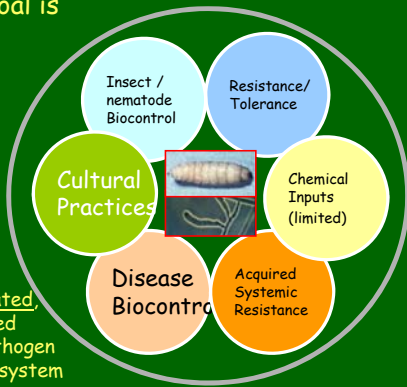
	1 st application (Rate/acre)	2 nd application (Rate/acre)	3 rd application (Rate/acre)
1	Untreated	Untreated	Untreated
2	Assail 70 WP (3 oz) 4A*	Assail 70 WP (3 oz) 4A	Assail 70 WP (3 oz) 4A
3	Vacuum	Vacuum	Vacuum
4	Rimon 0.83 EC (12 fl oz) 15 + Brigade (16 oz) 3A	Met52 EC (16 fl oz) + Delbug Turbo (104 fl oz)	Met52 EC (16 fl oz) + AzaGuard (18 fl oz)
5	Sequoia (4.5 oz) 4C	Sequoia (4.5 oz) 4C	Vacuum
6	Pfr-97 (2 lb) + Neemix (9 fl oz)	Pfr-97 (2 lb) + Neemix (9 fl oz)	Vacuum
7	Vacuum	Sivanto (14 fl oz) 4D + Delbug Turbo (104 fl oz)	Rimon 0.83 EC (12 fl oz) 15 + Brigade (16 oz) 3A
8	Sivanto (14 fl oz) 4D	Sivanto (14 fl oz) 4D	Vacuum
9	Sequoia (4.5 oz) 4C	Sivanto (14 fl oz) 4D	Beleaf 50 SG (2.8 oz) 9C
10	<i>B. bassiana</i> +neem (1qrt)	<i>B. bassiana</i> +pyrethrum 3A+neem (1qrt)	<i>B. bassiana</i> +pyrethrum 3A (1qrt)
11	<i>B. bassiana</i> +pyrethrum 3A (1qrt)	<i>B. bassiana</i> +neem (1qrt)	Beleaf 50 SG (2.8 oz) 9C
12	<i>B. bassiana</i> +pyrethrum 3A (1qrt)	Vacuum	Rimon 0.83 EC (12 fl oz) 15 + Brigade (16 oz) 3A

IPM with microbial in chrysanthemums

- Week 1: introduce *Dacnusa* for leafminers
- Week 2: early in week, *Beauveria* for thrips
late in week, apply fungicides
- Week 4: Apply predaceous mites
Apply *Beauveria*
- Week 6-8: Spray *Beauveria* weekly
Apply Bt for lepidoptera, as needed
Apply cinnamaldehyde (cinnamon oil)
only to mite "hotspots"
- Week 8: Introduce *Diglyphus* for leafminers

And this program was designed by a farmer

Ultimate goal is





An integrated
bio-based
pest and pathogen
management system



Thank you for your attention

*Do not ask for whom the Beauveria smiles,
It smiles for Thee.*

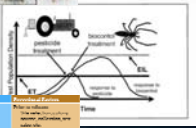


IMPROVING THE EFFICIENCY OF BIOLOGICAL CONTROL (OR HOW TO BUILD A BETTER BC AGENT)



Jeff Littlefield
Department of LRES
Montana State University

KEY POINTS

- What is biological control?
- The Players involved
- Types of biocontrol
- Our goals
- Failure matrix

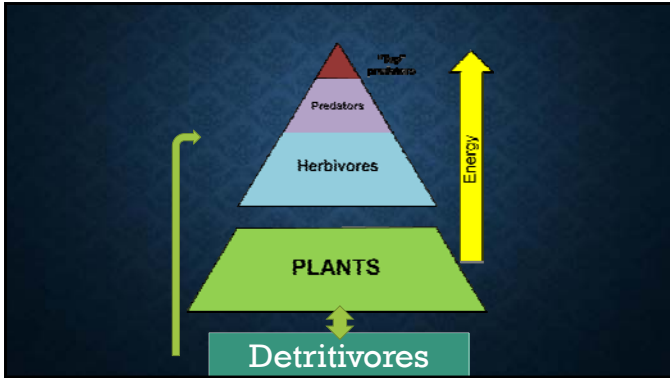


Failure Matrix
Host Specificity - Inadequate host range - Inadequate host range - Inadequate host range
Dispersal/Establishment - Poor dispersal - Poor dispersal - Poor dispersal
Reproduction - Poor reproduction - Poor reproduction - Poor reproduction
Survival - Poor survival - Poor survival - Poor survival
Competition - Poor competition - Poor competition - Poor competition
Regulation - Poor regulation - Poor regulation - Poor regulation
Evolution - Poor evolution - Poor evolution - Poor evolution
Other - Poor other - Poor other - Poor other

WHAT IS BIOLOGICAL CONTROL?

Natural control vs Biological control

➤ **Natural control** – the regulation of a pest populations by naturally occurring enemies



WHAT IS BIOLOGICAL CONTROL?

Natural control vs Biological control


- **Biological control** – the manipulation of natural enemies to achieve desired levels of control

THE PLAYERS

- Predators
- Parasitoids
- Nematodes
- Pathogens
- Weed herbivores



GUILDS OF BIOCONTROL AGENTS



- Multiple & varied hosts
- Larger than host
- Fairly mobile
- Immature & adults may have similar feeding habits
- Generally operate under higher pest populations

PREDATORS

PREDATORS & PARASITOIDS

PREDATORS: FUNCTIONAL CLASSIFICATION

- Ambushers
- Searchers for passive prey
- Pursuers of active prey
- Nest-provisioners



ARKive
www.arkive.org

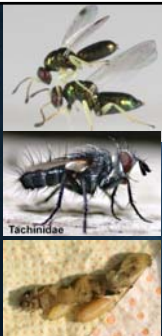
Ground Beetles



- Single host
- Smaller than host
- Narrow host range
- Fairly mobile only as adults
- Immature & adults have different feeding habits
- More effective under lower pest populations



PARASITOIDS

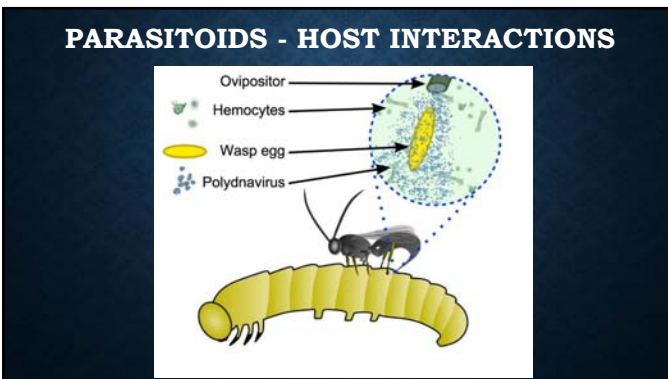


- Mostly Hymenoptera
- Some flies & other orders
- Solitary, gregarious, polyembryonic
- Multiple parasitism vs superparasitism
- External vs internal


PARASITOIDS



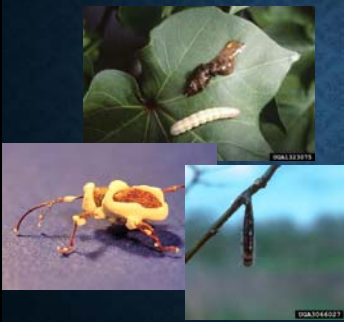




- Small, clear, worm-like
- Three families: Steinerhmatidae, Heterorhabditidae & Rhabditidae
- Wide host range
- Cruiser vs ambush spp
- Mutualistic bacteria
- Kill host 24-48 hr
- Sensitive to low moisture
- Optimal temperature



NEMATODES



- Applied as "biopesticide"
- Include bacteria, fungi, & viruses
- Broad host range
- Limited shelf life
- Sensitive to environmental conditions & UV

PATHOGENS

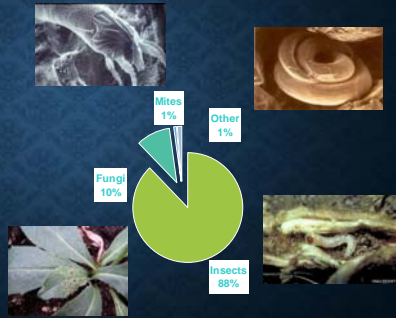
ENTOPATHOGENS

- Nosema locustae
- Milky spore
- Beauveria bassiana
 - Thrips
 - Whiteflies
 - Aphids
- Bacillus thuringiensis (BT)
 - Lepidoptera larvae
 - Fungus gnats or black flies
 - Beetles



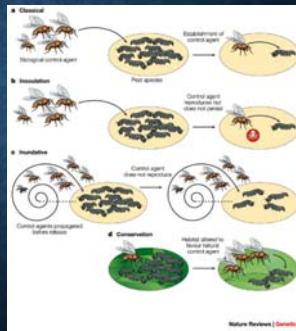
WEED HERBIVORES

- Mostly insects – weevils, moths, tephritid flies, etc.
- Host specific or narrow host range
- Largely internal feeders or gall makers
- Immatures & adults may or may not have different feeding habits
- Used primarily in classical BC for exotic weeds



TYPES OF BIOLOGICAL CONTROL

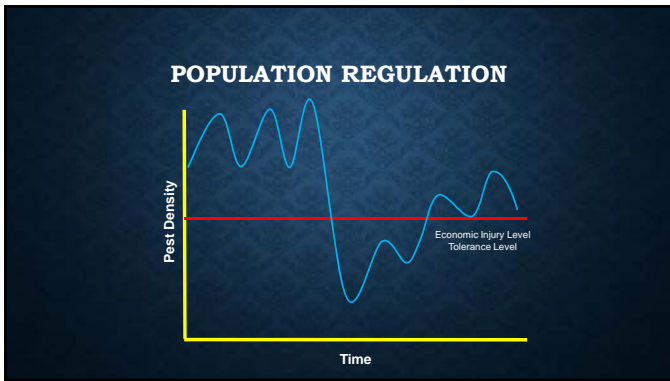
- **Classical/ Inoculation** – initially small numbers of natural enemies are released in target pest areas for long-term control.
- **Augmentative/ Inundative** – large numbers of natural enemies are released to control a target pest for a short amount of time.
- **Conservation** – changing environmental conditions to aid in natural enemy survival.

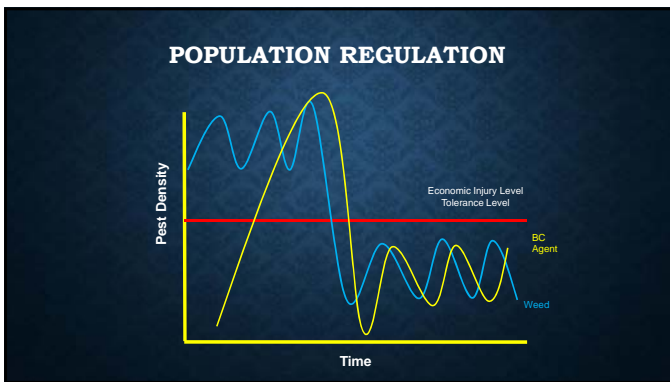


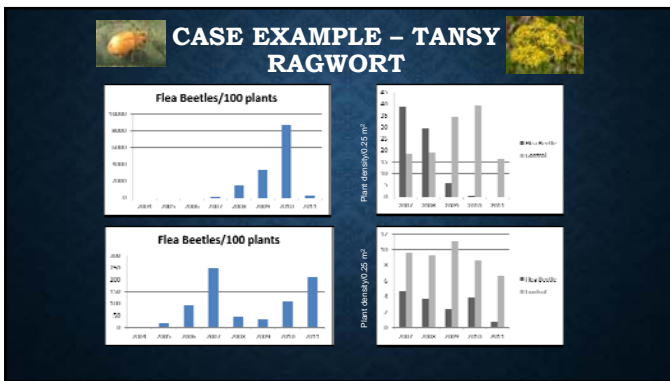
GOALS?

Dependent upon:

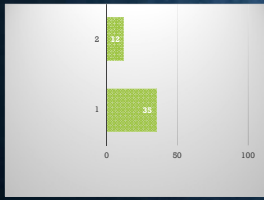
- Type of pest
- Level of control required
- Speed of control
- Scale of control



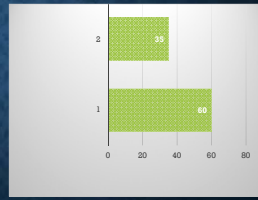




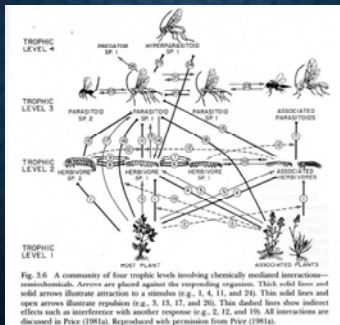
ARE WE SUCCESSFUL?

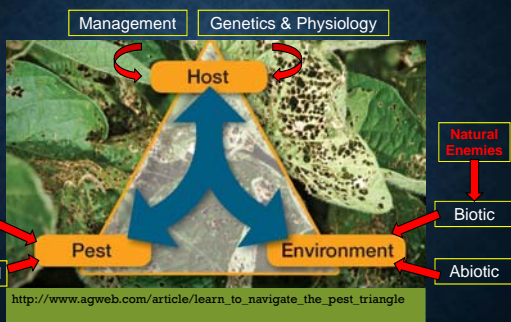


Insect Biocontrol



Weed Biocontrol





FAILURE MATRIX (WEED BC)

Biotic Factors	Abiotic Factors	Procedural Factors
Plant Community Host density Succession	Climate Temperature Precipitation	Prior to release Site selection, colony source, collection, sex ratio, etc.
Interactions Predation/parasitism Competition	Site Characteristics Soil, slope, aspect Shade, moisture	Release Methods, wrong agent or host, timing, life stage, etc.
Biocontrol organism Synchronization Physiology Fecundity & behavior Genetic diversity Emigration	Elevation Temperature Precipitation	Post release Site management, agent detection, vandalism, disturbance
	Latitude Seasons, day length Disturbance Fire, flood, cultivation	Personnel Training, experience, prioritization, follow-up

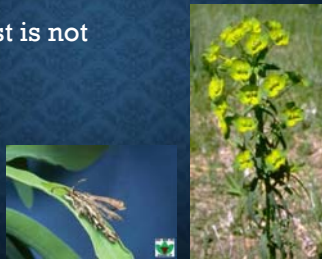
Modified from Coombs OR Dept. Ag.

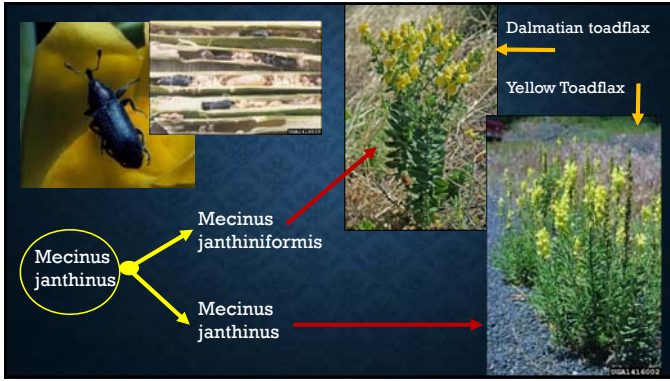
TOP 10 REASONS FOR BIOCONTROL FAILURE



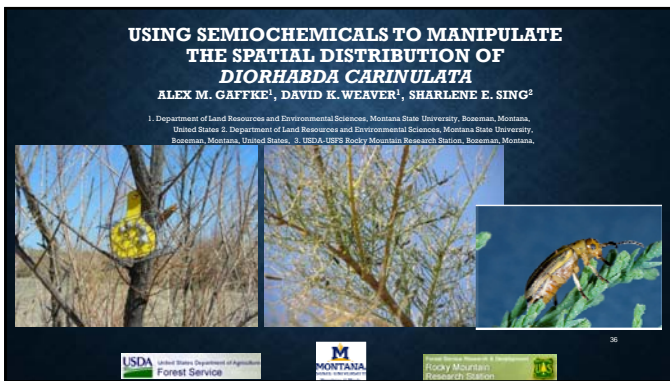
TOP 10 REASONS FOR BIOCONTROL FAILURE

10 - My pest is not the **PEST**









WHY MANIPULATE AN AGENT

- Increase establishment
- Easier to monitoring
- Control agent's distribution
 - Increase herbivory on target plant
 - Manage spread to avoid critical habitat



37

SEMIOCHEMICALS

- Male produced aggregation pheromone
 - (2E, 4Z)-2,4-heptadien-1-ol (Cossé et al. 2005)
- Aggregation causing green leaf volatiles (Cossé et al. 2006)
 - (E)-2-hexenal
 - (Z)-3-hexenal
 - (Z)-3-hexen-1-ol
 - (Z)-3-hexenyl acetate



38

TOP 10 REASONS FOR BIOCONTROL FAILURE

8 - You call this a niche?

Seedhead Feeders

Flies:
Urophora affinis
Urophora quadrifasciata
Urophora sp.
Chamaecha aeneiparis

Moth:
Megastoma pascuiparvula

Beetles:
Leptus nitidulus
Leptus albipes
Pezomachus fuscus

Root Borers

Moths:
Agrotis prosopea
Plutella maculipennis
Plutella maculipennis

Beetles:
Cyathodermus scutellus
Sphenophorus signatus



Larinus minutus attacks diffuse knapweed stems & destroys seeds

Both agents have strong individual effects

Plant performance reductions are independent

No competitive effects

Cyphocleonus achates attacks diffuse knapweed roots

zoology.ubc.ca

SEEDHEAD FEEDERS

Seedhead Feeders

Flies:
Urophora affinis
Urophora quadrifasciata
Senecella cineraria
Chaetorellia acrolophi

Moth:
Metzneria paucipunctella

Beetles:
Larinus minutus
Larinus obtusus
Bangasternus feustii

INSECT BIOCONTROL

(a)

Mean proportion established \pm SE

Number of natural enemies released per project

○ Homoptera
 ● Lepidoptera

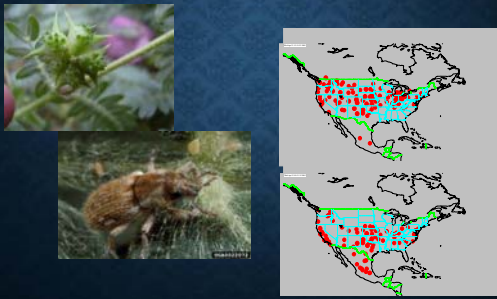
Mills, 2006, Trophic and Guild Interactions in Biological Control, 191-220.

TOP 10 REASONS FOR BIOCONTROL FAILURE

7 – Too HOT - Too COLD – Too Dry - Too WET (The Goldilocks Syndrome)



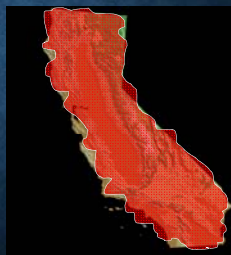
PUNCTUREVINE BIOCONTROL



THE COTTON CUSHION SCALE



California introduced 1880
State-wide 1886



NATURAL ENEMIES FOUND



Rodolia cardinalis



Cryptochaetum iceryae

NATURAL ENEMY RELEASED



1890: Cotton Cushion scale eliminated



OTHER IMPORTED NATURAL ENEMY OF THE COTTON CUSHION SCALE



CLIMEX - SOFTWARE TO PREDICT THE EFFECTS OF CLIMATE ON SPECIES

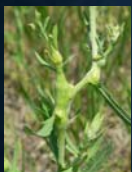


TOP 10 REASONS FOR BIOCONTROL FAILURE

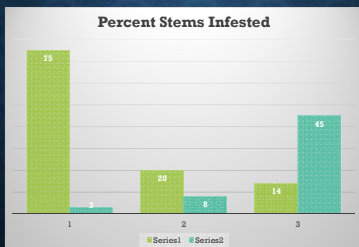
6 – Hey! Some *#@\$#!
sprayed (set fire to, cultivated,
flooded, etc.) my plots



RUSSIAN KNAPWEED & GRAZING



Aulacidea
Aa

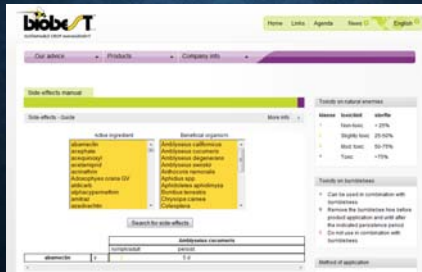


Japiella
ja

PESTICIDES SIDE EFFECTS DATA BASE

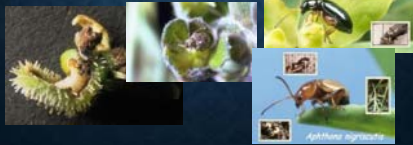
<http://www.biobest.ca/>

<http://side-effects.koppert.nl/>



TOP 10 REASONS FOR BIOCONTROL FAILURE

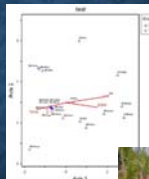
5 - Wrong place – wrong time
(I should have checked my Outlook calendar)



SITE SELECTION & RELEASE

Site selection

- Suitable habitat
- Sites not prone to fires, flooding, or other natural disturbances
- Adequate plants
- Small vs. large infestations



Release methods

- Timing
- Numbers released



TOP 10 REASONS FOR BIOCONTROL FAILURE

4 - When the consumers are consumed







Month	% Galls Eaten
June	~10
Oct.	~40



Month	% Parasitism
June	~10
Sept.	~10



Month	% Parasitism
June	~50
Oct.	~80

TOP 10 REASONS FOR BIOCONTROL FAILURE



3 - "The Little moth ... beetle ... fly... that couldn't (or Too little or Too late)

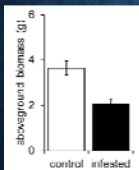




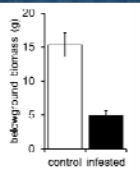
Impact studies

CABI - July 2015

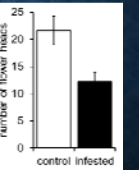





Group	aboveground biomass (g)
control	~4.0
infested	~2.0



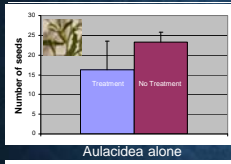
Group	belowground biomass (g)
control	~15.0
infested	~5.0



Group	number of flower heads
control	~22.0
infested	~13.0

→ Significant impact on biomass and number of flower heads

Interactions among biocontrol agents



Aulacidea alone



Jaapiella alone

TOP 10 REASONS FOR BIOCONTROL FAILURE

2 – The penguin effect – What we have here is a lack of diversity!

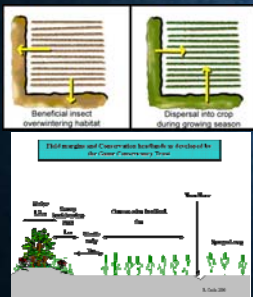


TOP 10 REASONS FOR BIOCONTROL FAILURE

2 – The penguin affect



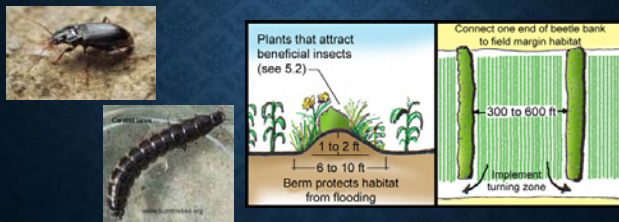
INCREASING DIVERSITY & STRUCTURE



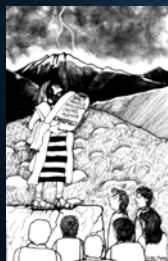
Key Design Considerations

- Provide plant diversity and structure in the buffer.
- Protect buffer from disturbances (e.g., pesticides, tillage).
- Predation of insect pests generally increases with the percentage of buffer habitat in the area.
- Locate buffers throughout the fields and landscape to encourage dispersal of beneficial insects.
- Buffers may provide habitat for some pest insects but this can be reduced by selecting appropriate plants.
- Beetle banks are long, planted berms that provide habitat for beneficial insects .

Beetle Banks

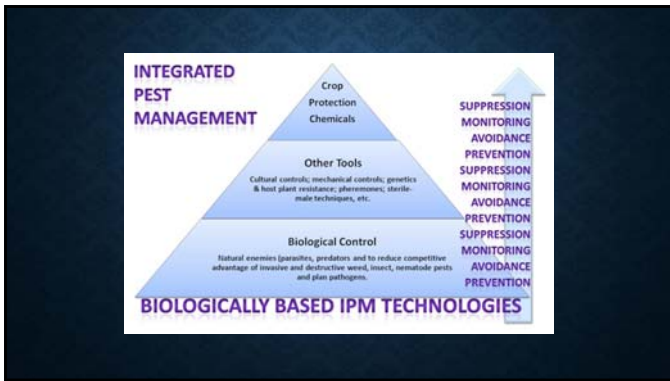


TOP 10 REASONS FOR BIOCONTROL FAILURE



1 – Stupid peoples (we have met the enemy and they are us)





ADDITIONAL REFERENCES



The Xerces Society: Farming with Native Beneficial Insects.
<http://www.xerces.org/conservationbiocontrol/>

Conservation Biological Control – Pedro Barbosa, Academic Press

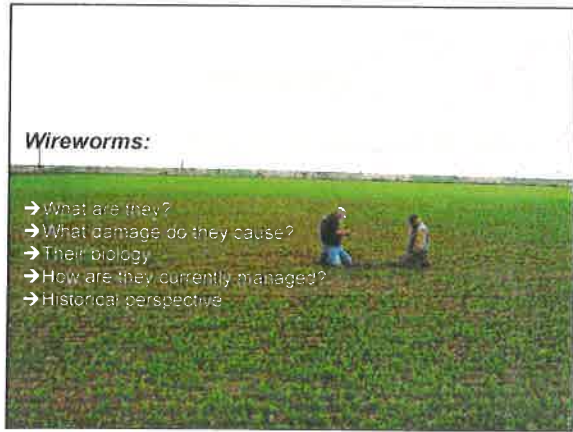
Michigan State U.: <http://nativeplants.msu.edu/>

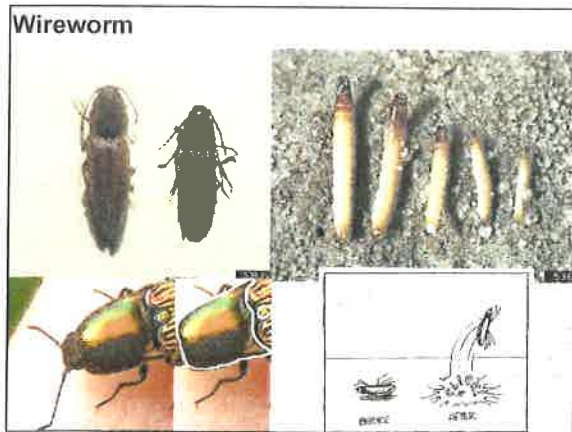
British Columbia Ministry of Forests, Lands & Natural Resource Operations (Publications & manuals):
<http://www.for.gov.bc.ca/hra/Plants/publications.htm#manuals>

EDDMapS Biological Control of Invasive Plants (USFS Manuals):
<http://www.eddmaps.org/biocontrol/>

Jeff Littlefield Department of LRES, MSU, PO 173120, Bozeman, MT 59717 (406) 994-4722 e-mail: JeffreyL@Montana.edu

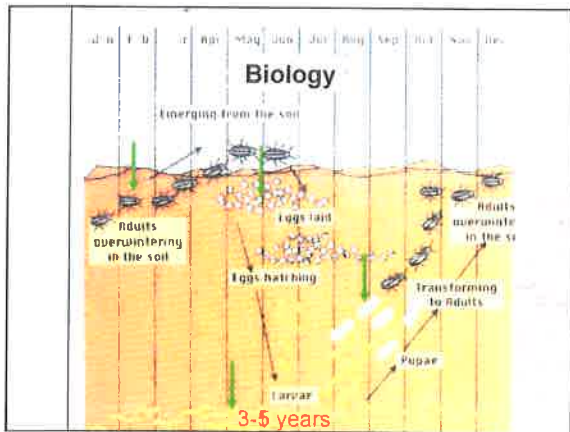


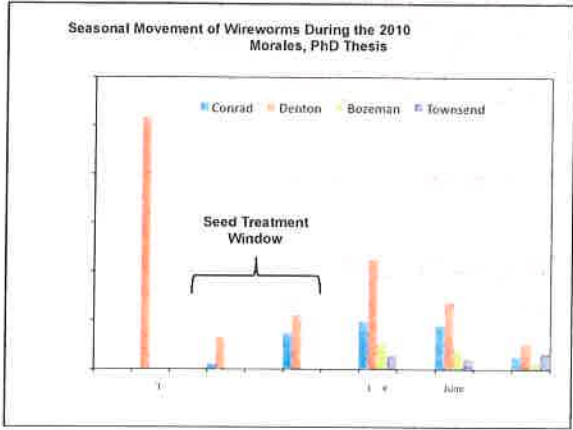


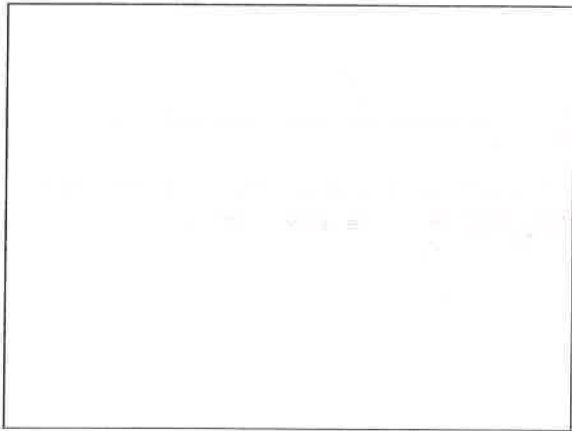








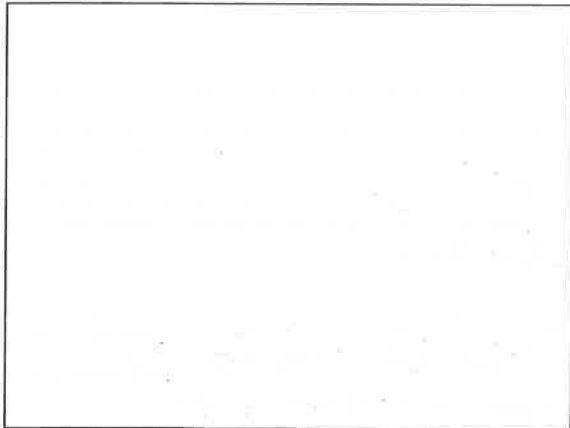











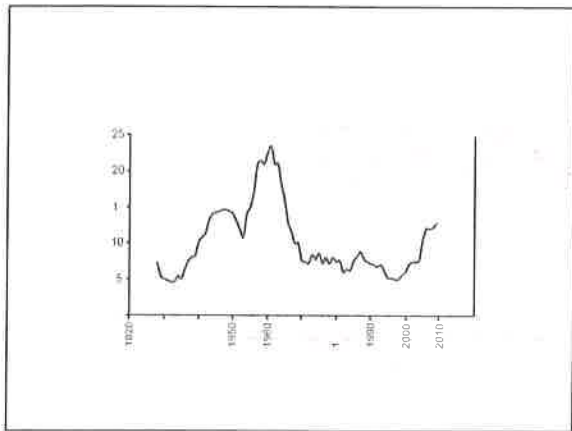


History



seed

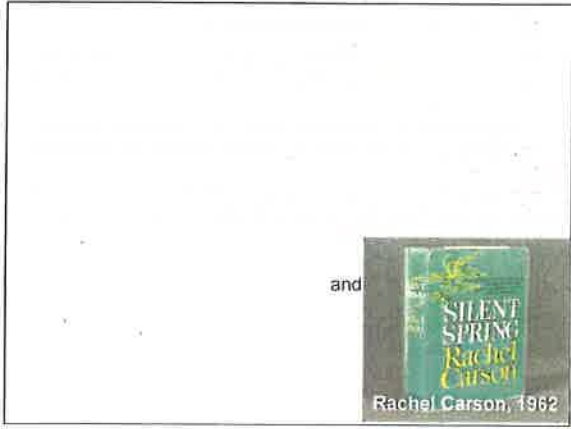
'We
to

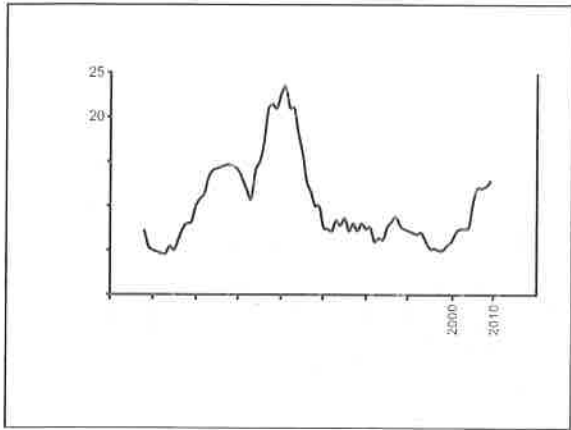


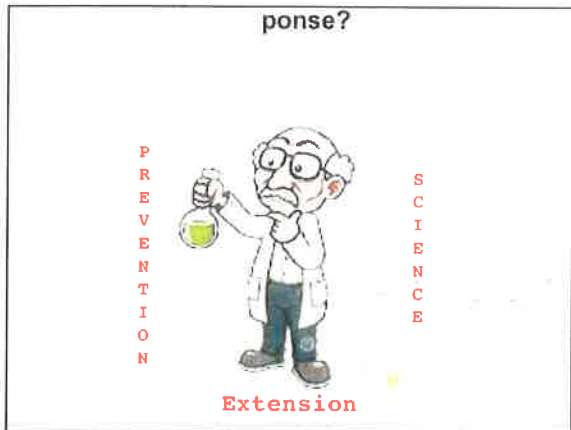
What Happened ?

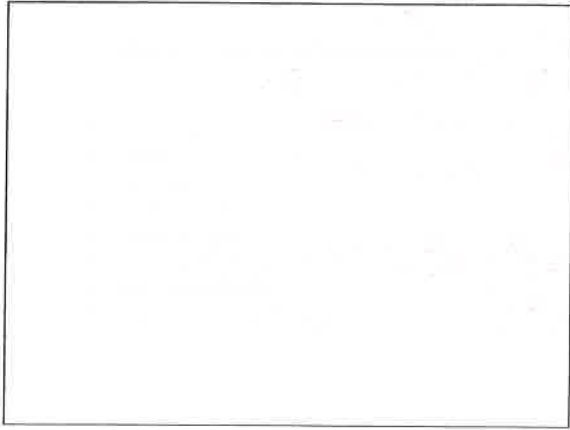
TABLE 1 Chemicals in the 1950s and 1960s

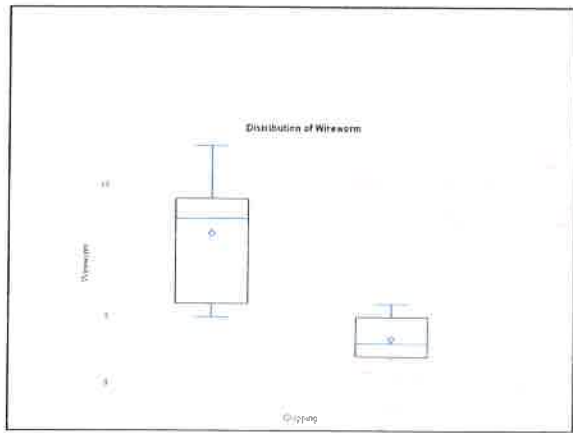
Chemical	Year	Source	Regulation	Notes
Organochlorines	1940s	DDT, dieldrin, heptachlor, chlordane, heptachlor epoxide, mirex, toxaphene, aldrin, dieldrin, endosulfan, heptachlor, mirex	1972 (DDT), 1973 (mirex), 1974 (toxaphene), 1975 (aldrin, dieldrin, endosulfan)	100% ban on DDT, 90% on others
Organophosphates	1950s	Parathion, diazinon, malathion, chlorpyrifos, phosphamidon, phosalone, phosmet, phosalone, phosalone, phosalone	1972 (parathion), 1973 (diazinon), 1974 (malathion), 1975 (chlorpyrifos), 1976 (phosphamidon), 1977 (phosalone), 1978 (phosmet)	Partial bans, some still in use
Organotin	1950s	Triphenyltin chloride, dibutyltin dilaurate, dibutyltin diacetate, dibutyltin diacrylate, dibutyltin diacrylate, dibutyltin diacrylate	1972 (tributyltin chloride), 1973 (dibutyltin dilaurate), 1974 (dibutyltin diacetate), 1975 (dibutyltin diacrylate)	Partial bans, some still in use
Organotin	1950s	Triphenyltin chloride, dibutyltin dilaurate, dibutyltin diacetate, dibutyltin diacrylate, dibutyltin diacrylate, dibutyltin diacrylate	1972 (tributyltin chloride), 1973 (dibutyltin dilaurate), 1974 (dibutyltin diacetate), 1975 (dibutyltin diacrylate)	Partial bans, some still in use

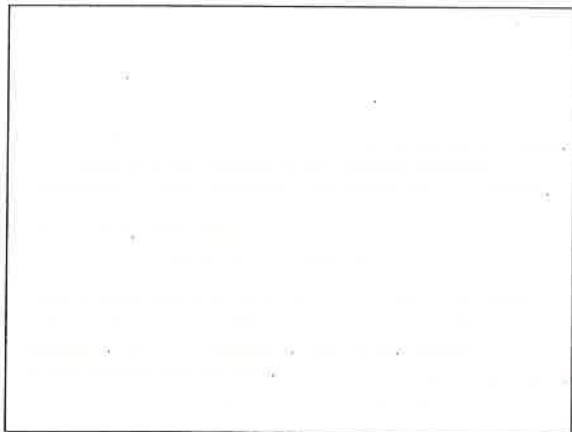












Other Cultural Options

John Henry Comstock (1849-1931)
Professor of Entomology, Cornell University



METHODS OF EXPERIMENTATION.
The soil in each cage of apparatus was first sterilized
by boiling it in water for 24 hours. It was
then sown with the wireworms to be experimentally
tested. The cages that were
to be used in an experiment were first
sterilized in this manner. The time of
the experiment was 10 days to 100 days.

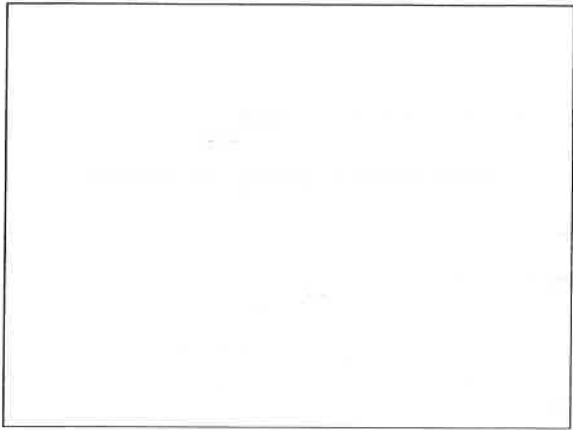
been kept growing. In cages 1 and 2 where no more vegetation
alive so long in cages containing clover and timothy.

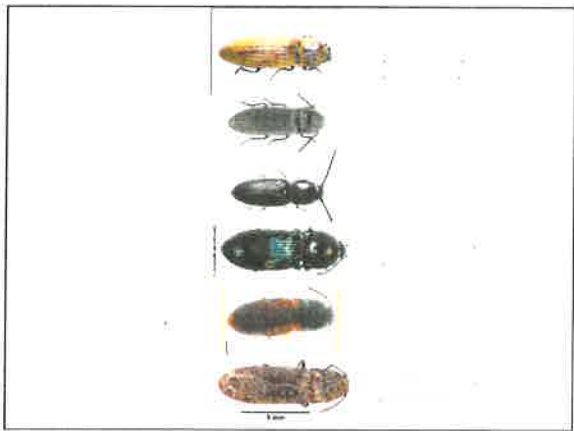
Therefore our experiments do not indicate that a crop of mustard
will render the soil so free from wireworms that the succeeding
crop will escape their ravages.

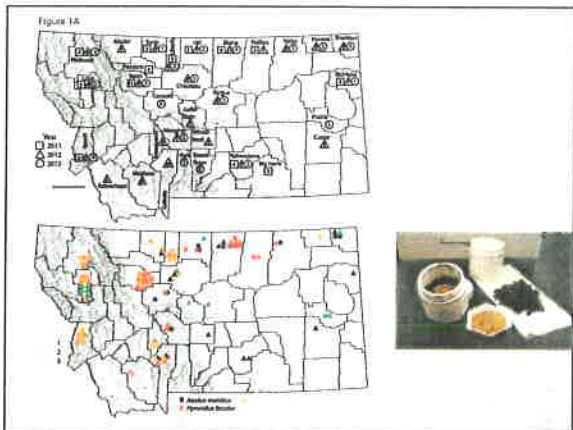
RAPE.
usually recommended for starving out the wireworms are buck-
wheat, mustard and rape.

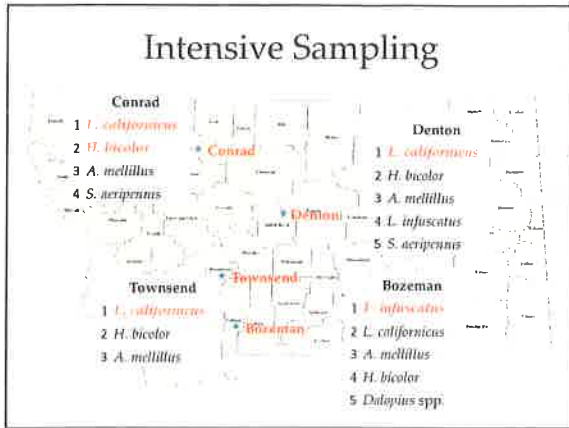
keep it free from all vegetation in the hope that he may thus
starve out the wireworms.

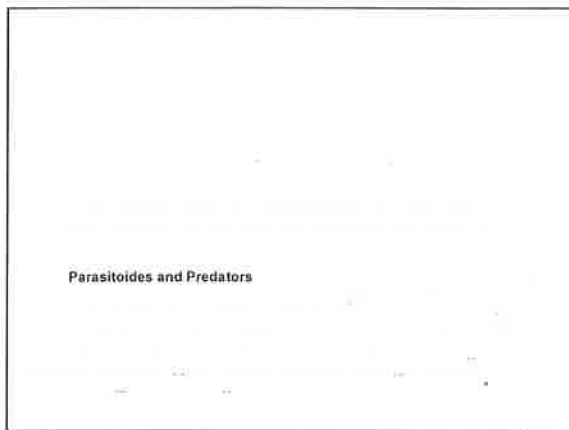
U.S. DEPARTMENT OF AGRICULTURE
BUREAU OF ENTOMOLOGY AND PLANT QUARANTINE
WASHINGTON, D. C.





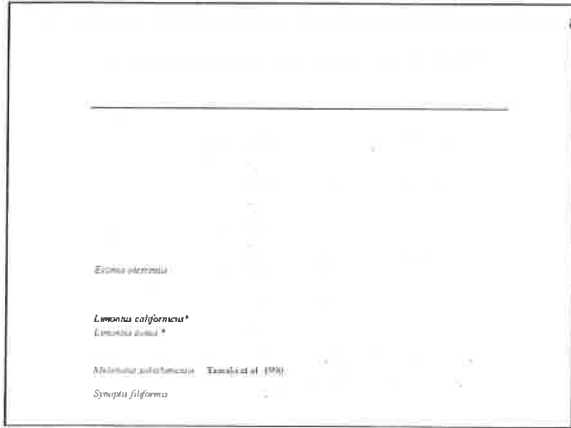




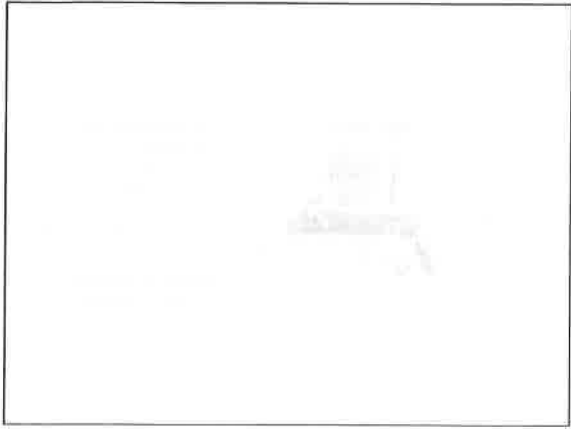















Ecology and Biological Control of Alfalfa Weevil

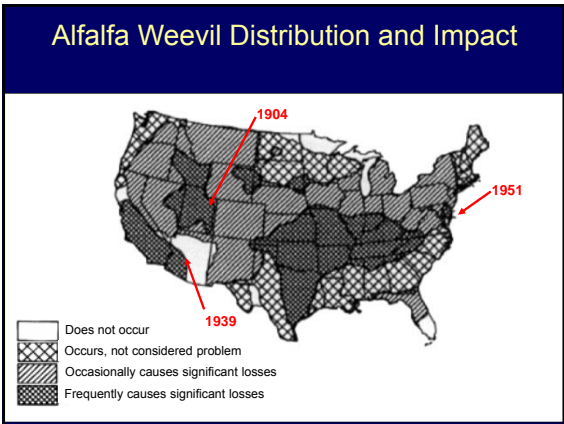
Tatyana A. Rand

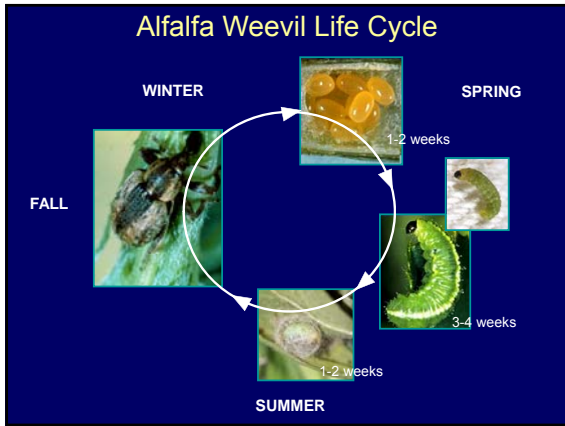
USDA ARS NORTHERN PLAINS AGRICULTURAL RESEARCH LABORATORY
Bozeman, Montana



Outline

- Alfalfa weevil biology/ecology
- Biocontrol research at USDA-ARS-NPARR
 - Identifying key biocontrol parasitoids
 - Assessing the role of generalist predators
 - Identifying field and landscape scale drivers of biological control success





Damage

Larval feeding results in:

- Reduced yield (1st cutting)
- Delayed regrowth (2nd cutting)
- Reduced plant vigor
- Decreased stand density

Determining Infestation Levels

Larval sampling:

- Sweep sampling: 38 cm diam net, 10-sweeps, 5 locations, count larvae; ET = 20 larvae/sweep
- Cut 6 stems, shake in 5-gallon bucket, count dislodged larvae, repeat at 5 locations ET = 2 larvae/stem

Plant inspections:

- Examine 10-20 stems at 5 locations / field
- Assess % damaged plants

Cultural and Chemical Control (NDSU-Extension)



Cultural Control

- If close to cutting, best strategy is to cut early
- After cutting, monitor carefully for damage to crowns / delayed regrowth



Insecticides

- 1st Cutting: If weevils infest crop at early stage (<15 inches), 7-10 days from harvest, treat if 2 larvae / stem and/or 35 to 40% plants show tip feeding
- 2nd cutting: Treat if 50% of the crowns damaged, re-growth delayed 3-6 days

Biological Control

- The action of parasites, predators or pathogens in maintaining pest numbers at lower average levels than would occur in their absence
- DeBach, 1964
- Restore ecological balance
- Can provide self-sustaining and environmentally friendly control of pest populations


National alfalfa weevil biocontrol program

- National biological control program carried out by USDA, 1957-1988; 9 parasitoid wasps established in US
- Parasitoid wasps and disease highly effective at controlling weevils in the eastern US
- Many parasitoid species re-distributed throughout the US in the 1980's (including in MT, ND)

Study Goals


- Characterize the parasitoid complex of alfalfa weevil in the region
- Determine whether habitat characteristics (dryland vs. flood irrigated) affect parasitoid effectiveness
- Assess the role of host density in driving levels of parasitism

Study Design



- Large scale surveys (>30 sites) over 2 years
- Irrigated and dryland systems
- Reared larval and adult weevils to ID parasitoids and calculate parasitism levels

Survey Results

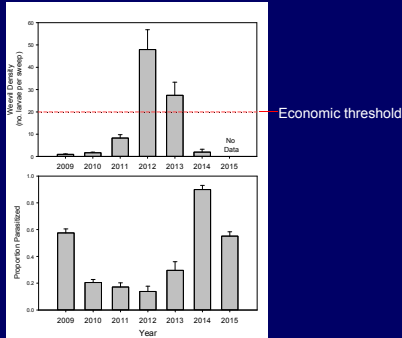


Oomyzus incertus

Bathyplectes curculionis

- Weevils present in every field every year surveyed
- Parasitoid present in >90% of fields surveyed
- 2 larval parasitoids recovered
- No Adult parasitoids recovered

Variability in alfalfa weevil numbers and parasitism through time

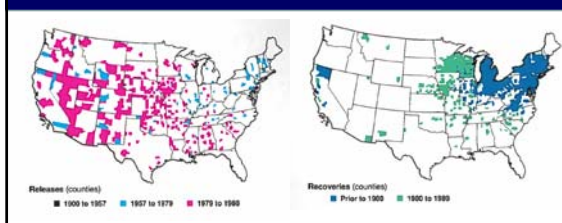


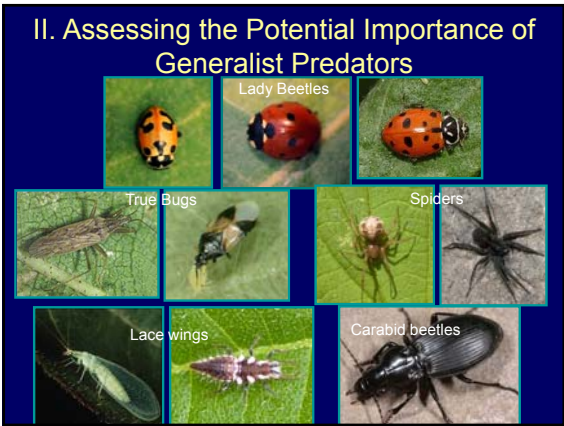
I. Conclusions

- Larval parasitoids of alfalfa weevil are common in the region, and parasitism pressure can be high in some sites and years
- Parasitoids seem to lag behind weevils, not keeping up in outbreak years
- Parasitoids of adult weevils are lacking, but might significantly complement the exiting parasitoid complex

Future Directions

- 4 State (MT, WY, ND, SD) survey for adult parasitoids
- Identify potential source populations in MN





Many predators described, lack of field based measures of impact (Barney & Armbrust 1981)

Table 1.—Insect predators of the alfalfa weevil, with information on the role of each predator

Species	Prey stage ^a	Importance ^b	Observation ^c	Reference
Hemiptera				
Nabidae				
<i>Nabis alternatus</i> Parshley	Larva	Minor	Both	19, 22, 15
<i>N. americanus</i> Carayon	Larva	Minor	Both	9, 22
<i>N. fervus</i> (L.)	Larva	Minor	Lab	15
Lygaeidae				
<i>Grocotus pallens pallens</i> (Stal)	Larva	Minor	Lab	9
Reduviidae				
<i>Sinea rhodema</i> (F.)	Larva	Minor	Field	12
Pentatomidae				
<i>Epidelus maculiventris</i> (Say)	Larva	Major	Field	22
<i>P. placidus</i> Uhler	Larva	Minor	Field	22
<i>Sternus anthracinus</i> (F.)	Larva	Uncertain	Both	18
Coleoptera				
Cicindelidae				
<i>Cicindela pusilla imperfecta</i> LeConte	Larva	Uncertain	Both	21
Carabidae				
<i>Pachymachus elongatus</i> LeConte	Adult	Minor	Both	3
Meloidae				
<i>Colletes bipunctatus</i> (Say)	Larva	Major	Both	21, 9, 15
<i>C. albicollis</i> Fall	Larva	Minor	Field	16
<i>C. quadrimaculatus</i> (F.)	Larva	Uncertain	Field	20
<i>C. vittatus</i> (Say)	Larva	Minor	Lab	15
Coleoidea				
<i>Colletes maculatus</i> (DeGeer)	Larva	Minor	Both	23, 22
<i>Hippodamia convergens</i> Guerin-Meneville	Egg, larva	Major	Both	21, 1, 16, 23, 9, 22, 1
<i>H. glaucalis</i> (F.)	Larva	Minor	Field	22
<i>H. parvithorax</i> (Say)	Larva	Minor	Lab	23



Study Goals

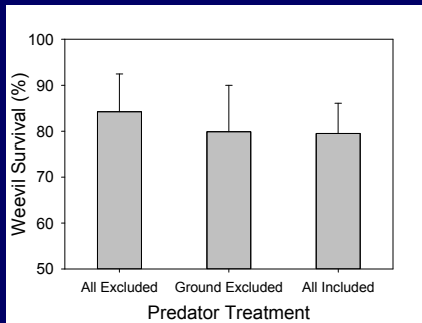
- Examine the potential impact of different enemy groups (ground vs. foliage foraging) on larval weevil survival and plant damage
- Predictions:
 - Predators will reduce weevil numbers and damage
 - Foliage foragers will have the greatest impact

Study Design

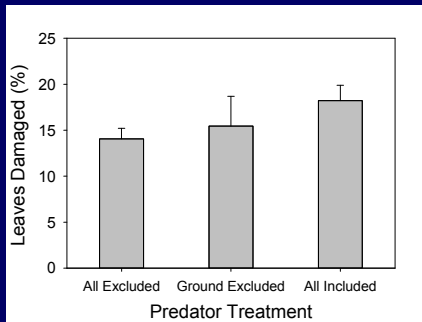


- 50 1st instar weevil larvae added to caged alfalfa plants
- 3 cage treatments imposed for ~3 weeks
- Responses: weevil survival, plant damage

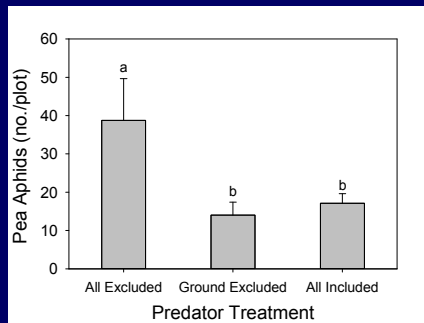
Predator Impacts on Weevil Survival



Predator Impacts on Plant Damage



Predator Impacts on Pea Aphids



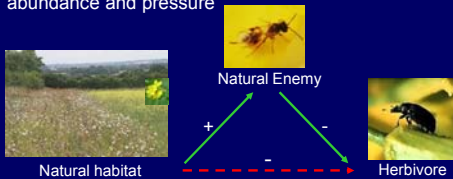
II. Conclusions

- Predators had minimal effects on alfalfa weevil larval survivorship and thus levels of plant damage
- In contrast, predators appear to have major effects on pea aphid densities
- Effects on alfalfa weevil may be higher at other sites or years
 - Quantify predator densities across sites
 - Repeat experiment in more years

III. Effects of field and landscape variables on alfalfa weevil numbers and biological control

Semi-natural habitats promote conservation biocontrol

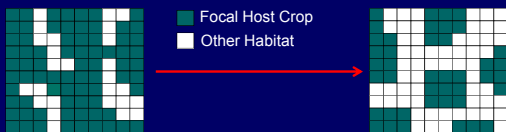
- Smaller crop fields, and higher edge-area ratios, increase enemy colonization from adjacent natural habitats promoting biocontrol
- Increasing natural habitat cover at landscape scales (500-3000 m) is associated with increased enemy diversity, abundance and pressure



– Landis et al (2000) Ann Rev Ent; Chaplin-Kramer et al (2011) Ecol Lett; Zumoffen et al (2012) Agr Ecosys Env; Veres et al (2013) Agr Ecosys Env

“Fragmentation” of host plant (e.g. focal crop) area can negatively impact specialized insect pests

- Reduced habitat area results in increased emigration and extinction, reduced colonization → reduced population densities
- Herbivore pressure declines in smaller, or more isolated, patches and with reductions in habitat cover at landscape scales



– Veres et al (2013) Agr Ecosys Env; Martinson and Fagan (2014) Ecol Lett; Rand et al. (2014) Agr Ecosys Env

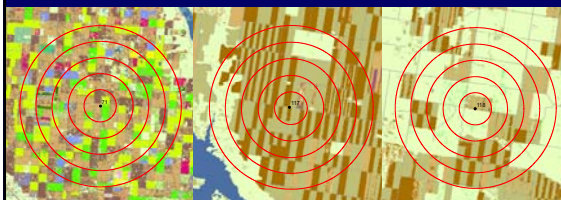
Landscape studies on pest control lack longer-term view:

- Lack of multi-year studies prevents our understanding of landscape effects on insect population dynamics and system stability

– Chaplin-Kramer et al (2011) Ecol Lett; Veres et al (2013) Agr Ecosys Env

- Landscape complexity is predicted to increase temporal stability in insect numbers and biological control

– Tscharrltke et al. (2012) Biol Rev; Zhao et al (2015) Basic and Appl Ecol



Slide 34

TR7 is the time required for an ecosystem to return to an equilibrium or steady-state following a perturbation (which is also defined as stability by some authors).

Tatyana Rand, 11/10/2015

Slide 37

TR9 Hypotheses to explain negative density area relationships

1. Lower predation rates
2. Lower emmigration rates
- 3.

Tatyana Rand, 11/9/2015

TR8 For each response variable, the y-axis represents residuals from a model removing effects of other significant predictors

Tatyana Rand, 11/13/2015

Slide 39

TR10 Tatyana Rand 11/6/2015

y-axis represents residual parasitism rate after removing effects of other block and significant predictors

Tatyana Rand, 11/9/2015

III. Results

- Increasing natural habitat cover was not significantly related to either mean levels of, or variability in, pest densities or parasitism across years
- Instead, crop pest and natural enemy dynamics were strongly linked with local and landscape patterns in the focal crop
- Increasing field size was associated with higher alfalfa weevil densities
- Increasing host crop area at landscape scales was associated with reduced weevil densities, reduced outbreak duration, and increased stability in parasitism



Final Summary

- Alfalfa weevil larval parasitoids are widely established, and likely play a role in reducing weevil numbers on average (parasitism levels exceeded the 30% critical threshold in 4 of 7 years surveyed to date)
- However, parasitism is negatively related to host density across sites and years (i.e. parasitoids can't keep up in areas or years of very high weevil density). Re-introducing additional parasitoids (e.g. those that attack adults) could be a solution
- Generalist predators do not appear to be important agents of mortality for alfalfa weevil larvae
- Planting designs that reduce individual field sizes and spread fields more evenly across the landscape could both reduce weevil numbers directly, and promote the stability of biological control

Acknowledgements

Funding: USDA-ARS

Field and Lab Assistance: Deb Waters, Amelia Jurkowska, Megan Greenwood, Alyssa Kessell, Jana Seright, Rhonda Lawhead, Joyce Wick, Anna Petroff, Lance Turner, Morgan Manger, Sarah Mason, Ellen Titus

Numerous alfalfa growers in the Yellowstone valley for permission to sample their fields over many years

**Biological Control of Orange Wheat Blossom Midge
(Diptera: Cecidomyiidae)**

Gadi V.P. Reddy, Brian Thompson, Dan Picard and Govinda Shrestha
Montana State University
Western Triangle Ag Research Center
9546 Old Shelby Rd., P. O. Box 656
Conrad, MT 59425, USA

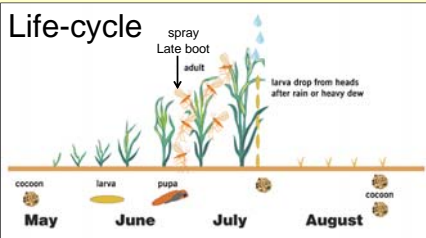


**Orange Wheat Blossom Midge, *Sitodiplosis mosellana*
(Diptera: Cecidomyiidae)**

- First Reported in Flathead County during 2006 and has continued to be a significant pest.
- MSU Extension Pondera County Agriculture and 4-H Agent have installed pheromone baited traps in wheat fields for the past 5 years to determine if midge was present in Pondera county.
- Positive ID made in 2011.
- During 2012, detected in irrigated spring wheat fields near Valier MT and in an irrigated field west of Conrad.

*Threshold = 1 midge/4 - 5 wheat heads
**Pesticide only recommended before crop reaches anthesis. After = No effect





* In Central Montana this life-cycle is shifted two weeks into late June early July.

Importation of *Macroglenes penetrans* (Hymenoptera: Pteromalidae) from Alberta, Canada for release into the environment of Montana

- In 2008 Northwestern Agricultural Research Center, MSU obtained *Macroglenes penetrans* from NDSU and released them at Kalispell.

Midge carries 4-105 eggs (mean 80) per female while *M. penetrans* carries an average of 205 eggs per female.

No sign of establishment until 2014.



2mm

4

OWBM Parasitoid Project

- In 2014, Team of Entomologists from WTARC, headed up a team to collect *M. penetrans* from the Lethbridge area. The collection was released at Conrad and Kalispell.



Biocontrol...

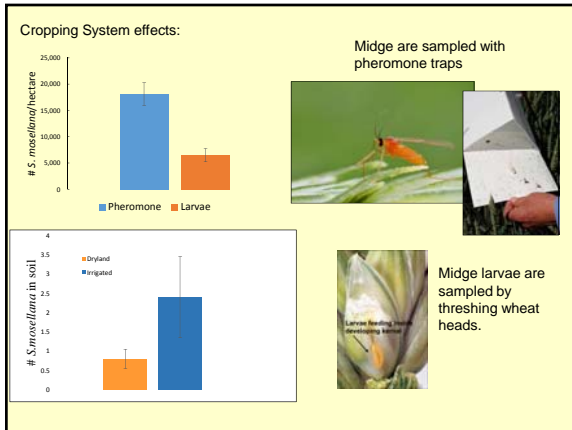
About 700 *M. penetrans* was collected from Alberta and released in Pondera and Flathead County in Montana on July 10, 2014.

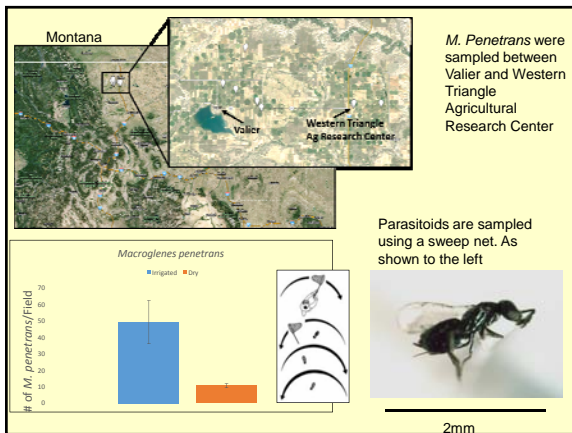
M. penetrans: 40 - 80% control

Another 20-40% control: Two parasitoids



Collection site Sweep net survey Release site





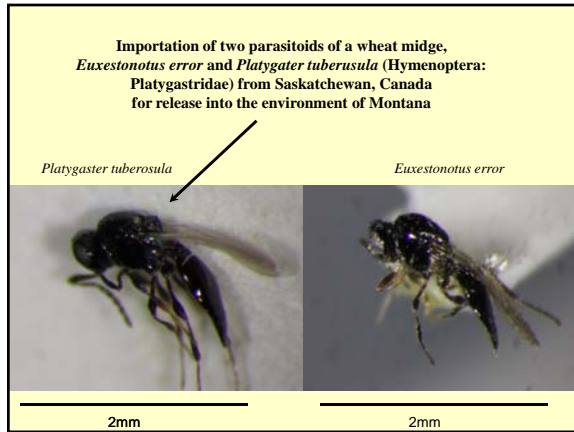
What is being done:

Monitoring = (pestweb.montana.edu)

Resistant wheat = (Egan)

Biological control = (*Macroglenes penetrans* &...)

Platygaster tuberosula and *Platygaster tuberosula* is a new parasitoid we are introducing to help *M. penetrans* control midge in Montana.

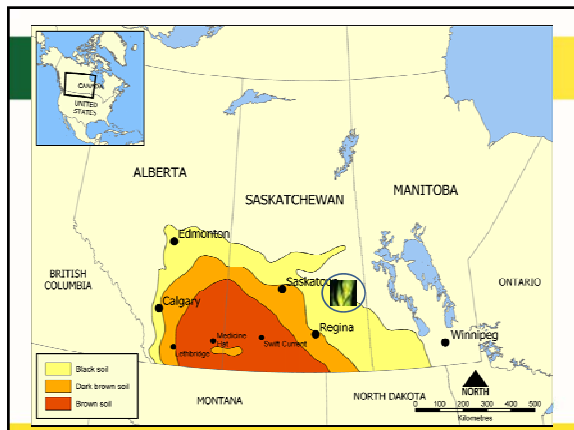


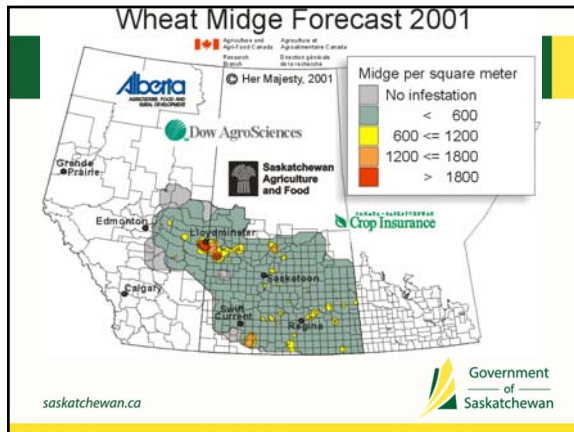
Wheat Midge Parasitism in Saskatchewan

S.A.R.E Workshop
Great Falls, Montana
March 2, 2016

Scott Hartley P.Ag.
Saskatchewan Ministry of Agriculture
Dr. Owen Olfert
Agriculture and Agri-Food Canada

saskatchewan.ca Government of Saskatchewan





Wheat Midge

- Wheat midge surveys were introduced in the late 1980s and expanded with the increasing distribution of the wheat midge.
- A survey is conducted annually, in the fall, to determine the density and distribution of wheat midge cocoons over-wintering in the soil in Saskatchewan.
- All cocoons are extracted and dissected to determine parasitism levels - parasitized larvae are deducted from total number of cocoons to estimate viable midge larvae.
- Only the viable midge are depicted on the forecast maps.

saskatchewan.ca Government of Saskatchewan

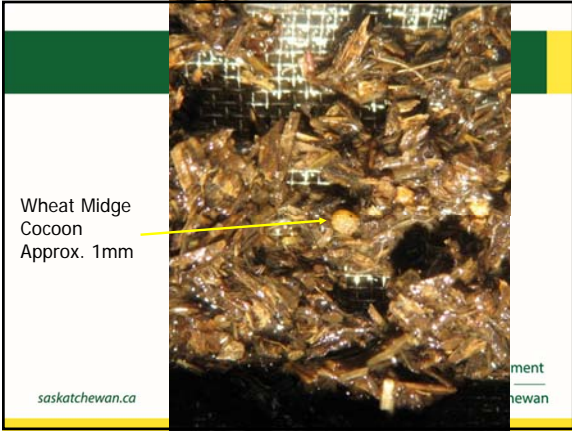
Wheat Midge Forecast 2016

Soil samples were collected in the fall of 2015, from 421 sites in Saskatchewan to produce the 2016 Forecast map.

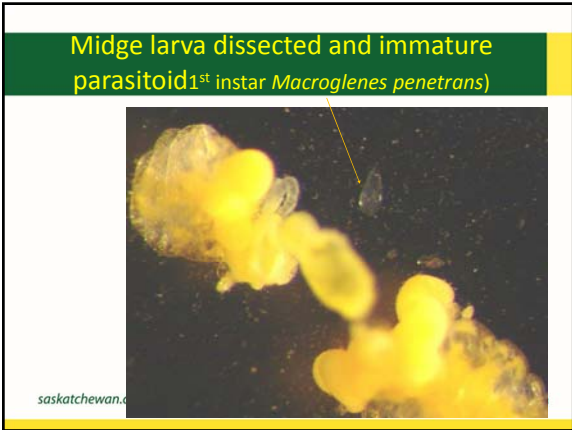
Midge / m²

- No infestation
- < 600
- 600 ≤ 1200
- 1200 ≤ 1800
- > 1800
- Not surveyed

saskatchewan.ca Government of Saskatchewan









Wheat Midge - Parasitism


- Adult wasp (native)
 - *Macroglenes penetrans* (female)
- Surveys : average parasitism
 - 1998 - 42%
 - 1999 - 36%
- Higher levels in established areas, lower in new areas




saskatchewan.ca 

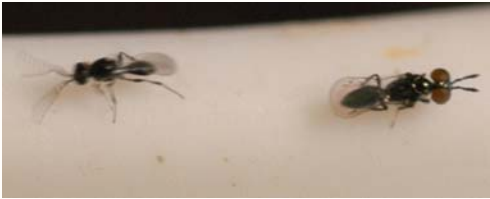
Wheat Midge - Parasitism

- *Macroglenes penetrans* (adult male)
 - a small 1 to 2 mm long parasitic wasp




saskatchewan.ca 

Parasites of Wheat Midge



Introduced species *Platygaster tuberosula* (left) native *M. penetrans* (right)
- actual size approx. 2 mm in length

saskatchewan.ca 

Wheat Midge Management

- Chemical Control
 - Insecticide application during advanced stages of flowering is discouraged for the following reasons:
 - By late flowering, wheat heads are no longer susceptible to damage (damage is less than 1%)
 - Treatments applied during advanced flowering are not effective because larvae have already hatched and caused damage
 - **Sprays are also discouraged during advanced flowering because it would have a negative impact on midge parasites**

saskatchewan.ca



Wheat Midge Parasitoid – Success Story



- ✓ *Macroglenes penetrans* discovered in SK in 1984
- ✓ *Platygaster tuberosula* and *Euxestonotus error* were introduced from Europe in 1990s
- ✓ Monitoring tool was developed to estimate populations of both pest and parasitoid and to track their expansions

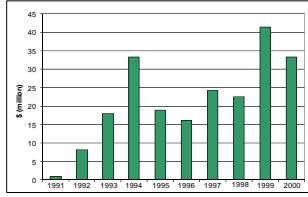
Benefits (\$) – Wheat Midge Biocontrol

Objective: To estimate the value of parasitoids we assessed the area of wheat production over 10 years that did not require an insecticide application

- ✓ Quantify areas where viable wheat midge cocoons in the soil were controlled by the parasitoid to below economic threshold levels (<600/m²)
- ✓ The study did not include environmental benefits nor increases in crop yield



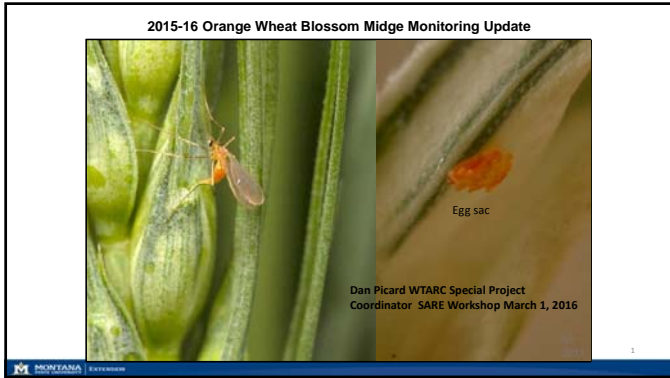
Farm Savings Due to Parasitoid

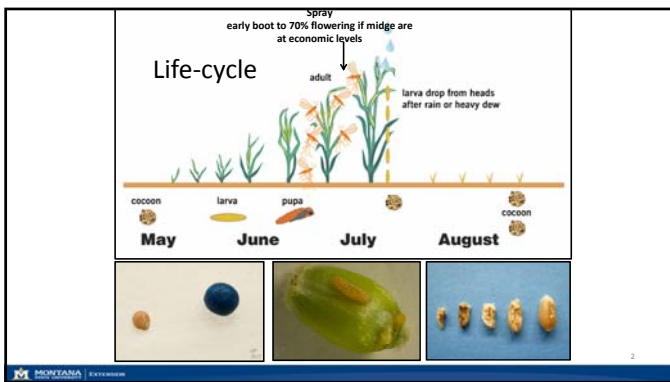


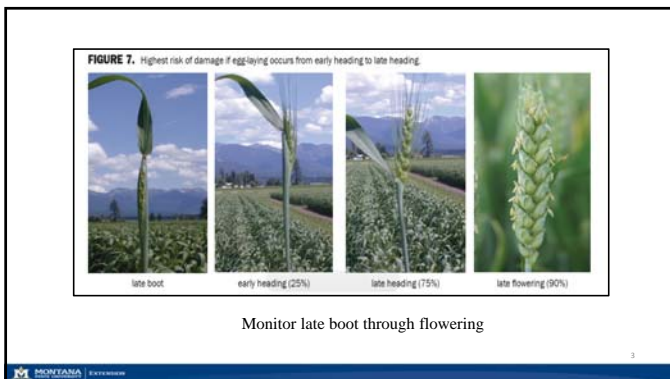
- ✓ 1991-2000, there were ~15.5 million ha of wheat that did not require a pesticide application because of parasitism
- ✓ At a cost of \$16.00/ha, the total saving in pesticide costs alone were over \$200 million in the 1990's

Acknowledgements Montana Wheat and Barley Committee









2016 Statewide OWBM Monitoring Program

- Presentations-speakers available on OWBM for various grower meetings, conventions, and field days.
- Pheromone traps/supplies are available at no cost to 8 Research Centers and 27 or more MSU Extension Agents.
- Additional supplies for Pheromone traps may be ordered through 3 Extension offices for growers and consultants.
- Detailed trap use, placement recommendations, and reporting record sheet will be handed out with each trap order.
- Cooperators were urged to report trap location and counts on Pestweb-0 is a number.

2015 OWBM Observations

- Midge starting flying on June 16th in Pondera County
- Peak emergence in Pondera County was at 1,149 degree days (base 40) on June 29th. Flathead was on June 23 at 1,196 degree days
- Traps with medium to high counts do not always correlate to economic midge levels being detected while scouting
- Currently, midge are being found primarily in northwestern, northcentral and northeastern counties.
- Counties, with midge detected, in one or more years, went from six in 2013 to twenty two in 2015 in part due to the expanded monitoring effort.
- White Delta Traps needed to painted green and put out by June 10th

Sm1 Gene-Midge Resistant Spring Wheat

- Dr. Luther Talbert, MSU Spring Wheat Breeder, is heading up the effort in Montana
- New spring wheat variety-Egan is available for growers to seed this spring. (District 1 only)
- Several lines with Sm1 plus solid stems are in yield trials-will take a couple of years
- Very important industry and growers maintain 90-10 refuge mix to maintain effectiveness of SM1 gene.

 Agriculture and Agri-Food Canada / Agriculture et Agroalimentaire Canada



Biological control of field crop insect pests in the Canadian Prairies.

Héctor Cárcamo

Lethbridge Research and Development Centre



Collaborators

- Arash Kheirodin
 - Current Ph. D. student with Alejandro Costamagna (U of MB)
- Catalina Fernandez (MSc student)
 - With Rob Laird (University of Lethbridge)
- Swaroop Kher, Meghan Vankosky
 - U of Alberta, With Lloyd Dossdall
- Vince Hervet
 - Master student (2010, France) with L. Dossdall, U. of Alberta
- John Gavloski, Manitoba Ag (Carman)

2


Outline

- Background
 - Cropping systems in the Canadian Prairies
 - Major insect pests
- 3 case studies
 - *Cereal leaf beetle*
 - *Lygus bugs*
 - *Sitona weevils (pea leaf weevil)*
- Future needs

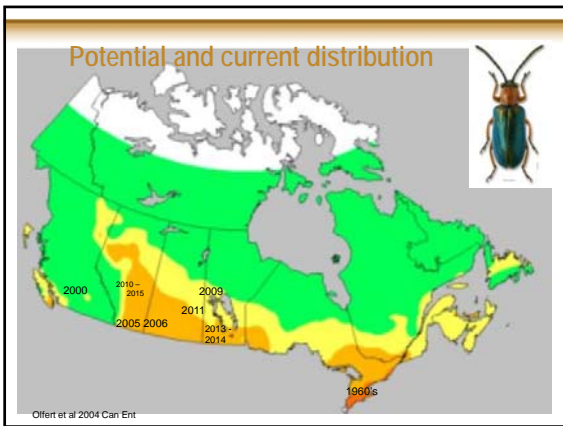
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Cereal leaf beetle

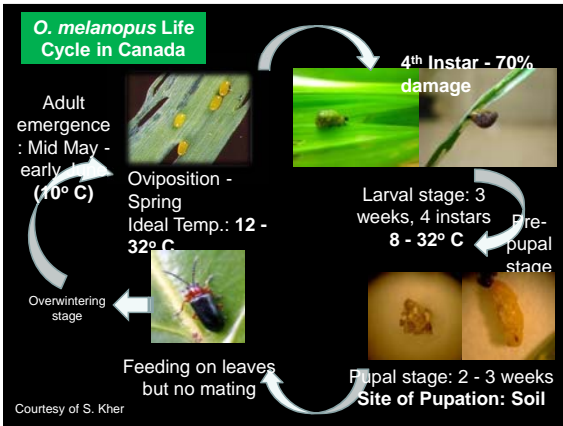
- Family: Chrysomelidae
 - Includes flea beetles, colorado potato beetle
- Genus: *Oulema*
- Species: *Oulema melanopus*
 - Only species of genus in Canada?
- From Eurasia
- Serious pest of most cultivated cereals in temperate world
- Quarantinable pest in Canadian Prairies up to 2009
 - Trade barrier, e.g. California



7





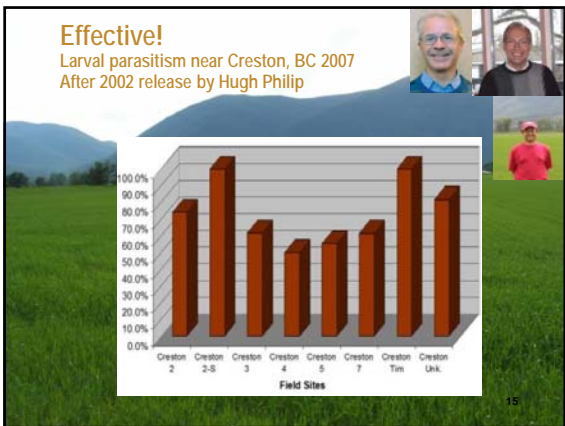


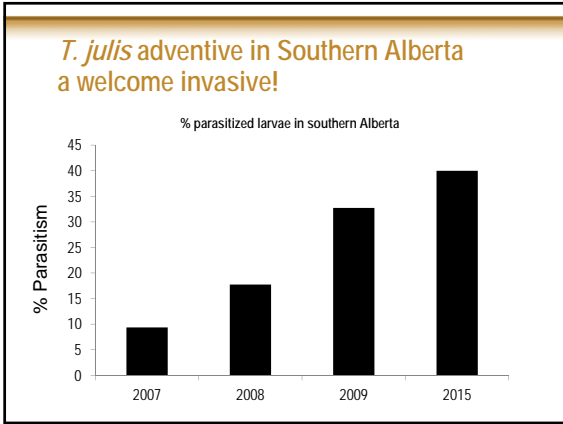








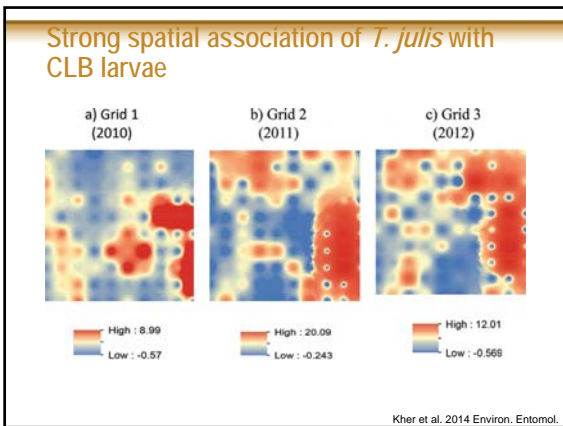




Host specific!

- Lily leaf beetle
- Twelve-spotted asparagus beetle
- Colorado potato beetle
- Three-lined potato beetle
- *Galerucella californiensis*
- *Gastrophysa polygoni*
- *Cassida azurea*


Hervet, V., Carcamo, H., Dossall, L., Kher, S. 2016. Biological Control. In Press



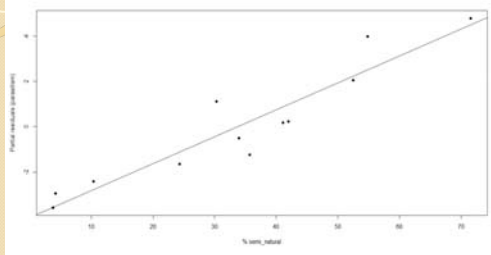
Landscape analysis

Arash and Alejandro – U of MB

- Select landscapes ranging from simple to complex.
- GPS to record geographical information
- Digital map by ARC MAP 10. proportion of different habitat and diversity of these habitats.
- Around 60 sites mapped (2014-15): CLB and *T. julis* data



Positive effects of semi natural habitats on parasitism rate (preliminary analysis)



Semi_natural consist of: native pasture, cultivated pasture, road ditches, riparian and trees.

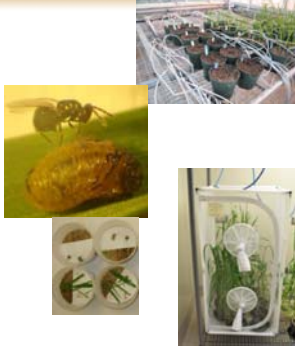
Relocation of *T. julis* throughout Prairies

- Enhance biological control of Cereal Leaf Beetle in the Prairies
 - Identify areas with beetle pest that lack parasitoid or have low levels of parasitism
 - Relocate *T. julis* to those areas

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Method


- Laboratory rearing
 - Expose CLB larvae to parasitoid and rear adult wasps



22

Method

- Field collection
 - Collections around southern AB
 - Focus on areas with high levels of parasitism
 - Rear adult wasps
 - Also second generation overwintering larvae in beetle cocoons



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Best way to get *T. julis* is to collect larvae in the field and rear in green house in large cage

source	CLB larvae	<i>T. julis</i>
Buckets in field	400	65
lab exposed larvae dishes	630	12
lab exposed larvae cages	500	300
field coll. Larvae dishes	450	926
field coll. Larvae - Cages in GH		8,770
Sweeps for <i>T. julis</i> adults		1,000
total	1980	11073

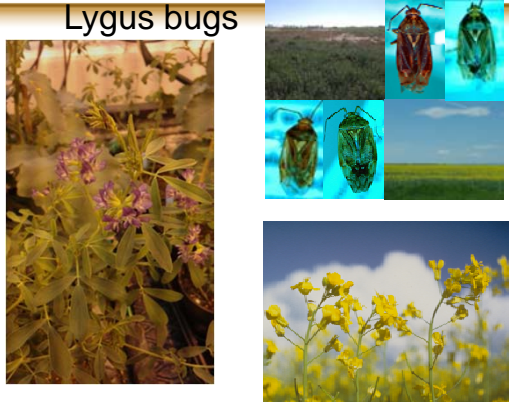
24

Future related work

- Dynamic Economic Threshold - including natural enemies to discourage spraying insecticides in fields with low damage
- Continue working with plant breeder to diversify IPM tool box


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Lygus bugs







Lygus management

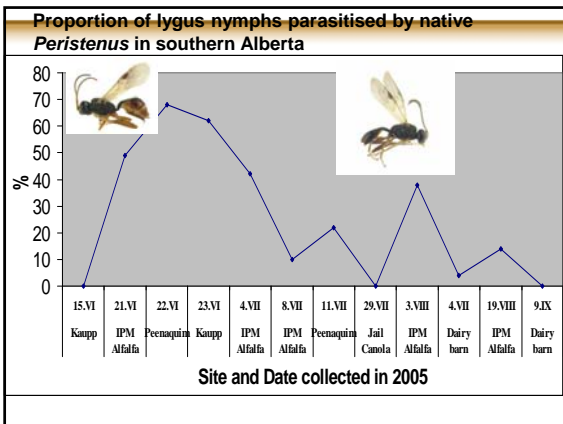


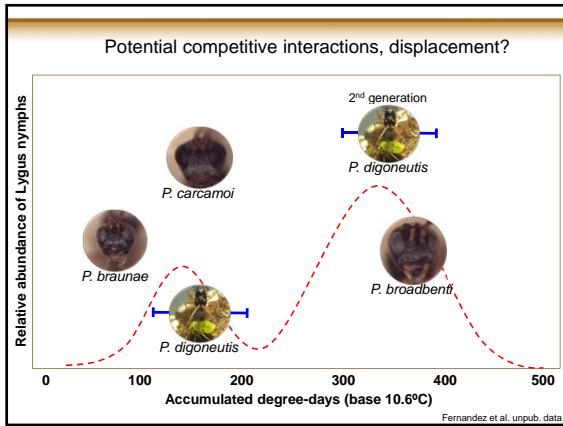
- Insecticides only
- Economic thresholds:
 - 1 per sweep at end of flower to early pod Or 2 per sweep at mid pod
- Biocontrol possibilities?
 - For a native pest?
 - Neo-classical biocontrol

Parasitoid effective in NE USA

- *Peristenus digoneutis* (European)
- Introduced to NE USA
- Reduced lygus in alfalfa and other crops
- Adventive migration to Ontario and Quebec
- Relocate west?






Introduction of an exotic wasp to control native lygus pests – a risky strategy?


- Competition lab studies in progress and ongoing field studies in Ontario (Mason & Lachance).
- Considered low risk in eastern Canada and North East USA.
- Benefits thought to outweigh risks in California (more aggressive *Peristenus stygicus* introduced there)

Pea Leaf Weevil



- Sitona (broad nosed weevils)
 - ~23 spp in Canada, native and introduced
- *Sitona lineatus* (native to Europe, N. Africa)
- Adult 5 mm long
- Light brown stripes extend to wing covers



- Larva milky white body and dark head
- grub-like, curved shape
- legless



Life Cycle in Alberta




Alfalfa & tree shelters	Pea fields	Pea roots	Pea field harvest
			
			
Sept-May	May-June-July	June-July-August	August-Sept

Host plants

- Feeding hosts
 - Adults feed on many leguminaceae (e.g. alfalfa, beans, clover, lentils, lupins, vetch) but generally don't cause economic damage
- Reproductive hosts
 - Peas
 - Faba beans

Damage- Adults

- Notching of leaf margins in June-July
- Plants can compensate

Field peas	
Faba beans	
Lupins	


Damage- larva

- Larvae feed on root nodules
- destroy *Rhizobium*



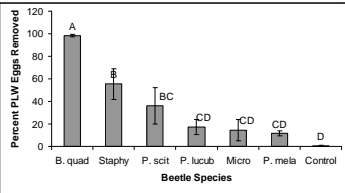
Biocontrol of *Sitona* species

- A few releases targeting sweet clover weevil or alfalfa weevil adults
 - Microctonus* and *Perilitus* (Braconidae)
- Parasitoids were reared on pea leaf weevil
- No parasitoids of eggs or adults found by Vankosky (2010)
- Extensive survey lacking
- Endemic predators...




Carcamo & Vankosky 2013

Egg predators of Pea Leaf Weevil



Beetle Species	Percent PLW Eggs Removed
B. quad	~100 (A)
Staphy	~55 (B)
P. scit	~35 (BC)
P. lucub	~15 (CD)
Micro	~15 (CD)
P. mela	~10 (CD)
Control	~5 (D)



Ground beetle – egg predator

Vankosky et al. 2010. J. Appl Entomol.

Conservation practices

- Practice Integrated Crop Management
 - Include **perennial** legume forages in **rotation** to build up beneficials
 - Select varieties or crops resistant to pests
 - Choose big seeds to get strong seedlings and uniform stand
- Spray only if absolutely necessary – i.e. follow economic thresholds
 - Means sampling for pest and keeping an eye on beneficial insects
- If beneficials abundant, give them a chance!
 - Choose products that are "softer" on beneficials,
 - e.g., microbial biopesticides if available, seed treatments,
 - Time spraying to avoid beneficials
 - Larval parasitoids active a little later than pests
 - Less active early am or late pm

Conservation practices

- Use a **trap crop** to concentrate pest
 - e.g., cabbage seedpod weevil
- **Less tillage**
 - Low soil disturbance key for cereal leaf beetle parasitoid survivorship
 - Not for all species!
- **Taller stubble**
 - Benefits sawfly parasitoid
- Some flowers in or near field may enhance beneficials by providing nectar fuel – **plant diversity** through intercropping
 - Variety blends or solid/hollow wheat may enhance sawfly parasitoids

Future



- Biocontrol research lags far behind other strategies such as chemical and host plant resistance
- Research needs
 - Inventory of beneficial arthropods (basic biodiversity studies)
 - Basic ecological information on seasonal activity, relationship of natural enemies with pest (impact studies) and plants
 - Habitat features that enhance their populations and efficacy
 - Integration into current or modified agronomic practice
 - Exotic natural enemies, other biocontrols such as microbial pesticides

Acknowledgements

- A-base/matching funds of AAFC
- Pest Management Centre, Insecticide Risk Reduction Program of AAFC
- Universities of Alberta, Lethbridge, Manitoba
- Alberta Crop Industry Development Fund, Canola Council of Canada, Alberta Canola Producers Commission, Alberta Pulse Growers Commission



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**Conservation Biological Control of Wheat Stem Sawfly:
Applying Agroecology to Enhance
Integrated Pest Management**

David K. Weaver¹, Scott B. Meers², Brian L. Beres³, Micaela Buteler⁴,
Megan L. Hofland¹, Justin B. Runyon⁵, Ryan J. Bixenmann⁶, Dayane A. Reis¹,
and Perry R. Miller¹

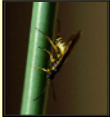
(1)Land Resources and Environmental Sciences, Montana State University, Bozeman, MT; (2)Crop Diversification Centre South, Alberta Agriculture and Rural Development, Brooks, AB, Canada; (3)Lethbridge Research Centre, Agriculture & Agri-Food Canada, Lethbridge, AB, Canada; (4)Instituto de Investigaciones en Biodiversidad y Medio Ambiente (INIBIOMA), Consejo Nacional de Investigaciones Cientificas y Tecnicas (CONICET), Bariloche Rio Negro, Argentina,(5)Rocky Mountain Research Station, USDA Forest Service, Bozeman, MT, (6) STEM Education and Communication, Government Relations, AAAS, Washington DC

OUTLINE

- **Background**
 - Distribution and Damage
 - Field and Crop Ecology
 - Attempts at Management
- **Endemic Native Parasitoids**
 - Characterize Benefits
 - Conservation Biocontrol in Fields
- **“Trap” Crops**
 - The “Trap” Idea
 - Benefits of Field Periphery
 - Source-Sink
 - The Return of Nectar
- **Summary**

Wheat Stem Sawflies

- Early members of Hymenoptera
- Cephidae - specialists on grasses
- Semi-arid regions
- Crop associations (*Cephus*, *Trachelus*)
- Diversity greatest at origin of wheat cultivation
- Occur in more than 50% of world wheat crops
- Exclusively in the Northern Hemisphere
- Estimated losses - > **\$2 billion** annually

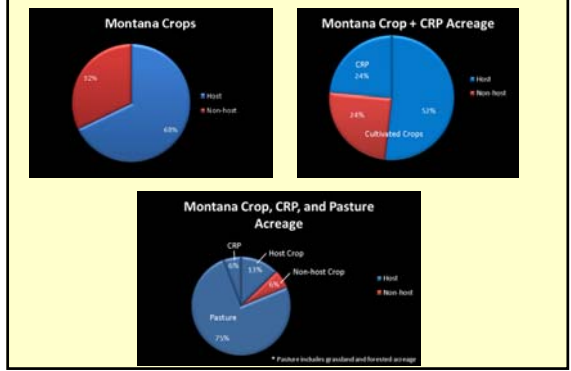


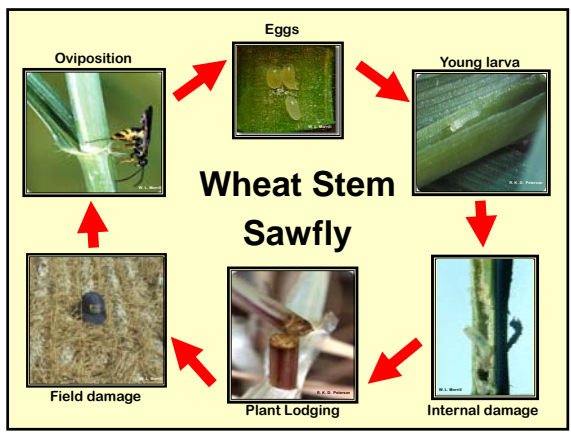
Cephus cinctus
(Norton)

Shortgrass Prairie & Alpine Meadows



Metapopulation





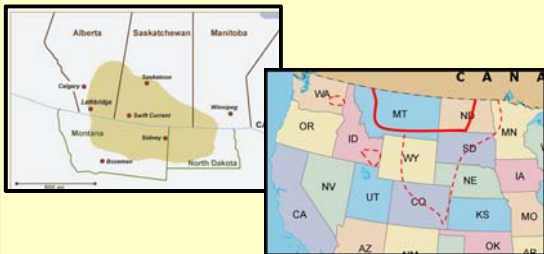
Data Collection



Losses in Montana
\$ 45 - 80 Million Annually
2008 - 2014



Population Expansion



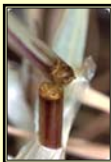
≈ \$350 million losses annually
Damaging populations since 1910

Damage

All immature stages are in the stem
All damage occurs within stem



Internal Feeding
10 - 20% Loss



Plant Lodging
10 -15% Loss (or more)



Greater Lodging
Compounds Losses

Lodging Challenges



- Decreased snow retention
- Slower harvest
 - harvest one way
 - more fuel; swathing
- Equipment damage
- *Extra equipment

- Volunteer wheat
 - uses water
 - herbicide



Management Attempts


Swathing to "Save" Crop



- Saves money in heavy infestation
- Continues or worsens infestation
- Money lost in kernel weight
- 10 - 30% with no correction
- *Reduces larval survival only with losses

*Goosey 1999

Tilling Residue



**Survival in exposed, bare crowns-
10 - 15%**


**Survival in soil associated crowns-
85 - 90%**

Goosey 1999

Burning Residue

**No appreciable mortality
in stubble**

**No change in mortality in
stubble with straw added
to increase fuel load**



Criddle 1907, Ainslie 1920

Natural Enemies

Braconid Parasitoids

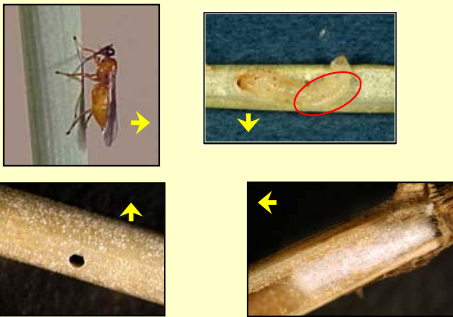


Bracon cephi

Bracon lissogaster

Sympatric, congeneric, bivoltine larval idiobionts
Specialists, uncertain ovigeny, egg maturation

Braconid Parasitoids



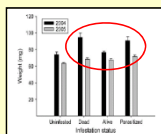
Braconid Parasitoids



Bracon lissogaster is also gregarious



Retain Yield



Buteler et al. 2008

Table 4. Correlative matrix of insect-related variables, grain yield, yield components, and grain protein in spring wheat grown in grass prone to wheat stem sawfly (WSS) infestation.

Variable	Stem infestations	Previous vegetation conditions						
		Rate of penetration by WSS	Stem dieback by WSS	Grain yield	Plant establishment	Spikes density	Spikes per plant	Grain protein
Stem infestations	1	-	-	-	-	-	-	-
Rate of penetration	-0.41	1	-	-	-	-	-	-
Stem dieback by WSS	0.77	0.23	1	-	-	-	-	-
Grain yield	-0.49	-0.33	-0.33	1	-	-	-	-
Plant establishment	-0.49	-0.42	-0.28	-0.42	1	-	-	-
Spikes density	-	-	-	0.71	0.70	1	-	-
Spikes per plant	-	-	-	-0.22	-0.83	0.30	1	-
Grain protein	-	-	-	-	-	-	-	1

† ** P < 0.05, all other values presented at P < 0.01.

Beres et al. 2011

MONTANA STATE UNIVERSITY EXTENSION **PESTICIDE NEWS**

Thimet 20-G[®] for Wheat Stem Sawfly Management
 Dr. Kevin Wanner, MSU Extension Specialist, Crop Entomology
 Cecil Tharp, MSU Extension Pesticide Education Specialist

RESTRICTED USE PESTICIDE
 Due to acute oral, dermal and inhalation toxicity and avian hazards
 For retail sale to and use only by Certified Applicators or persons under their direct supervision,
 and only for those uses covered by the Certified Applicator's certification

THIMET[®] 20-G
 Lock 'n Load[®] Closed Loading System

EPA Reg. No. **5481-530**
 EPA SLN No. **MT-150001**

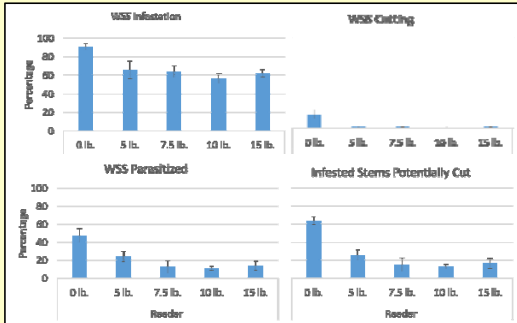
FIFRA 24(c) SPECIAL LOCAL NEED LABELING
FOR USE ON WHEAT IN MONTANA TO CONTROL WHEAT STEM SAWFLY LARVAE

MONTANA STATE UNIVERSITY EXTENSION **PESTICIDE NEWS**

Thimet 20-G[®] for Wheat Stem Sawfly Management
 Dr. Kevin Wanner, MSU Extension Specialist, Crop Entomology
 Cecil Tharp, MSU Extension Pesticide Education Specialist

- Apply at 5 lb acre - granular, post-emergence
 - Soil incorporated at 1 inch below surface
 - 85 day pre-harvest interval (critical)
 - Systemic, post-emergence timing
-
- Yield recovery 5.4 bu (spring) – 6.1 bu (winter)
 - 64 - 100% reduction in stem cutting
 - This is due to combined parasitoids and product

INSECTICIDE



Parasitoid Conservation



Zero-till - Herbicide



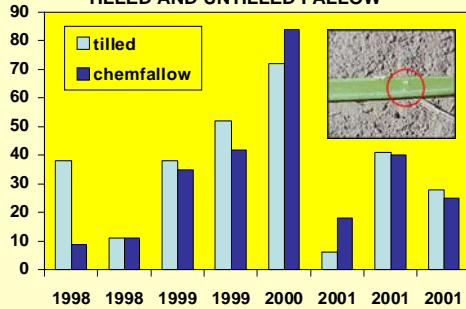
Light Tillage



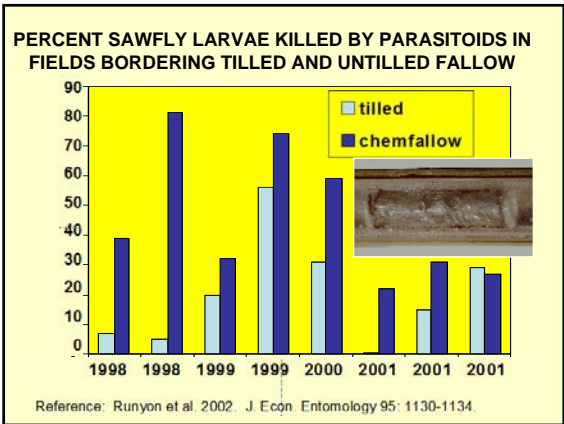
Heavy Tillage

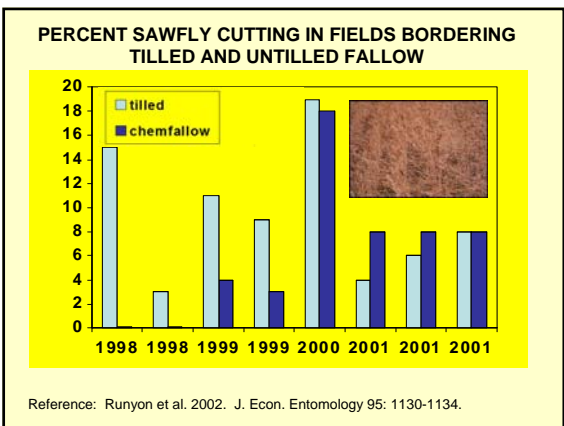


**SAWFLY INFESTATION IN FIELDS BORDERING
TILLED AND UNTILLED FALLOW**





Reference: Runyon et al. 2002. J. Econ. Entomology 95: 1130-1134.







Impact of Tillage

- Heavy tillage has more impact on parasitoids than sawflies
 - Sawfly overwinter in underground stubs – adapted to emerge from soil
 - Parasitoids overwinter in above-ground part of stems – not adapted to emerge from soil

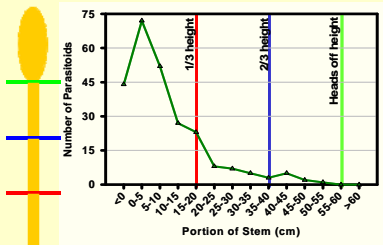
Impact of Harvest

- Where are the parasitoids in the stem interior?
- What is the impact of cutting heights?

Parasitoid Distribution

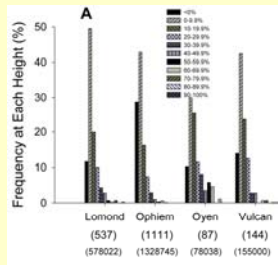
14,556 stems, 63% infested
31% parasitized*



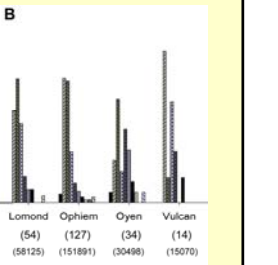
*Data collected from spring wheat fields in Alberta and Montana over 2 years. Image is for one field.

Parasitoid Distribution - 2002

A



B

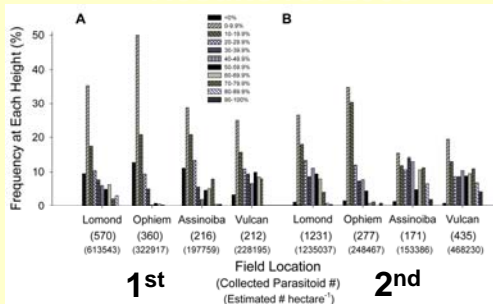


1st

Field Location
(Collected Parasitoid #)
(Estimated # hectare)

2nd

Parasitoid Distribution - 2003

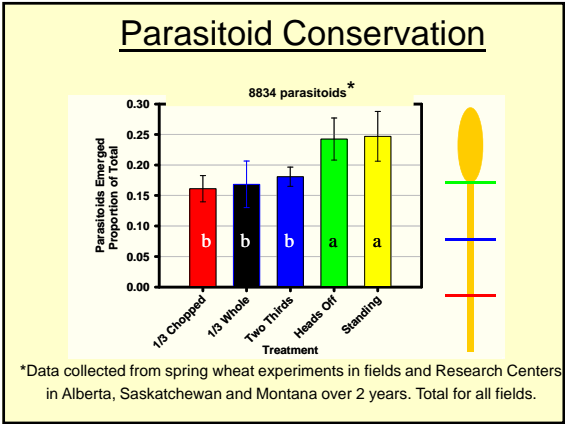


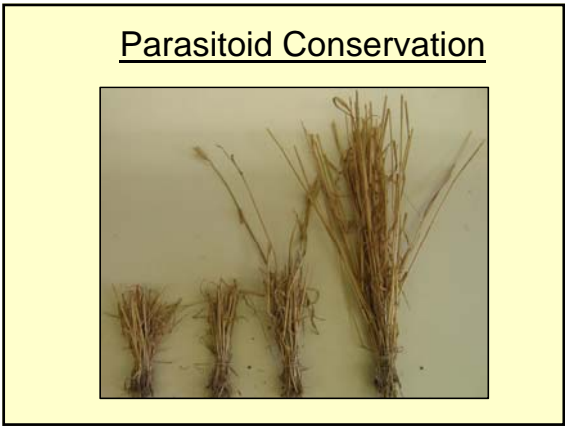
Parasitoid Conservation

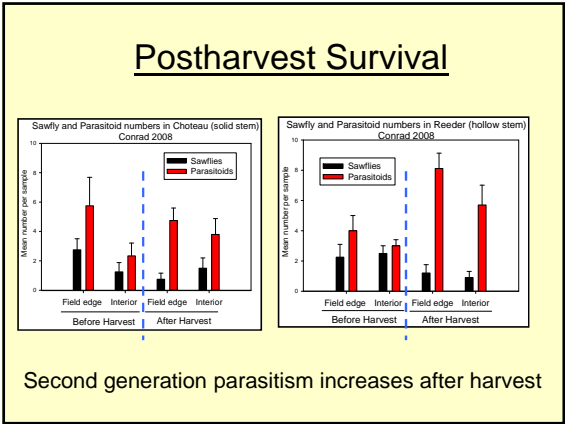


Parasitoid Conservation










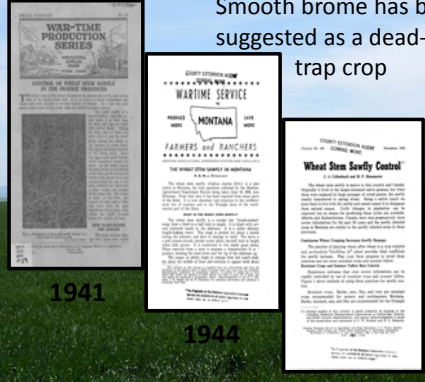
“Trap” Crops

Smooth Brome (*Bromus inermis*)

- Non-native, perennial, cool-season, bunch grass
- Stems 30-130 cm tall and 1-5mm at first node
- Common along roadsides and disturbed areas throughout North America



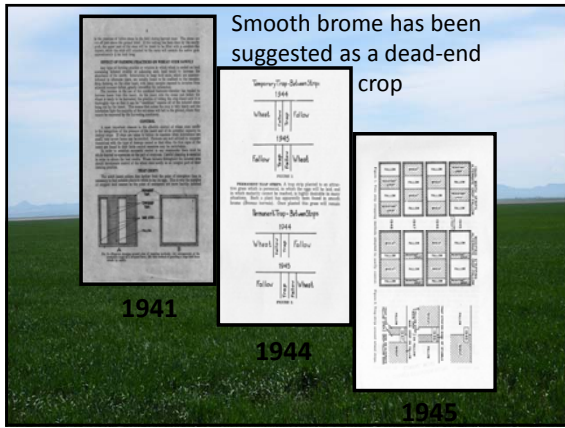
Smooth brome has been suggested as a dead-end trap crop

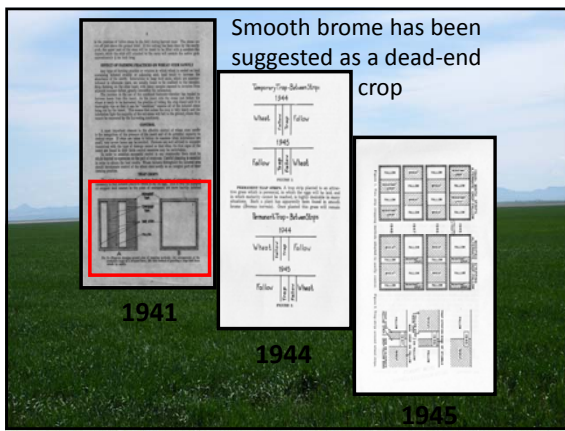


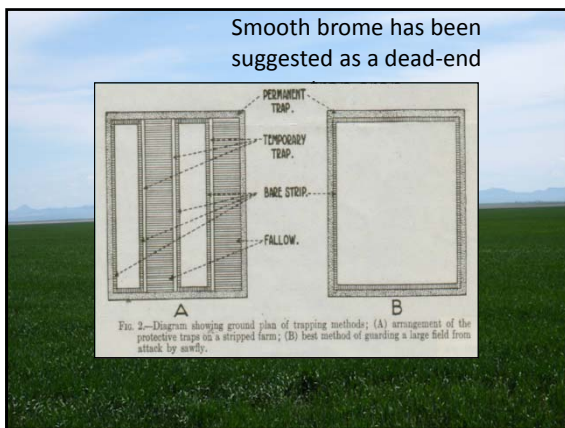
1941

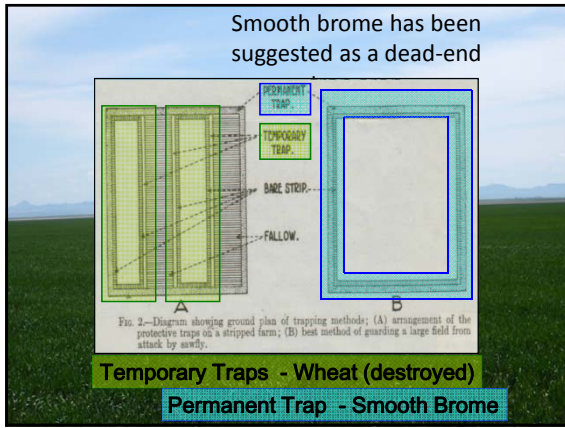
1944

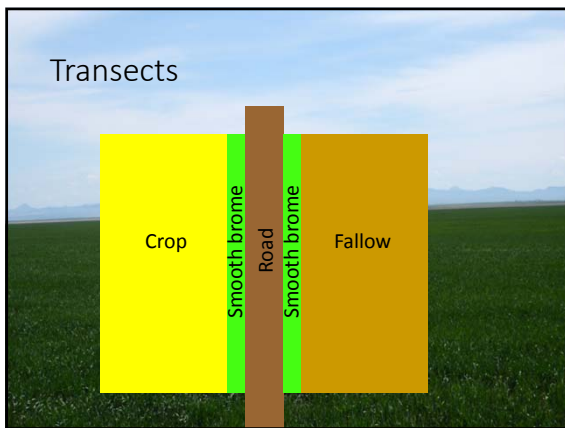
1945

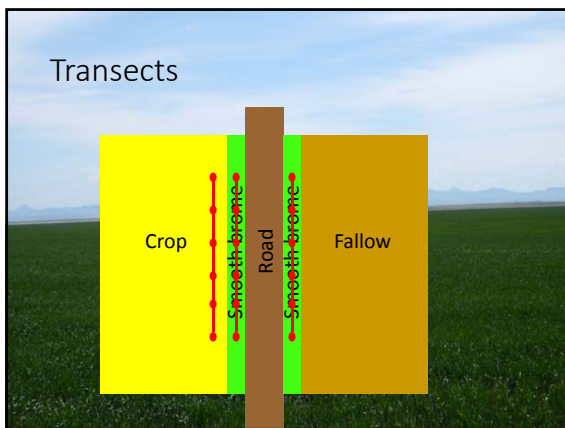


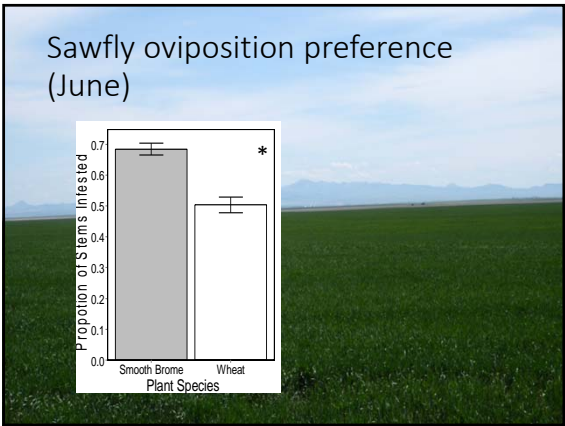


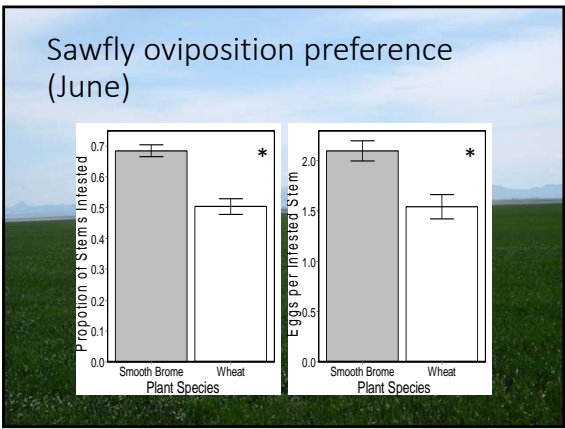


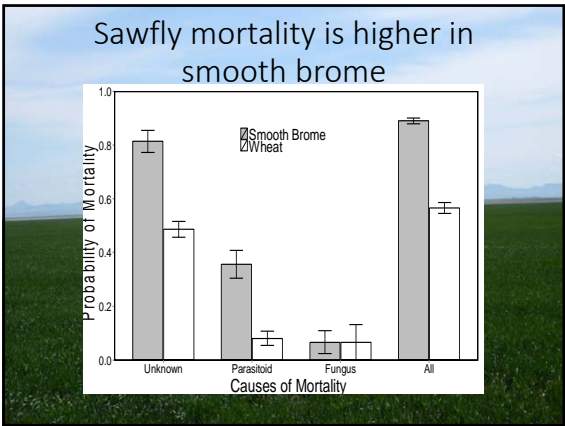


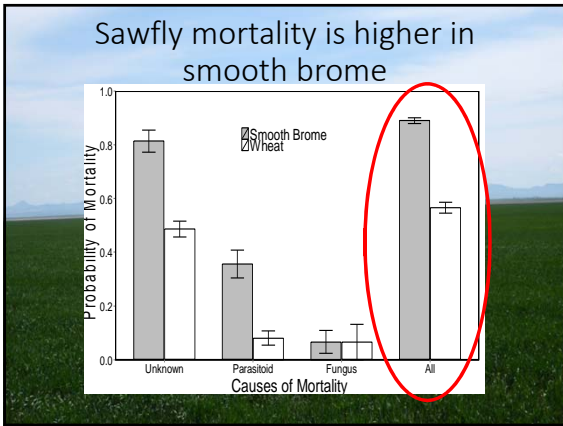


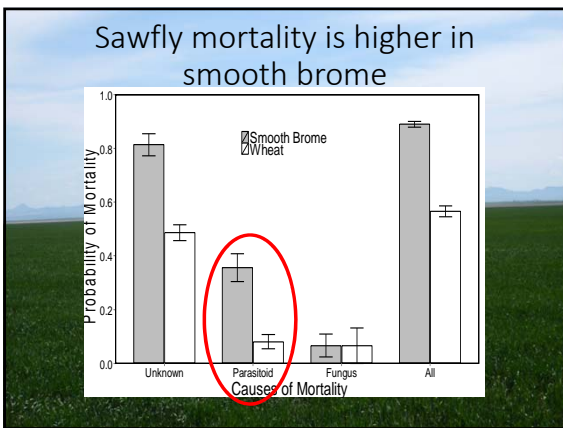


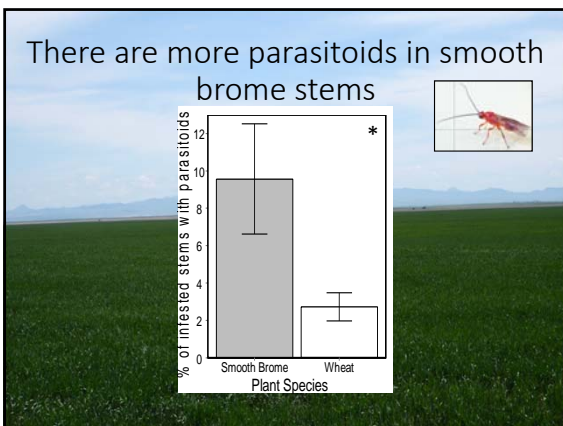


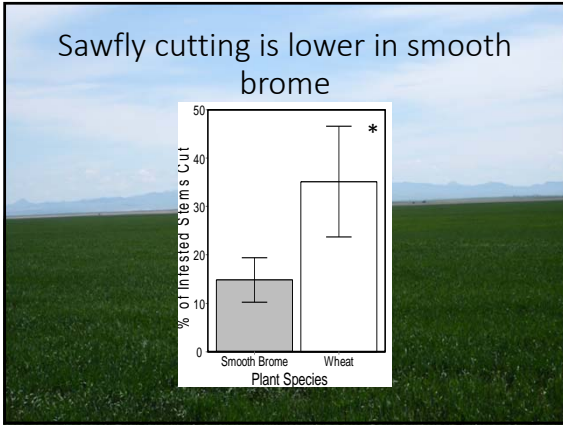












Smooth brome is a source of parasitoids and a sink for wheat stem sawflies.

A grass periphery can have greater value than expected, even surrounding a wheat field

Trap Crops

Terms Used

Antixenosis – unattractive
(not recognized or repellent)

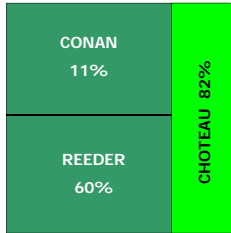
Antibiosis – causing mortality
(typically chemically-based)

Antibiosis and Antixenosis

Sawfly Control

Sawfly Life Cycle

Sawfly Trap



Antixenosis is rare

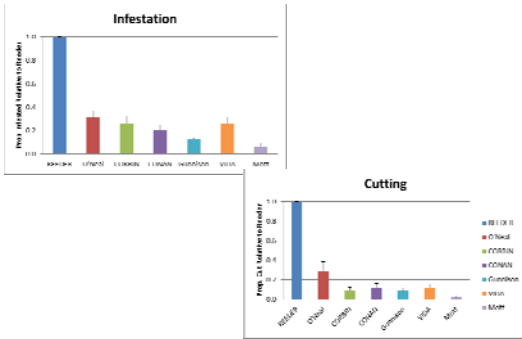
Grower Trap Crops



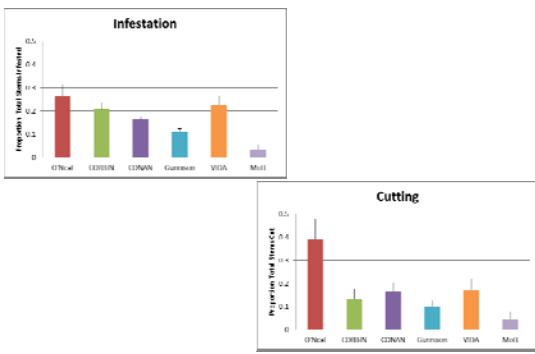
Hill Plots

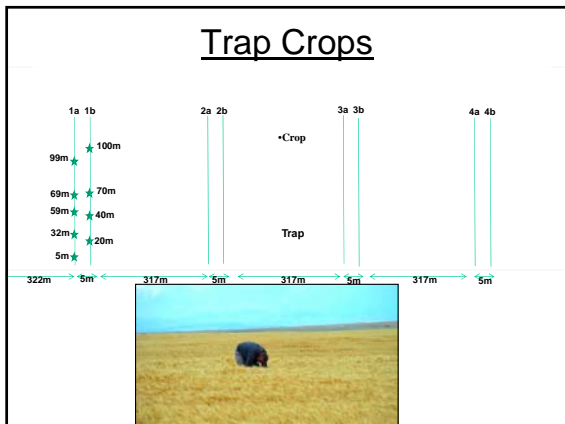


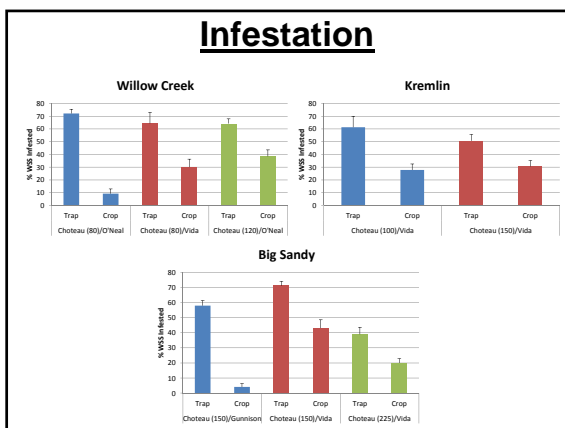
Unattractive Spring Wheat Varieties

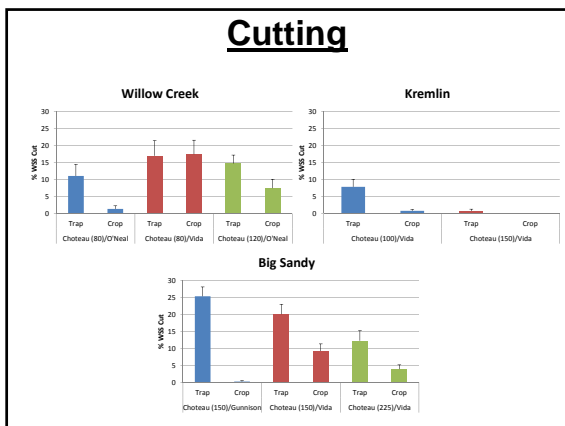


Unattractive Variety Comparison







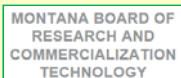


Summary

1. Native parasitoids and host pest species have moved into cultivated wheat in Montana
2. This represents a change from perennial to annual monoculture vegetation that cannot be restored
3. Strategies that further sustainability and resilience can be applied to conservation biological control of wheat stem sawfly by native species
4. Considerable complexity exists that must be understood by ecological observation and experimentation
5. Demonstrating an immediate benefit in yield from parasitism events is compelling to growers
6. New crop rotations, cover crops and attention to ecosystem services promise continued gains in IPM

Acknowledgments

Funds supporting this research were provided by competitive and earmarked State, Regional, and Federal sources, in addition to the Montana Ag Experiment Station.



The projects described above involve a number of researchers, graduate students, skilled laboratory staff, and many undergraduate student laborers. Implementation of the research relies heavily on collaboration across research disciplines, plus critical support from Ag Research Centers, County Extension Personnel, and numerous wheat growers statewide.

Questions?



Phyllobaenus dubius

Incorporating classical biological control into integrated management of invasive weeds

Sharlene E. Sing

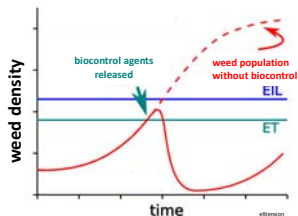
Western SARE Professional Development Program Workshop
Best Western Plus Heritage Inn – Great Falls, MT
March 1-2, 2016

What makes it classical biological control?

- the target weed is **not** a native species
 - weed’s native range is on another **continent** - *not* North America
 - weed was moved **intentionally** or **accidentally** from native range
 - weed became **established** and then **invasive** in North America
- weed’s natural enemies are also **not native** to North America

Goal of weed biocontrol:

- same rationale as for all weed control approaches - to safely suppress weed population below an **economic** or **ecological** threshold



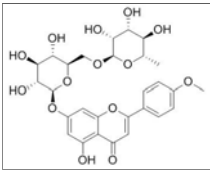
Goal of weed biocontrol:

NOT

- growing lots of bugs on weeds
- causing **cosmetic** damage only to weeds
- impacting only individual weeds

Why use foreign bugs?

- all plants produce **phytoprotectants** - biologically active and often toxic compounds, to protect themselves from predation or infection



linarin

Why use foreign bugs?

- all plants produce **phytoprotectants** - biologically active and often toxic compounds, to protect themselves from predation
- insect herbivores evolve adaptations through selection or spontaneous mutation
- this allows them to withstand the negative effects of ingesting or contacting phytoprotectants

Why use foreign bugs?

- adaptation is very 'costly' so insect herbivores often specialize on only a few related plant species
- adaptation occurs through lengthy host plant - herbivore association in their shared native range

Why use foreign bugs?

- the target weed's native range is often the best place to find candidate biocontrol agents that:
 - **want to** eat/use the weed
 - **can safely** eat/use the weed
- this chemistry is also the basis of **host specificity**:
 - degree to which a potential biocontrol agent is restricted in the number of plant hosts utilized

Integrating biocontrol into weed management programs:

I. determine if weed biocontrol will complement land use activities and management objectives for the affected area

– use the following questions to help determine if biocontrol will be appropriate:

Integrating biocontrol into weed management programs:

- 1. What are the short- and long-term land use objectives for the affected area, and how have they changed?**
 - did the weed infestation follow a significant change in land use:
 - resource extraction – grazing vs. timber harvest vs. gas/oil wells vs. mining?
 - road or other construction?
- 2. How urgent is the weed problem?**
 - long standing, chronic infestations of established, widespread weed species more suited to biocontrol than
 - new invaders (Priority 1A, 1B)
 - species invading previously uncolonized areas (Priority 2A)
 - small-sized or isolated infestations of well established weed species (Priority 2B)
- 3. How tolerant will neighbors or the public be if target weed abundance is slow to decline?**

Integrating biocontrol into weed management programs:

II. determine the scope of the weed problem

1. develop a weed distribution map at a scale that allows you to address the problem in a manner consistent with your overall land management objectives and your available weed management resources
2. in large landscapes with significant weed infestations and limited mapping resources, identify priority areas for additional survey and weed management efforts

Integrating biocontrol into weed management programs:

III. define overall goals of the IWM program

- **goals** broadly define the “what” or desired outcome of management
 - goals state general land use decisions or targets over large areas and/or extended periods of time
 - manage for something specific, instead of generically against weeds
- **objectives** define the “how” or specific activities through which desired outcomes can be achieved
 - objectives must be SMART: specific, measurable, achievable, realistic, and timely
- by defining what you want to achieve, you will be able to determine if, when, and where you can integrate biological control

Integrating biocontrol into weed management programs:

IV. understand the control methods available for accomplishing your IWM goals

1. review available weed control methods (biological control, physical treatments, cultural practices, and herbicides)
 - determine the conditions (when, where, if, etc.) under which it might be appropriate to use each method or combination of methods
 - be realistic about control method benefits and limitations
2. identify resources that will be available for weed management activities
 - determine if they will be consistently available until you meet your weed management program objectives

Biocontrol advantages

- selective
- sustainable - agents generally do not have to be reintroduced once established
- public acceptance is generally higher than with other weed control methods
- most economical option for large infestations

Biocontrol disadvantages

- perceivable changes in target weed density are slow, especially for showy (or particularly annoying) weed species
- population level impacts of biocontrol on the target weed, not to mention effects of biocontrol on the wider plant community, are often difficult to detect through observation, and complicated to measure
- some risk of undesirable effects on nontarget plants
- permanent; cannot be undone
- not successful in all situations
- treatment efficacy is not predictable

Interaction of biocontrol with other weed control methods:

- **Physical treatments:**
 - hand pulling, mowing, tilling, etc.
 - not recommended for weed species that reproduce from stem or root fragments, or if germination increases following disturbance
 - labor intensive, difficult on rough terrain
 - removes or destroys agents developing in weed stems or roots
 - removes stage-specific food resources of agents
 - biocontrol can be applied to large, main infestations while physical treatments can be used on surrounding small, satellite weed populations
 - mowing can be compatible with agents that develop in the roots of the target weed

Interaction of biocontrol with other weed control methods:

- **Cultural control:**
 - flooding, burning, grazing, seeding with competitive species, etc.
 - not recommended for weed species adapted to specific cultural control methods
 - fire adapted species: toadflax
 - flood adapted species: saltcedar
 - grazing can remove or destroy agents developing in weed stems, flowers or seeds
 - may not have a negative impact on root dwelling agents
 - compatible with bison grazing (selective grass feeders)
 - strategically timed grazing can increase attractiveness of target weeds to biocontrol agents, and increase competitive ability of desirable plant species

Interaction of biocontrol with other weed control methods:

- **Chemical control:**
 - most effective on small infestations, including newly established populations and recently established satellite patches arising from nearby older, larger infestations
 - may also be useful on the leading edge of large, advancing infestations
 - often too costly to be of practical use in treating extensive infestations
 - impractical in hard-to-access and environmentally sensitive areas
 - repeated applications may be required over time
 - potential for nontarget damage to associated vegetation

Interaction of biocontrol with other weed control methods:

- **Chemical control:**
 - all approved classical biological control agents are host specific
 - herbicide treatments reduce the local availability of target weed stems, leaves, and flowers
 - agents relying on food and shelter resources provided by affected weeds may not survive if herbicides are applied when they are unable to move on to hosts in an untreated area
 - chemical and biological control can be successfully integrated when agents are released on large infestations and herbicides are applied to control smaller satellite patches
 - herbicide can be used to reduce dense above ground biomass to make weed patches more hospitable for biocontrol agents

Developing, implementing, and managing IWM programs:

I. selecting appropriate release sites

1. infestations of at least 4 acres (1.6 hectares) are typically the minimum size recommended for biological control releases - larger infestations are even more desirable
2. consider the site's ease of accessibility, terrain, and slope
3. consider land use and rare vs. chronic disturbance factors
4. survey for presence of biological control agents

Developing, implementing, and managing IWM programs:

II. select appropriate biocontrol agents to release

1. agent efficacy
 - what will work well under your field conditions?
 - are your weed patches seasonally flooded?
 - under snow until mid or late July?
 - subject to heavy wildlife or livestock grazing at 'sensitive' points in agent's life cycle?
2. agent availability
 - do you know where you can get high quality agents?
3. match weed distribution and density to preferences of agent(s)
 - are your weed patches too sparse, too dense, or too far apart?

Developing, implementing, and managing IWM programs:

III. documenting releases

- permanently mark release sites
- record GPS coordinates
- generate a map and written directions to release site
- select a photo point with a unique, permanent landmark in the background
 - used to visually document changes in weed infestations and the plant community over time following the release of biocontrol agents
 - avoid capturing images that mostly show only the target weed without reference points that can identify specific release sites!

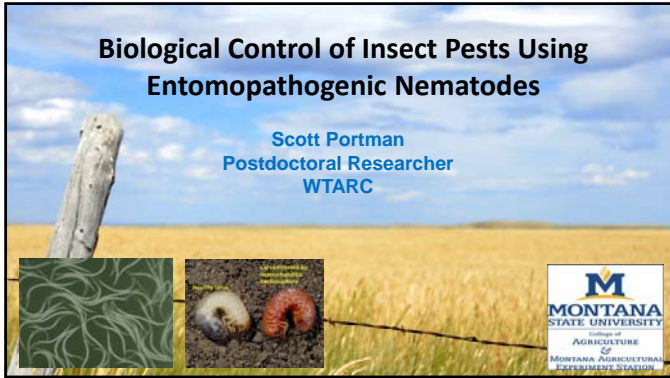
Developing, implementing, and managing IWM programs:

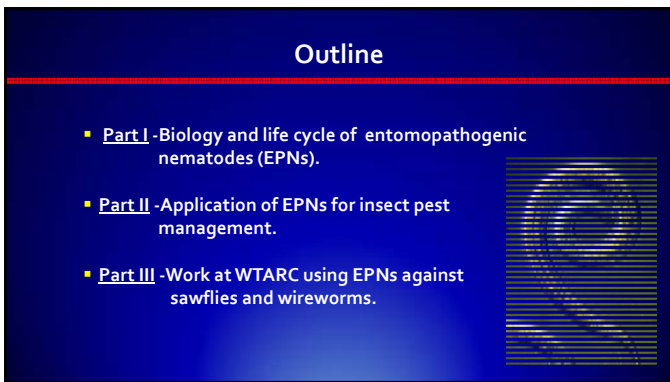
IV. monitoring the success of the program

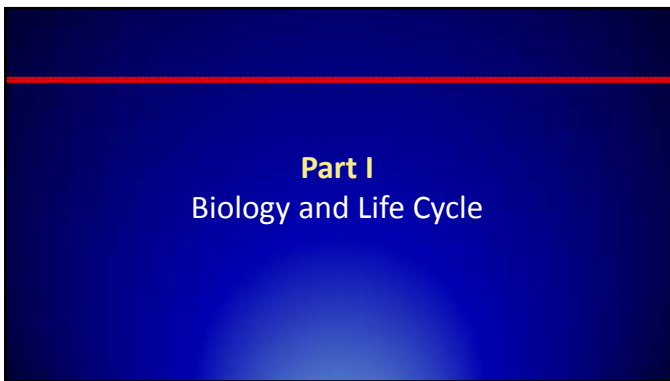
- field observations of agent specific damage on the target weed, annual photo points of the release site, or simple monitoring transects to assess year-to year changes in weed and agent populations, and plant community, to determine:
 1. where is it working?
 2. are unanticipated ecological interactions positively or negatively affecting control?
 3. where is establishment or realized control lower than expected, and why?

Standardized Impact Monitoring Protocol (SIMP)

- systematic monitoring approach to assess changes in the densities of both weed biocontrol agents and the target weed
- can be easily modified to meet personal or agency needs for monitoring most weed biocontrol releases
- Available online at: http://www.agri.state.id.us/AGRI/Categories/PlantsInsects/NoxiousWeeds/Bio_Control.php







Nematode trophic ecology



insect pathogenic vs. plant pathogenic

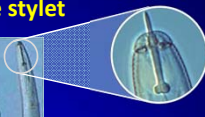
Plant pathogenic nematodes have stylet



Entomopathogenic



Phytopathogenic



Close-up of stylet

What are EPNs?

- EPNs are round worms -Phylum Nematoda
- EPNs are microscopic.
- Inhabit a variety of soil environments.
- Exclusively prey on insects.

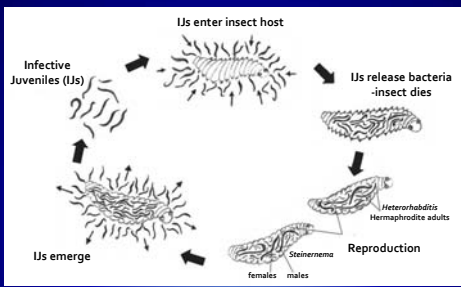


Family: Heterorhabditidae

Genus: *Heterorhabditis*

- 1. *H. bacteriophora*
- 2. *H. megidis*
- 3. *H. zealandica*
- 4. *H. indica*
- 5. *H. argentinensis*
- 6. *H. brevicaudis*
- 7. *H. marelatus*
- 8. *H. poinari*
- 9. *H. downesi*

EPN Life Cycle

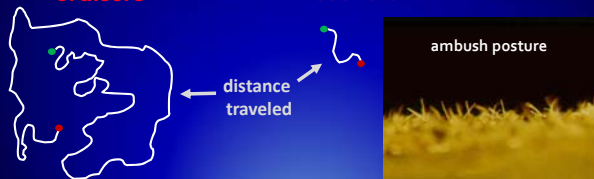


Host finding

Infective juvenile movement

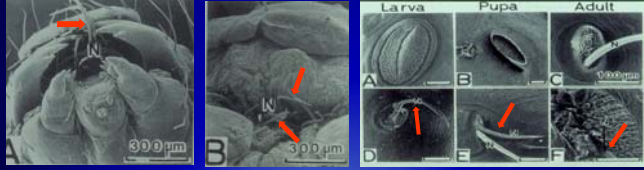
Cruisers

Ambushers



Getting inside the insect

Typically EPNS enter in one of 3 locations




Mouth Anus Larva Pupa Adult

Spiracles

Detailed description: This slide features a composite of scanning electron micrographs (SEMs). On the left, a close-up of an insect's mouthparts shows a red arrow pointing to a potential entry point. In the center, another SEM shows the anus with a red arrow. On the right, a grid of six images (A-F) shows the entry of EPNS through spiracles in larval, pupal, and adult stages, with red arrows indicating the entry points. Scale bars of 300 μm are present in the mouth and anus images.

Getting inside the insect

EPNs can also exploit weak points in the cuticle



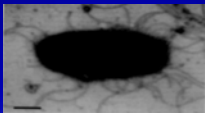
Detailed description: Two scanning electron micrographs (SEMs) show the insect cuticle covered in fine hairs. Red arrows in both images point to specific locations where EPNs are seen exploiting weak points or gaps in the cuticle for entry.

Symbiotic bacteria

EPNs form mutualistic symbiosis with specific bacteria

Steinernema sp. *Heterorhabditis sp.*

Xenorhabdus *Photorhabdus*



Enterobacteriaceae, Gram negative

Detailed description: This slide illustrates the mutualistic symbiosis between EPNs and specific bacteria. It shows *Steinernema sp.* associated with *Xenorhabdus* and *Heterorhabditis sp.* associated with *Photorhabdus*. A central micrograph shows a bacterium, identified as Enterobacteriaceae, Gram negative.

Releasing symbiotic bacteria

Photorhabdus -entire gut *Xenorhabdus* -gut vesicle

The image shows two micrographs. The left one, labeled 'Photorhabdus' and magnified 118x, shows a wax worm's gut glowing green. The right one, labeled 'Xenorhabdus' and magnified 390x, shows a close-up of a gut vesicle containing green bacteria. Labels 'pharynx' and 'symbiotic bacteria' point to specific parts of the worm's anatomy.

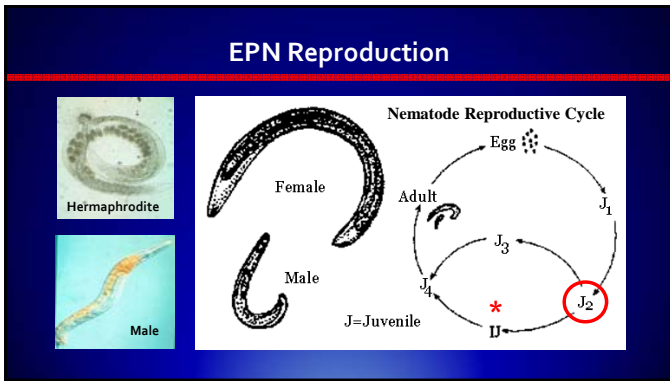
Bacteria kills the insect

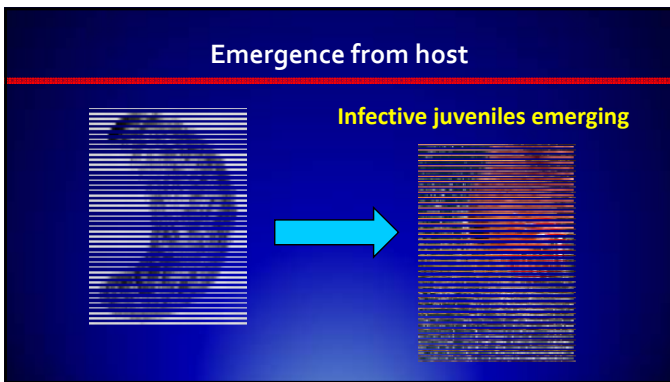
A healthy wax worm is shown on the left, and an infected wax worm is shown on the right. A red arrow labeled '24 - 48 hrs' and 'Death' with a skull and crossbones icon points from the healthy worm to the infected one.

Bioluminescence

Bacteria causes the dead insects to glow!

Four wax worms infected with *Photorhabdus luminescens* are shown glowing green.













Part II

Applications of EPNs

EPNs have a broad host range



termite Isoptera		mole cricket		white grub Scarabaeidae	
vine weevil Curculionidae		fungus gnat		army worm Noctuidae	
wax worm Pyralidae		Carabid beetle		fruit fly Drosophiliidae	

Pests controlled by EPNs

Sector	Nematode	Target pest
Citrus	<i>S. riobravae</i> <i>H. bacteriophora</i>	Citrus root weevil
Turfgrass	<i>S. carpocapsae</i> <i>S. scapterisci</i>	Hunting billbug, Black cutworm Sod webworm, Fall armyworm Mole crickets
Greenhouse	<i>H. bacteriophora</i> <i>S. feltiae</i>	White grubs Fungus gnats, Black vine weevil
Mushrooms	<i>S. feltiae</i>	Sciarid flies
Cranberries	<i>S. carpocapsae</i>	Cranberry girdler
Nursery	<i>H. bacteriophora</i> <i>H. megidis</i>	Black vine weevil
Pet vet	<i>S. carpocapsae</i>	Dog flea


Commercial production of EPNs

Production of beneficial nematodes in Israel





Consumer products containing EPNs


Variety of products targeted for different insect pests




white grubs



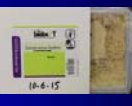
black vine weevil



soil dwelling pests



citrus root weevil



Lepidoptera

Suppliers of EPNs

Company	Address	Website	Nematodes
BioBest	2020 Fox Run Road, RR 4 Leamington Ontario N8H 3V7	www.biobest.be	Sc Sf Sk Hb
BioLogic Grows	P.O. Box 177 Wilson Hill, PA, 17211	www.biologicco.com	Sc Sf
BASF	821 Cooper Ave Ames, Iowa 50010	www.beckerrunderwood.com	Sc Sf Sk Hb
Hydro Gardens	P.O. Box 25845 Colorado Springs, CO 80936	www.hydro-gardens.com	Sc Sf Hb
Koppert	28465 Beverly Road Romulus, MI 48194	www.koppertonline.com	Sf Hb
Natural Insect Control	3727 Nettieby Rd Stevensville, ON L8S 1S0	www.naturalinsectcontrol.com	Sc Sf Hb
Sierra Biological	19750 Forest View Cir. Pioneer, CA, 95668	www.sierrabiological.com	Sc Sf Sr
Southeastern Insectaries	606 Bill Street P.O. Box 1546 Perry, GA 31069	www.southeasterninsectaries.com	Sc Hb Hi
The Environmental Factor	65 Chambers Drive, Unit 8 Apex, ON L1C 1E2	www.environmentalfactor.com	Sc Sf Sg

Application techniques -Sprayers

EPNs are generally mixed in water and applied by spraying.



backpack sprayer




towed spray array




self-propelled boom sprayer

Application techniques –Irrigation

EPNs can also be added to irrigation mixtures.



side roll system




pivot / linear system

Environmental conditions

Important to apply EPNs under the right conditions

- EPNs require moisture.
- Cannot survive direct sunlight.
- Apply at temperatures above 60°F.
- Require proper host insects.



Not a good time to apply nematodes! ☹️

Follow the directions!

For best results –

Follow the manufacturer's recommendations.

USER'S INSTRUCTIONS

biobVT
B-Green

CHECK! READ THESE INSTRUCTIONS BEFORE APPLYING



APPLICATION	
MODE	CURATIVE
DOSAGE RATE (MILLION/MP)	15-20
SOLUTION (ML/MP)	100
APPLICATION FREQUENCY	1-2X
APPLICATION INTERVAL	4 WEEKS
APPLICATION TIMING*	APRIL-SEPTEMBER

*Application timing depends from the geographical location, climate conditions and life cycle of the host. For chitin grade the application is usually done when grubs are above in the soil surface (August-September) and for black vine weevil when larvae are present (April-September) Once in the soil, nematode persist for up to 3 weeks.

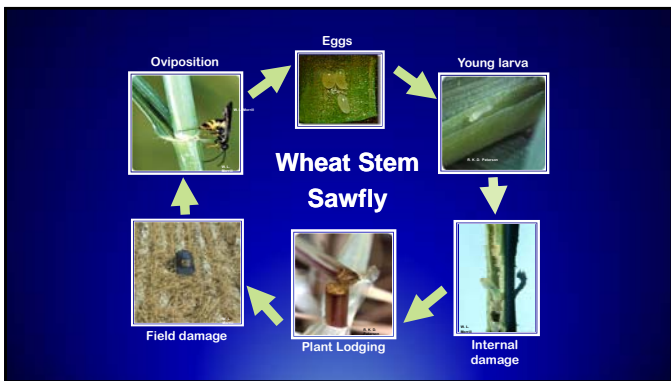
Part III

EPNs against sawfly and wireworms

Wheat stem sawfly (*Cephus cinctus*)





MT > \$30 Million Losses
Problem since 1920's
Dozens of Researchers



Sawfly larvae are well protected



Wheat stubble infected with sawfly



Insect protected from chemicals and natural enemies

Sawfly larvae are well protected

Wheat stubble infected with sawfly




Insect protected from chemicals and natural enemies

Exploit the plug

Combine EPNs with carrier solutions

Chemicals that counteract the plug's hydrophobic properties.

- Surfactants (Alypso)
- Detergents (Triton-X, Tween 80)
- Ionic Compounds (Urea)



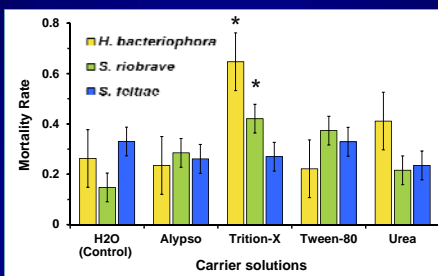
Different species of EPNs against sawfly

% Mortality <i>Cephus cinctus</i> larvae (N=20)						
Test Solution	<i>Steinernema feltiae</i>	<i>Steinernema riobrave</i>	<i>Heterorhabditis bacteriophora</i>	<i>Steinernema carpocapse</i>	<i>Steinernema glaseri</i>	<i>Steinernema krauseli</i>
H2O	31.3	11.1	26.3	0.0	0.0	16.7
Alypso	22.2	44.4	5.9	6.3	15.4	12.5
Triton-X	22.2	47.1	61.1	0.0	0.0	11.8
Tween 80	23.5	52.9	22.2	0.0	0.0	5.6
Urea	10.5	20.0	41.2	0.0	5.9	6.7

Different species of EPNs against sawfly

% Mortality <i>Cephus cinctus</i> larvae (N=20)						
Test Solution	<i>Steinernema feltiae</i>	<i>Steinernema riobrave</i>	<i>Heterorhabditis bacteriophora</i>	<i>Steinernema carpocapse</i>	<i>Steinernema glaseri</i>	<i>Steinernema krauseli</i>
H2O	31.3	11.1	26.3	0.0	0.0	16.7
Alypso	22.2	44.4	5.9	X	X	X
Triton-X	22.2	47.1	61.1	X	X	X
Tween 80	23.5	52.9	22.2	X	X	X
Urea	10.5	20.0	41.2	0.0	5.9	6.7

Mortality of sawfly larvae



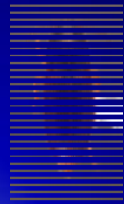
Wireworms (Family: Elateridae)



Two problem species for Montana grain growers

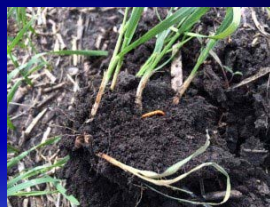
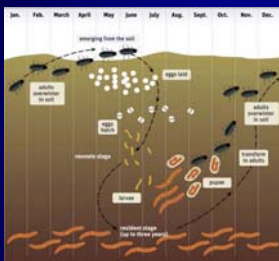


Limonius californicus



Hypnoidus bicolor

Wireworm lifecycle



wireworm feeding on wheat

Tenebrio larvae similar to wireworms



Tenebrio molitor larvae



wireworm larvae

Tested three EPN species

Added ~2400 EPNs to deli cups containing 10 immature *T. molitor*.

H. bacteriophora

S. feltiae

S. riobrave

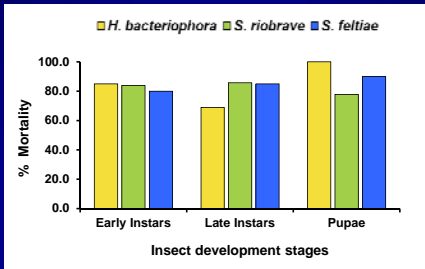


Experimental design

Tested EPNs with different development stages of *T. molitor*

development stage	Number of <i>T. molitor</i>		
	<i>Heterorhabditis bacteriophora</i>	<i>Steinernema feltiae</i>	<i>Steinernema riobrave</i>
Early Instar	20	20	20
Late Instar	20	20	20
Pupa	10	10	10

Mortality of *Tenebrio molitor*



Acknowledgements

SARE Organizers



Dr. Gadi VP Reddy



Dan Picard