From identification to implementation of a biocontrol agent in China

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Outline

1. A brief history of utilization of biocontrol agents in China
2. An overview on current utilization of biological control agents in China
3. Three cases for application of biological control agents

1. A brief history of utilization of biocontrol agents in China

- They used the whole plants that could repel insect pests, particularly for storage of cereals.

- Many records on utilization of predators for control in ancient time.

- **In Weijin Dynasty (A.D.220-581）**
- **Jinhong Tao (A.D. 452-536） described a predator, *Rhynchium bruneum* in a book, and determined the relationship between insect predator and insect prey.**

Rhynchium bruneum in Zhejiang province, China, this predator was used to control insect pests by hanging bamboo tubes in crop field. This predator distributes in Beijing, Zhejiang, Jiangxi, Sichuan, Guangxi, and Fujian provinces.

- **Xun Liu (A.D. 889-904） recorded that persons sale ants for control insect pests of orange trees.**
- **This was the first record for utilization of natural enemies to control insect pests.**

The ant is *Oecophylla smaragdina*, distributing in Guangdong, Fujian, Sichuan, and Guangxi provinces. It can predate more than 20 species of insect pests, including long horn beetle, moths and so on.

2. A brief overview of biological control agents in China

- Now, China has identified many biocontrol agents from plant, microorganism, and insect.
- But few are commercially widely distributed.

- Parasitoid wasps: about 130 species were studied as biocontrol agents.
- Parasitoid flies: 18 species were studied as biological control agents.
- Ladybirds: near 260 species were recorded.
- Chrysopa: 109 species, were recorded.
- Predacious mites: 16 species were studied as biocontrol agents
- Predacious spiders:1500 species were recorded

2.1. List for selected important parasitic wasps

- *Dibrachys cavus*
- *Pteromalus puparumi*
- *Tetrastichus schoenobii*
- *Aphelinus mali*
- *Aphytis yanonensis*
- *Anicetus ohgushii*
- *Ageniaspis testaceipes*
- *Litomactix* sp.
- *Anagrus nilaparvatae*
- *Brychymeria lasus*
- *Torymus* sp.
- *Grehspimpla kuwanae*

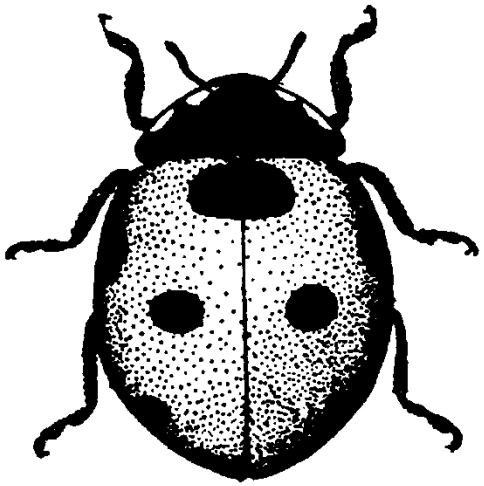
- *Campoletis chlorideae*
- *Eriborus terebrans*
- *Apanteles cypris*
- *A. chilonis*
- *Macrocentruis linearis*
- *Habrobracon hebetor*
- *Bracon greeni*
- *Aphidius gifuensis*
- *A. ervi*
- *A. avenae*
- *Diaeretiella rapae*
- *Telenomus theophilae*
- *T. rondotiae*
- *T. cirphivorus*
- *Scelio uvarovi*
- *Haplogonatopus japonica*
- *Pseudogonatopus flavifemur*

2.2. List of parasitic flies

- *Blepharipa tibialis*
- *B. schineri*
- *Actia nigroscutellata*
- *Baumhaueria goniaeformis*
- *Cuphocera varia*
- *Carcelia excisa*
- *C. kockiana*
- *Drino inconspicua*
- *Exorista amoena*
- *E. civilis*
- *E. japonica*
- *Halydaia luteicornis*
- *Lydella grisecens*
- *Phryxe vulgaris*
- *Pseudoperichaeta nigrolinea*
- *Turanogonia chinensis*
- *Nephotettix cincticeps*

2.3. List of ladybirds studied as biological control agents

- *Rodolia cardinalis*
- *Coccinella septempunctata*
- *Cryptolaemus montrouzieri*
- *Leis axyridis*
- *Adonia variegata*
- *Propylaea japonica*
- *Scymnus hoffmanni*
- *Stethorus punctillum*
- *Rodolia rufopilosa*
- *Stethorus siphonulus*



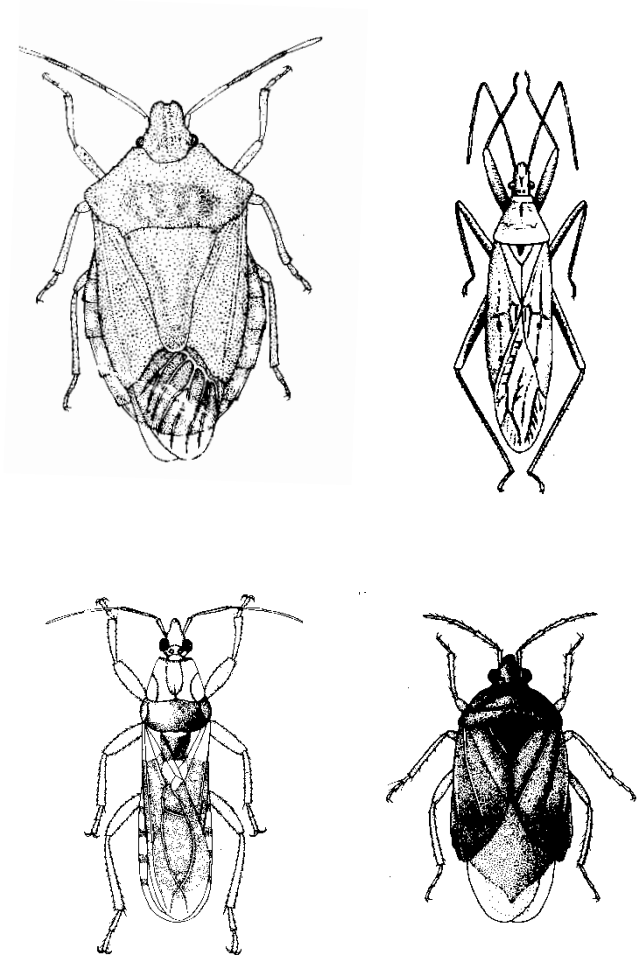
2.4. List of predatory mites as biological control agents

- *Amblyseius newsami*
- *A. nicholsi*
- *A. eharai*
- *A. orientalis*
- *A. longispinosus*
- *A. pseudolongispinosus*
- *A. okinawanus*
- *A. aizawai*
- *Euseius ovalis*
- *Agistemus exsertus*
- *Tydeus* sp.
- *Allothrombium ignotum*
- *A. pulvinum*
- *Anystis baccarum*
- *Gnorimus chudhrii*

2.5. List of other predatory insects

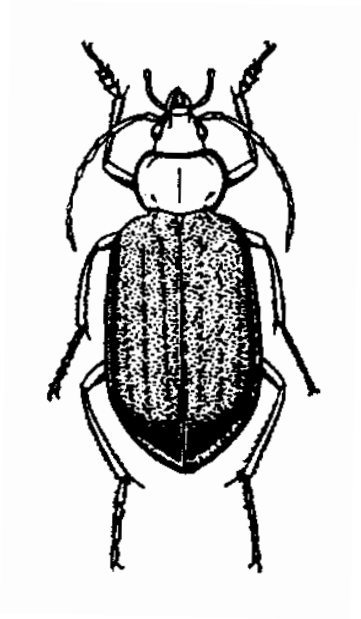
Homoptera

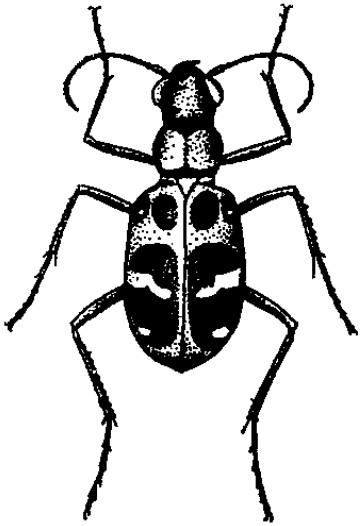
- *Orius minutus*
- *O. similis*
- *O. sauteri*
- *Cyrtorhinus lividipennis*
- *Nabis sinoferus*
- *Dortus chinai*
- *Compylomma chinensis*



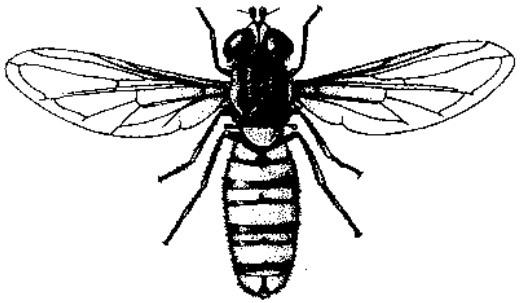
Coleoptera

- *Calosoma* sp.
- *Pterostichus* sp.
- *Chlaenius bioculatus*
- *Calathus halensis*
- *Parena rufolesacea*
- *Paederus fuscipes*

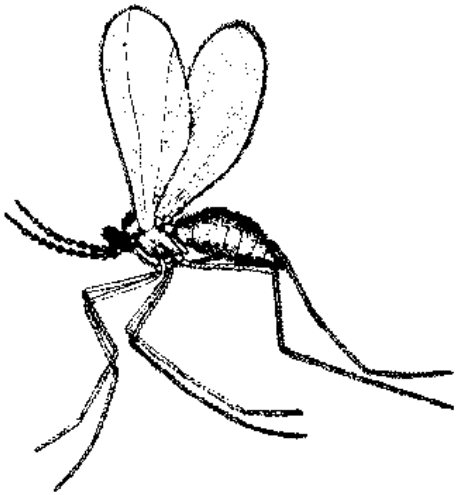


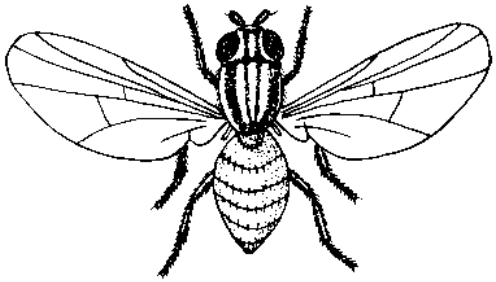


Diptera

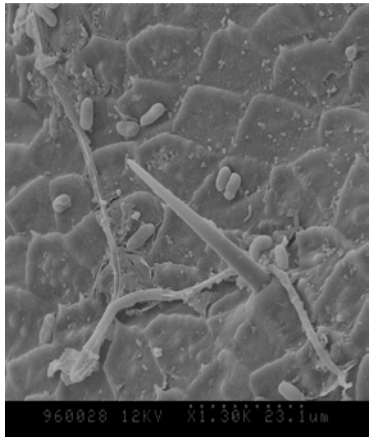


- *Cophinopoda chinensis*
- *Dasypogon aponicum*
- *Leucopis* sp.
- *Metasyrphus corollae*
- *Epistrophe balteata*
- *Lasiopticus pyrastri*
- *Aphidoletes aphidimyza*

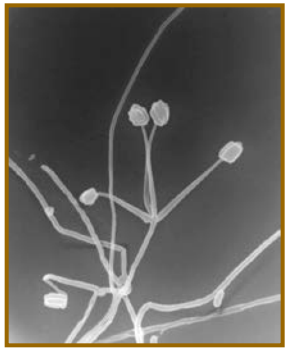


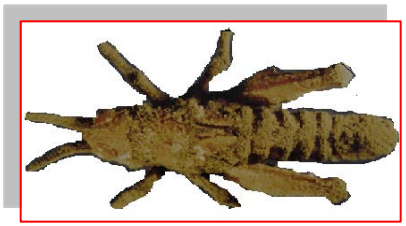


2.6. List of selected fungi as biocontrol agents



- *Beauveria bassiana*
- *B. brongniatii*
- *Metarhizium anisopliae*
- *Paecilomyces spp.*
- *Aschersonia spp.*
- *Hirsutella thompsonii*
- *Entomophaga spp.*
- *Nomuraea rileyi*
- *Verticillium lacanii*
- *Pandora spp.*
- *Zoophthora spp.*
- *Neozygites spp.*

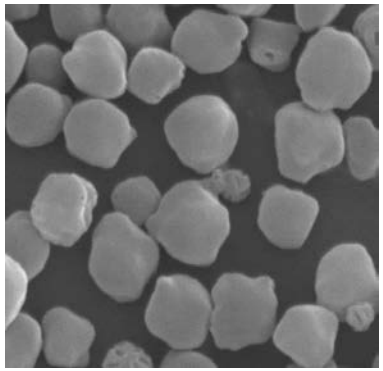


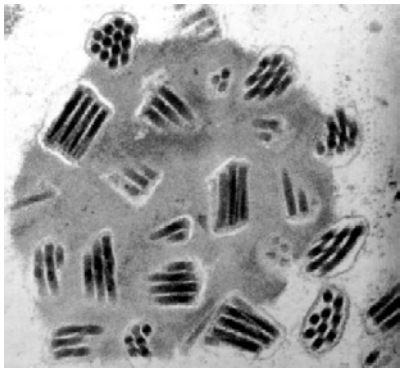


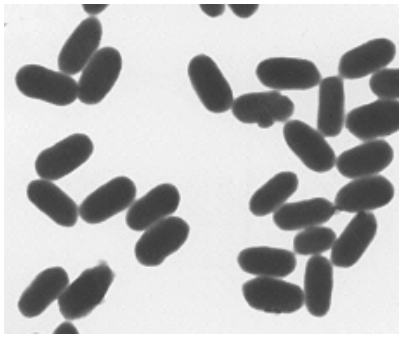
In China more than 200 species

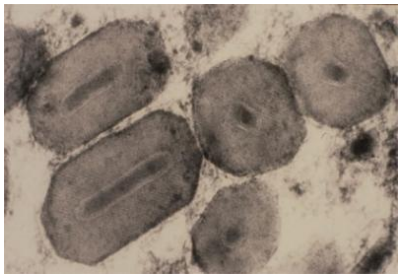
2.7. List of selected viruses as biocontrol agents

- *Heliothis armigera* NPV
- *Pieris rapae* GV
- *Spodoptera litura* NPV
- *Dendrolimus punctatus* CPV
- *Plutella xylostella* GV
- *Malacosoma neustia* NPV
- *Plusia agnata* NPV
- In China more 20 species of virus have been demonstrated in field,more than 243 strains of entomopathogenous virus.







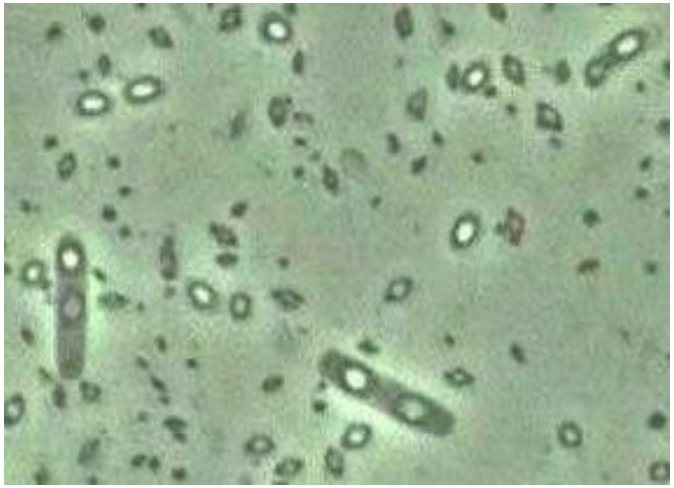


2.8. List of selected bacteria as biocontrol agents

Bacillus sphaericus

Bacillus thuringiensis

Bacillus popilliae



2.9. List of selected protozoa as biological control agents

- *Nosema* sp. Infects Heliothis
- *Nosema furnacalis* infects Corn borer
- *Nosema* sp. Infects *Laphygma exigua*
- *Vairimorpha* sp.
- *Nosema locustae*

2.10. Some commercialized biocontrol agents in China

Name	No. Product
Microorganisms	
Pseudomonas fluorescens	2
Bacillus sphaericus	2
Bacillus thuringiensis	193
Beauveria spp.	7
Metarhizium spp.	7
Granulovirus	3
Nucleopolyhedrovirus	38
Nosema locustae	1
Extracts of plant	
Sophora spp.	25
Azadirachta indica	7
Rotenone	19
Pyrethrum cinerariifolium	16
Other plant extracts	56
Natural enemies	
Trichogramma spp.	2

3. Examples for utilization of biocontrol agents

3.1. A case for A Parasitoid

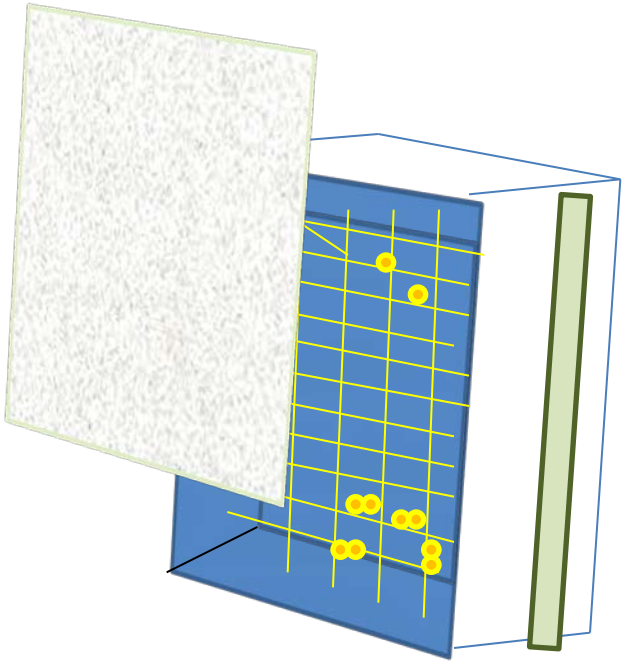
- *Anastatus japonicus*

- *Tessaratoma papillosa*

- **Eupelmidae**

- Morphology: Adult: size, about 4mm; black; antennae, 13 segments; forewings, yellow, covered with short hairs, in middle, there is a cross transparent stripe; abdomen, black, and basal part has white circle.
- Biology: 8 generations per year; 220 eggs/female; under 20°C,larvae into diapause, bisexual reproduction, but parthenogenesis produce only ♂ .
- The parasitoid can parasize the eggs of *Bombyx mori* and *Antheraea pernyi*.

- Mass produce
- *A. pernyi* as host to produce the parasitoid:
- Box: like mini poly-window. 10 eggs/mini-box, total about 2200 eggs/ box, 600 individuals/box as parents, change the host eggs every 2 days, till 20days.



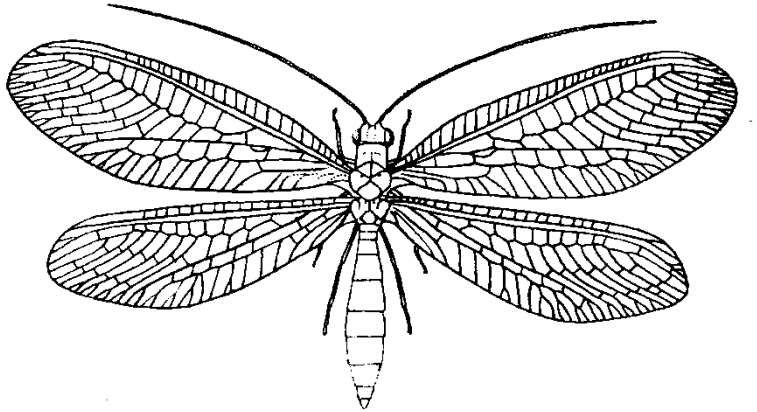
Release:

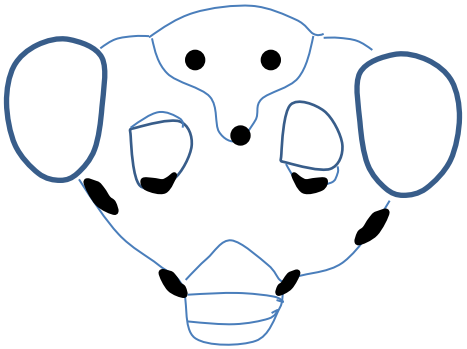
- 1000 individuals/ tree, two times in early spring, hanging the egg cards on trees. The percentage of parasitized host above 85%.
- Widely demonstrated in Guangdong, Fujian.

3.2. A case for predator

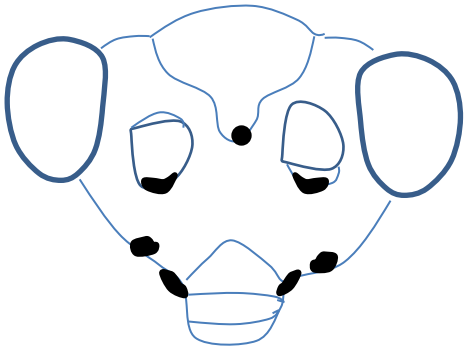
Chrysopa about 109 species in China

- *Chrysopa septempunctata*
- *C. farmosa*
- *C. phyllochroma*
- *C. kulingensis*
- *C. yatsumatsui*
- *C. boninensis*
- *C. albolineata*
- *C. carnea*
- *C. sinica*

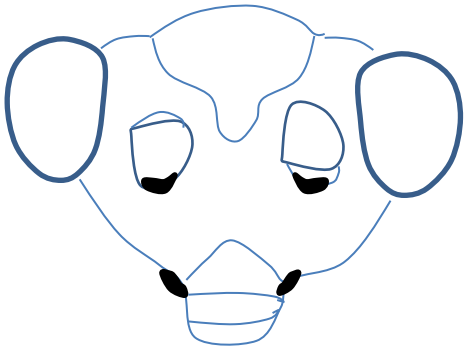




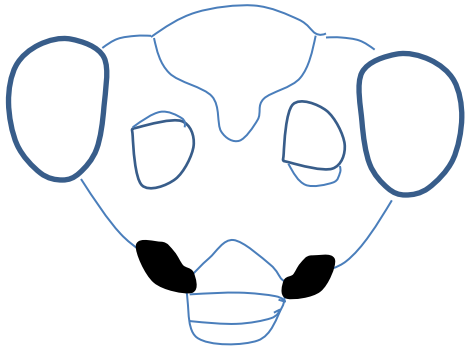
C. farmosa



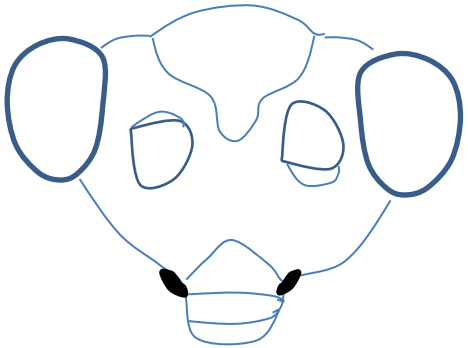
C. septempunctata



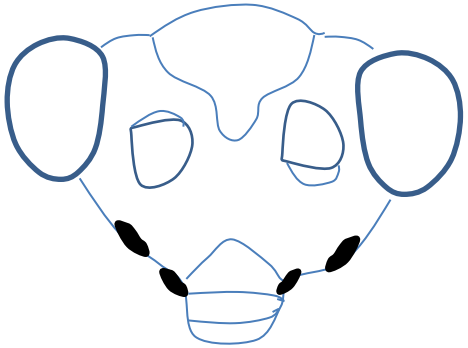
C. septempunctata



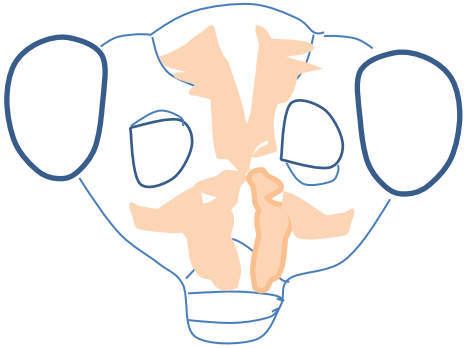
C. boninensis



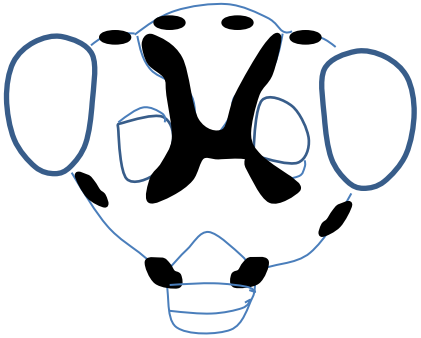
C. albolineata



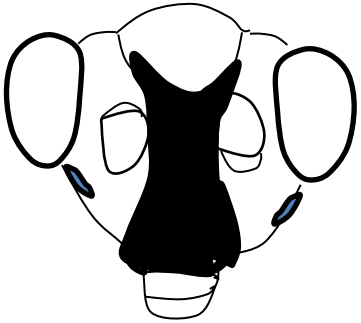
C. sinica



C. Yatsumatsui



C. intima



C. Kulingensis

1. Mass produce

(1) Natural diets

- aphids
- Eggs of moth (*Corcyra cephalonica*)

(2)Semi-artificial diets

For adults

a. Yeast + sugar.

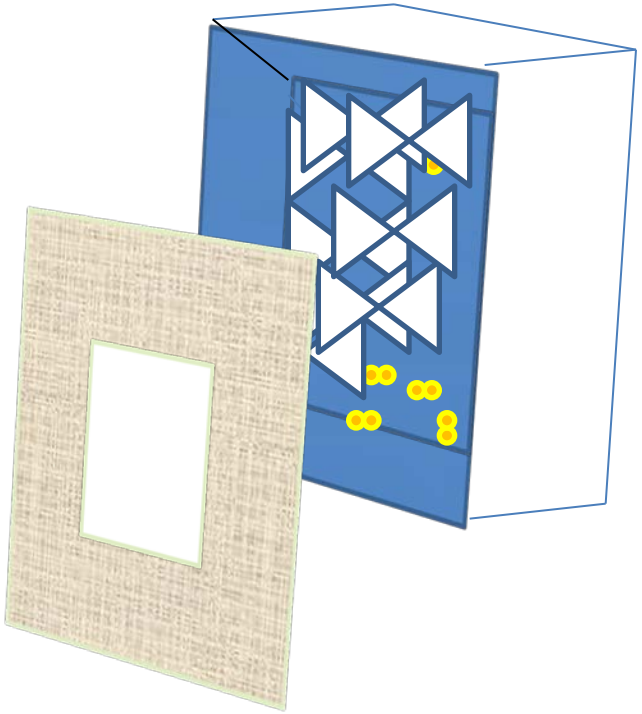
b. Powder of pig liver + yeast + honey.

For larva

a. Egg + honey + yeast + sugar.

Mass produce and release

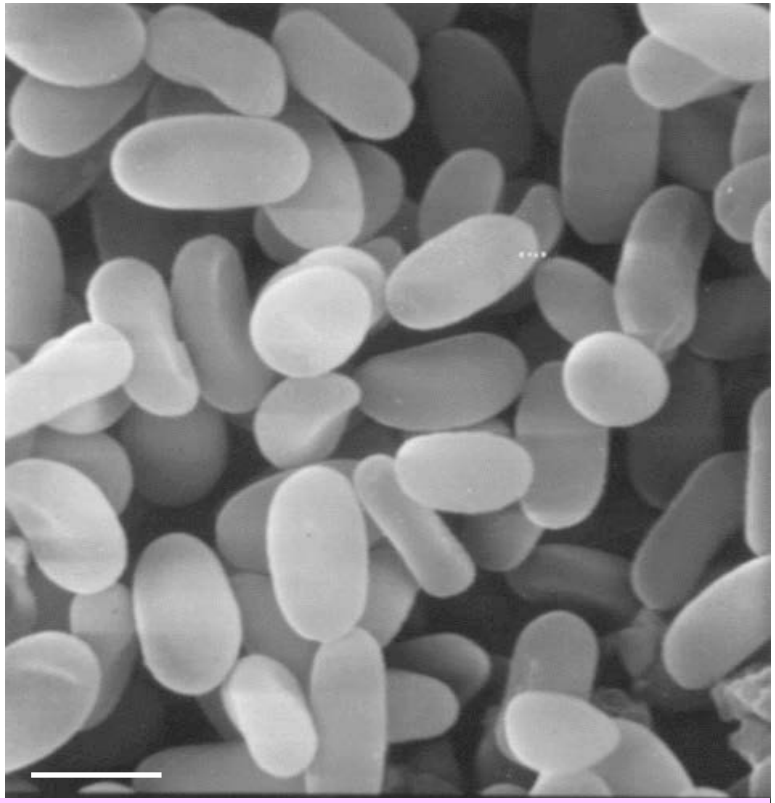
- Box for rearing larva
- Add barrier to avoid cannibalisms
- Cage for rearing adults
- Egg cards
- And release of larva, or egg cards
- Control white fly
- Control spider mites
- Control aphids
- Control corn borer



3.3. A case for pathogen

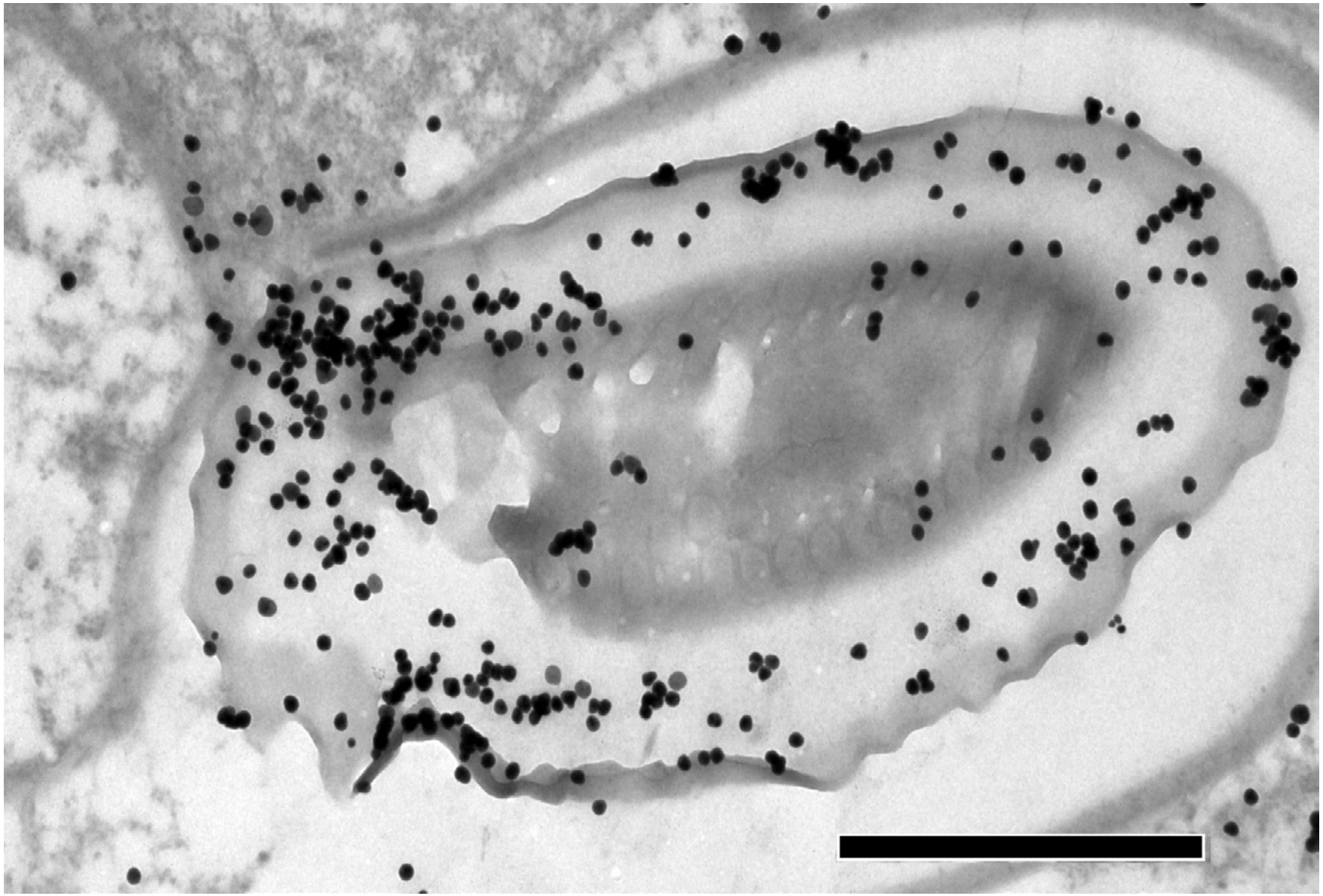
- Protozoa
- *Nosema locustae*

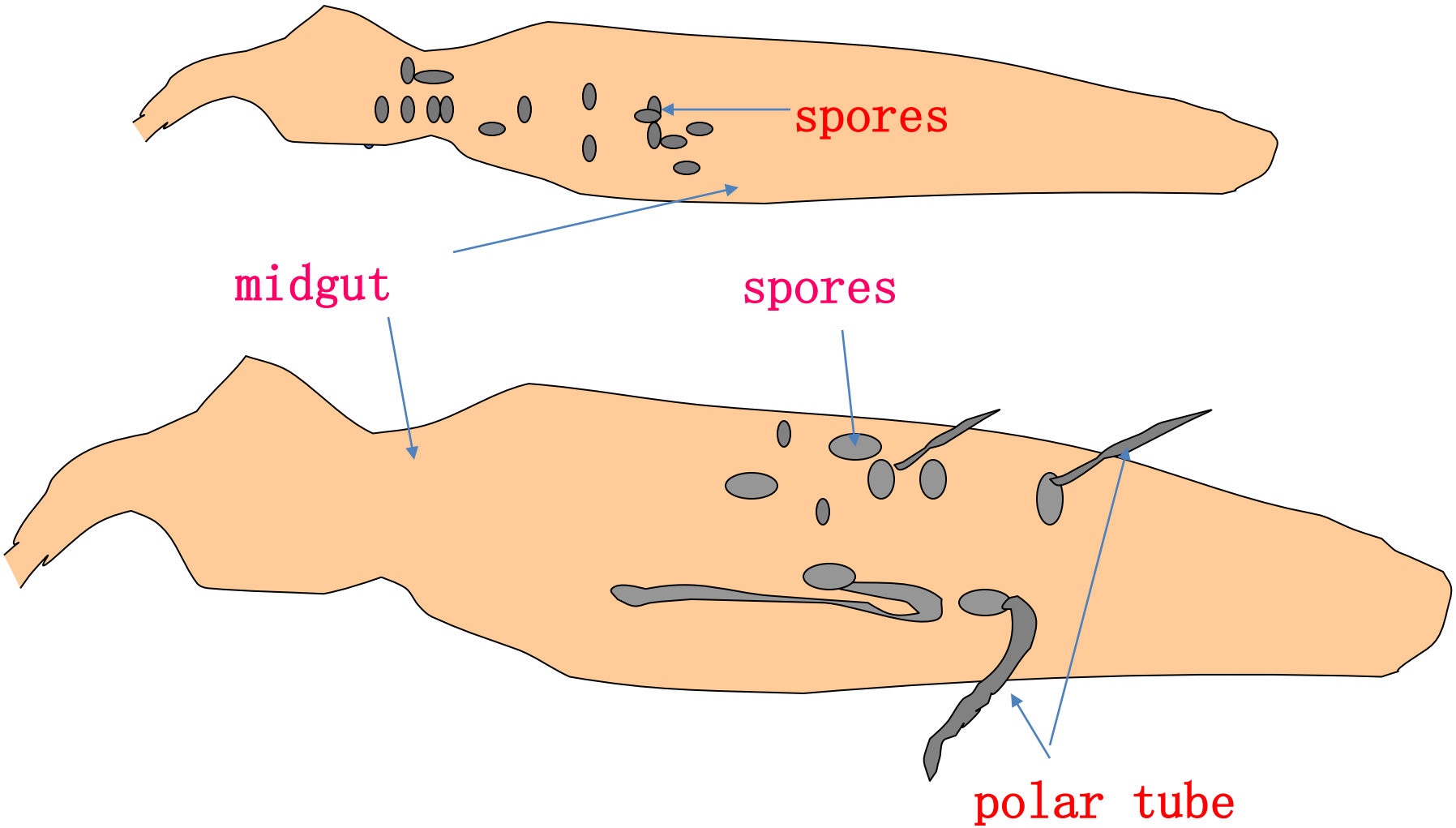
The application of *N. locustae*

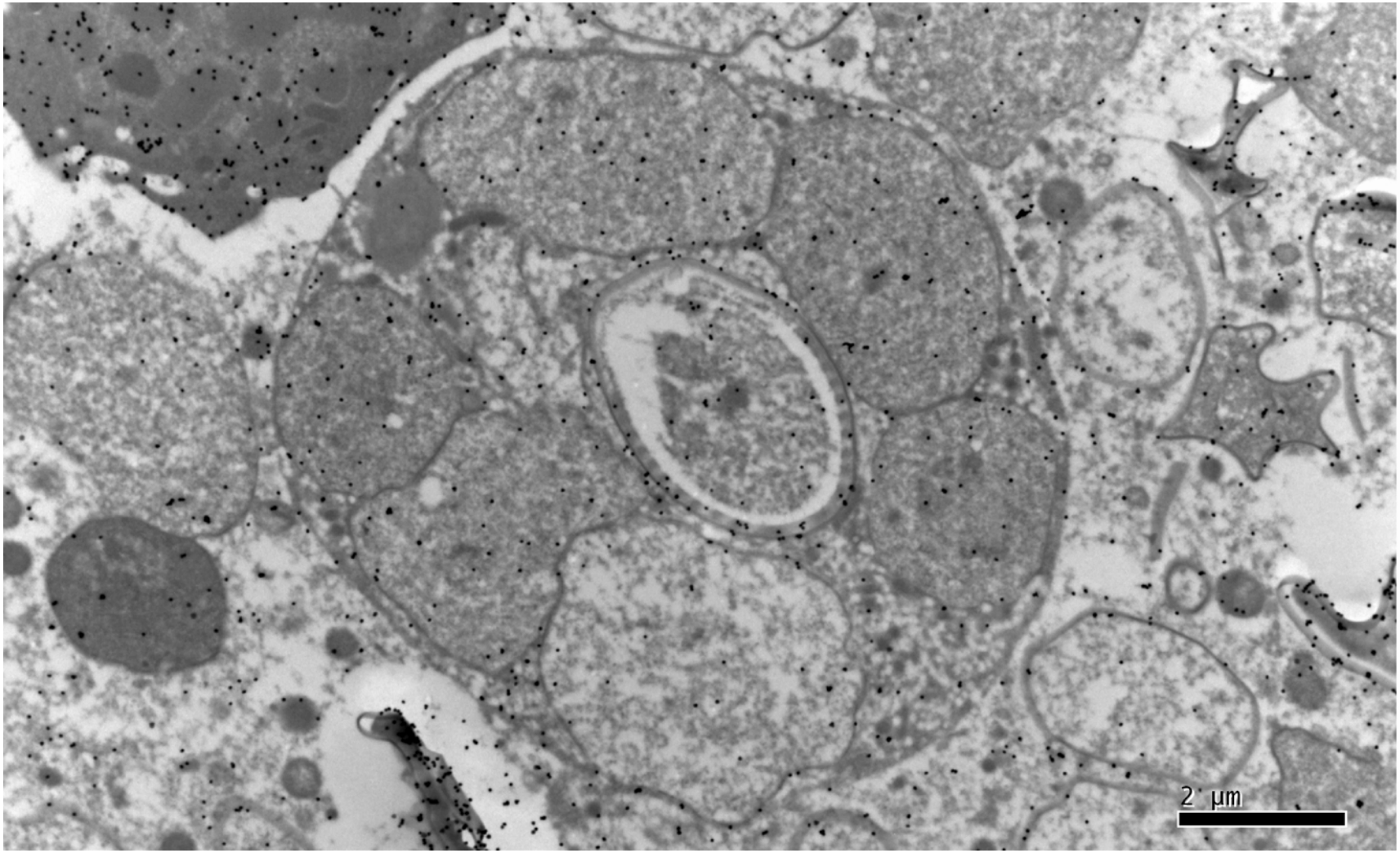


Spores of *N. locustae* were observed with SEM, Bar =60nm.

- The first large scale application of biological control agent was *Nosema locustae*, a protozoa based upon the effective jobs by Henry and his colleagues (Henry 1981). The agent has been widely applied in the world, including China.
- Our lab have researched and developed the strategies and technologies for application of *N. locustae* to management of locust and grasshoppers in China.




spores
midgut
spores
polar tube


2 µm

The application of *N. locustae*

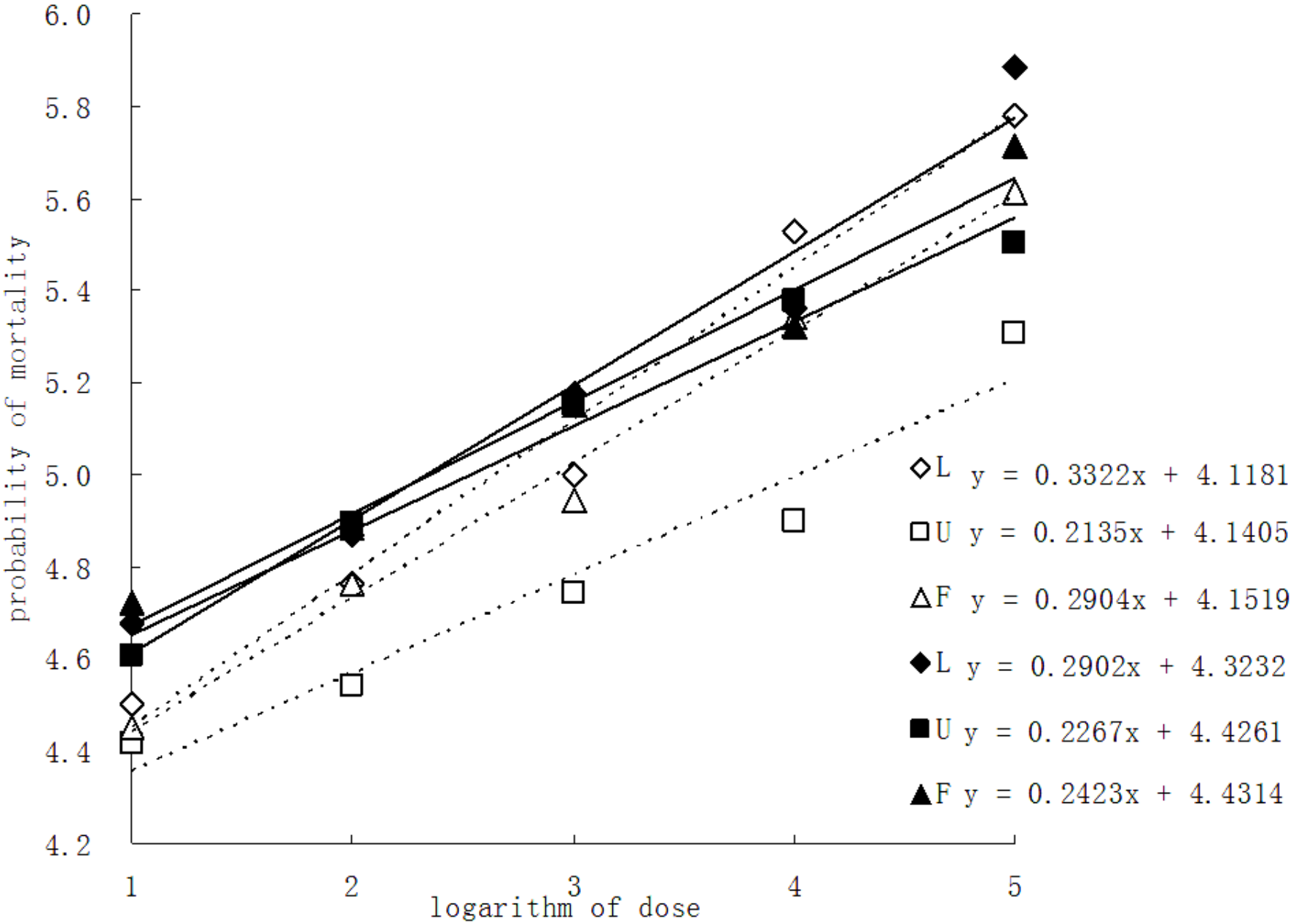
Host species of *N. locustae* and virulence

Locusta migratoria migratoria
L. m. manilensis,
Oxya spp.
Myrmeleotettix palpalis
Calliptamus italicus
Dociostaurus tartarus
D. kraussi kraussi
Gomphocerus sibiricus turkestanicus
Oedaleus asiaticus
O. infernalis infernalis
Dasyhippus barbipes
Chorthippus fallax
C. dubius
Bryodema tuberculatum dilutum
B. luctuosum luctuosum
Epacromius coerulipes
Angaracris rhodopa
A. Barabensis
Furhstorferiola tonkinensis
Ceracris spp.

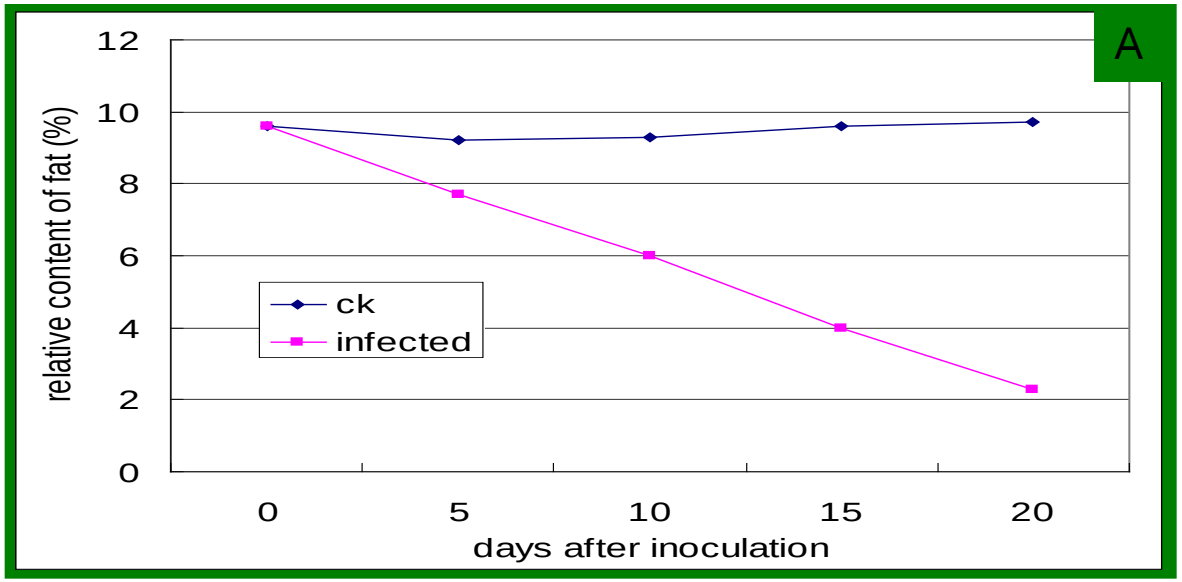
The LD_{50} of *N. locustae* to *L. m. manilesis* is about 1.88×10^3 spores/3rd instar nymph.

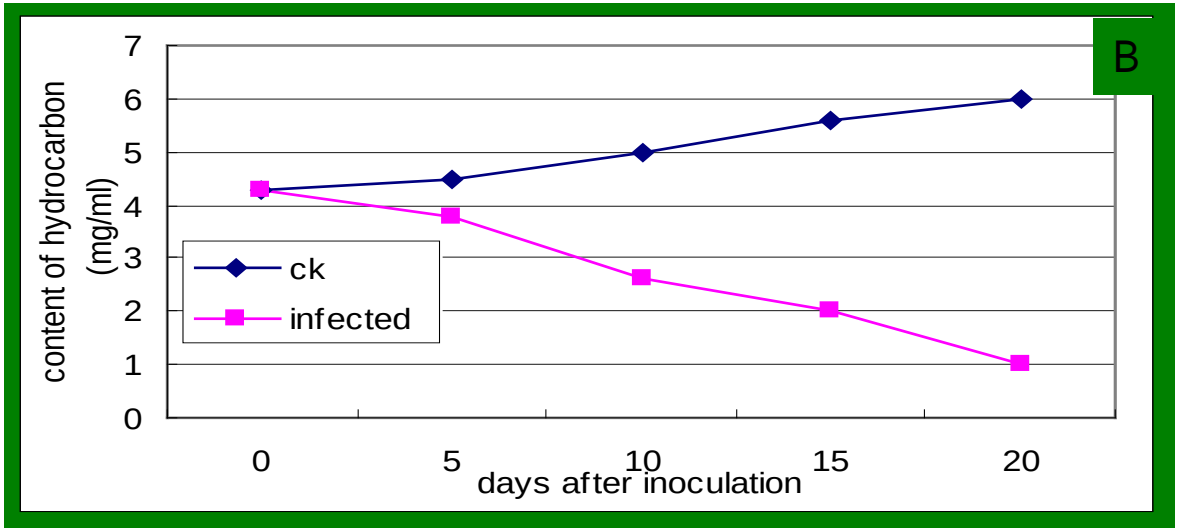
The intersegmental membrane was enlarged

Virulence of strains selected



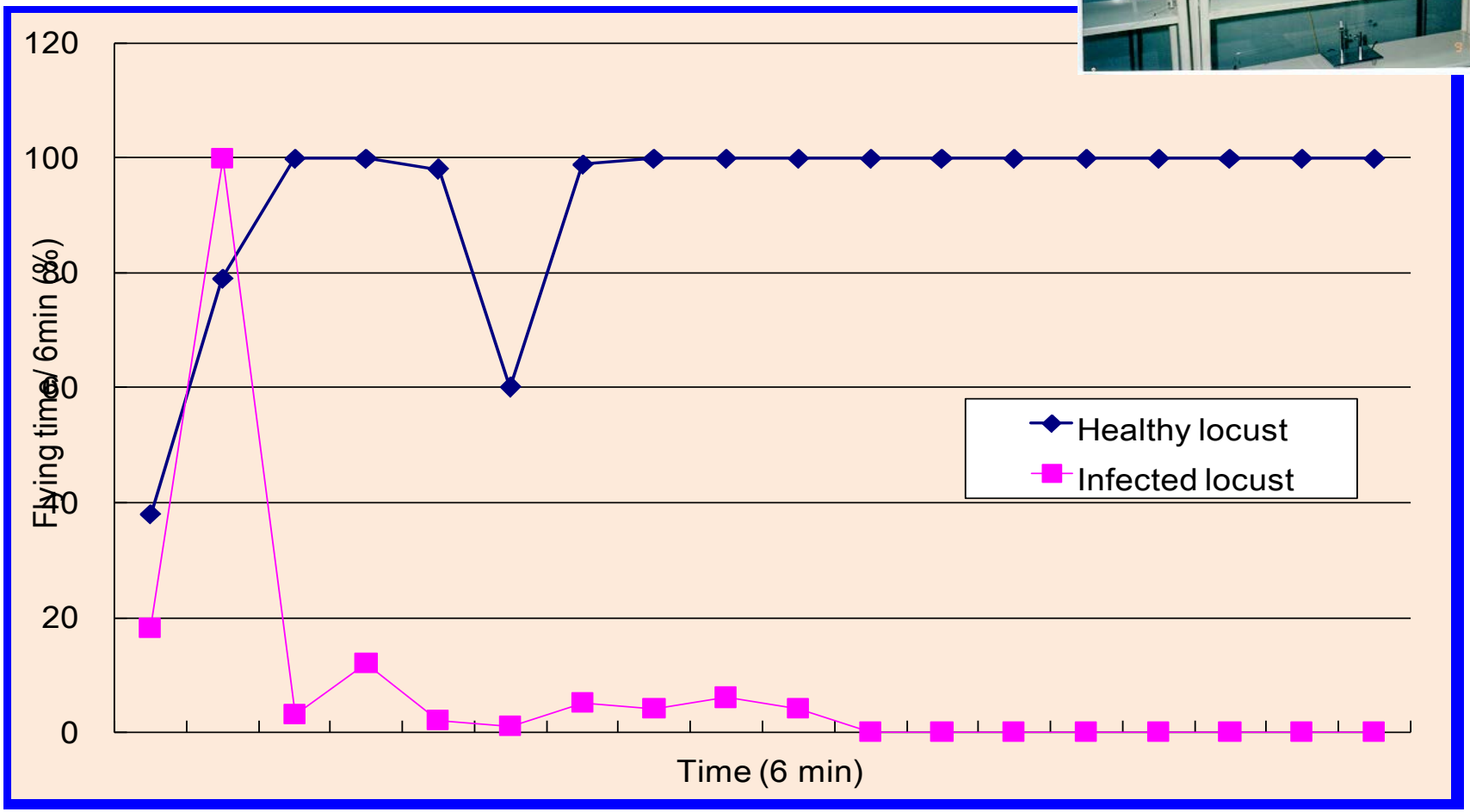
The physiological characters of infected locust





- ✓Infected locusts changed their physiological characters lot.
- ✓The contents of fat (A), hydrocarbon (B) in infected locusts were much lower than those in healthy ones.
- ✓Ck---Healthy locusts, 4th instar locusts were inoculated with *N. locustae*.

N. locustae reduced the flying ability of locust

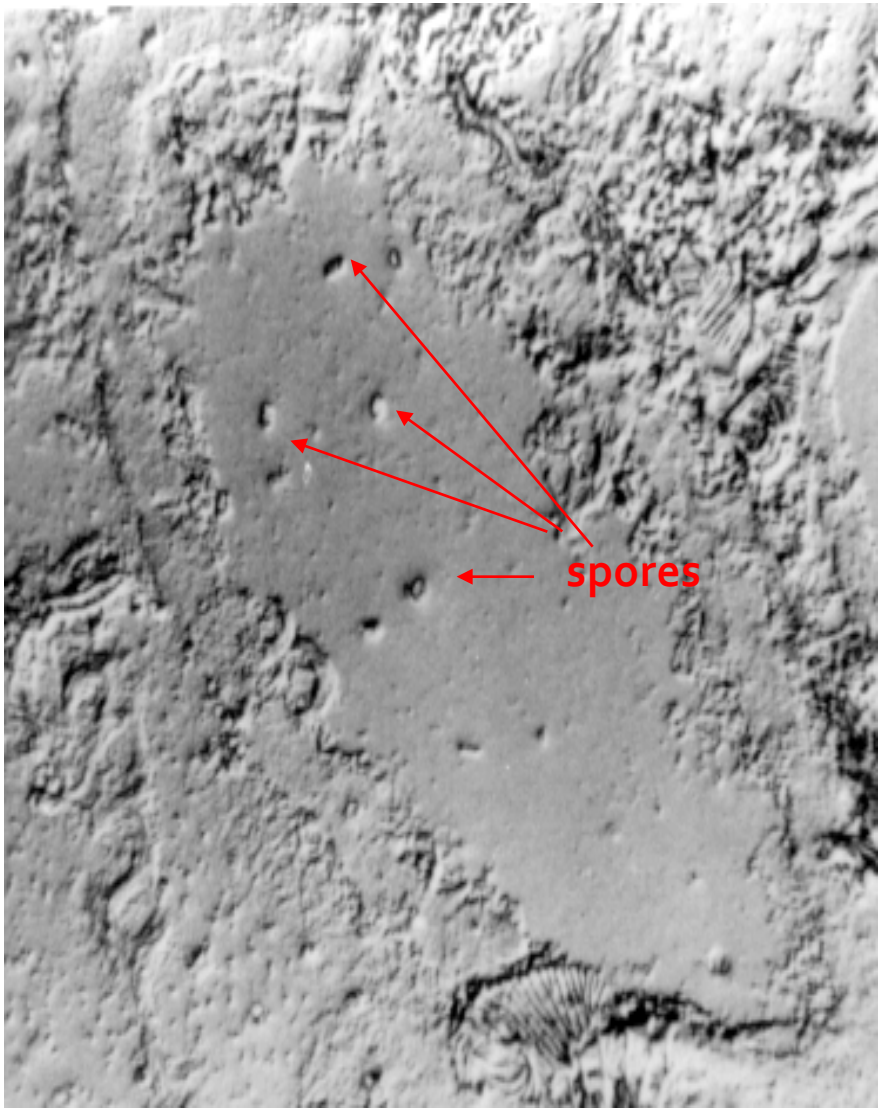


The infected locust adults only flied in the first 12 minutes, then decreased very fast, contrast to the healthy ones were flying continuously for 18 hours.

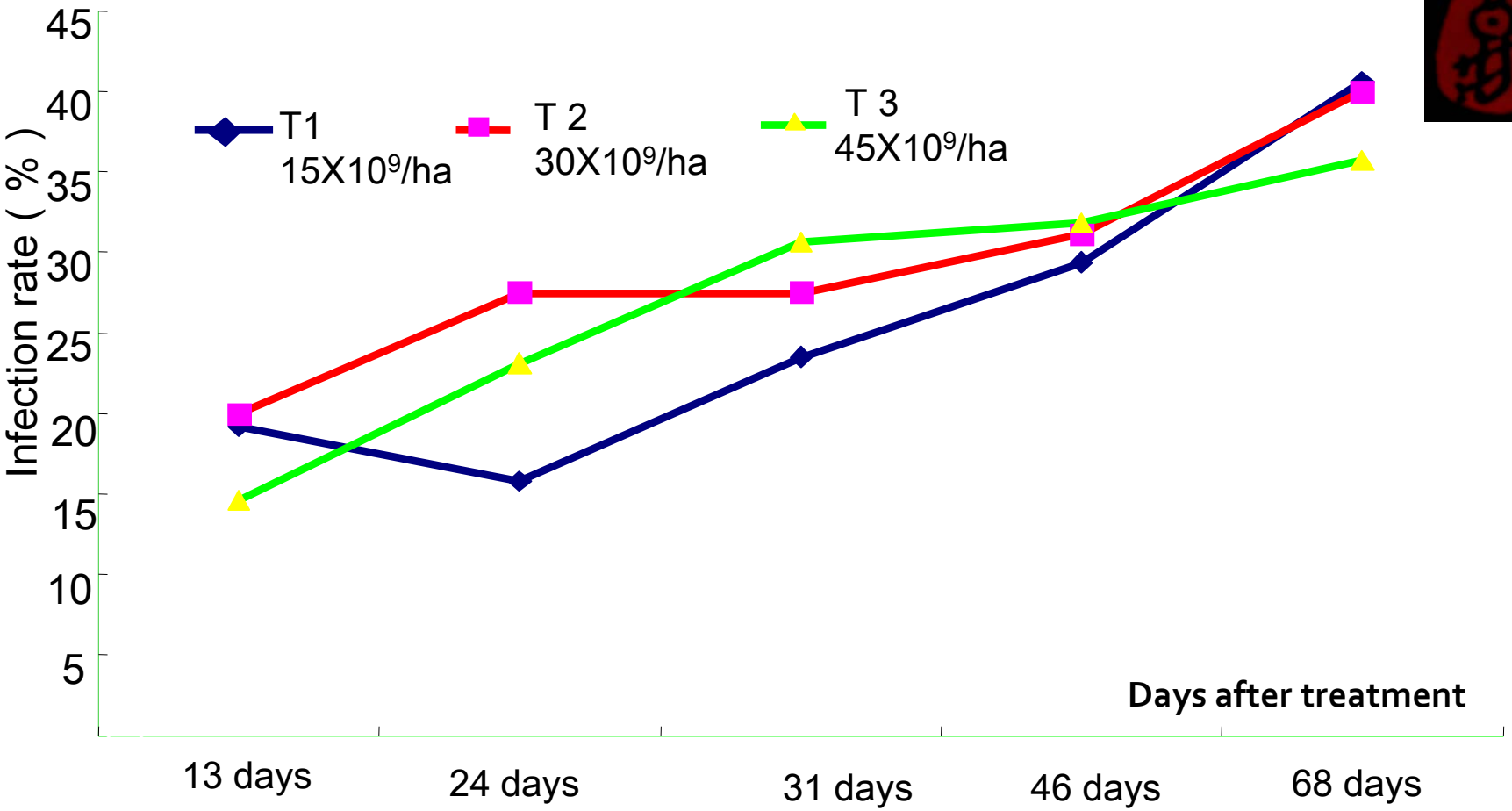
Epizootiology of *N. locustae* Disease

①Vertically transmission

- In oocytes of locust female infected with *N. locustae* the spores of *Nosema* were observed. And this means this pathogen could be transmitted vertically.
- The results of examination of eggs of infected female locusts, showed that about 10% eggs with *Nosema* spores.

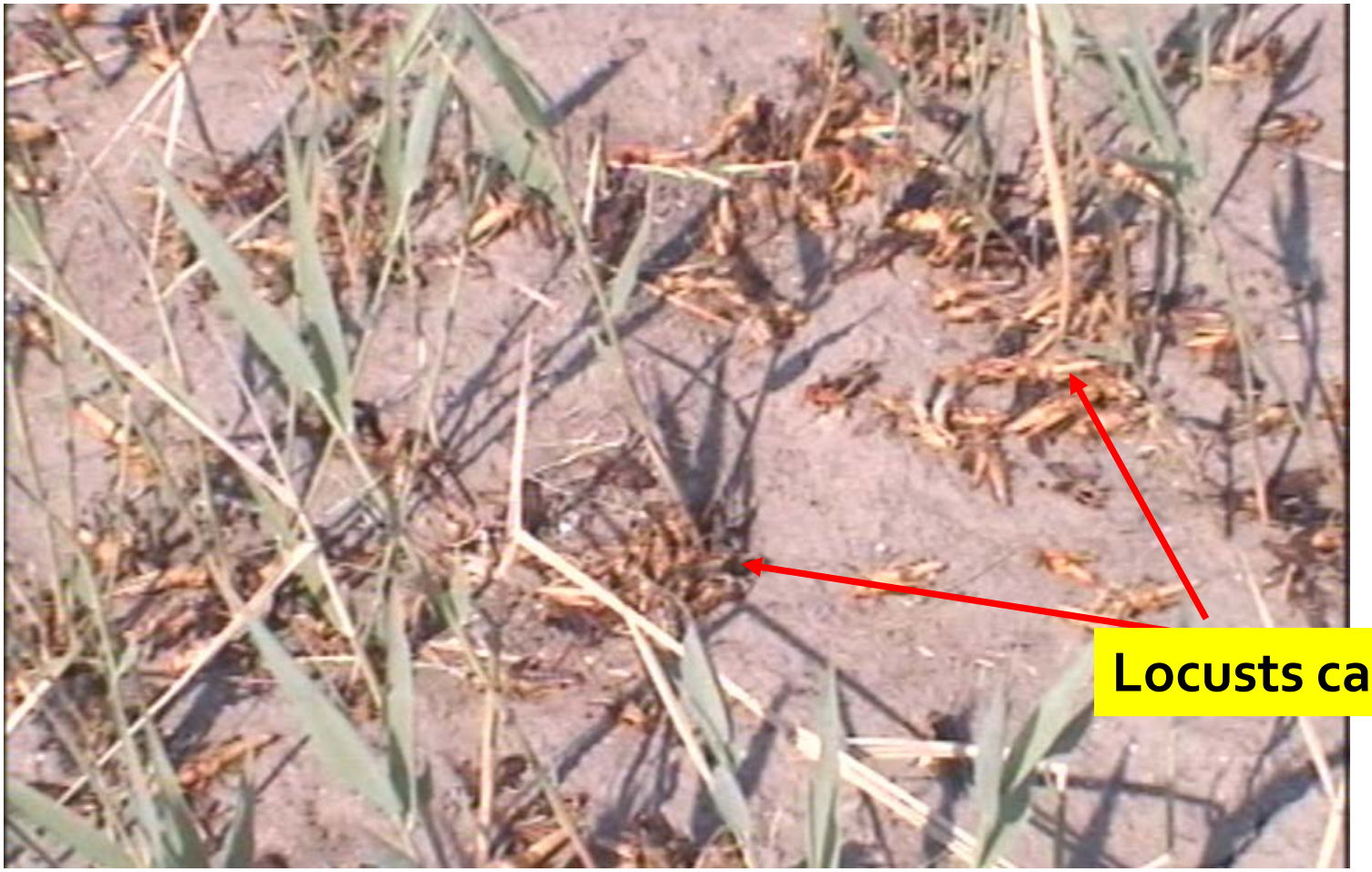


②Horizontally transmission

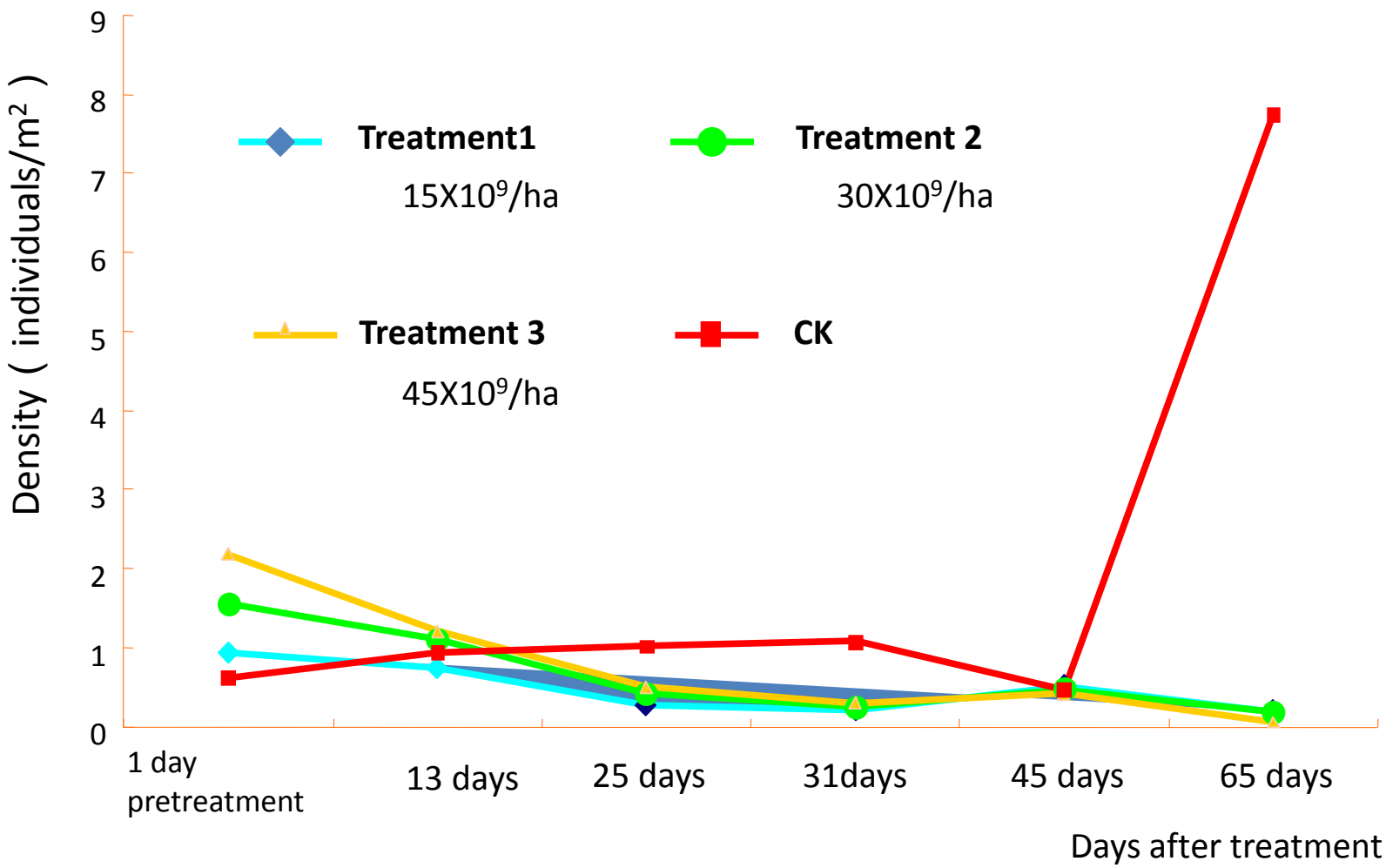


The infection rates of locust in different treatment plots with *N. locustae.*
The infections of locusts with *Nosema* in three treatments were increased after treatment, and lasting the 3rd generation (treated at 2nd generation).

The mortality caused by application of *N. locustae* in field

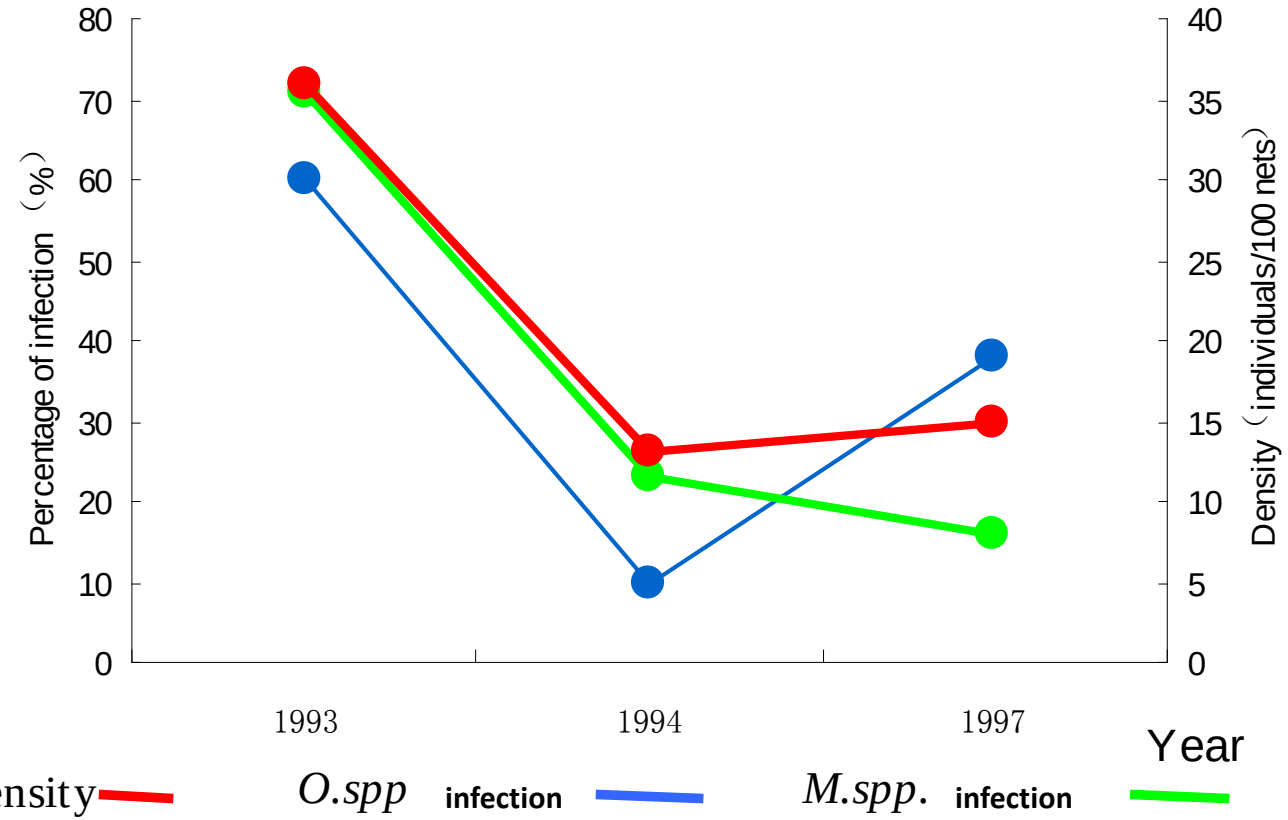


The mortalities of locusts were more than 80% before emergence in the plots treated with *A. locustae,* application rate was $30X10^9$spores/ha, treated time at the 2nd-3rd instar nymphs.



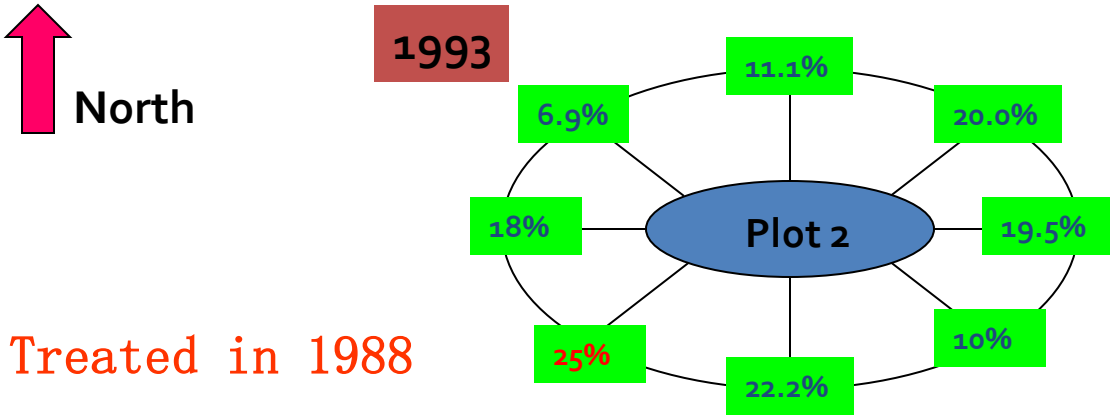
The comparison between densities of locust population in the plots treated with *N. locustae* and control plot, Haitou county, Hainan province.

③ Long term epizootiology

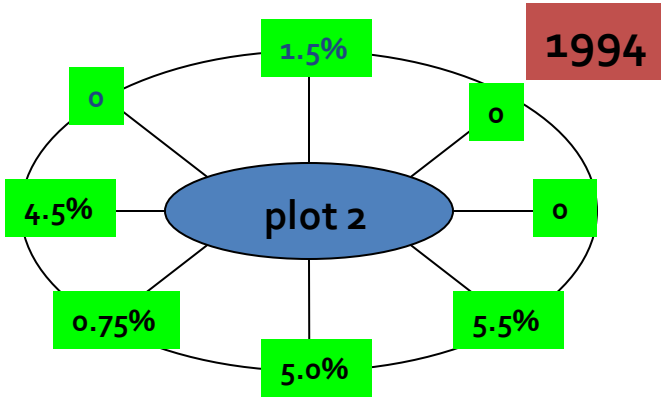


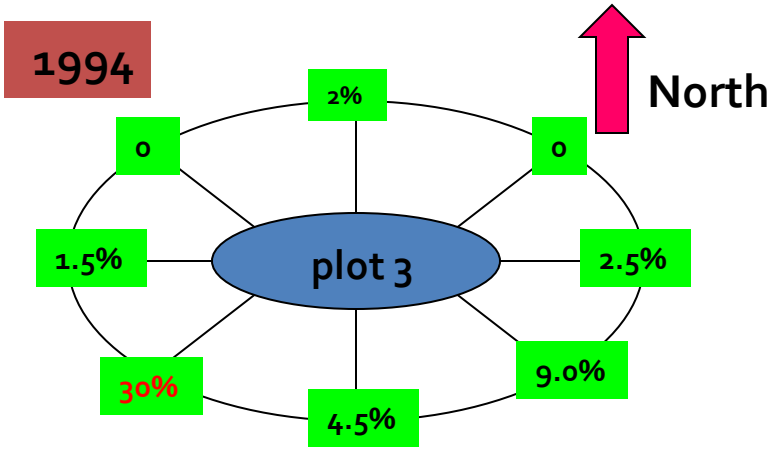
- The relationships between the density of mixture populations and infection rate of *N. locustae.*
- The place investigated was treated in 1989, with *A. locustae* bait, the application rate was
- 1.5 kg wheat bait with $7.5X10^9$ spores/ha. Investigations were conducted in the year 1993,
- 1994, and 1997 respectively. There was a positive relationship between the density and
- infections of two predominant species.

Dispersal pattern of *N. locustae* disease



Treated in 1988





Treated in 1989

The infections of grasshopper by *Nosema* at 500m distance in the 8 directions from the plot, treated with *N. locustae* in 1988 and 1989, and investigated in 1993 and 1994 again. Some time the infection rate was high 30%. Collected the grasshoppers with 100 nets in each sampling site.

- Application methods

Strategies for grasshoppers control

- Density of grasshoppers above 15 individuals/ /m^2 , reduce rapidly density and frequency of outbreak .

Integrative use insect growth regulator and *Nosema locustae* .

Application rate: IGRs 150ml/ha, and *Nosema* $15X10^9$spores/ha)

Application time: grasshoppers at 3rd instar.

They are separately applied in interval strips.

Strategies for grasshoppers control

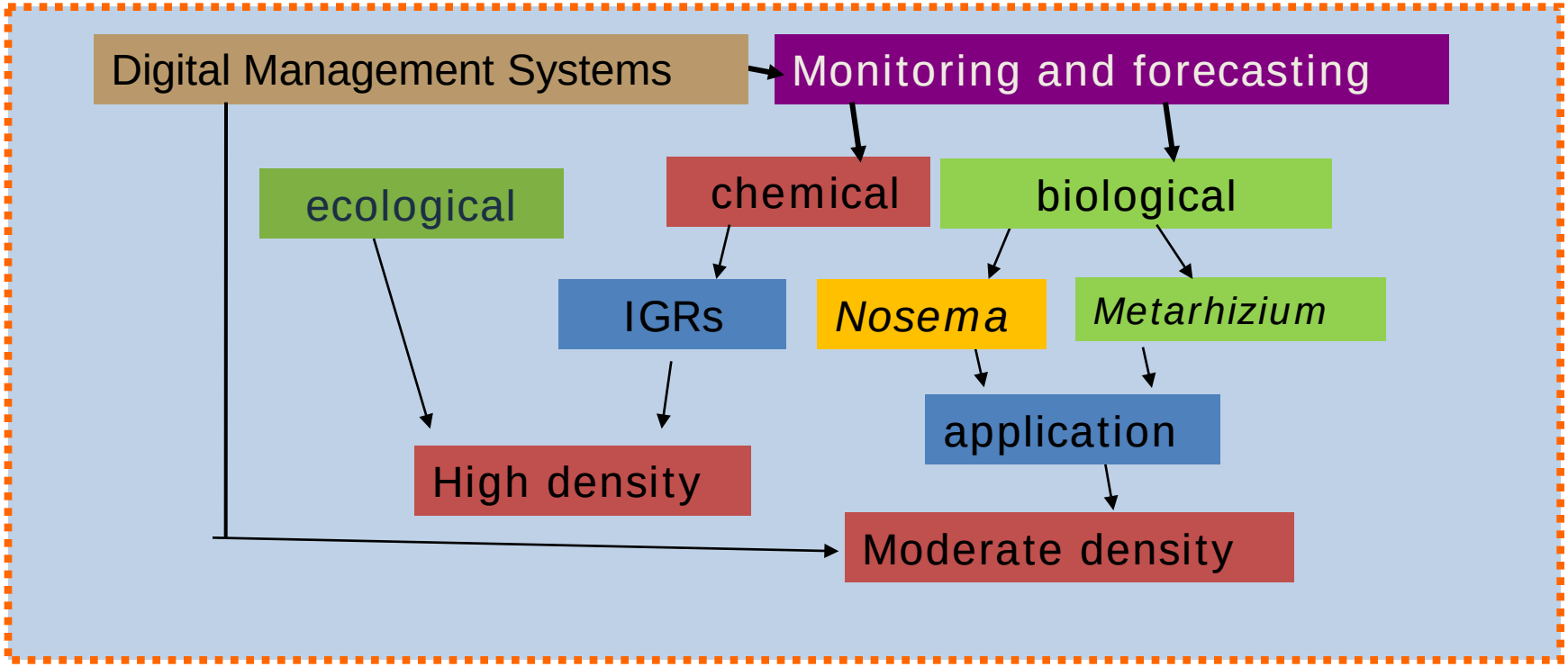
- Density of grasshoppers below 15 individuals/ /m^2 , reduce density and frequency of outbreak .

Use *Nosema locustae* only.

Application rate: *Nosema* 15X10^9spores/ha)

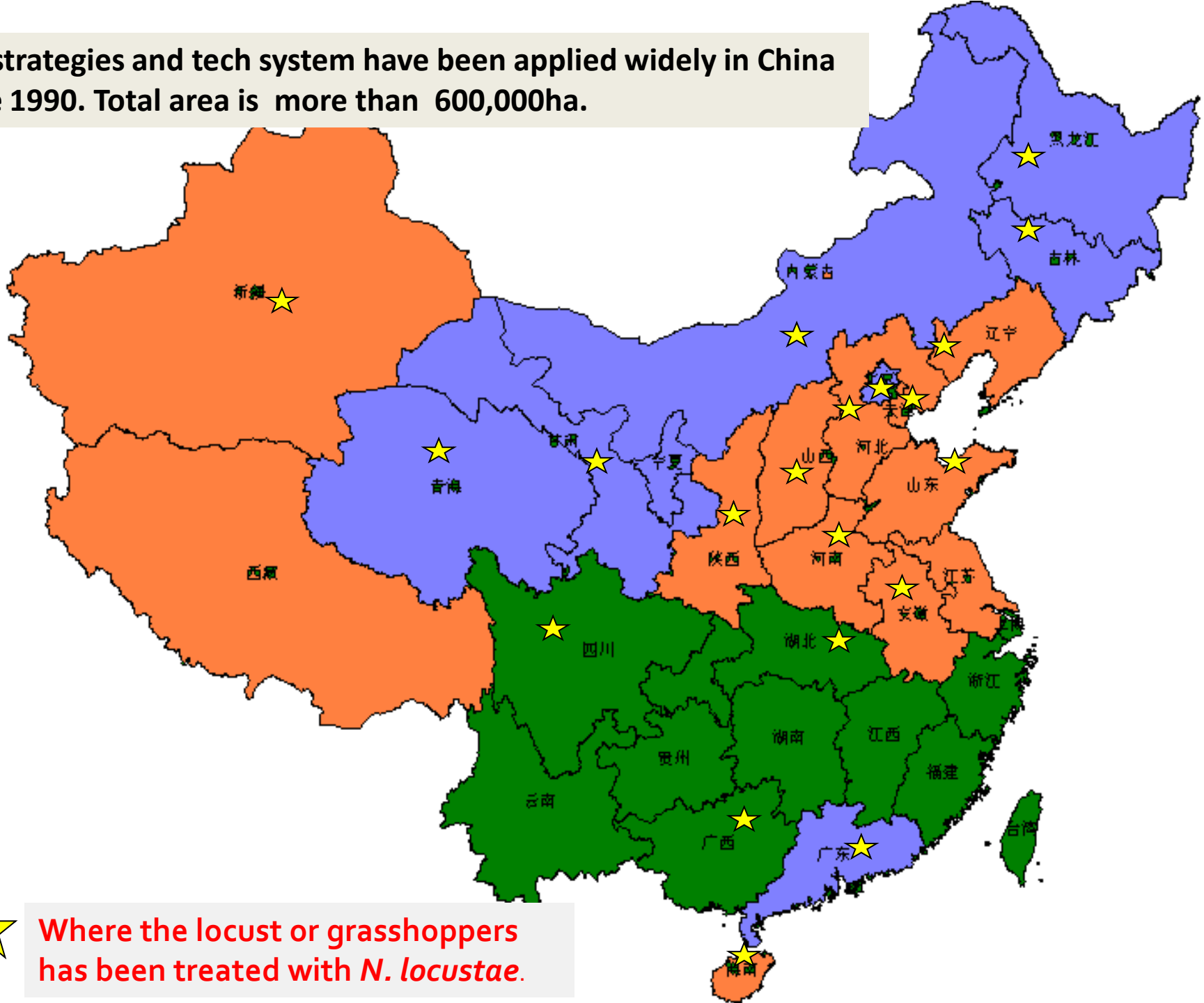
Application time: grasshoppers at 3rd instar.

Current IPM of locust and grasshopper program



Demonstration

The strategies and tech system have been applied widely in China since 1990. Total area is more than 600,000ha.



Where the locust or grasshoppers has been treated with *N. locustae*.

Thank

Ministry of Science and Technology, China
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International Centre for Research in Organic Food Systems, ICROFS