



“Climate changes and chemicals – the new sericulture challenges”

**Silkworm Diseases System
Management and Control**

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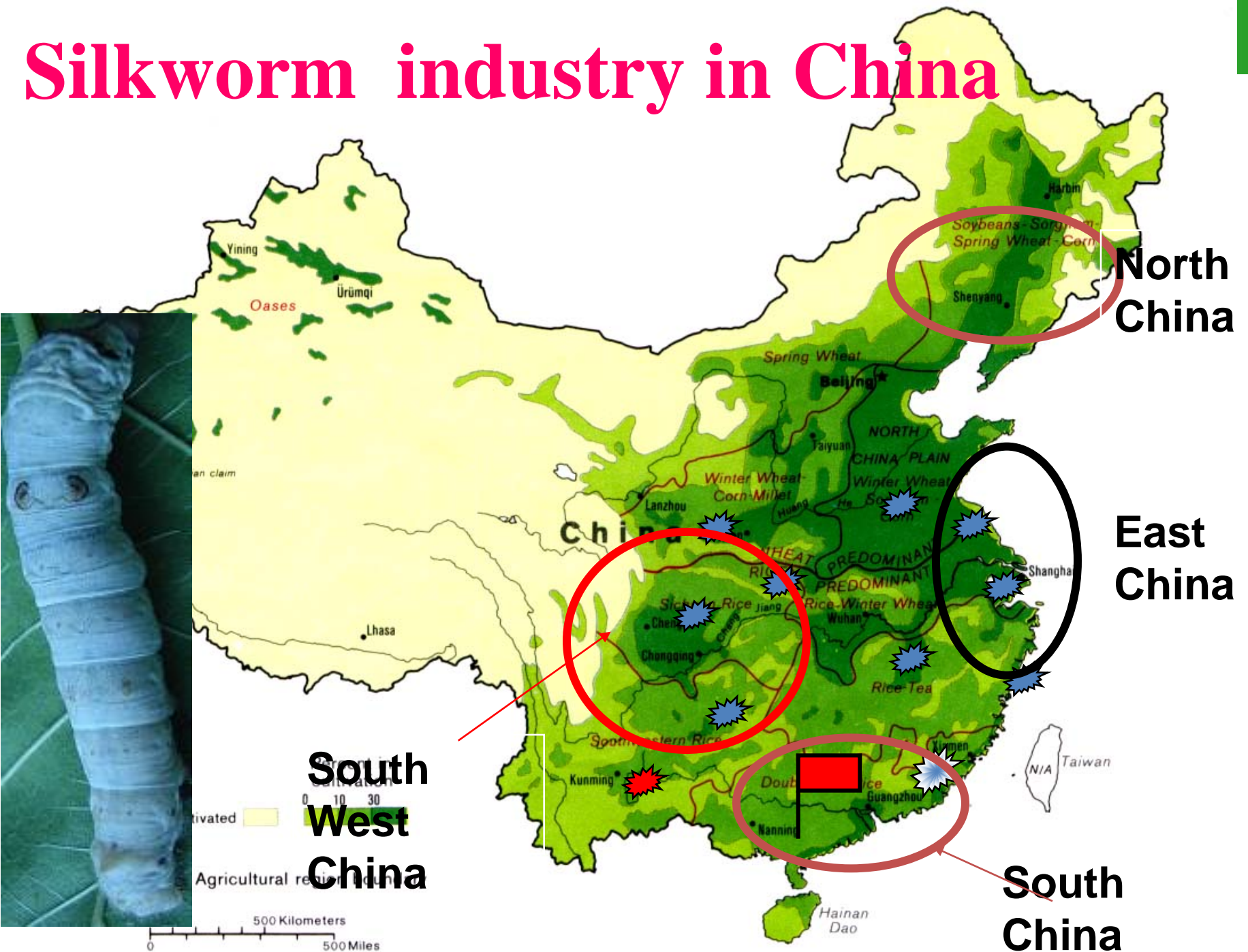
Sheki, Azerbaijan



BASCA in Turkey 2006-03



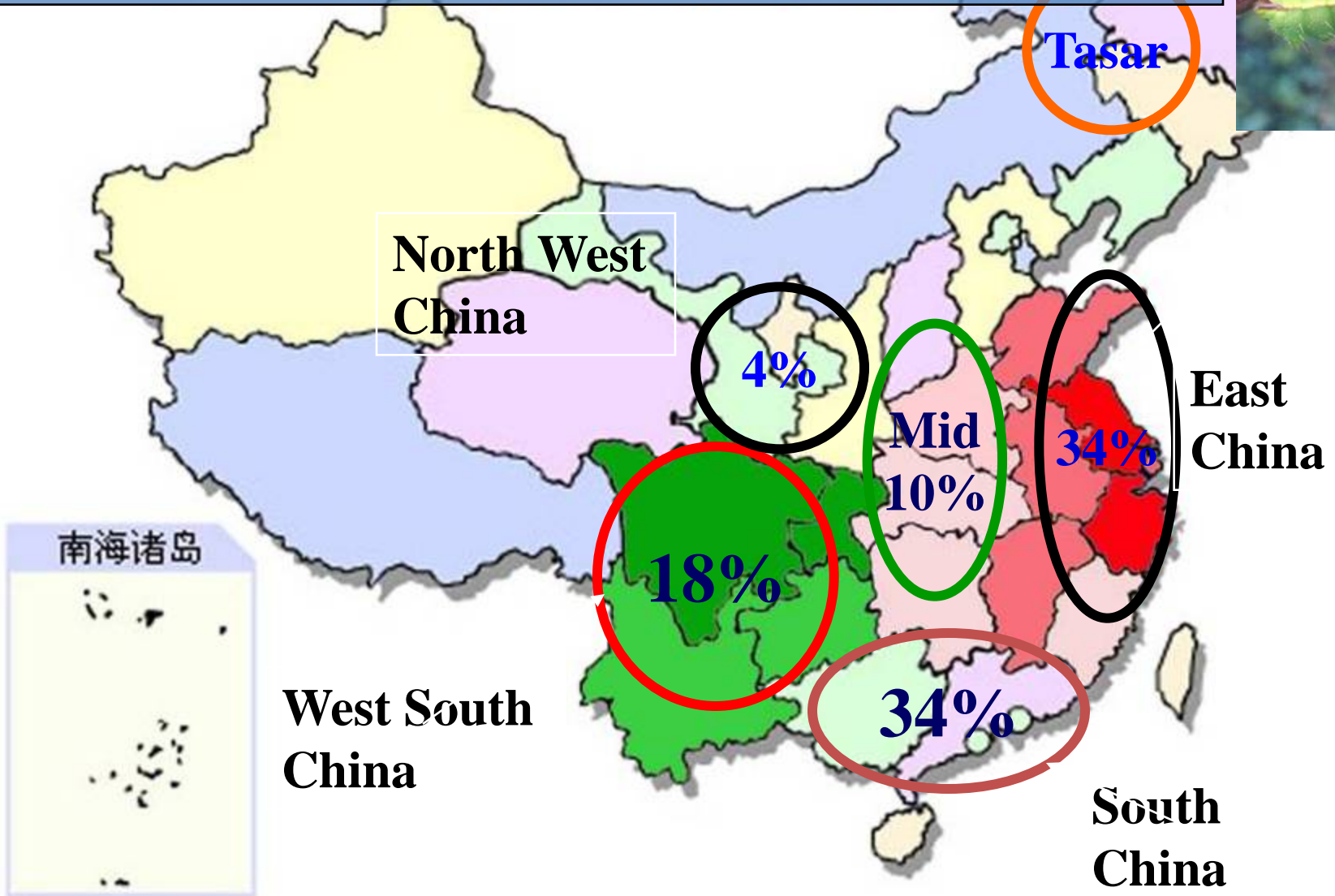
Silkworm industry in China



国家东桑西移

Silkworm industry distribution in China

产业布局调整战略示意图

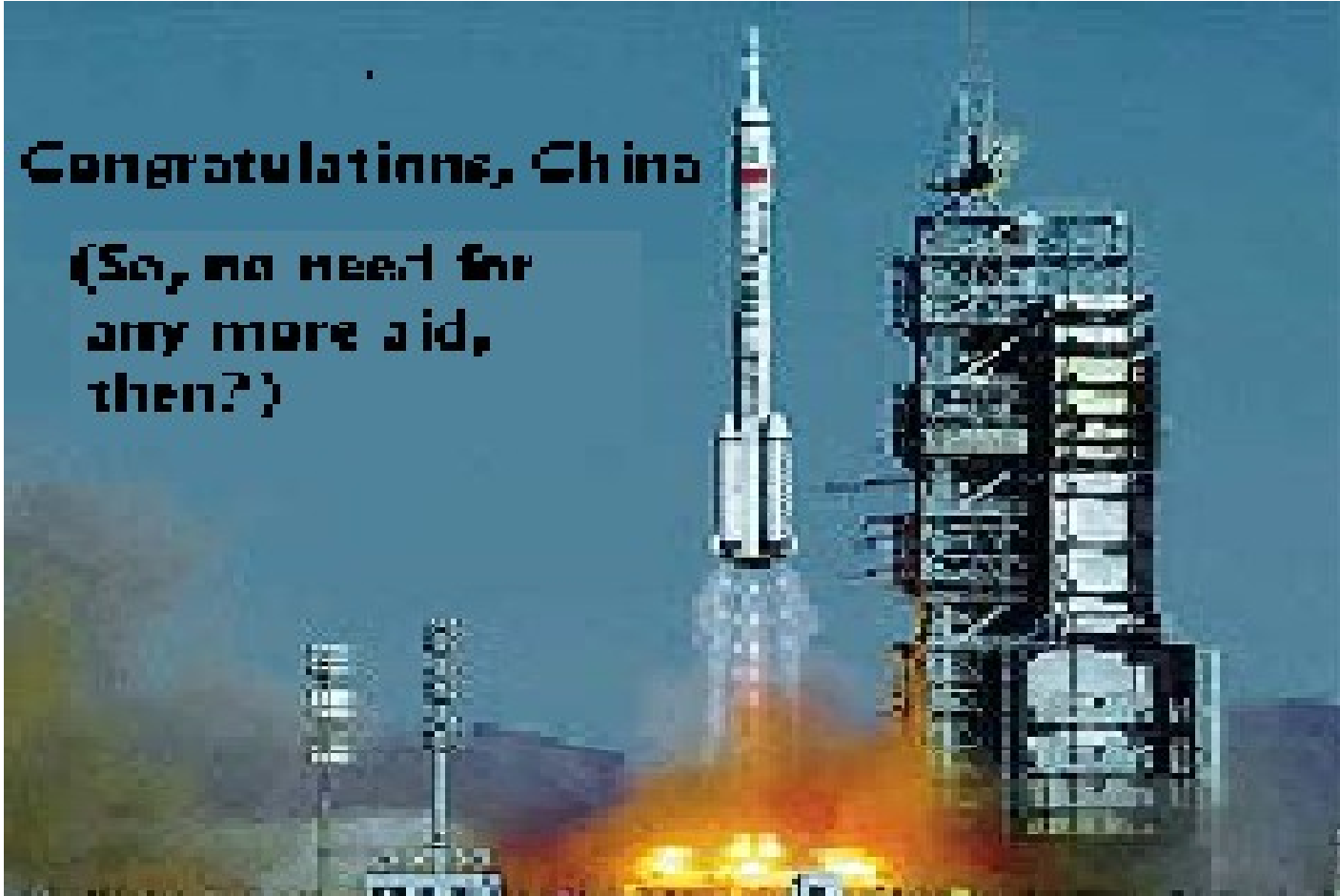




So, no need your any more aid ?

Congratulations, China

(So, no need for
any more aid,
then?)

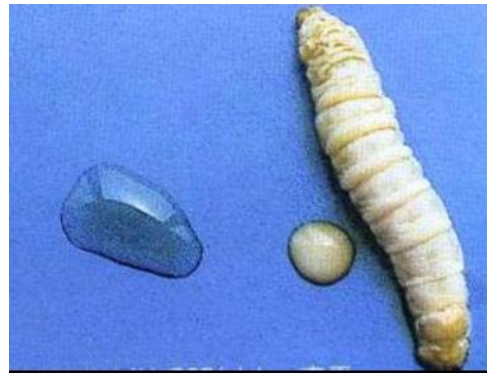






Climate changes and chemicals – the new sericulture **challenges**

How many diseases in silkworm?



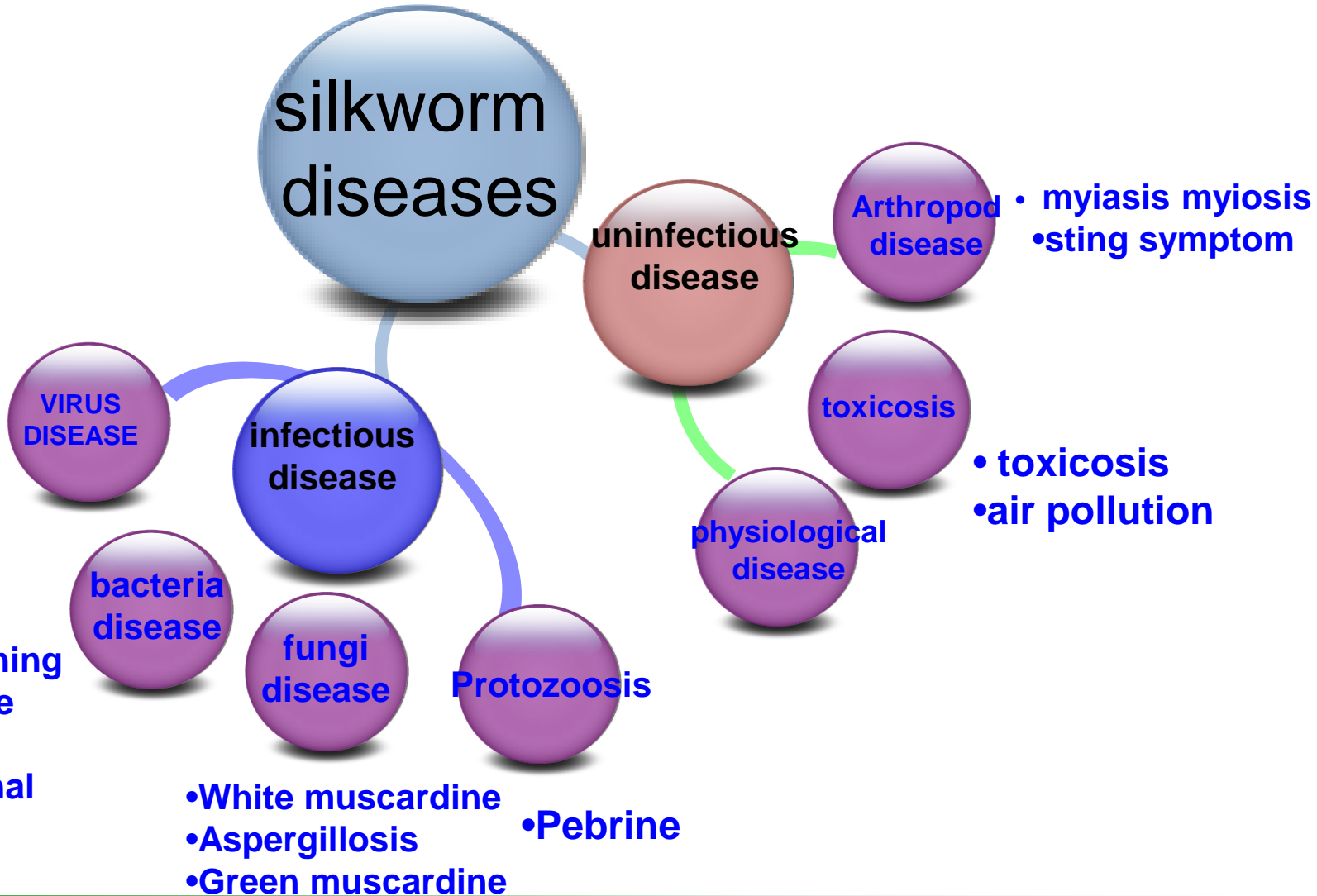


The types of silkworm diseases

- BmNPV
- BmCPV
- BmFV
- BmDNV

- Blood poisoning
- Sotto disease
- Bacterial gastrointestinal
- ,etc

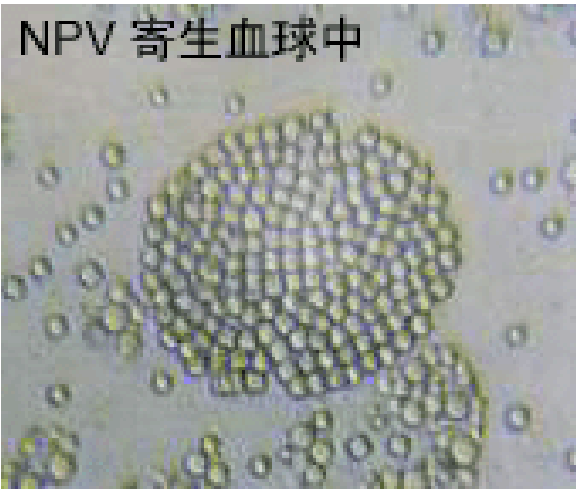
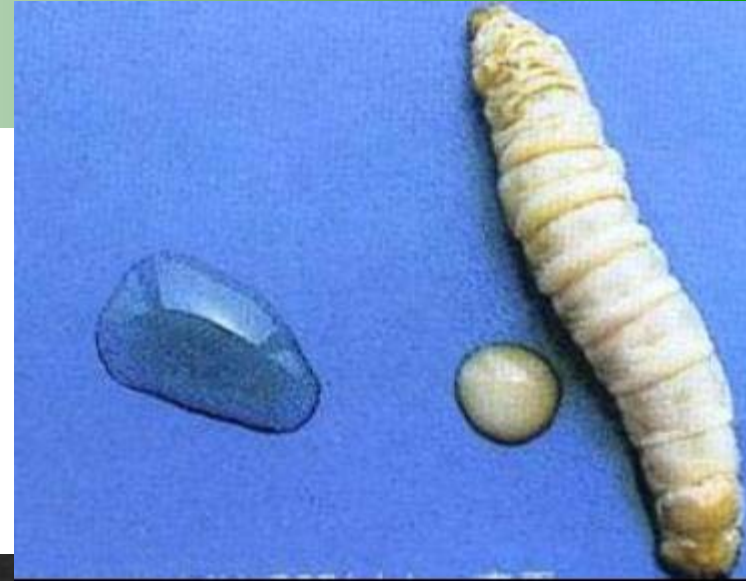
- White muscardine
- Aspergillosis
- Green muscardine
- Pebrine



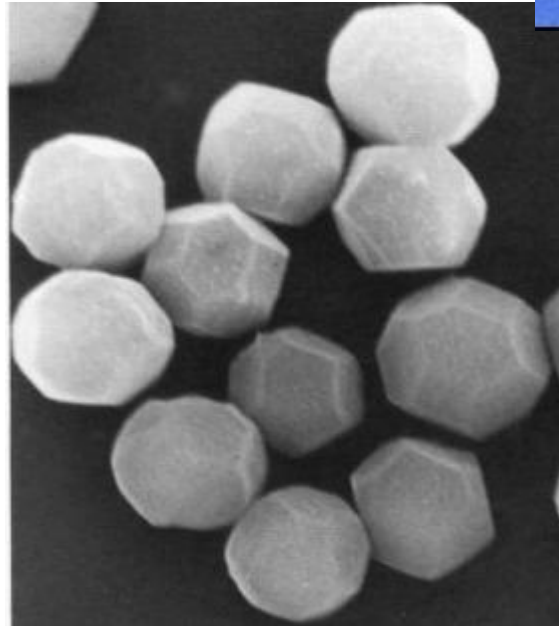


The main type of virus disease

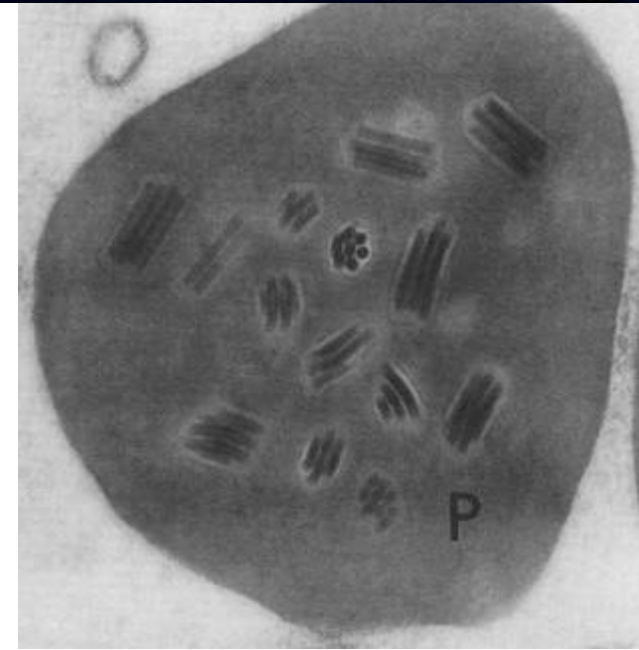
- 1) **BmNPV**



blood of sick silkworm
under light
microscope



NPB under e-scanning
electron microscope



virus particle under
transmission electron
microscope



silkworm virus disease prevalent in summer



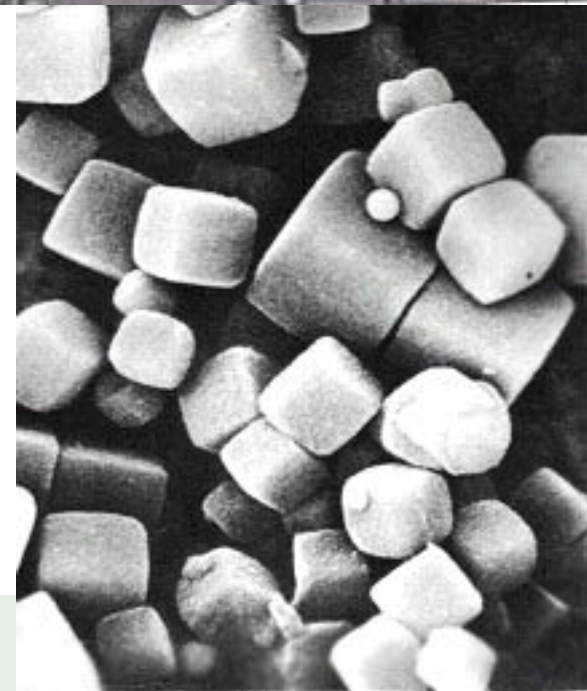
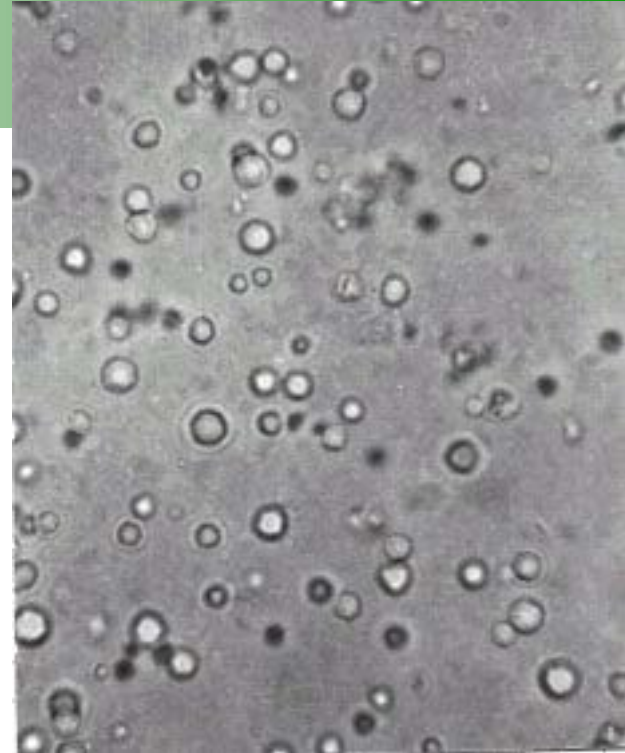


2、 BmCPV

the whole body and head are transparent . In this infected silkworm, BmCPV infection ration 75%, the rest of 25% flacherie or complication。



Iron silkworm?





The course of silkworm virus disease

Viral Disease	Instar	Time of Outbreak or Death	pathogenicity
NPV	1~3	3~4ds	acute
	4~5	4~6ds	subacute
CPV	1	4~7ds	chronic
	2~3	6~10ds	chronic
	4~5	8~12ds	chronic
DNV		7~12ds	chronic

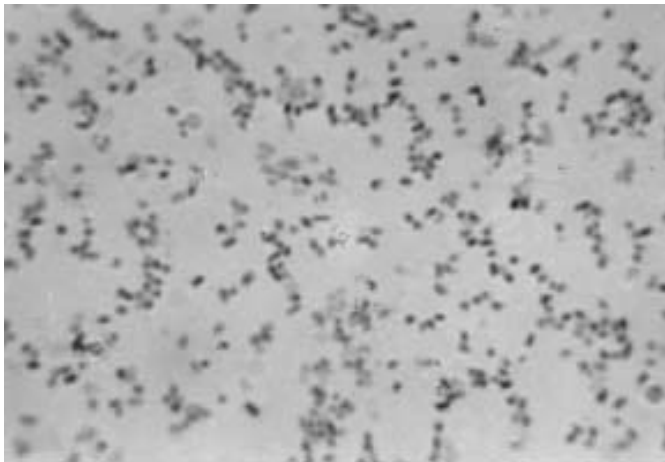
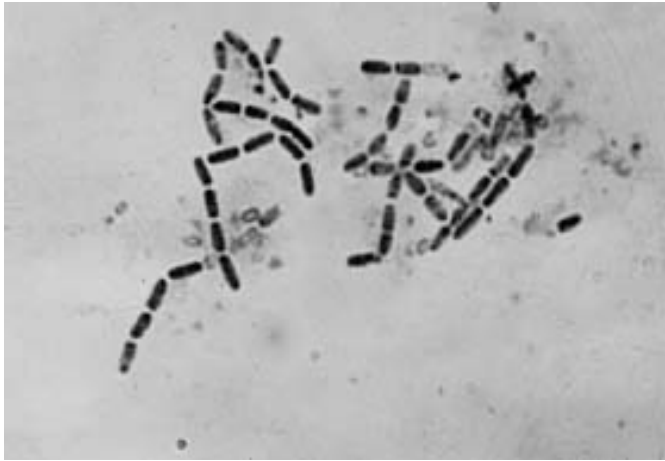


Bacterial disease





The silkworm bacterial disease



Bacillus thuringiensis (B. t.)

Blood poisoning



Fungi disease——muscardine disease

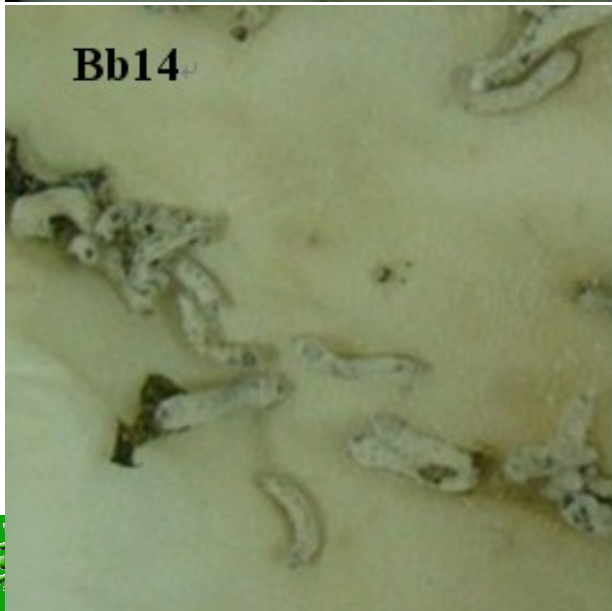
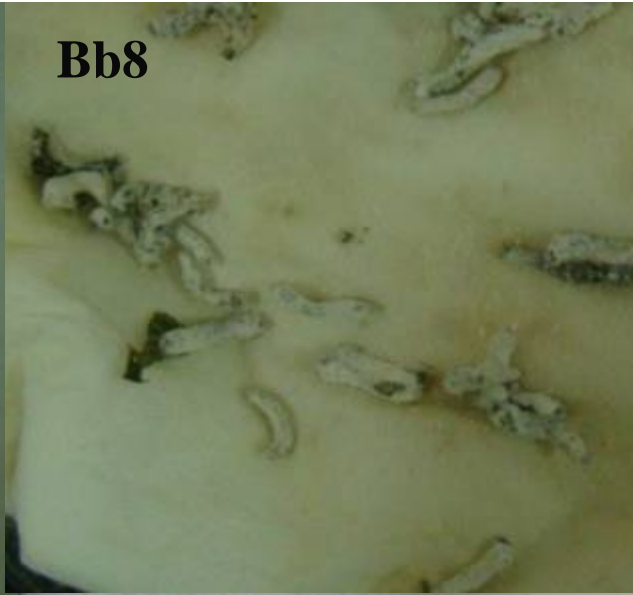


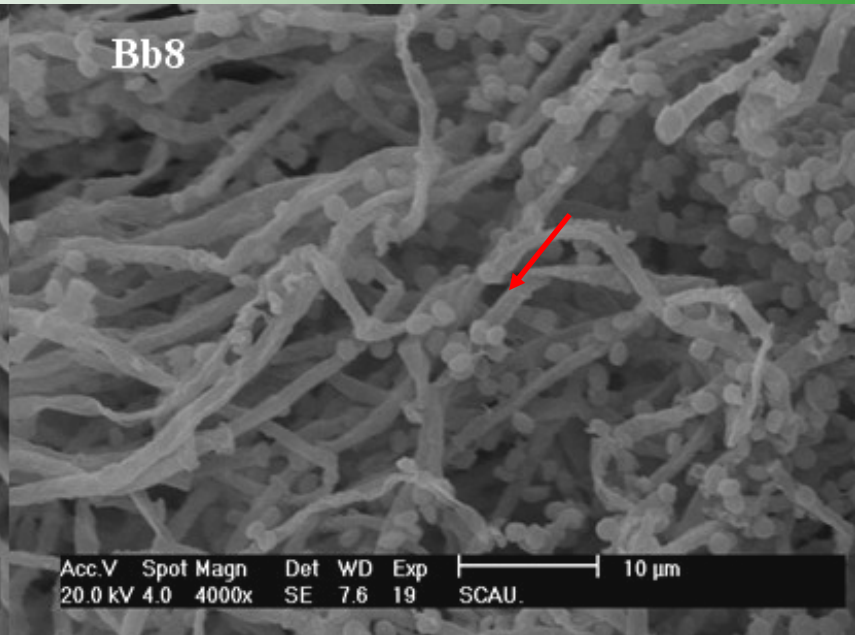
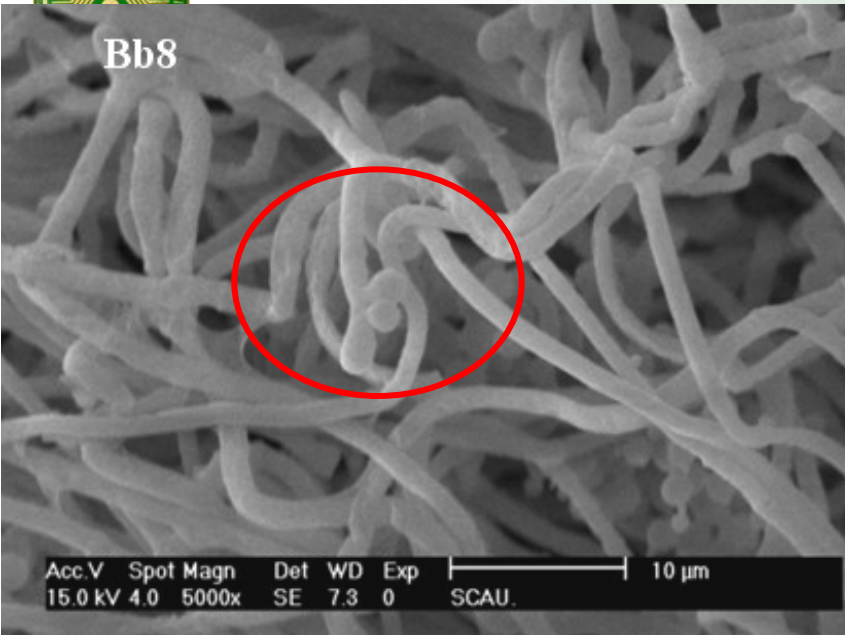


Aspergillosis

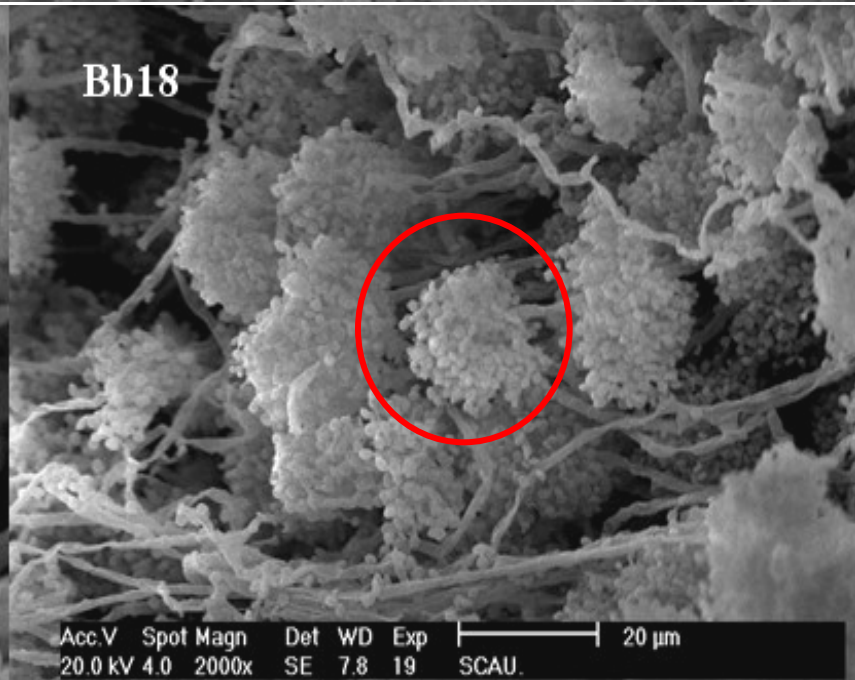
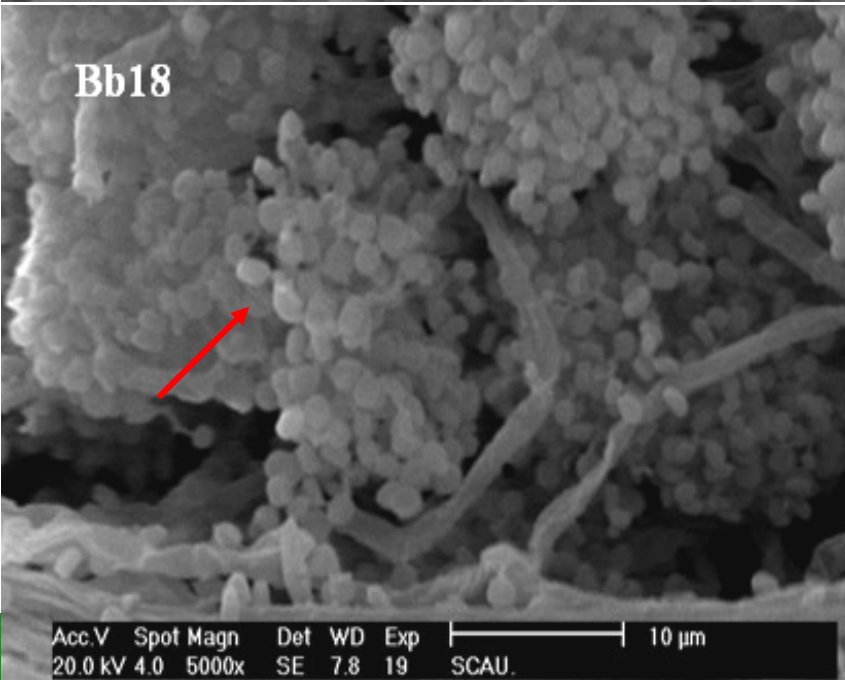
Aspergillus spp.

Symptoms in different isolates of *B. bassiana*





Bb8
B. bassiana



Bb18
Paecilomyces farinosus

B. bassiana Mixing infection in the silkworms

unit : head

No. of Mix with infections	Total	Day1	Day2	Day3	Day4	Day5	Day6	Total infection ratio %
		No. of 2 nd infections						
1	100	0	0	0	80	87	100	100%
2	100	0	0	4	98	100	0	100%
3	100	0	1	6	96	100	0	100%
4	100	0	2	11	100	0	0	100%
5	100	0	10	28	100	0	0	100%
CK	100	0	0	0	0	0	0	0

Note : 0 means no died



Pebrine disease in silkworms



not molting



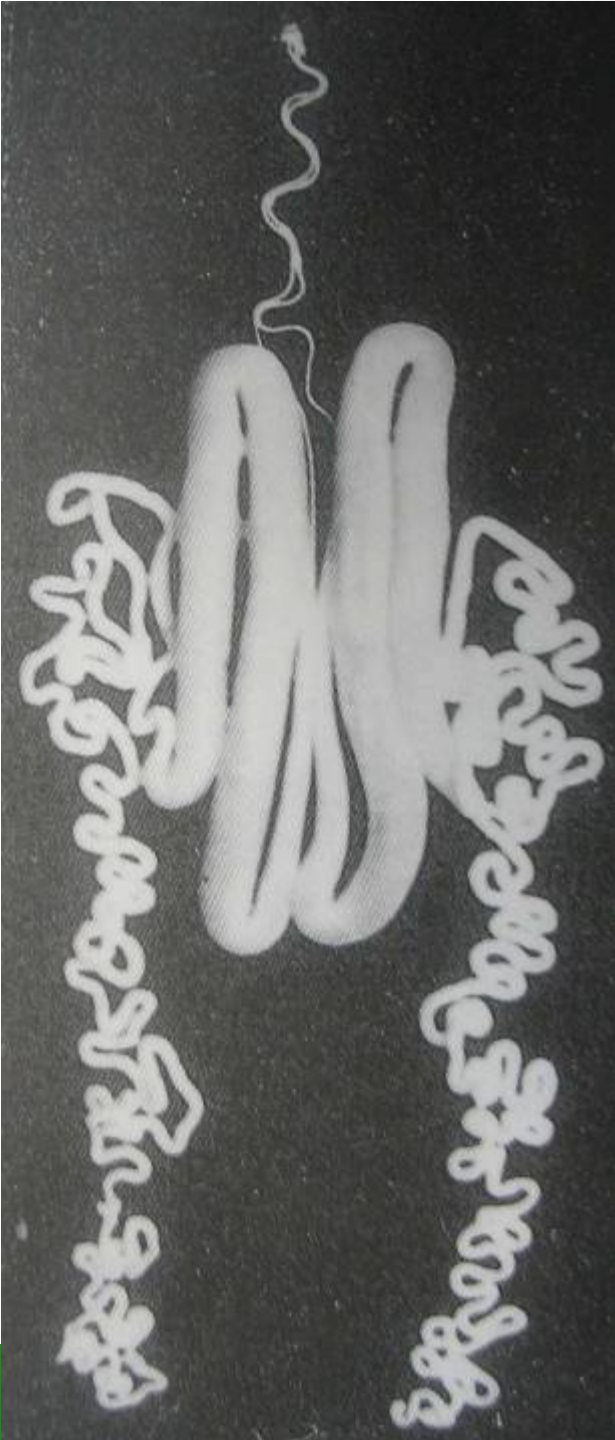
Pepper spot



batch of silkworms

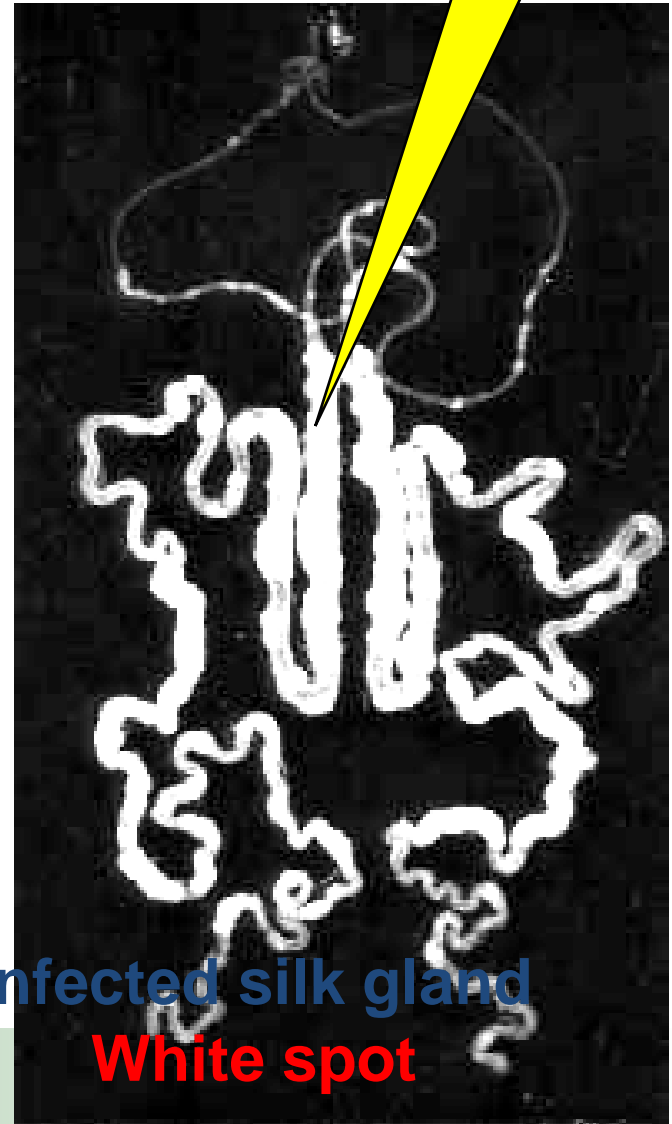


Infected silk gland



Normal silk gland

Infected silk gland



Infected silk gland
White spot

Silkworm pebrine is under control in Guangdong & Guangxi, South China





uninfectious silkworm disease——Arthropod

disease

myiasis



Exorista Sorbillans :Parasitic fly disease





myiasis



Exorista Sorbillans lay eggs on silkworm body

Fly larvae, pupae and worm hole cocoon





Chemicals challenges: nosotoxicosis & toxicity

- toxicosis
- air pollution
- factory fumes





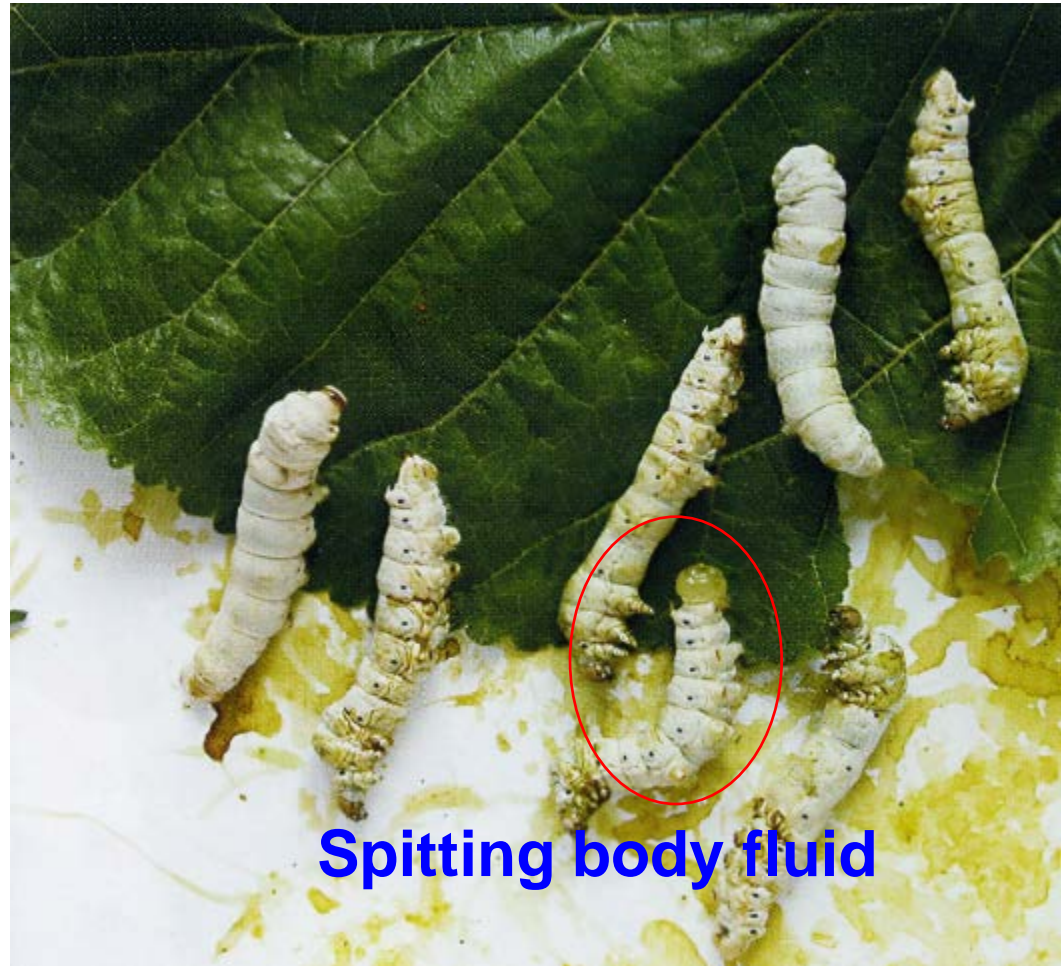
Industrial waste gas poisoning



the silkworm groups symptoms of Fluoride(**F & HF**) poisoning



the silkworm groups symptoms of sulfide(**SO₂**) poisoning





• **So, How we do**





Climate changes and chemicals – the new sericulture challenges our ideas and experience :



the influence factors of silkworm diseases incidence and their interrelationship
.....●



Integration of the silkworm diseases prevention and control
.....●



The practice of the system control of silkworm diseases
.....●

the basic conditions which influence silkworm disease occurring

1.Environment

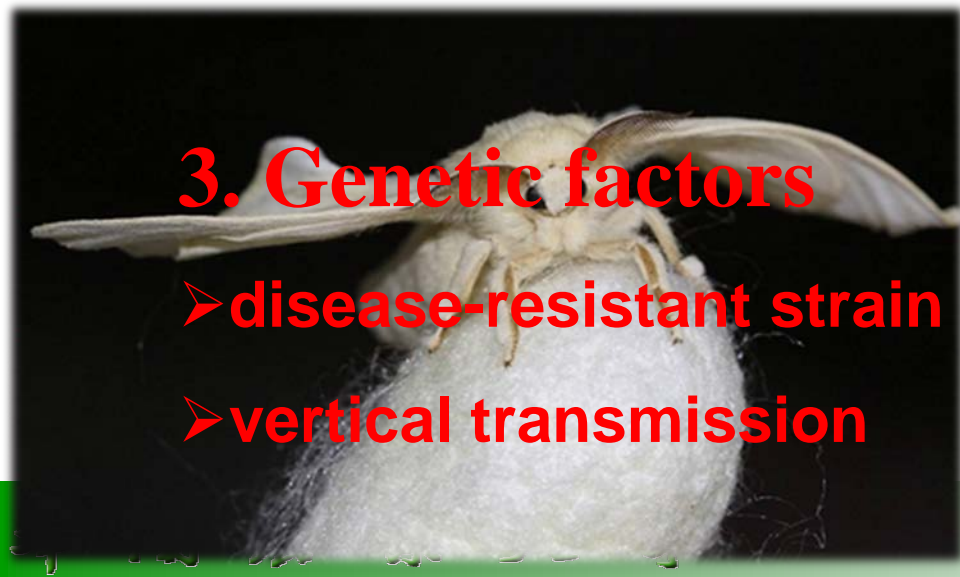
2.Breeding technology

- ◆ good mulberry and Satiation
- ◆ suitable meteorological environment

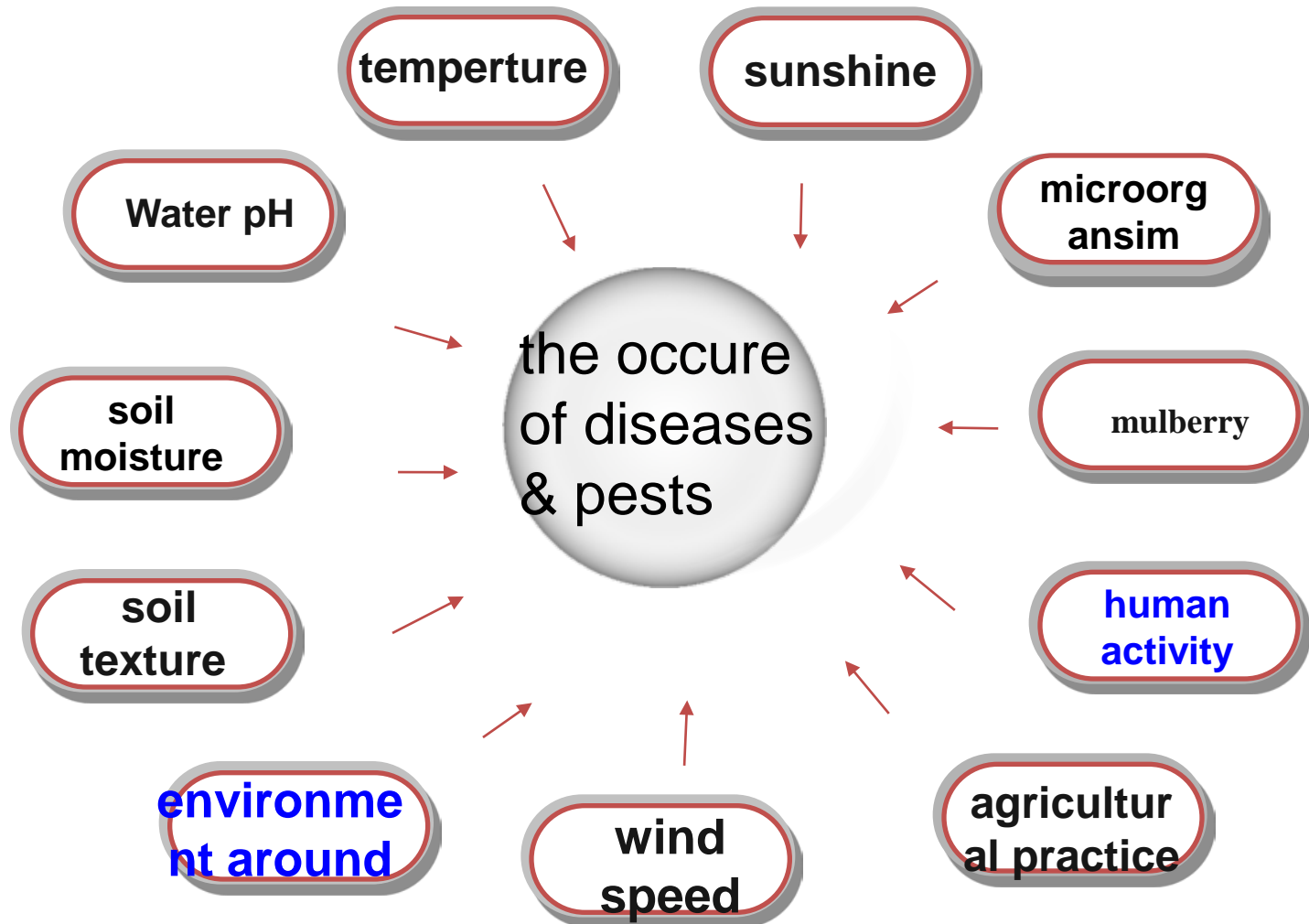


3. Genetic factors

- disease-resistant strain
- vertical transmission

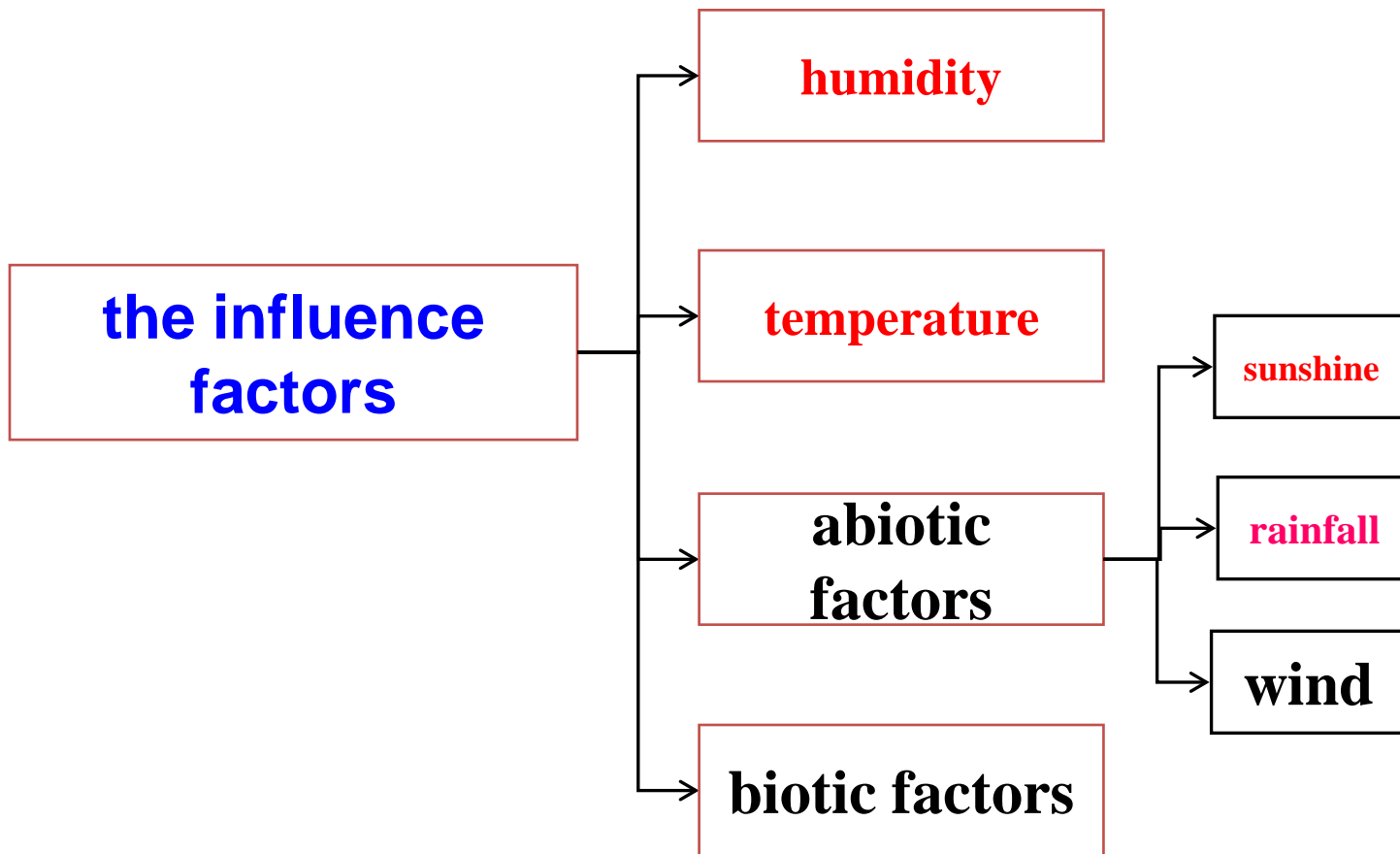


Environment condition influencing silkworm & mulberry diseases occurrence and prevalence





1.the influence factor of silkworm disease incidence and their interrelationship



the transmission route of pathogen in infected silkworm disease



Fig. the source and spread of silkworm pathogen

Transmission route of chemicals in infected silkworms

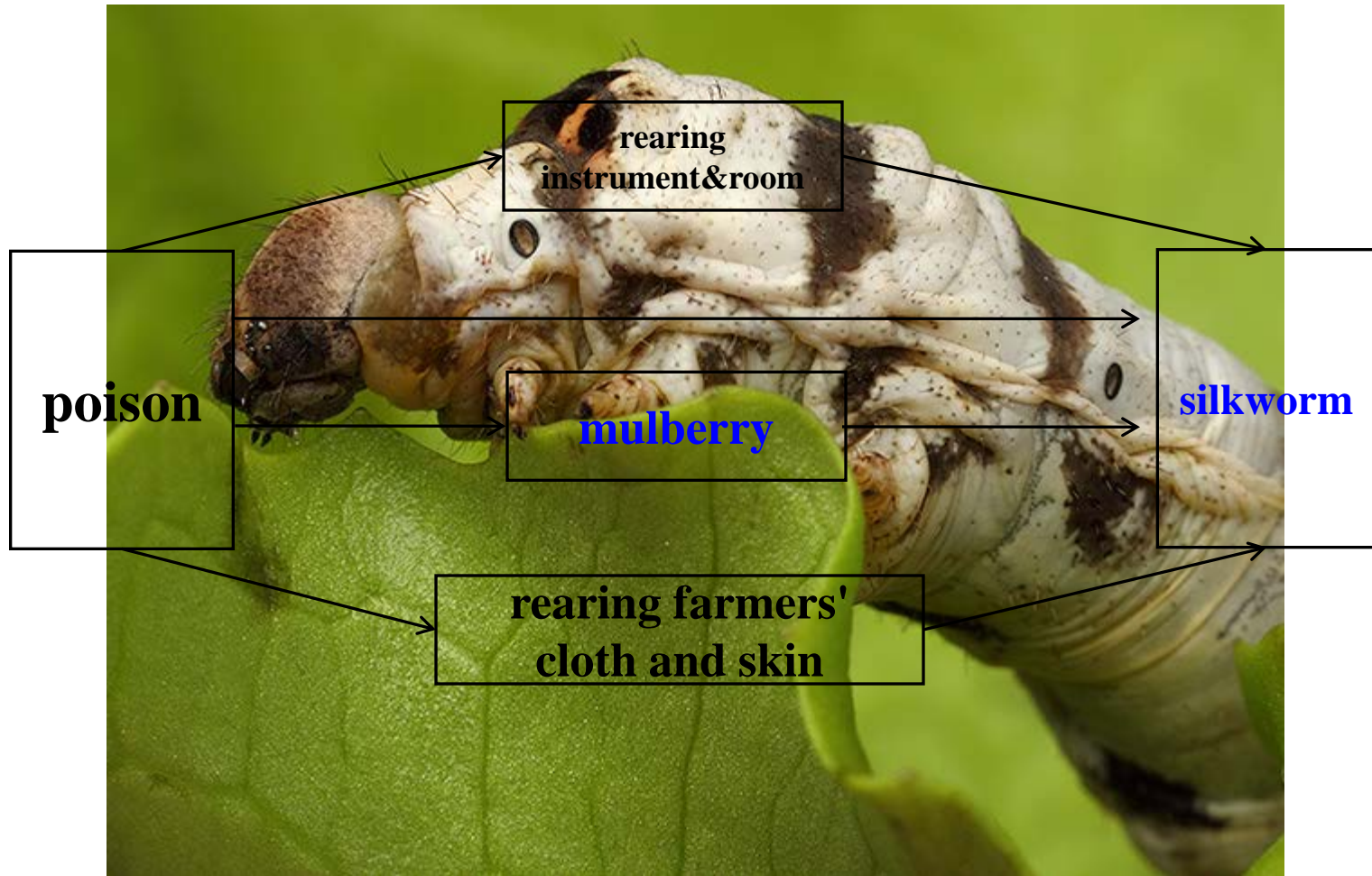


Fig. the main invade route of pesticide for silkworms



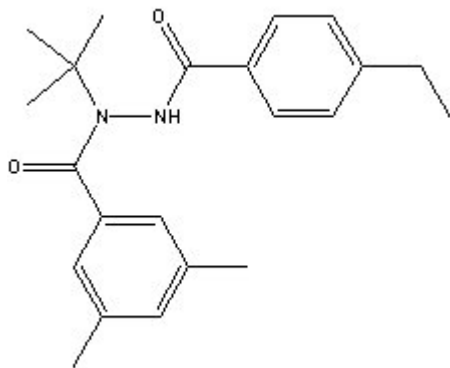
The hydrolysis half time period(in days)of 4 pesticides in different temperture and pH

pesticide	isofenphos- methyl	semiamitraz	pirimioxyph os	ethachlor
pH5, 25°C	374.6	188.4 too long!	37.3	320.9
pH7, 25°C	270.8	6.5	55.9	602.7
50°C	78.7	0.6	7.2	302.7
比值	3.44	10.83	7.76	1.99
pH9, 25°C	18.3	0.044	32.4	886.4

沈佐锐, 2009



the pesticide-resistance of the silkworm?



Tebufenozide(LC50 mg/Kg=0.0166)

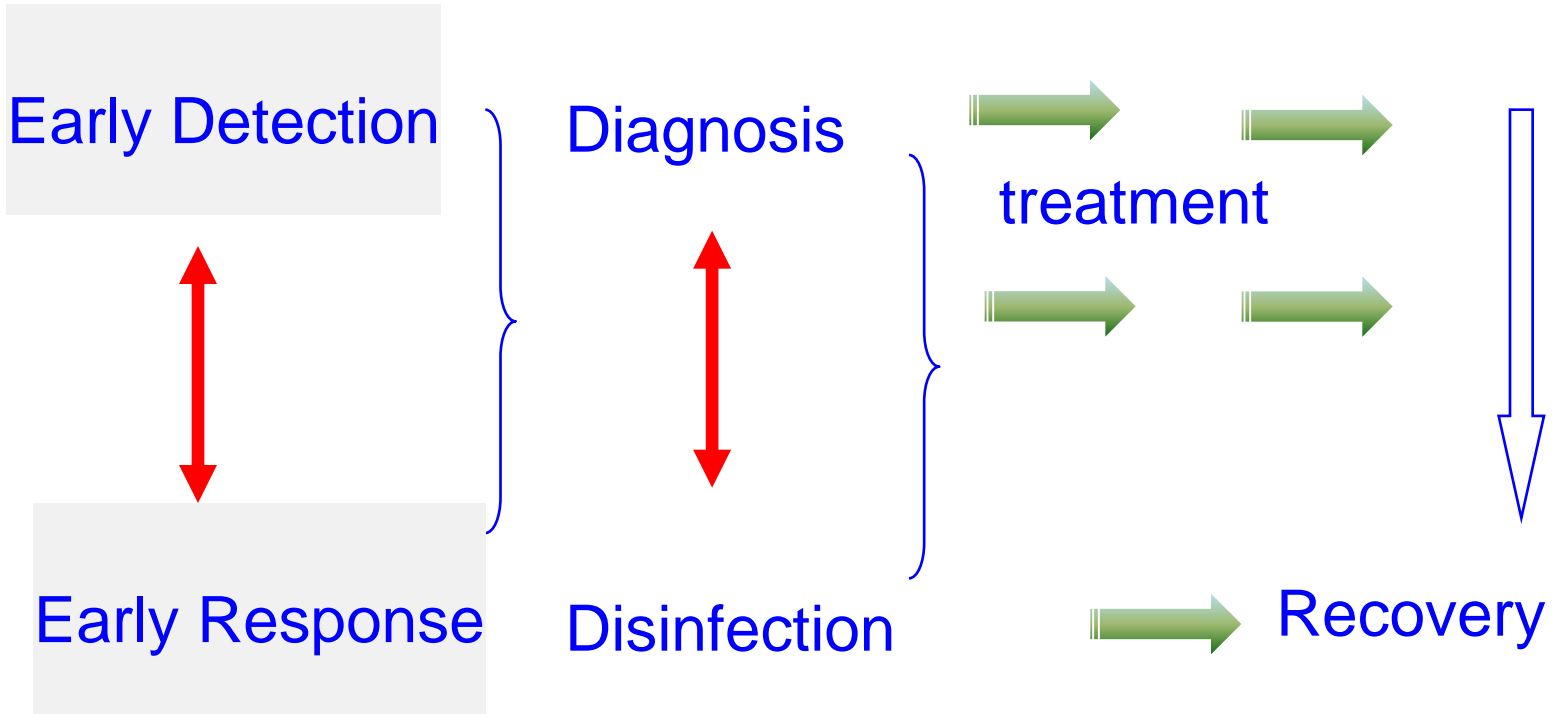
Tebufenozide suspension



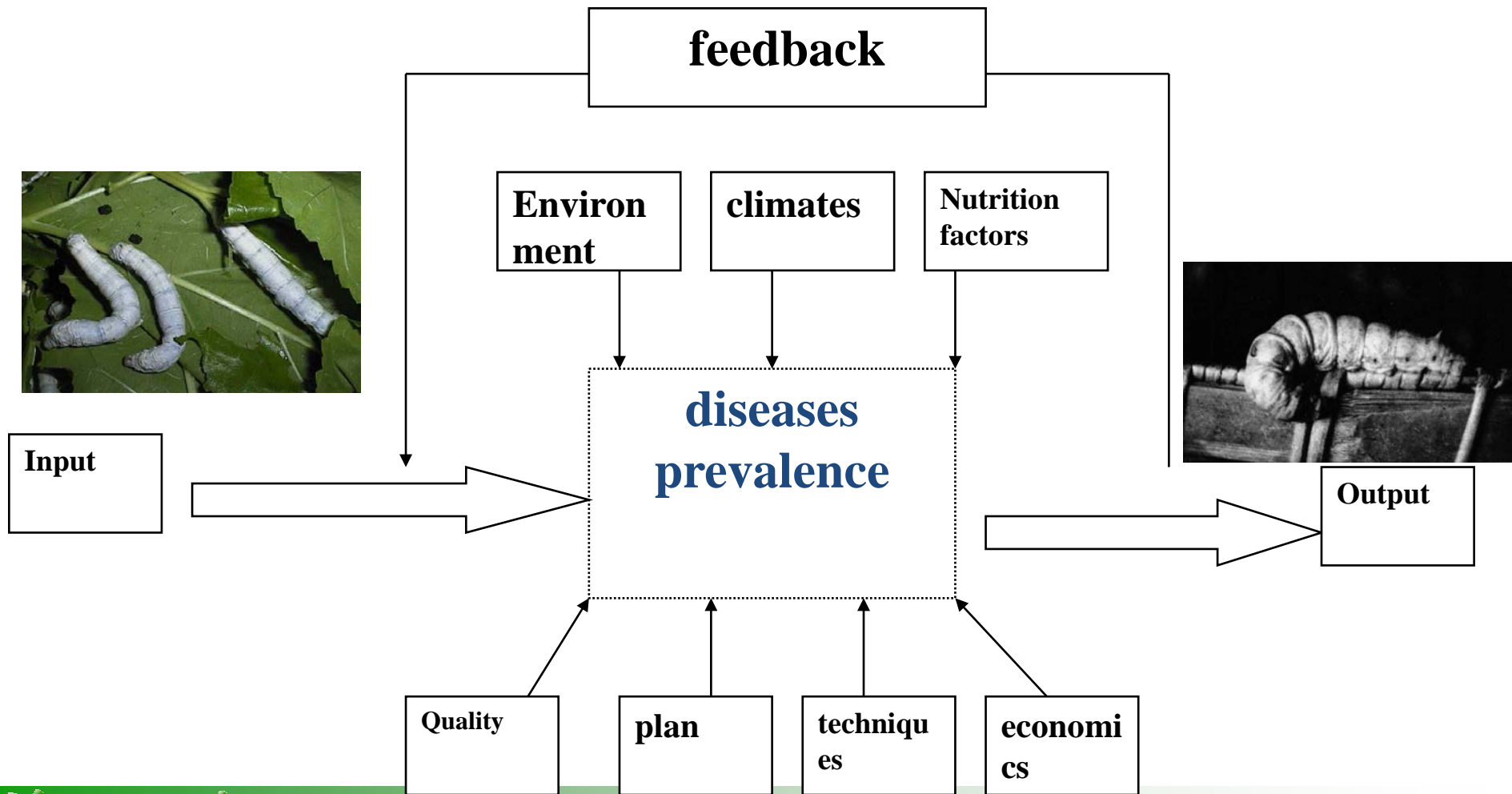
(孙新友等, 2011)



2. Integration of the silkworm disease prevention and control



2. Integration of the silkworm disease prevention and control' **Strategy: sysmatics control**





- 1)、 The practice of the system control of silkworm disease
- 2)、 Control standards and procedures
- 3)、 Silkworm disease detection technology
- 4)、 disinfect technology
- 5)、 the research of Treatment drug research
- 6)、 integration and development of Equipment



Rapid detection of environment pathogenic microorganisms

↓ Total number of colonies of the test piece

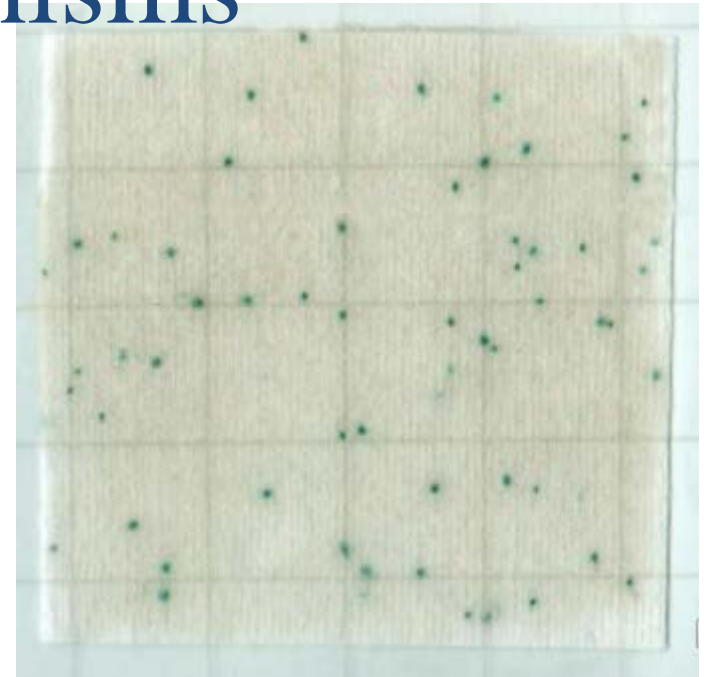
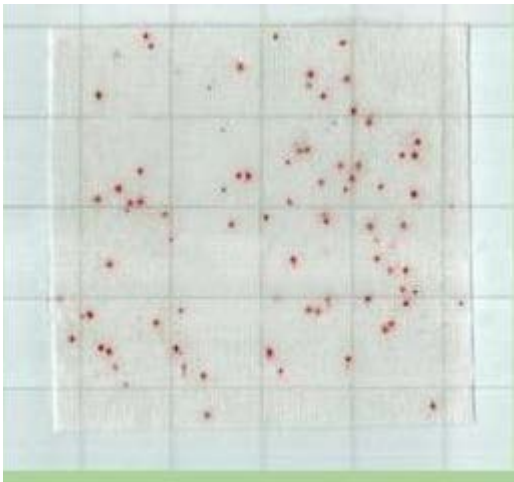


↑ Fungal yeast test piece



Rapid detection of environment pathogenic microorganisms

↓ Total number of colonies of the test piece



↑ 霉菌酵母测试片
Fungal yeast test piece

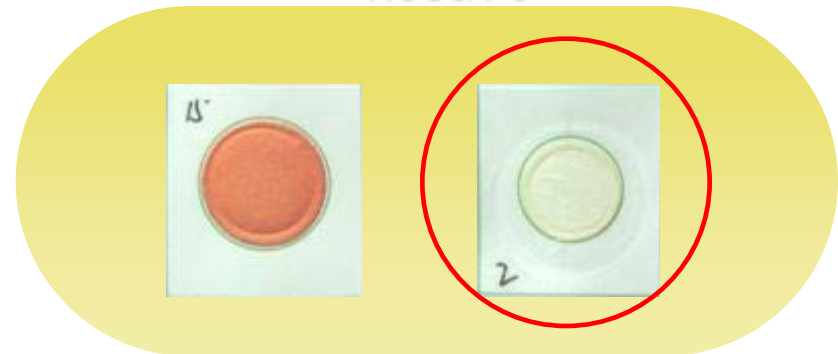


2) Mulberry pesticide residues detection

Kit for mulberry pesticide residues detection



Result





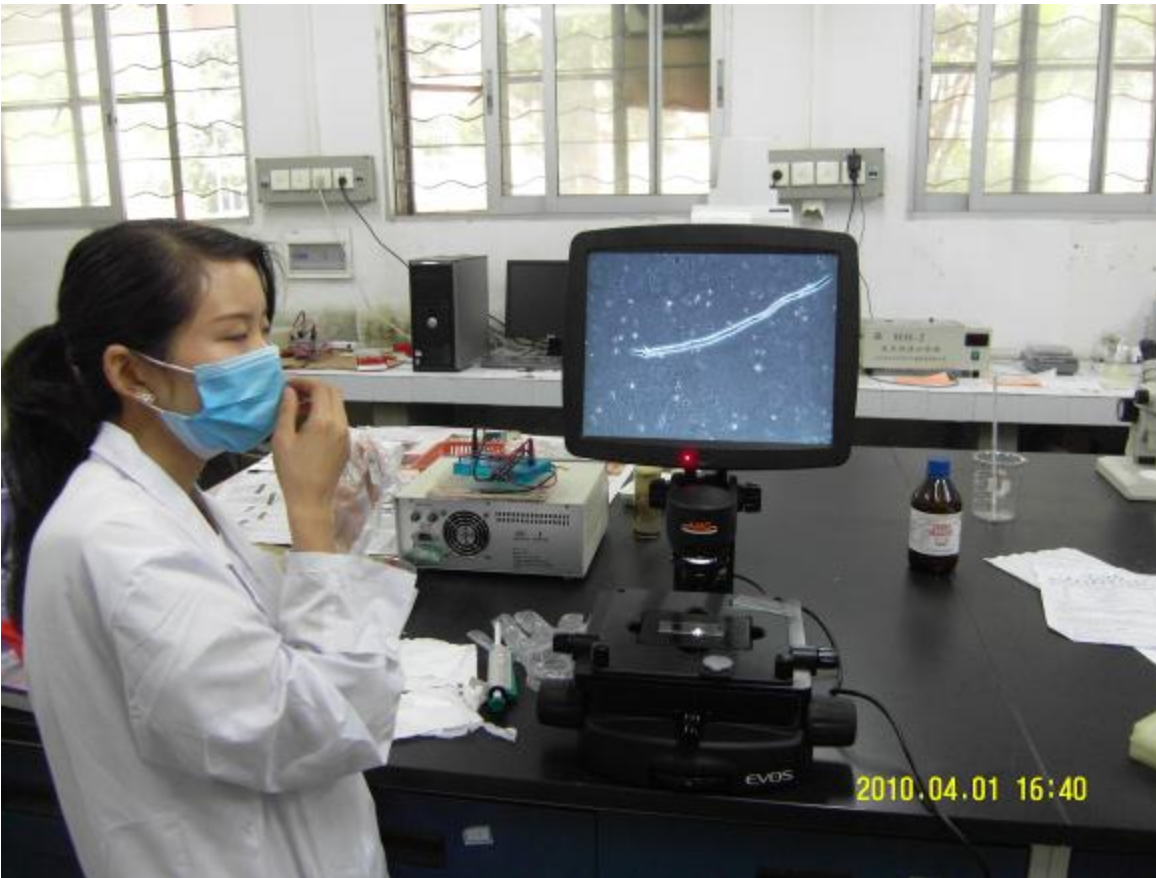
3) Research of visualization microscope techniques







USA EVOS Large screen digital inverted microscope



AMG (AdvancedMicroscopyGroup)
produced, \$100,000USD/unit





=



Biological Microscope

+



Stereo microscope

+



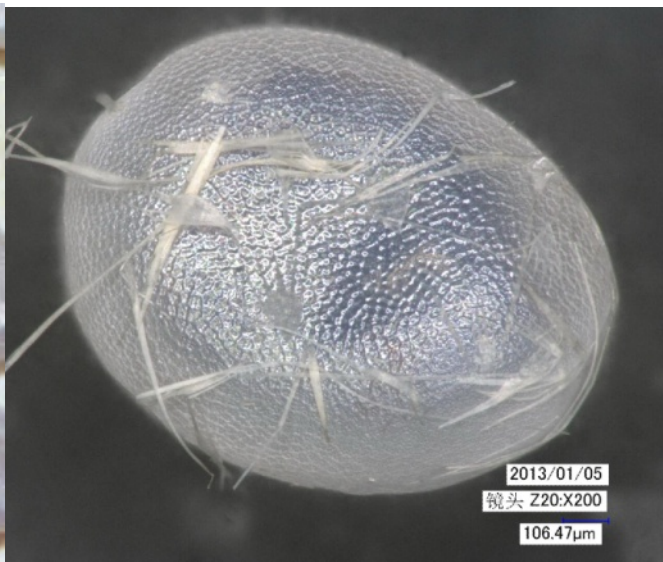
Fluorescence microscope

+



Computer

Multi-function LCD digital microscope observe the silkworm eggs





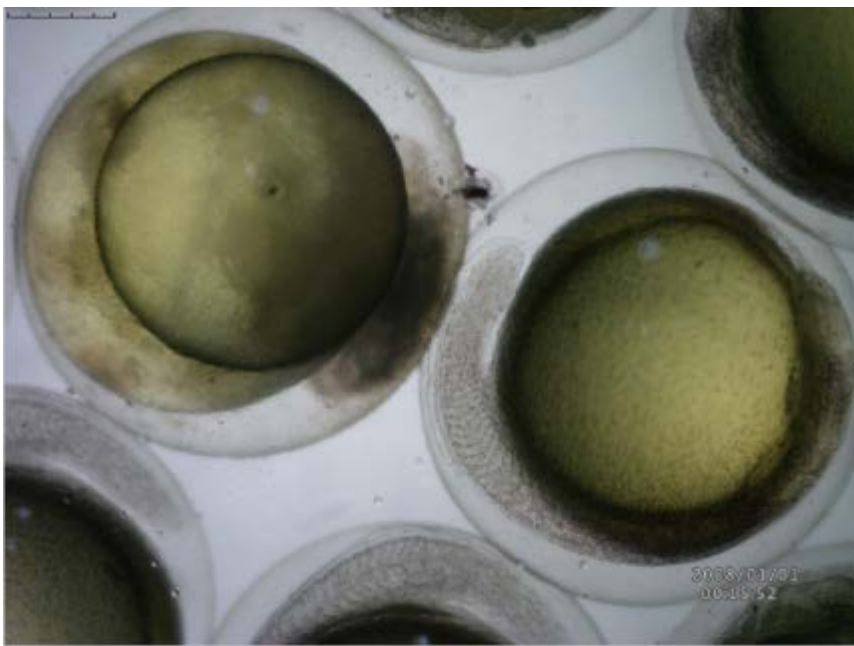
Observation of pests and disease



Powdery mildew



Digital Multifunction microscopy applied in aquaculture



Observation of zygote of fish



Fluorescence microscopy——Calcofluor White M2R Staining to identify Microsporidia



N.bombycis spore was stained Cyan fluorescent by Calcofluor White M2R



Observation

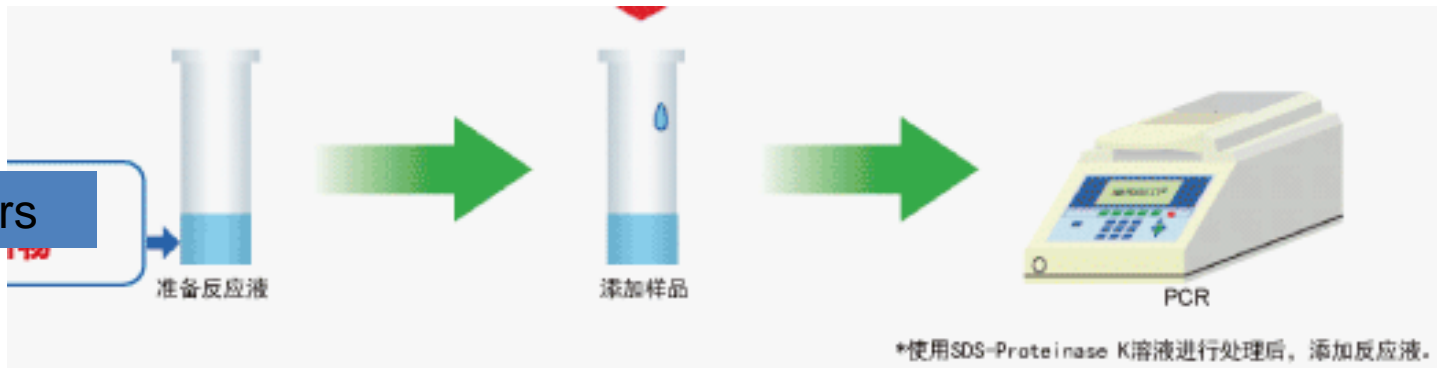


Observation of SILKWORM EGG



4) PCR & Loop Mediated Isothermal Application(LAMP)

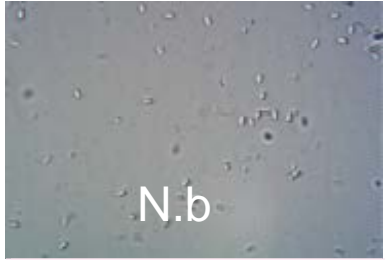
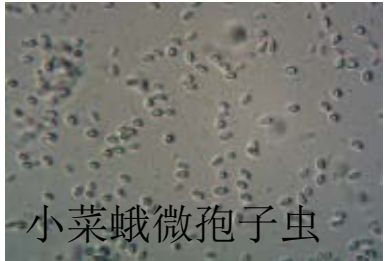
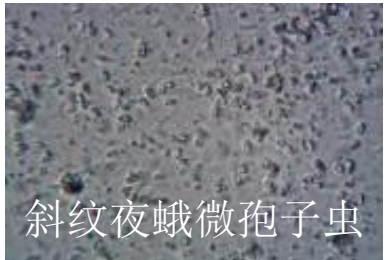
primers



the process of PCR amplification



A rapid method for extracting DNA of pebrine——Boiling method



适用于多种微孢子虫

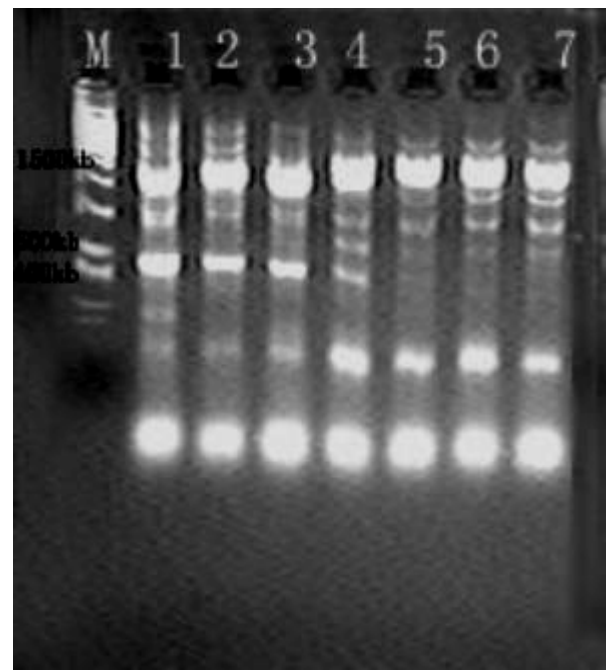


Figure Result of Electrophoresis of *Nosema bombycis* DNA

1 : positive ; 2 : 10^8 spore/mL ; 3: 10^7 spore/mL ; 4: 10^6 spore/mL ;
5: 10^5 spore/mL ; 6: 10^4 spore/mL ; 7 : **10^3 spore/mL** ; M : 100bp DNA Ladder.

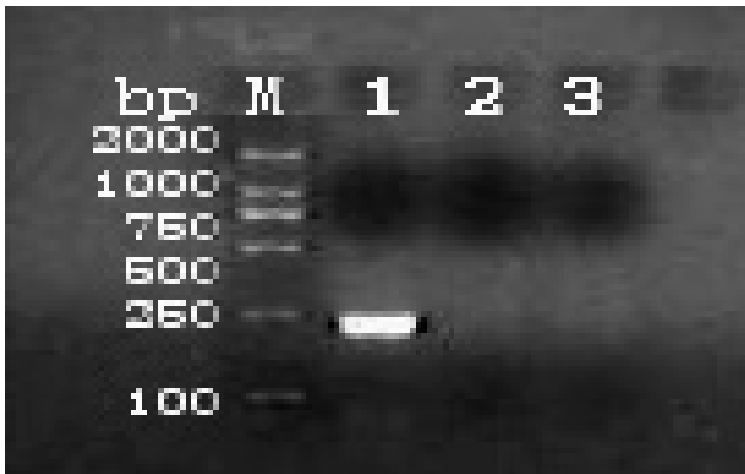
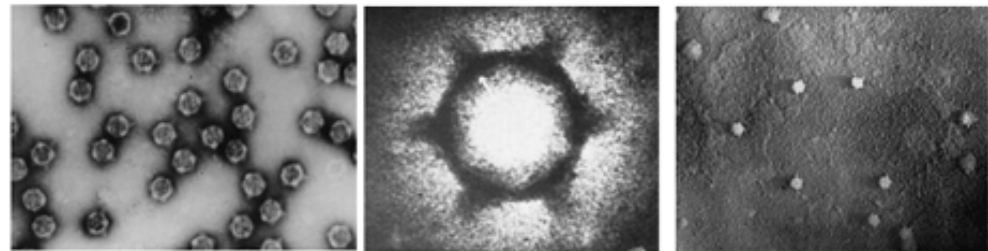


Figure RT-PCR result of BmCPV dsRNA

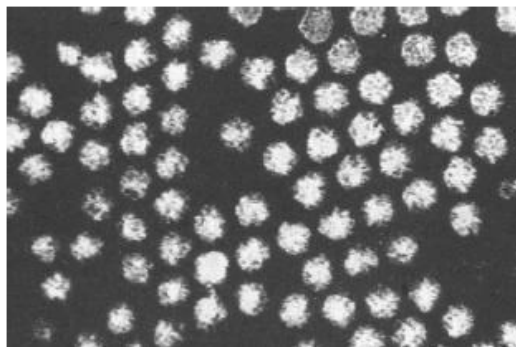
Note : M. DL2000 DNA ; 1. RT-PCR product of BmCPV dsRNA ; 2. RT-PCR product of healthy silkworm RNA ; ddH₂O

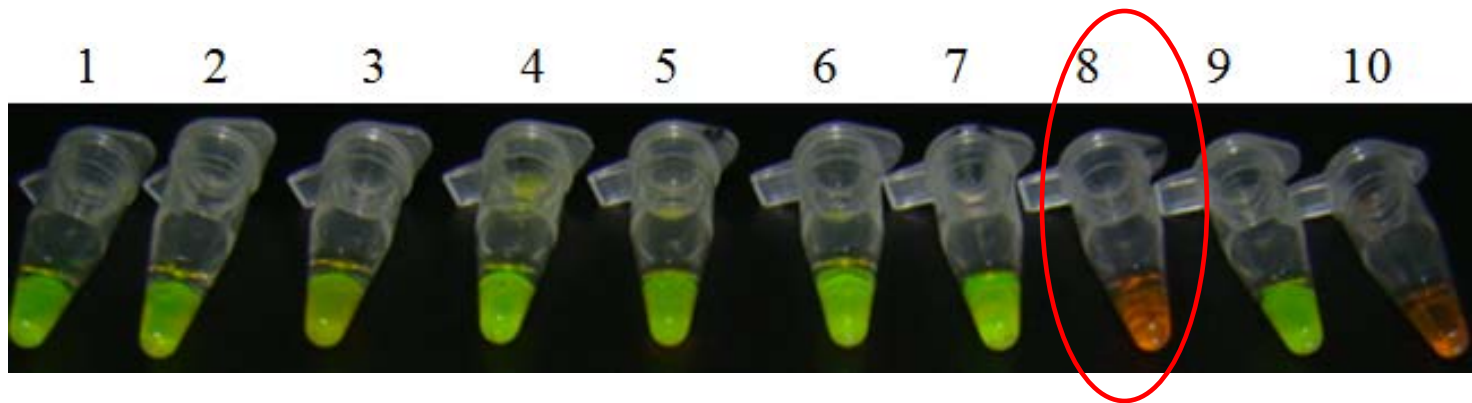




Patent:

- Target gene primers
- Positive template





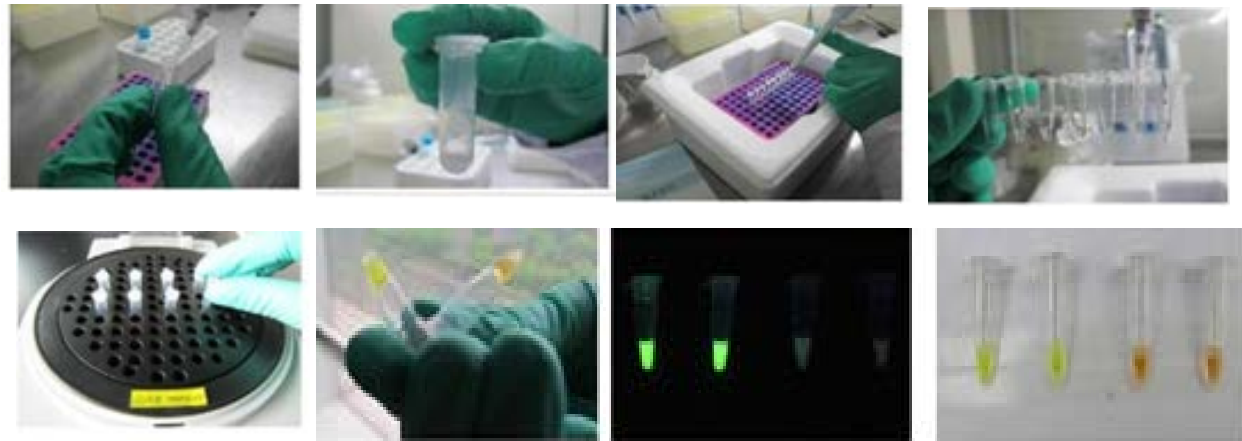
The sensitivity of BmDENV DNA test results

Note : 1-8 tubes were 1.5ng/ μ L、 0.15ng/ μ L、 15pg/ μ L、 1.5pg/ μ L、
0.15pg/ μ L、 15fg/ μ L、 1.5fg/ μ L、 0.15fg/ μ L BmDENV DNA ; 5
standerd ; 6 ddH₂O

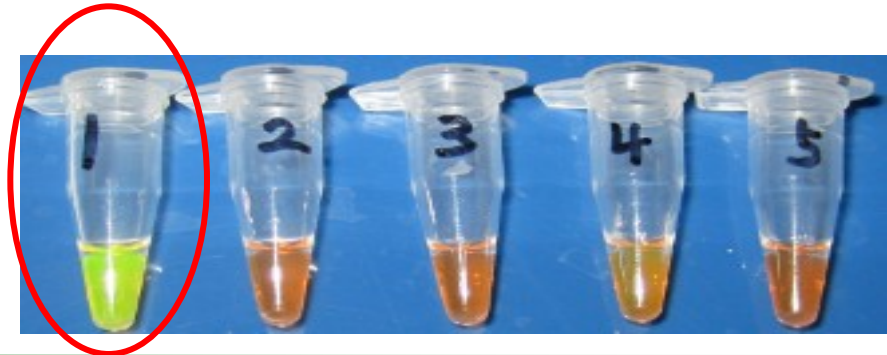


Detection of *Nosema bombycis* by LAMP

- LAMP diagnosis kit

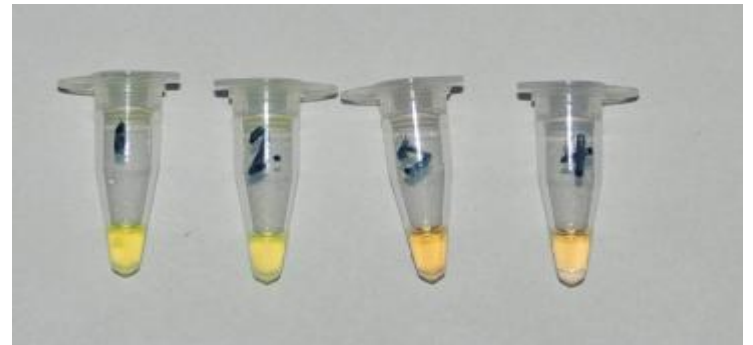


2.5hr





Boiling, mixing, observation? !





4、Developing of disinfection technology

- 1) Species of sericulture sterile
- physical methods (boil、hot air、sunlight、burn、compost)
- Chemical methods (Spraying, dipping, fumigation, disinfection powder)
 - Chlorine disinfectant: bleaching powder、calcium hypochlorite
 - foraldehyde
 - Lime
 - quaternary ammonium compounds
 - Sulfur
 - Antibacterial agents



- **1) Physical disinfection: temperature and humidity automatic control of the production environment**



Temperature and humidity automatic control equipment in silkworm rearing



4、Developing of disinfection technology

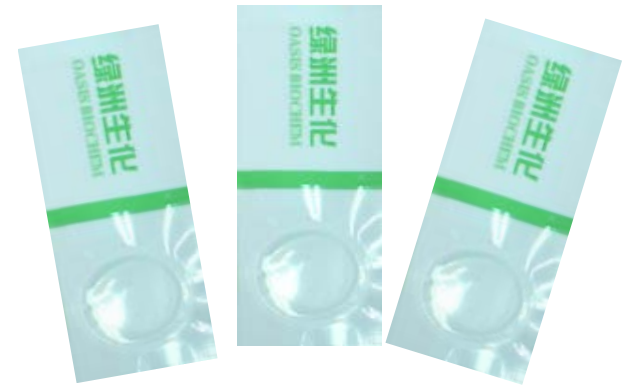
- Disinfectant drugs for silkworm



2009.04.11



Detection the chlorine of disinfectant



Suitable for detecting a variety of chlorine disinfectant formulated disinfectant chlorine concentration.



2) Disinfection equipment

①、② Ozone disinfection

③ Circulating air ultraviolet light, ozone, negative ions and air filtration and disinfection





- solar pest killing lamp





Captured by solar pest killing lamp



斜纹夜蛾

Spodoptera litura Fabricius



桑螟

Doaphnia pyloalis (Walker)



一点拟灯蛾

Asotacaricae Boisduval



粉蝶灯蛾

Nyctemera plagifera Walker



人纹污灯蛾

Spilarctia subcarnea Walker

Captured 32 kinds of insects, rate of microsporidia infection is 46.9%



Sanitation on Standard Operating Procedure (SSOP)

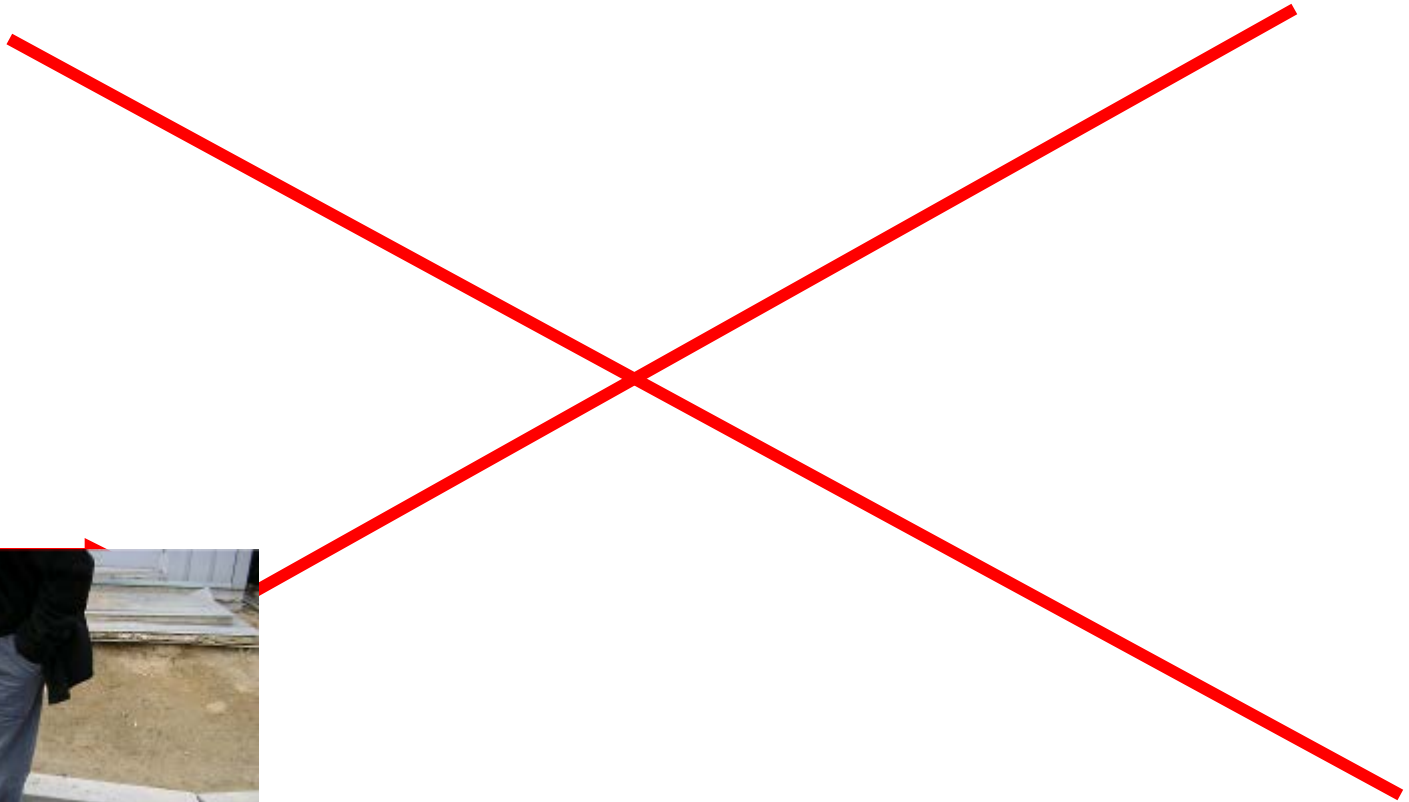


Disinfection pool

Disinfection of all tools and appliance



SSOP: changing your shoes, washing hands, etc..





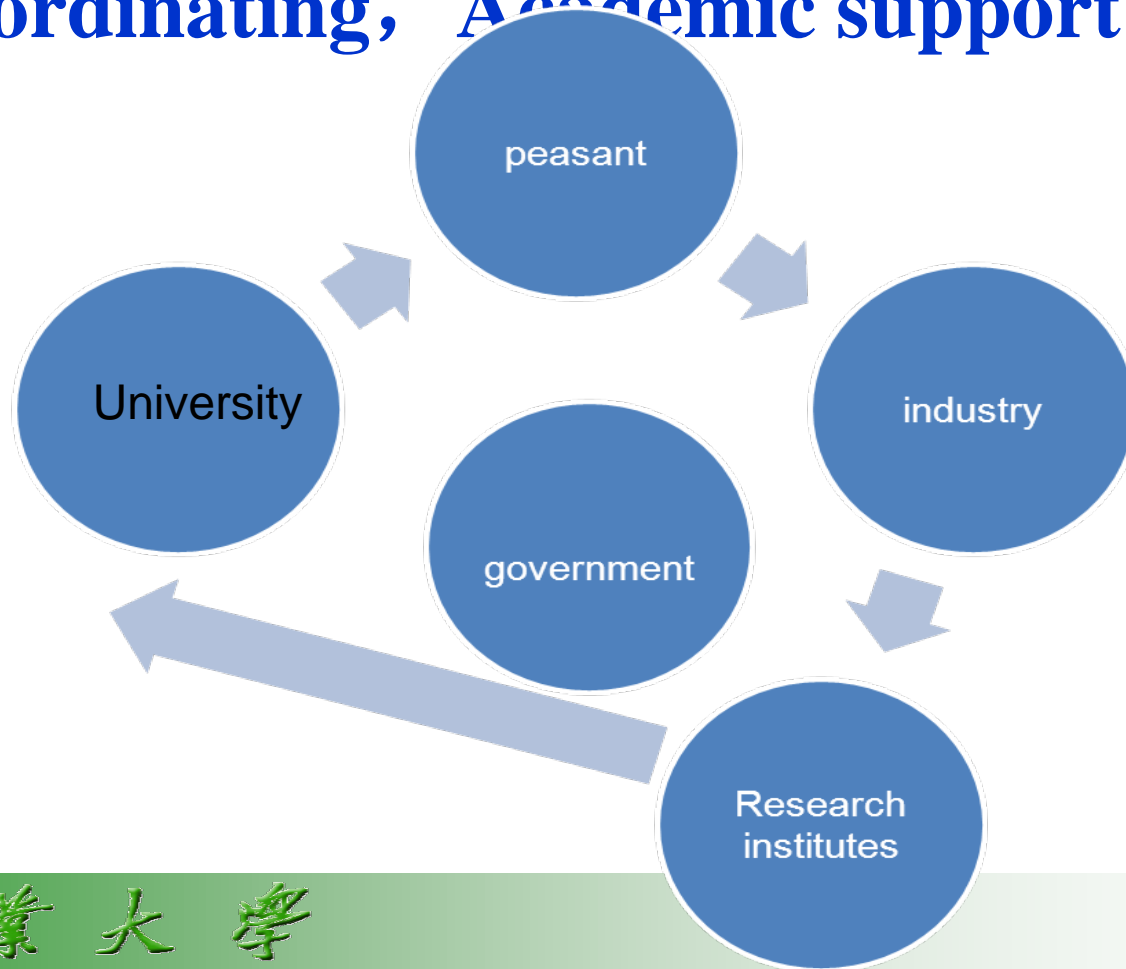
Disinfection on Standard Operating Procedure (DSOP)

消毒池



HOW to achieve effectively control of silkworm diseases?

Answer: government assist, enterprises coordinating, Academic support!





The main constrains and solution for sericulture development in BACSA Countries

1. Technology
2. Land productivity
3. Labour productivity
4. Machines
5. Marketing





Contact in RSTC _AP

Prof. CAO YANG, silkworm biology

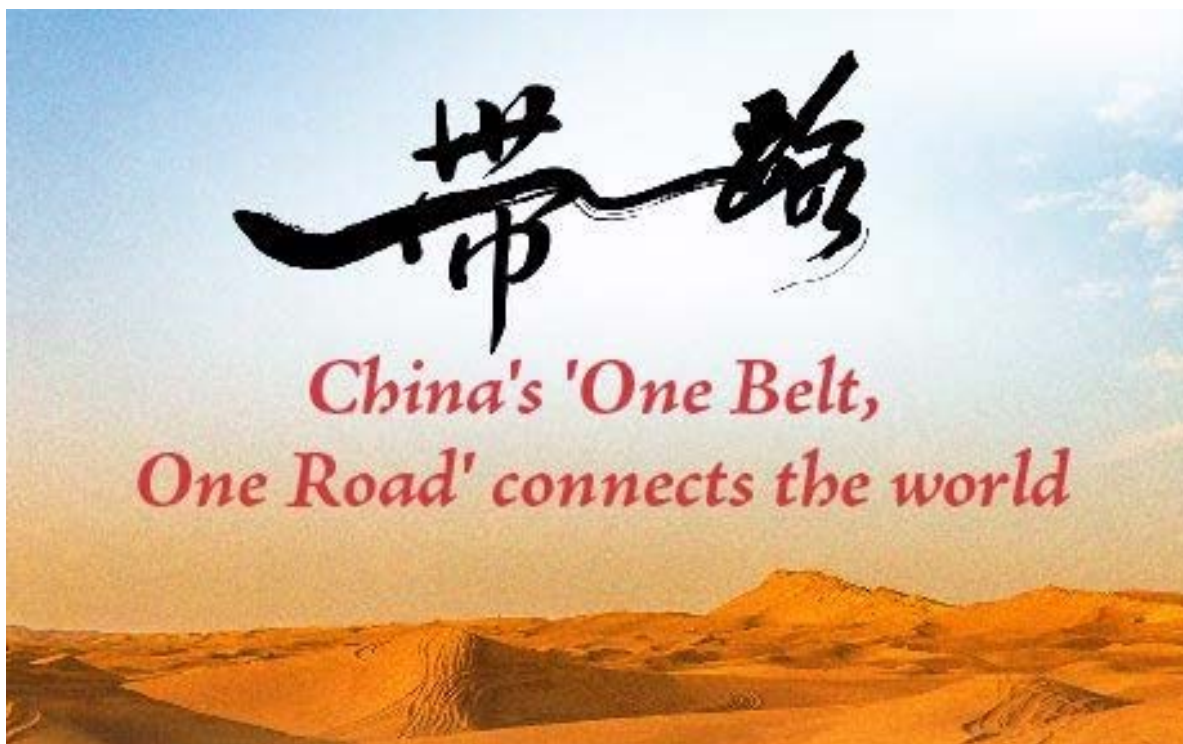
Prof. LIN JIANRONG, silkworm breeding

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Thank you for your attention !





21 Century's Silk Road

