

AFB: Control and Eradication

Oie



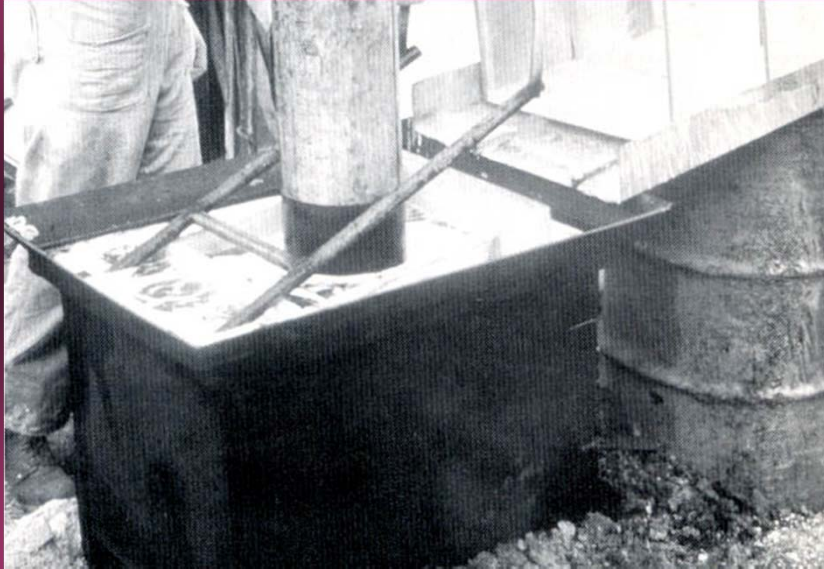
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Burning



Hot parafin wax



Antibiotic application: extender patties



Dusting



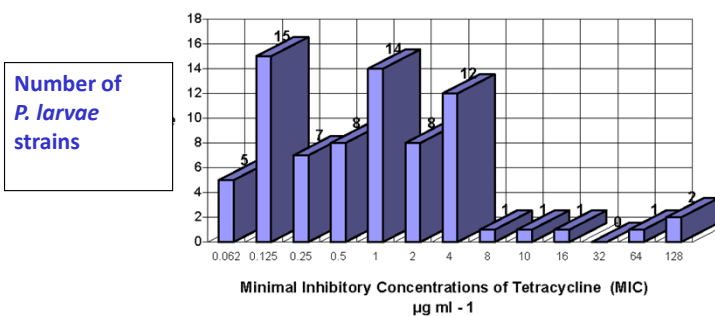
Management and Sanitation:

- Shaking bees
- Genetic Control
- Antibiotics



- New antibiotics with low residues in honey
- Biological Control Agents
- Integrated Pest Management

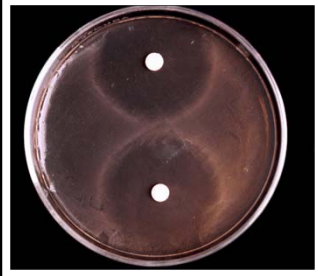
Tetracycline resistance



MIC values of 75 *P. larvae* strains from different geographical areas (0.062 – 128 $\mu\text{g/ml}$)

- MIC 50 : 1 $\mu\text{g/ml}$
- MIC 90: 4 $\mu\text{g/ml}$

Disc diffusion assay TC and OTC



NCCLS 5 µg discs

Resistant = ≤ 14 mm

Intermediate = 15 – 19 mm

Sensitive = ≥ 20 mm



NCCLS 30 µg discs

S = ≥ 19 mm

I = 15-18 mm

R = ≤ 14 mm

BSAC 10 µg discs

S = ≥ 20 mm

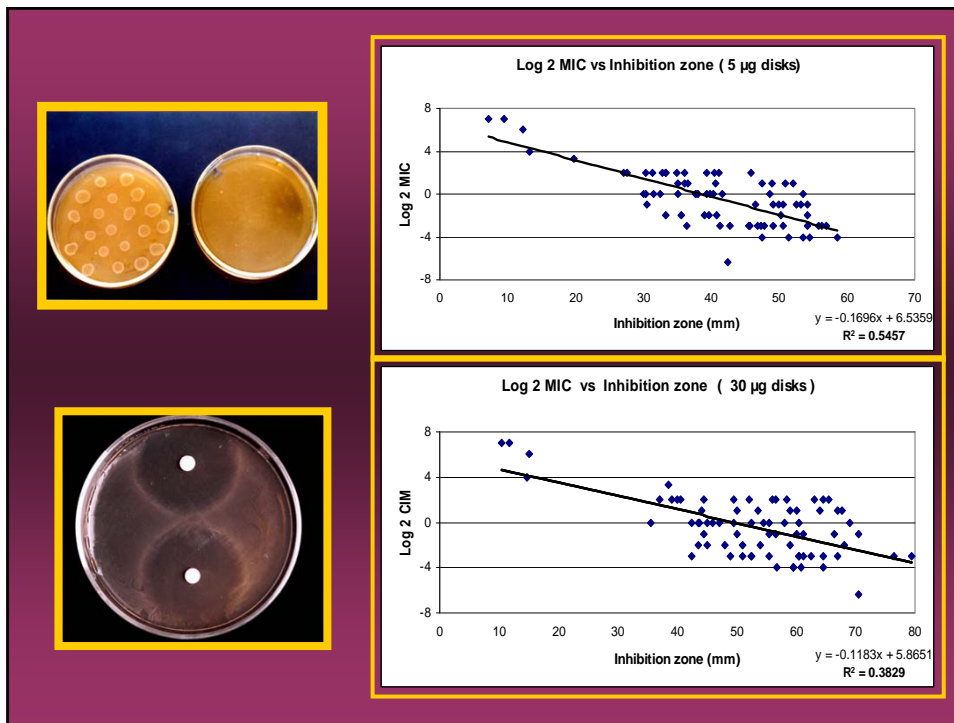
R = ≤ 19 mm

Minimal inhibitory concentrations (MIC values)



MIC's TC
NCCLS

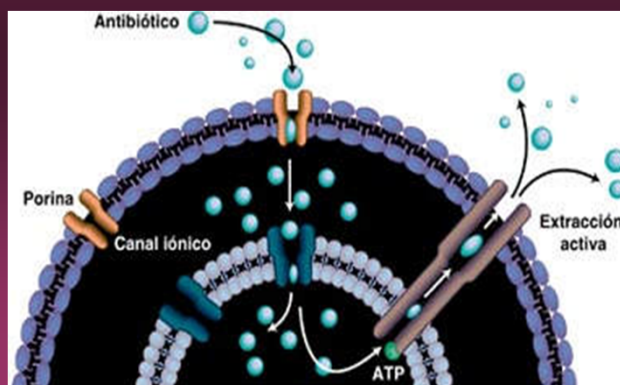
| | | |
|---|---|-----------------|
| { | S | ≤ 4 µg/ml |
| | I | 6 - 8 µg/ml |
| | R | ≥ 16 µg/ml |



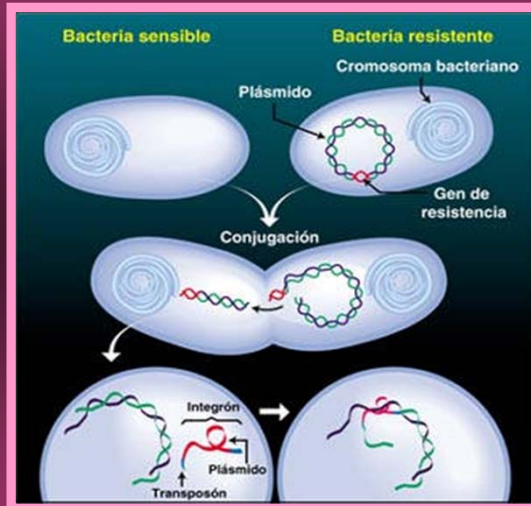
There are three main mechanisms underlying Tetracycline resistance:

- Energy-dependent efflux
- Ribosomal protection
- Enzymatic inactivation

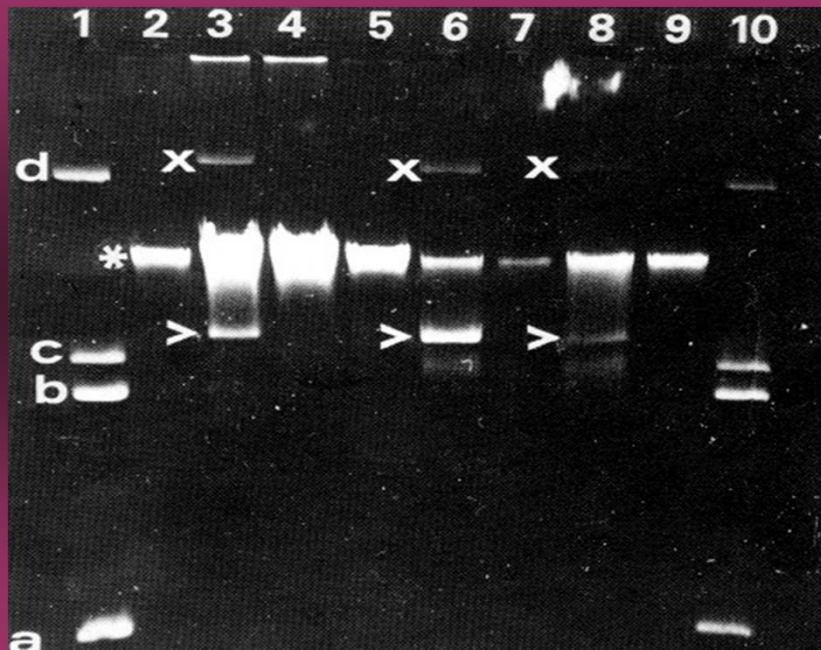
Being a) and b) the most widespread within bacterial populations. Resistance to tetracycline (Tc) is mainly due to the acquisition of *tet* determinants frequently associated with mobile elements. Approximately 40 different *tet* and *otr* genes have been reported in Gram positive bacteria, whereas in *Bacillus* and its related species, only the *tet(K)*, *tet(L)*, *tet(M)*, *tet(W)* and/or *otr(A)* determinants have been reported. Recently, natural *P. larvae* plasmids have been shown to confer Tc-resistance by *tetK* and/or *tetL* genes, respectively.



Antibiotic resistance

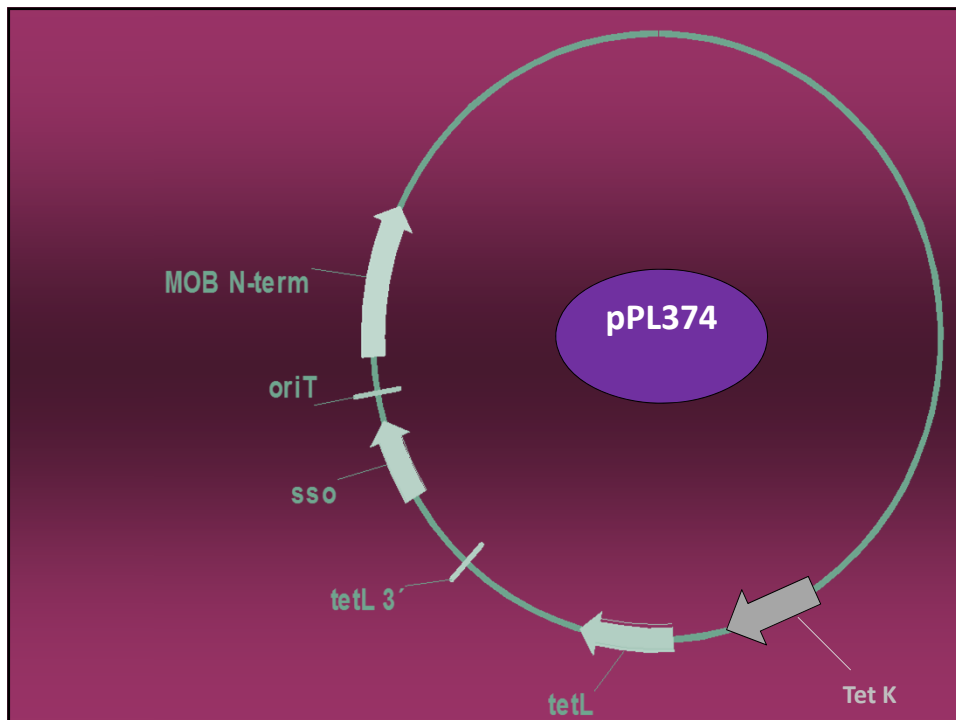


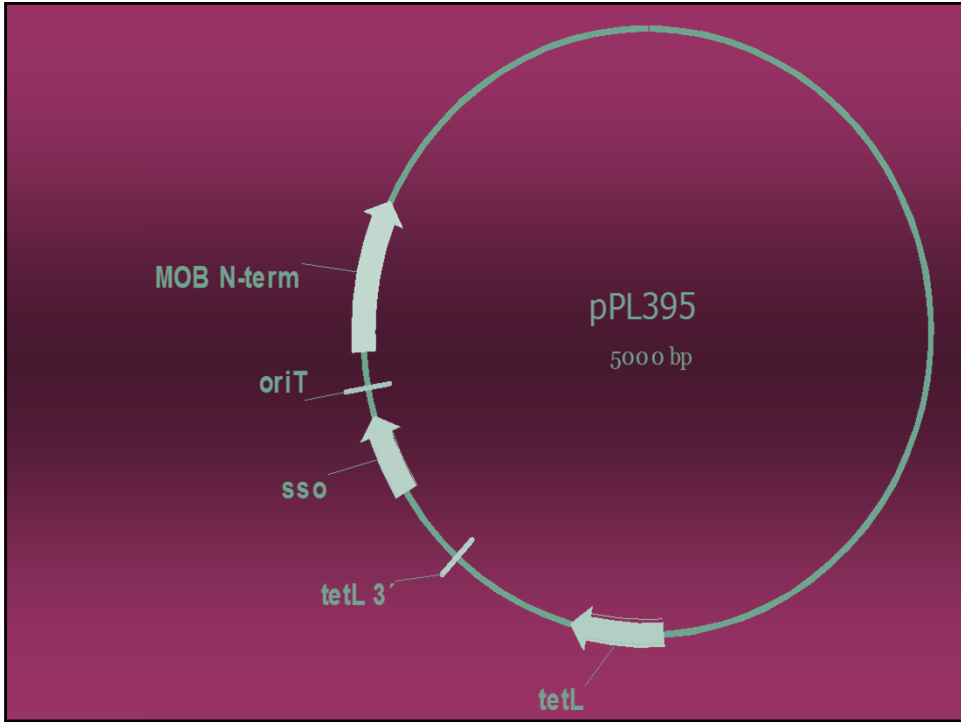
- Plasmidic DNA visualization



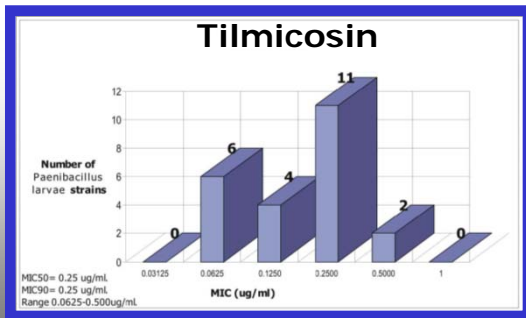
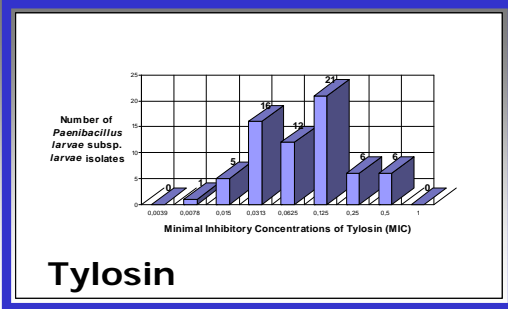
RESULTS

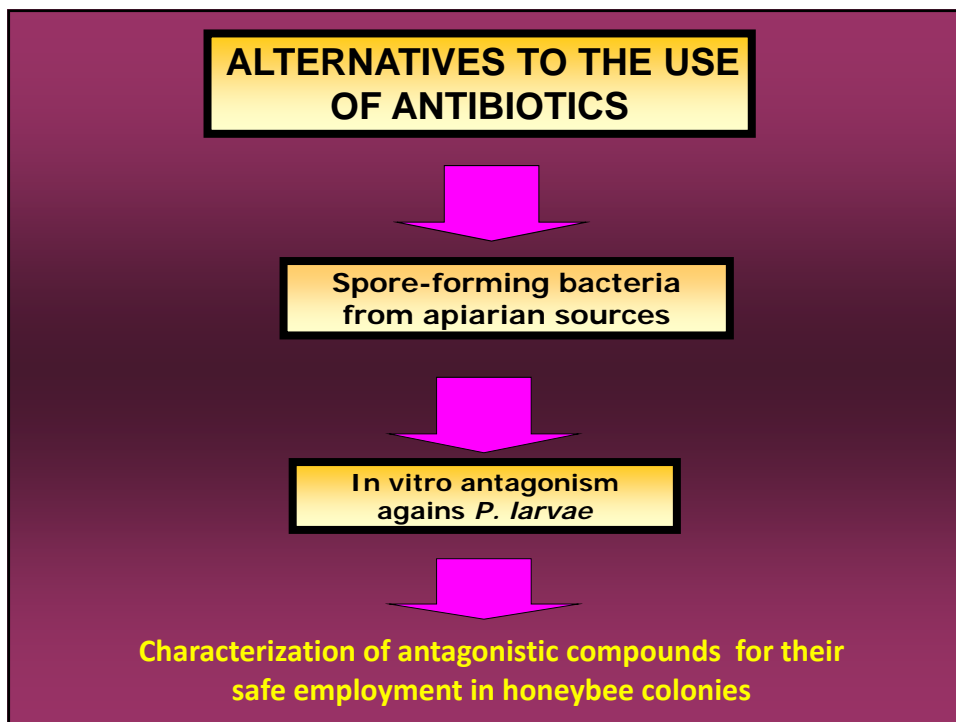
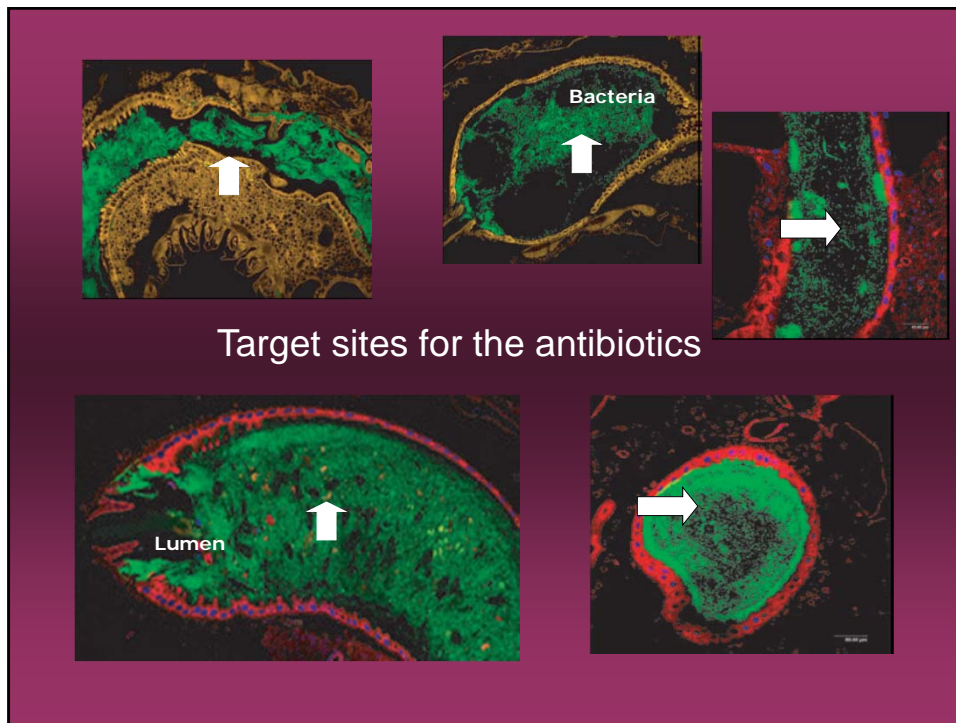
- First report of TC resistance horizontal transfer in *Paenibacillus larvae* by conjugation -**plasmid pPL374**- and also electroporation -**plasmid pPL395**-
- First report of *Tet (K)* determinant within *Paenibacillus* genus
- First report of *Tet (K)* and *Tet (L)* determinants on *P. larvae* plasmids
- Characterization of plasmid pPL374 : *Tet (K)* and *Tet (L)* resistance determinants – Mob genes – oriT region –
- Characterization of plasmid pPL395: *Tet (L)* – Mob – oriT region
- Complete sequence of plasmid pPL374



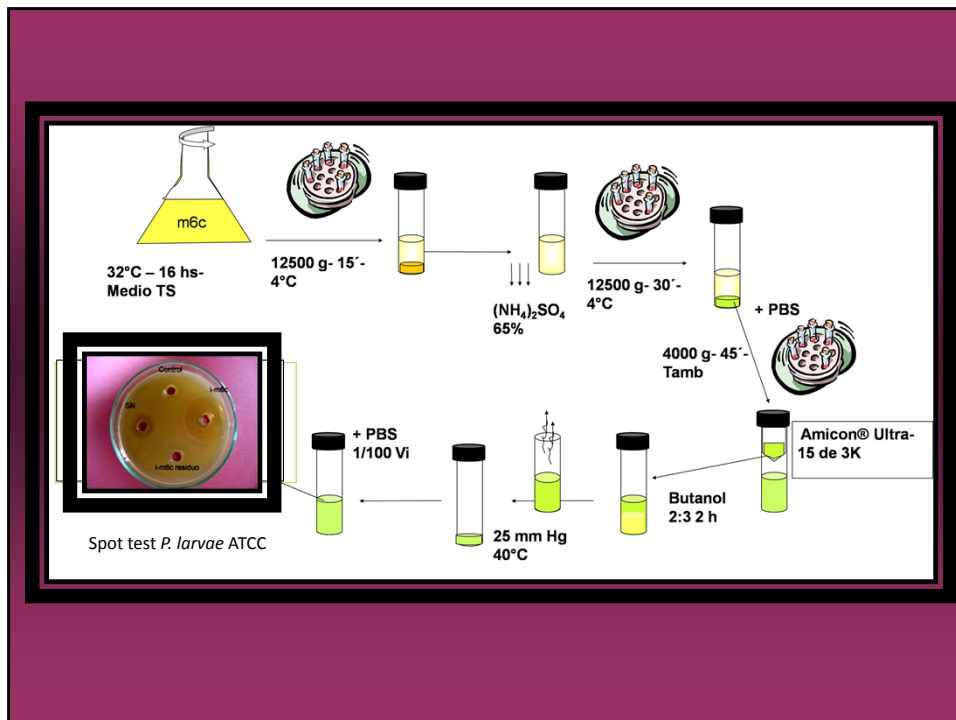


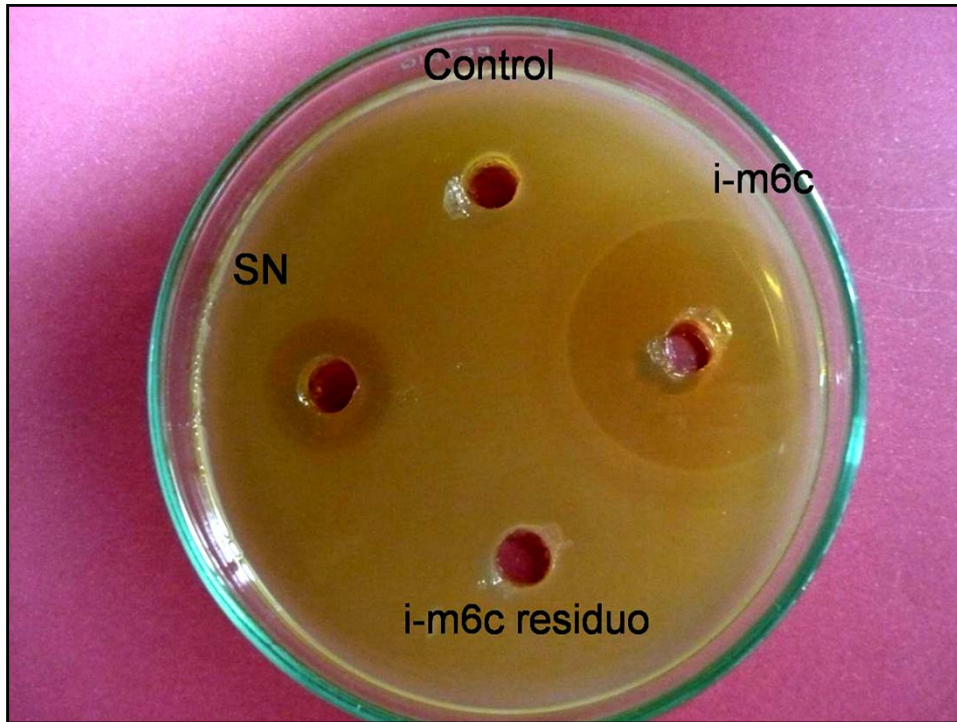
Antibiotics for AFB control





Bacterial antagonist





New strategies for AFB control

- **Biological control agents**
 - enzymes
 - antibiotics
 - antibiotic-like compounds
 - bacteriocins
 - lipopeptides
- **Stimulation of the bee's immune system**
- **Enhancement of the defense response of honey bees**



INTEGRATED PEST MANAGEMENT APPROACH (IPM)



Many thanks for your attention !!!