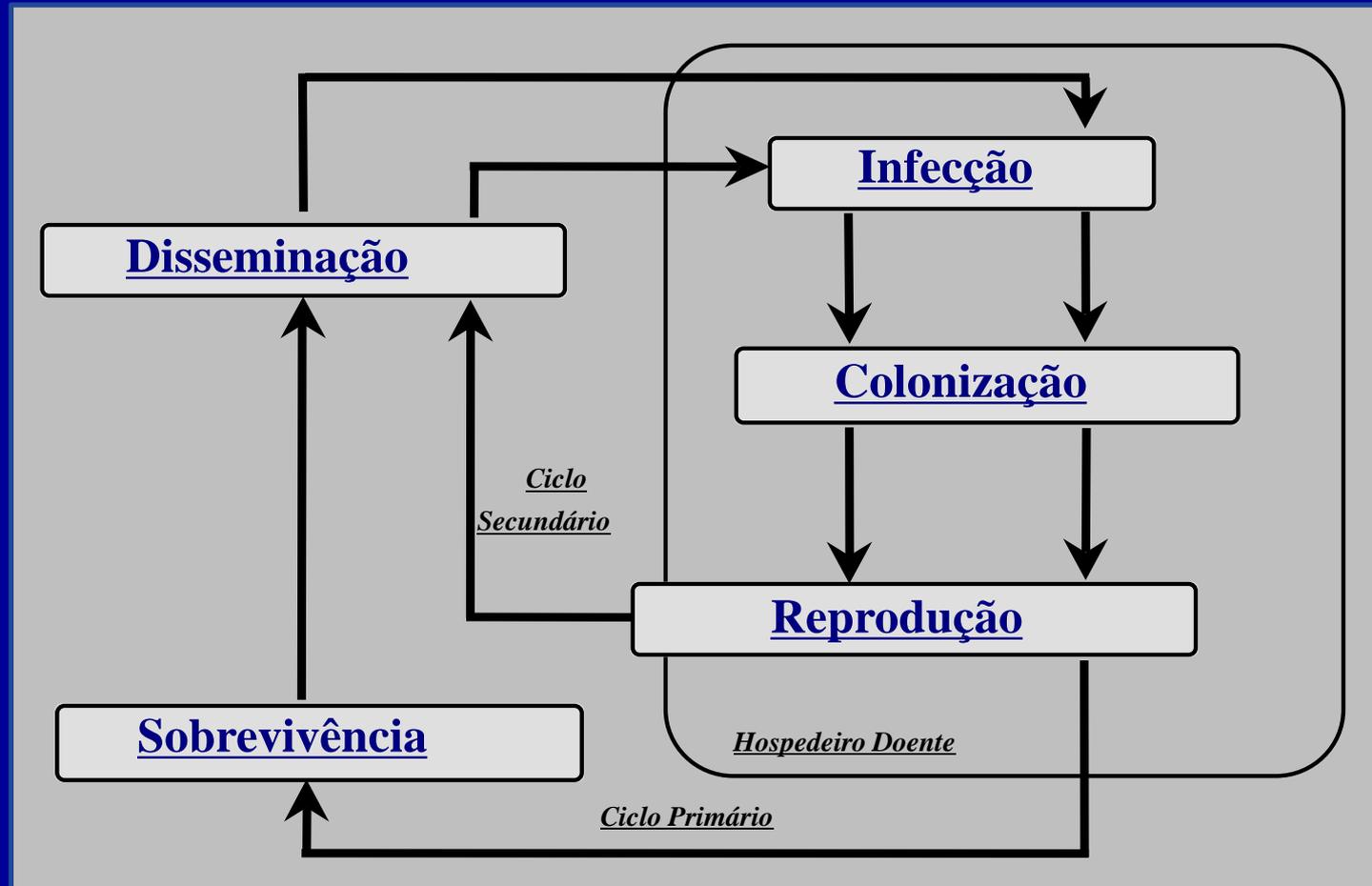
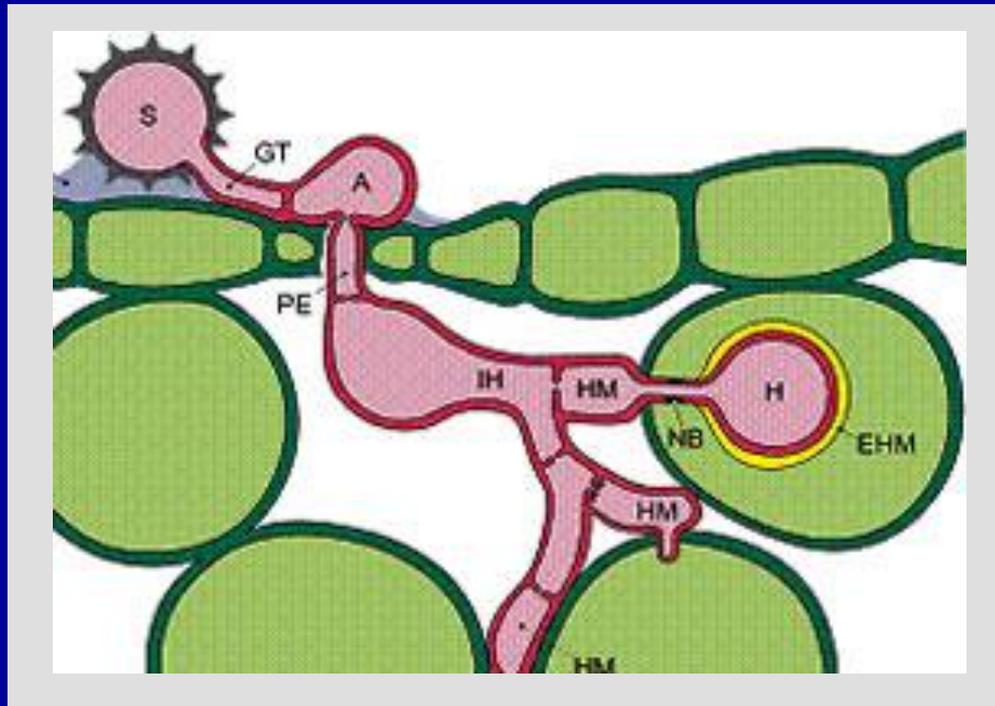


Ciclo das relações patógeno-hospedeiro



Infecção

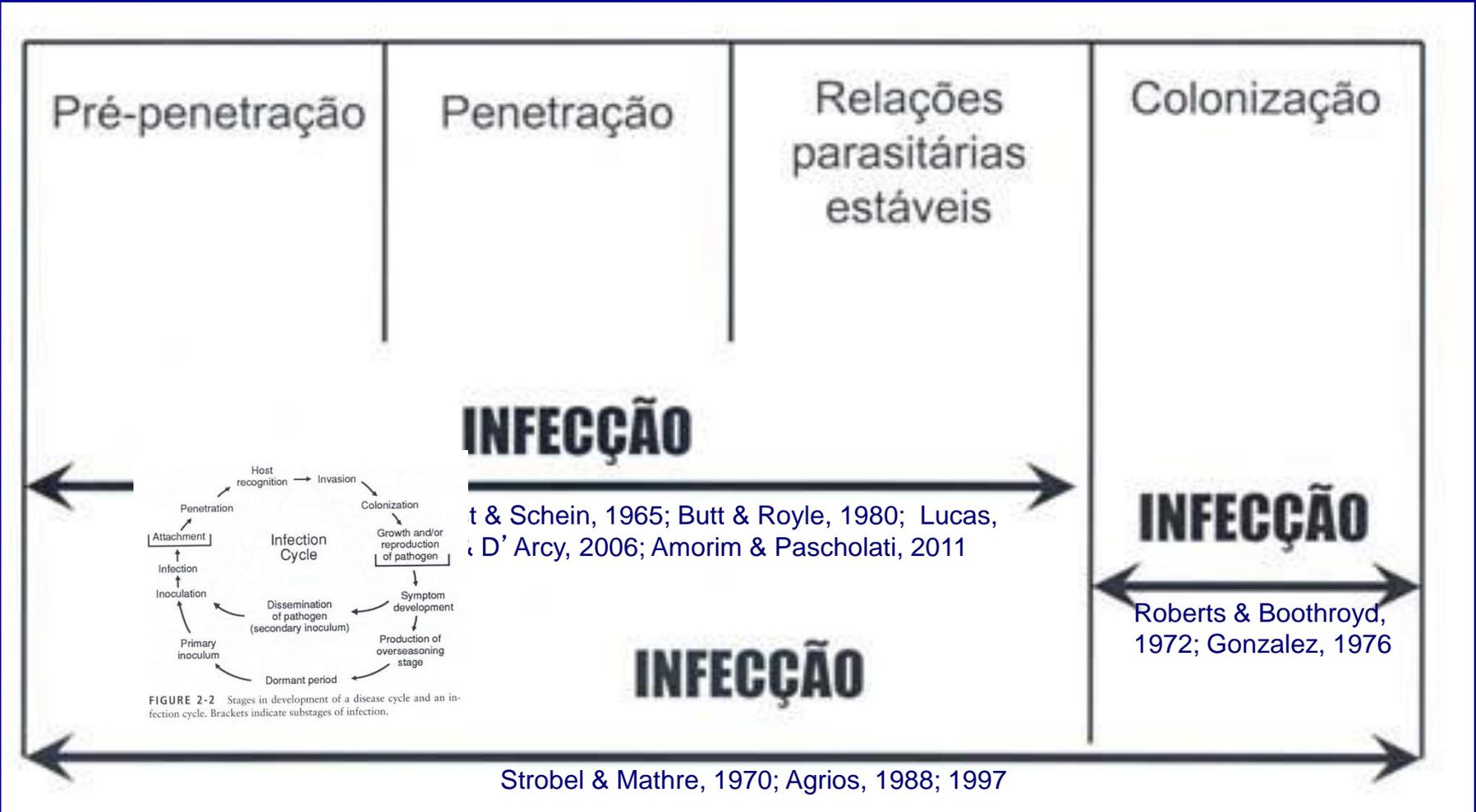


Legenda

S=esporo
GT=tubo germinativo
A=apressório
PE=peg de penetração
IH=hifa de infecção
HM=célula mãe do haustório
H=haustório

- **pré-penetração** adesão / movimento / germinação / crescimento
- **penetração** aberturas / ferimentos / direta
- **relações parasitárias estáveis**

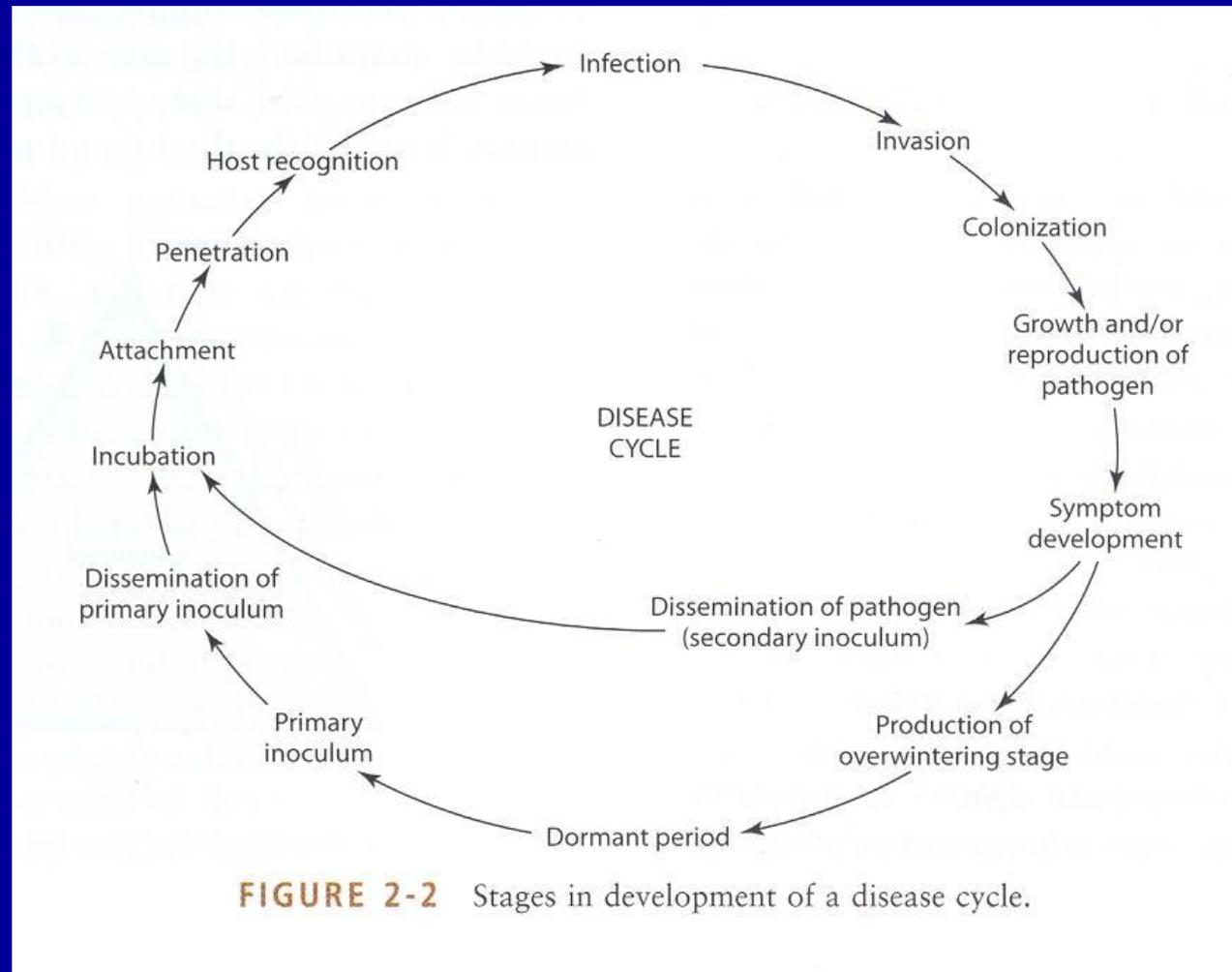
Infecção (Agrios, 2005; Trigiano et al., 2004)



“Infection is the process by which pathogens establish contact with susceptible cells of the host and procure nutrients from them. During infection pathogens grow, multiple, colonize and reproduces...” (Agrios, 1997).

Infecção

“Infection is the process by which pathogens establish contact with susceptible cells of the host and procure nutrients from them. Following infection pathogens grow...” (Agrios, 2005).

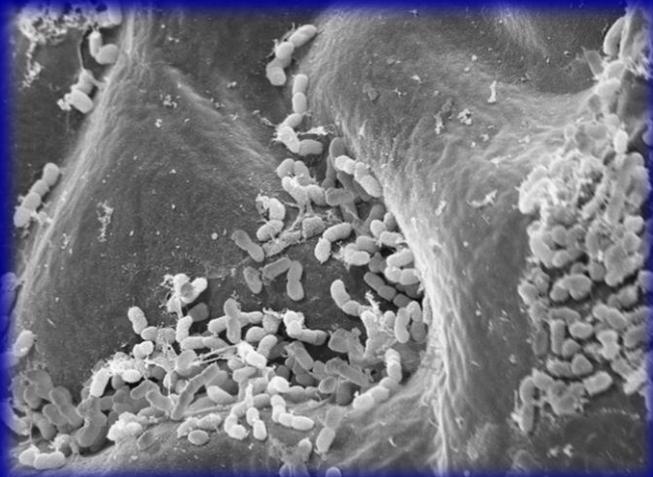
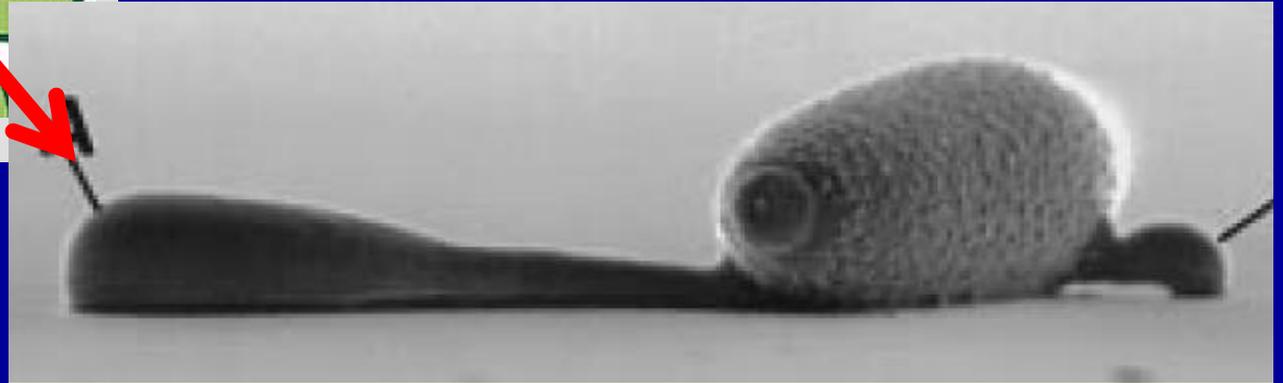
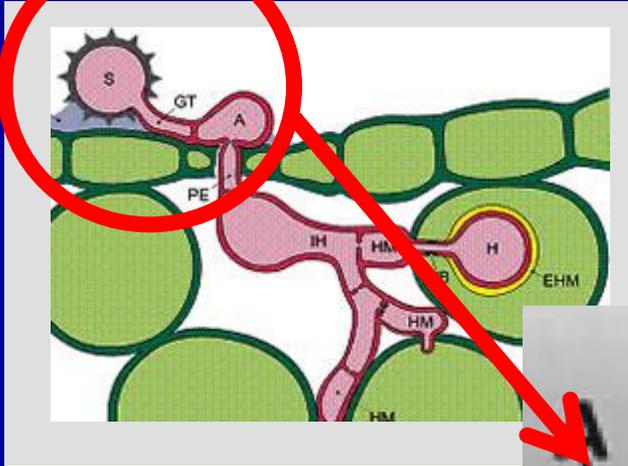


The steps of a disease cycle are inoculation, penetration, infection, invasion, reproduction and dissemination (Trigiano, R.N.; Windham, M.T.; Windham, A.S., 2004)

Infecção

Pré-penetração

adesão/ movimento
germinação/crescimento



Pré-penetração – adesão à superfície

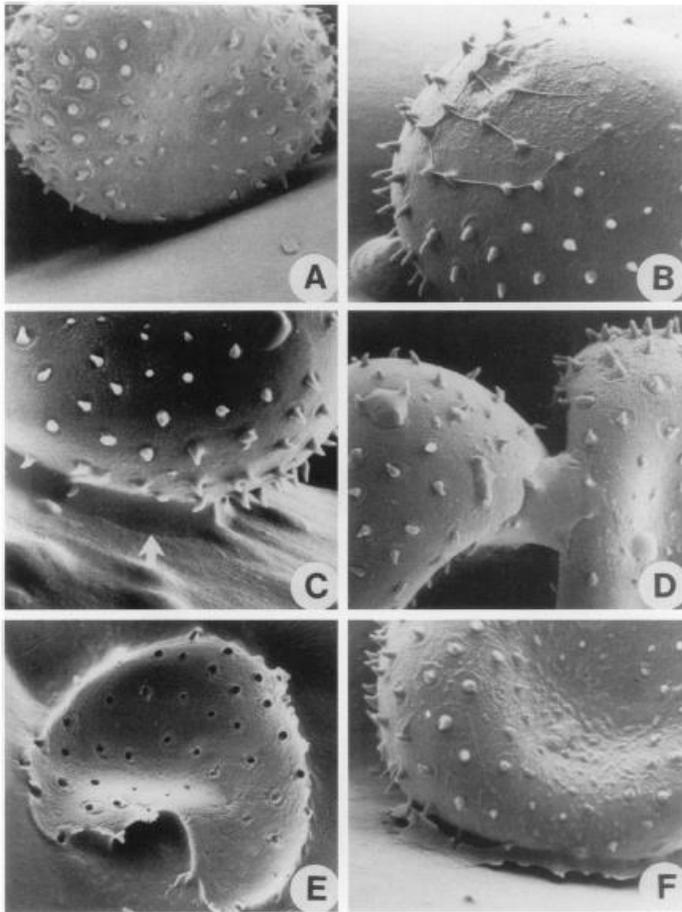


Figure 1. LTSEM Micrographs of Uredospores of *U. viciae-fabae* Showing Adhesion Pad Formation.

- (A) Dry uredospore, immediately after contact with the surface of a broad bean leaf ($\times 2900$).
 (B) Uredospore on the leaf surface misted with water and incubated at high humidity for 1 hr. Note the presence of a material that stretches between echinulations on the spore surface ($\times 3000$).
 (C) Uredospore misted with water, incubated at high humidity, and subsequently dried at ambient room conditions for 20 min. Note that the material that accumulated between the spore and the leaf surface (arrow) is still visible ($\times 4280$).
 (D) Spores treated as in (C). Note the presence of a material that appears to connect two adjacent spores ($\times 3300$).
 (E) Sample treated as indicated in (C) except that a spore was removed from the leaf surface with an adhesive tape revealing that an adhesion pad remains attached to the leaf surface ($\times 4000$).
 (F) Autoclaved uredospore treated as in (C) also exhibits the presence of an adhesion pad. Note the presence of a material between the spore and the underlying leaf surface ($\times 2680$).

Adesão de *Uromyces viciae-fabae* em superfícies diversas: formação passiva de substância adesiva após hidratação (esquerda) e presença de esterases apenas em esporos viáveis (direita, Deising et al., 1992)

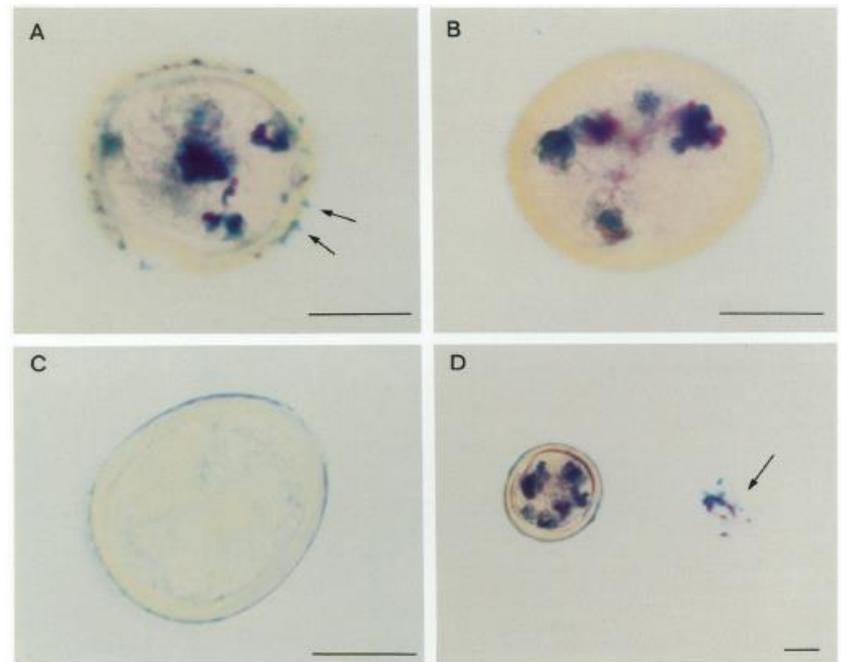


Figure 3. Histochemical Demonstration of Surface-Localized Esterase Activity.

- (A) Unwashed, native uredospore. Arrows indicate crystals of indigo blue that are present on the uredospore surface.
 (B) Esterase activity of uredospore washed to remove surface-bound esterases. Note the absence of crystals of indigo blue on the spore surface.
 (C) Autoclaved uredospore completely lacking indigo blue crystals.
 (D) Uredospore removed from the gelatin surface to expose underlying crystals of indigo blue (arrow).
 Surface-localized esterase activity was shown using indoxyl acetate as substrate as described in Methods and photographed by bright-field microscopy. Note that in both (A) and (B) crystals are present in fat bodies within the spores. Bars = 10 μm .

Pré-penetração – adesão à superfície

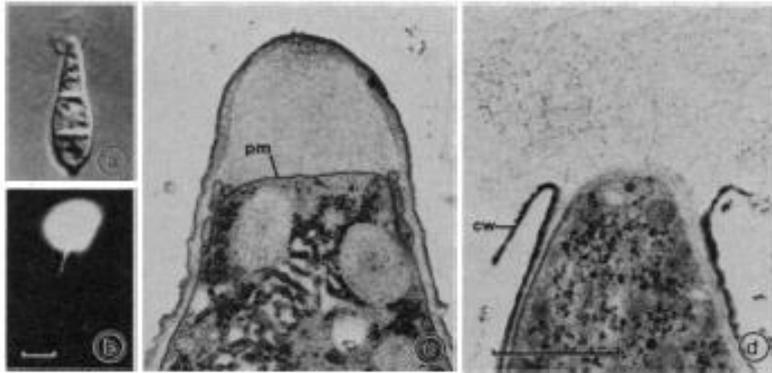
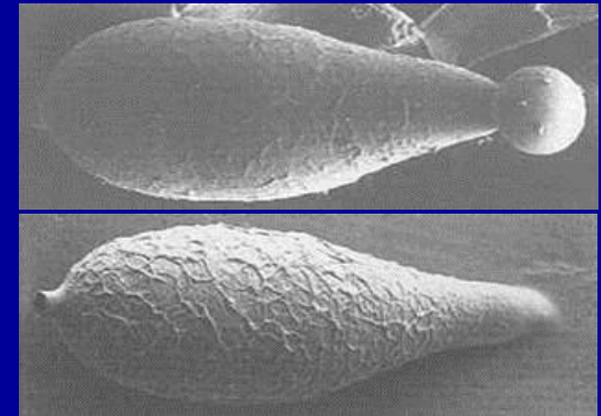


Fig. 2. Light (a and b) and electron (c and d) micrographs of *M. grisea* conidia before (c) and after (a, b, and d) hydration. (a and b) The same FITC-Con A-labeled conidium imaged with differential interference contrast optics and epi-fluorescence, respectively; scale bar, 5 μm . (c) A dry conidium apex contains a deposit outside the plasma membrane (pm). (d) When hydrated, the cell wall (cw) is broken at the apex and an extracellular fibrous matrix is released; scale bar, 1 μm . For light microscopy conidia were harvested in an aqueous suspension of FITC-Con A (1 $\mu\text{g}/\text{ml}$; Sigma) and viewed immediately. Electron microscope specimens were prepared by freeze substitution (13).



Adesão passiva de
Magnaporthe grisea
Substância lipo-protéica

“Apical attachment of the pyriform conidia produced by *M. grisea* allows them to resist the flow of water, and as water is essential for later stages of development in this fungus, this may be a useful adaptive mechanism to maintain water droplets around the attached conidium. Although similar mechanisms of spore attachment are likely to be widespread among pathogenic fungi, conidium attachment by the barley powdery mildew pathogen *Blumeria graminis* f. sp. *hordei* and many other powdery mildews differs markedly from the situation in *M. grisea*.” (Sara L. Tucker and Nicholas J. Talbot, 2001)

Pré-penetração – adesão à superfície

Morphogenesis of the powdery mildew fungus in water

149

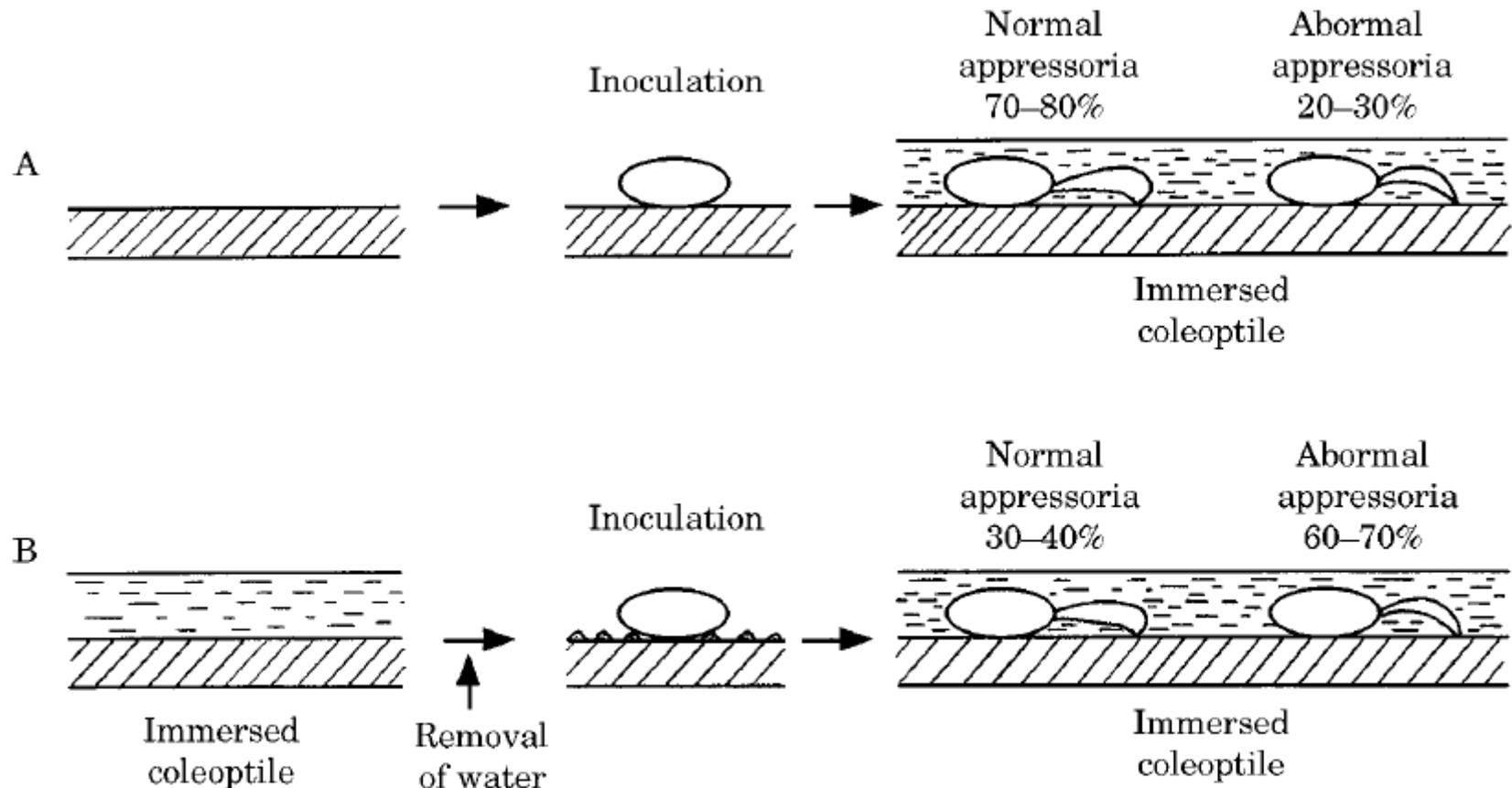


FIG. 2. Inoculation of *Erysiphe graminis* conidia onto the barley coleoptile surface without (A) or with (B) a water treatment for 20 min prior to inoculation and immersion in water. The percentage shows the frequency of normal or abnormal appressorium development in water.

Pré-penetração – adesão à superfície

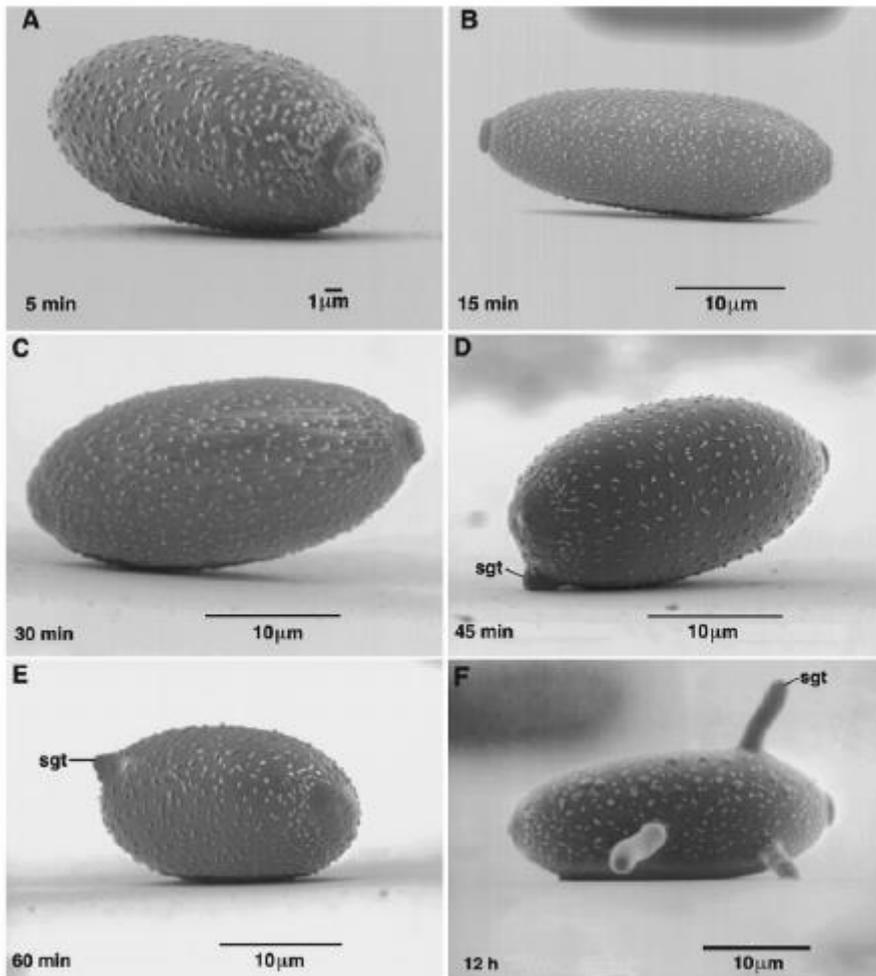


Fig. 2. Low angle, low temperature SEM images of *Blumeria graminis* conidia and germ tubes incubated on hydrophilic glass for various times after inoculation as indicated in the figure. sgt = subsidiary germ tube. Note in D we describe the emerging germ tube as an sgt because no ECM was evident beneath the germ tube.

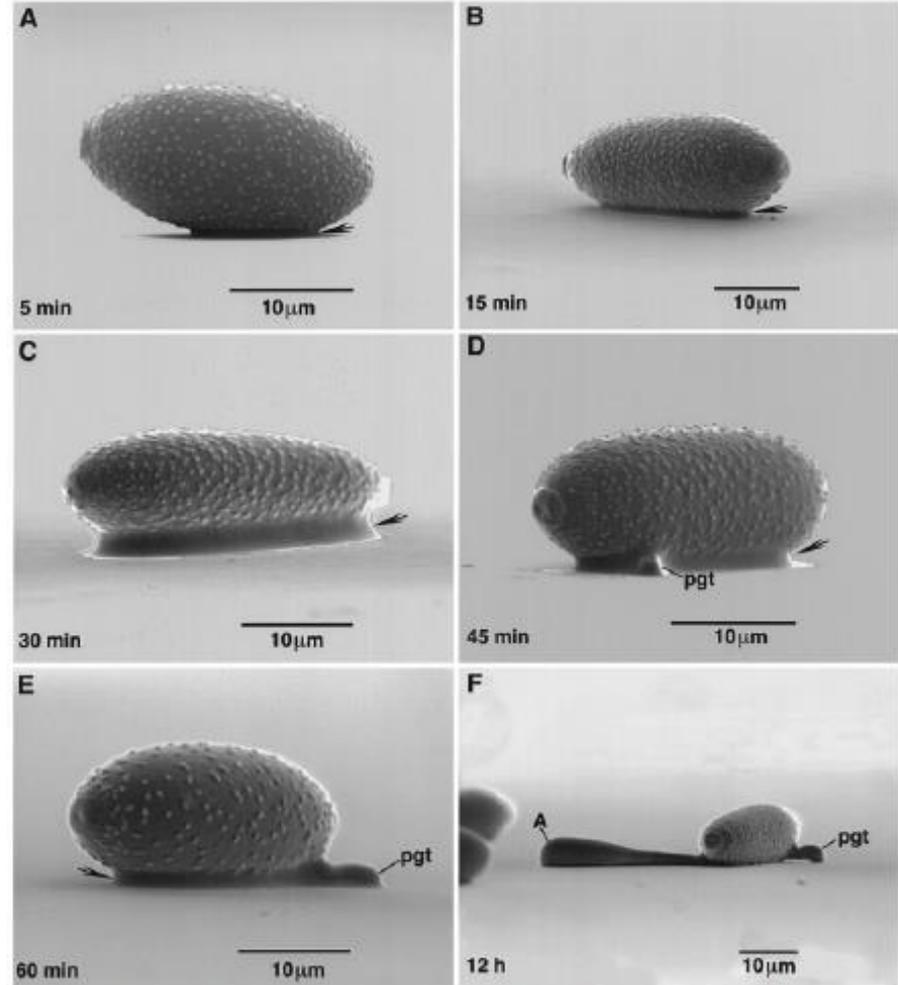


Fig. 4. Low angle, low temperature SEM images of *Blumeria graminis* conidia incubated on hydrophobic plastic for various times after inoculation as indicated in the figure. Arrows point to an accumulation of ECM material at the contact interface of the conidium with the substratum. A = appressorium, pgt = primary germ tube.

Pré-penetração – adesão à superfície

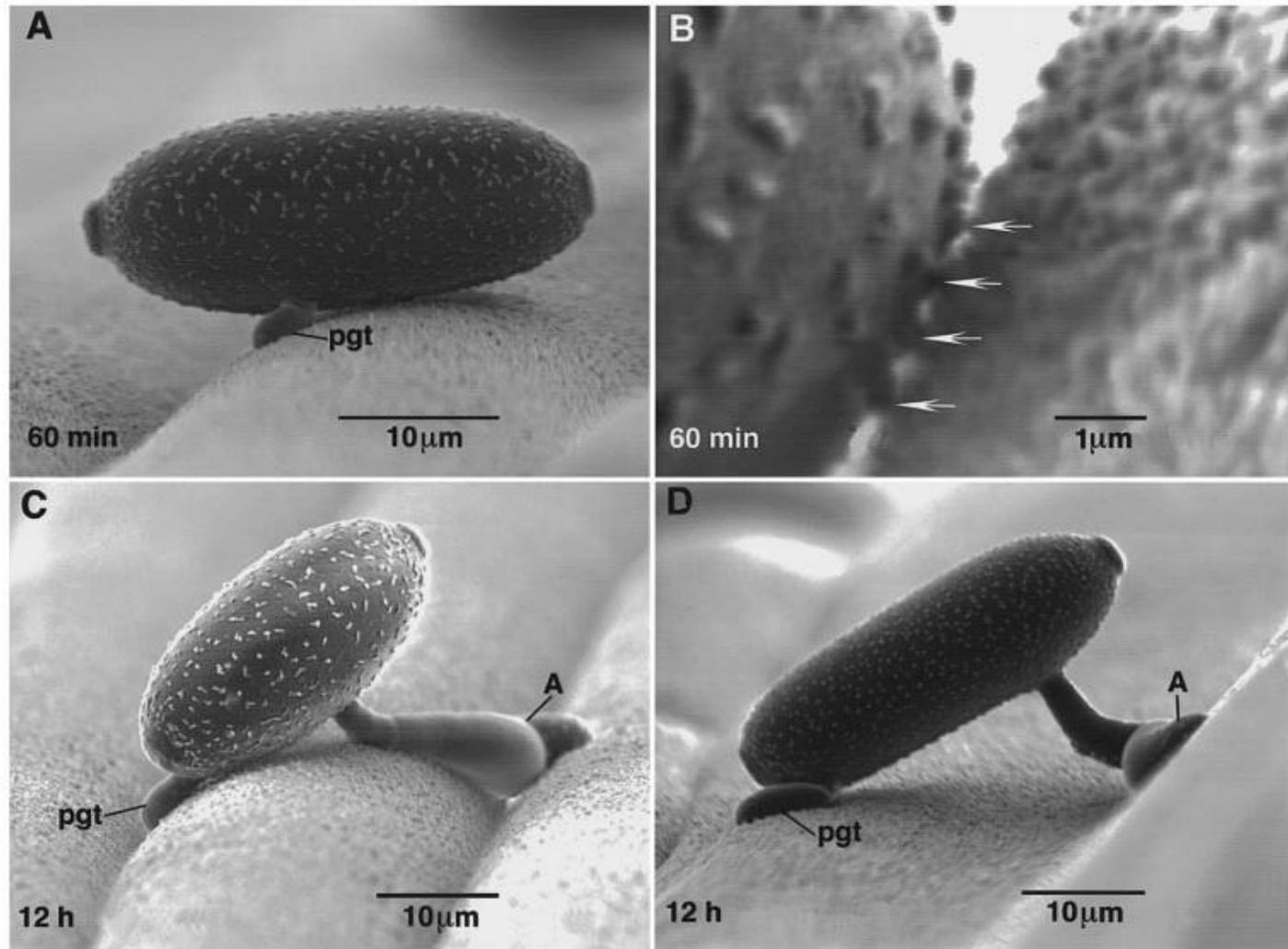


Fig. 9. Low angle, low temperature SEM images of *Blumeria graminis* conidia incubated on barley leaf surfaces with hydrophobic waxes in place. No ECM could be seen at the interface of conidia with the leaf surface. A and B, 60 min after inoculation. Arrows in B point to conidial surface projections that extend outward from the conidium (left of figure) to wax platelets on the surface of the leaf (right of figure). C and D, 12 h after inoculation. A = appressorium, pgt = primary germ tube.

Pré-penetração – auto-inibidores de germinação

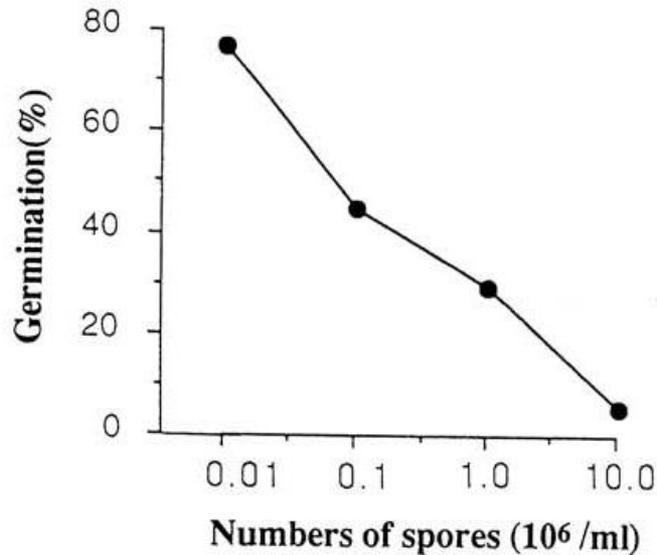


Fig. 1 Relation between germination percentage and spore density of *Colletotrichum gloeosporioides* f. sp. *jussiaea*.

Table 1. Effect of washing on germination of crowded conidia

Treatment	Germination (%)
	Conc.: 5×10^5 conidia/ml
Nonwashed	56
Washed 1 x	74
Washed 2 x	88

Table 4. Content of CG-SI 1 and 2 and detection of CG-SI 3 in cultures of various anthracnose fungi

Anthracnose fungus	CG-SI 1 ^a	CG-SI 2 ^a	CG-SI 3 ^b
<i>Colletotrichum gloeosporioides</i>	149 μ g	63 μ g	++
f. sp. <i>jussiaea</i>			
<i>C. gloeosporioides</i>	- ^c	-	-
f. sp. <i>aeschnomene</i>			
<i>C. gloeosporioides</i> isolate 7696	-	-	-
<i>C. malvarum</i>	-	-	-
<i>C. graminicola</i>	-	-	-
<i>C. dematium</i>	-	-	-
<i>C. fragariae</i>	38 μ g	5 μ g	+
<i>C. lagenarium</i>	-	-	-
<i>C. lindemuthianum</i> IFO 5260	-	-	-
<i>Glomerella cingulata</i> IFO 6445	-	-	-
<i>G. cingulata</i> IFO 7478	-	-	-
<i>G. fructigena</i> IFO 5951	-	-	-
<i>G. gossypii</i> IFO 6181	-	-	-
<i>G. glycines</i> IFO 7384	-	-	-
<i>G. tucumanensis</i> IFO 7395	-	-	-
<i>G. fusarioides</i> IFO 8831	-	-	-

^a Quantitative analyses were performed by HPLC-UV detection.

^b Spots on TLC-plates were detected with the van Urk-Salkowsky reagent.

^c “-” indicates below limit of detection (ca. 0.1 μ g for CG SI-1 and CG SI-2).

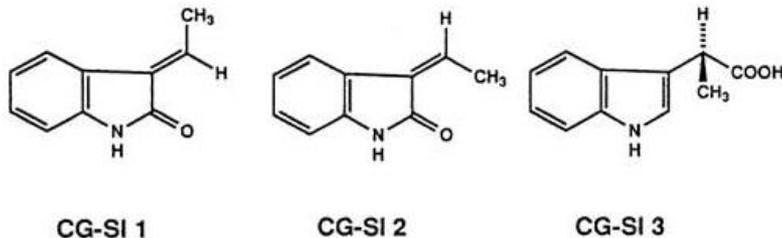


Fig. 3. Chemical structures of germination self-inhibitors from *Colletotrichum gloeosporioides* f. sp. *jussiaea*.

Pré-penetração – auto-inibidores de germinação

Table 2. Inhibitory activity of CG-SI 1 and 2 against conidial germination of various fungi

Fungus	Sample	Germination (%) ^a			
		Conc. (µg/ml)			
		10	3	1	0
<i>Colletotrichum gloeosporioides</i> f. sp. <i>jussiaea</i>	CG-SI 1	0	40	81	82
	CG-SI 2	0	68	80	82
<i>C. gloeosporioides</i> f. sp. <i>aeschnomene</i>	CG-SI 1	0	46	80	80
	CG-SI 2	0	62	81	80
<i>C. gloeosporioides</i> isolate 7696	CG-SI 1	0	47	83	81
	CG-SI 2	0	65	79	81
<i>C. malvarum</i>	CG-SI 1	5	60	84	81
	CG-SI 2	14	79	82	81
<i>C. graminicola</i>	CG-SI 1	96	94	96	95
	CG-SI 2	96	96	96	95
<i>C. dematium</i>	CG-SI 1	88	90	90	88
	CG-SI 2	87	84	85	88
<i>C. fragariae</i>	CG-SI 1	0	48	80	82
	CG-SI 2	0	60	82	82
<i>C. lagenarium</i>	CG-SI 1	13	64	95	95
	CG-SI 2	92	97	94	95
<i>Cochliobolus miyabeanus</i>	CG-SI 1	32	80	92	93
	CG-SI 2	85	91	90	93
<i>Cochliobolus sativus</i>	CG-SI 1	32	76	89	88
	CG-SI 2	80	89	89	88
<i>Fusarium oxysporum</i> f. sp. <i>cucumerinum</i>	CG-SI 1	3	65	91	94
	CG-SI 2	12	91	93	94

^a Germination percentages were measured by the microscope slide assay.

TABLE 1. Effect of washing the conidia of *Monilinia fructicola* on germination and appressorium formation on a polystyrene surface^x

Treatments	Conidial germination (%)	Appressoria formed by germlings (%)
Washed	92 a	93 a
Nontreated	92 a	5 b
Washing control ^y	93 a	4 b
Original supernatant ^z	94 a	3 b

^x Data are from a single internally replicated experiment and are representative of three experiments with similar results. Each value is the mean of three separate determinations. Values followed by the same letter within a column are not significantly different ($\alpha = 0.01$) according to one-way analysis of variance using Tukey's test.

^y Conidial suspensions that had been centrifuged repeatedly as washed conidia but no supernatants were removed or added.

^z The original supernatant removed after the first centrifugation was added back to the washed conidia, as described in Materials and Methods.

(Lee & Bostok, 2006)

Pré-penetração

Movimentação direcionada →

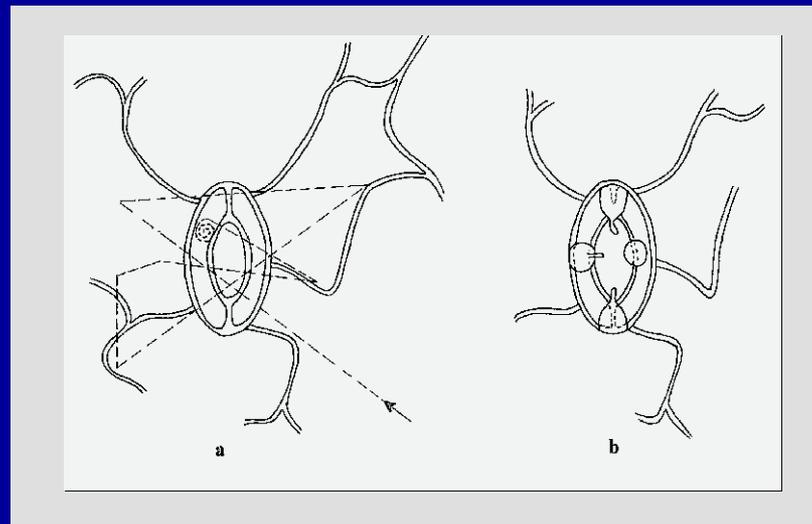
Tatismo

Respostas positivas ou negativas de um organismo (ou parte dele) a um estímulo

Crescimento direcionado →

Tropismo

fungos
bactérias
nematóides



tatismo de
zoósporos de
Plasmopara viticola

Tropismo & Tatismo

Estímulo	Resposta
substâncias químicas →	quimiotropismo quimiotatismo
cargas elétricas →	eletrotropismo eletrotatismo
topografia do hospedeiro →	tigmotropismo
água →	hidrotropismo hidrotatismo
luz →	fototropismo fototatismo

Pré- penetração

Quimiotatismo em nematóides

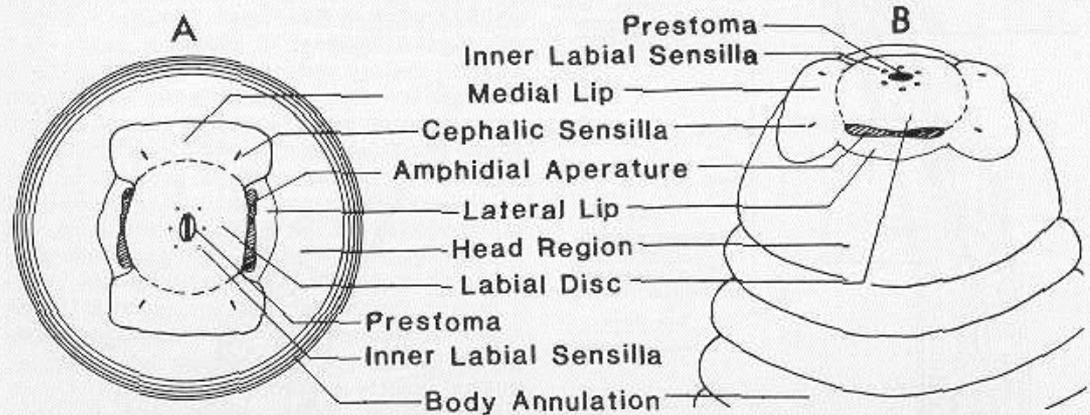


Fig. 4. External head morphology of a second-stage juvenile root-knot nematode in A) face and B) lateral views, as revealed by scanning electron microscopy [After Eisenback & Hirschmann (27)].

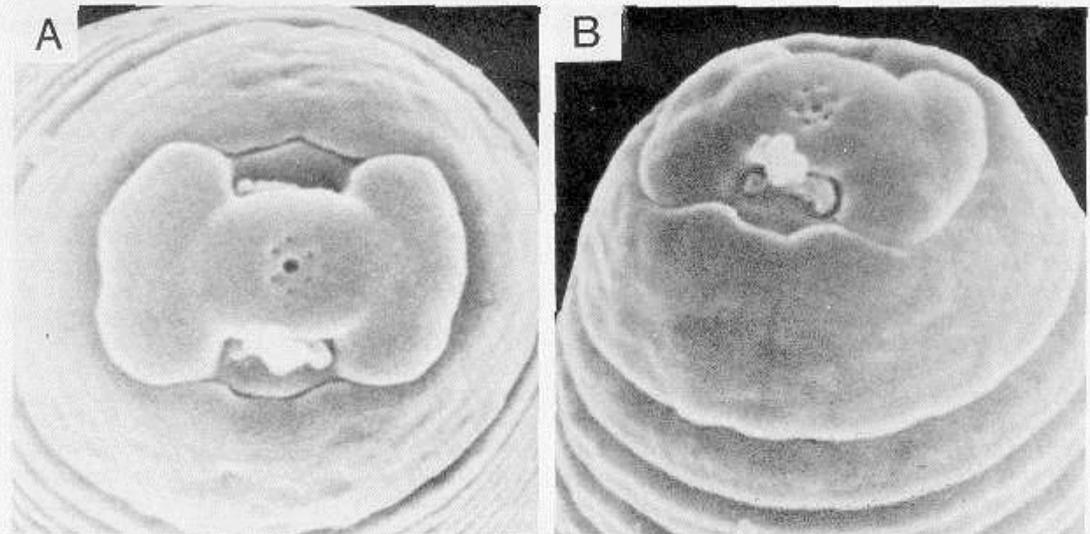


Fig. 5. Scanning electron micrographs of the head of a second-stage juvenile of *Meloidogyne hapta* in A) face and B) lateral views. [After Eisenback & Hirschmann (27)].

Pré- penetração

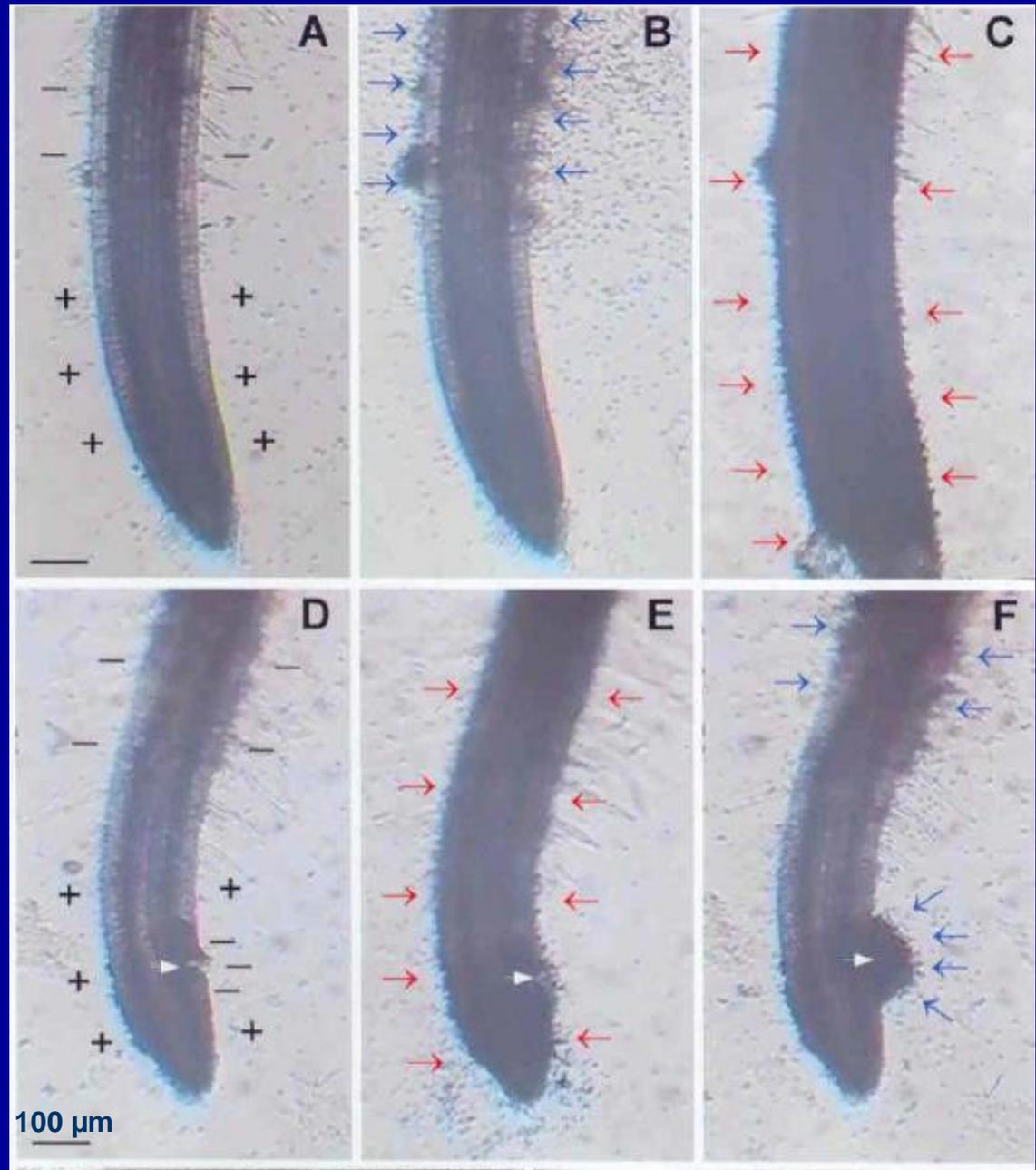
Eletrotatismo em *Pythium* *aphanidermatum*

Raiz intacta

A - *Pythium* 0 min

B - *Pythium* 10 min

C - *Phytophthora* 10 min



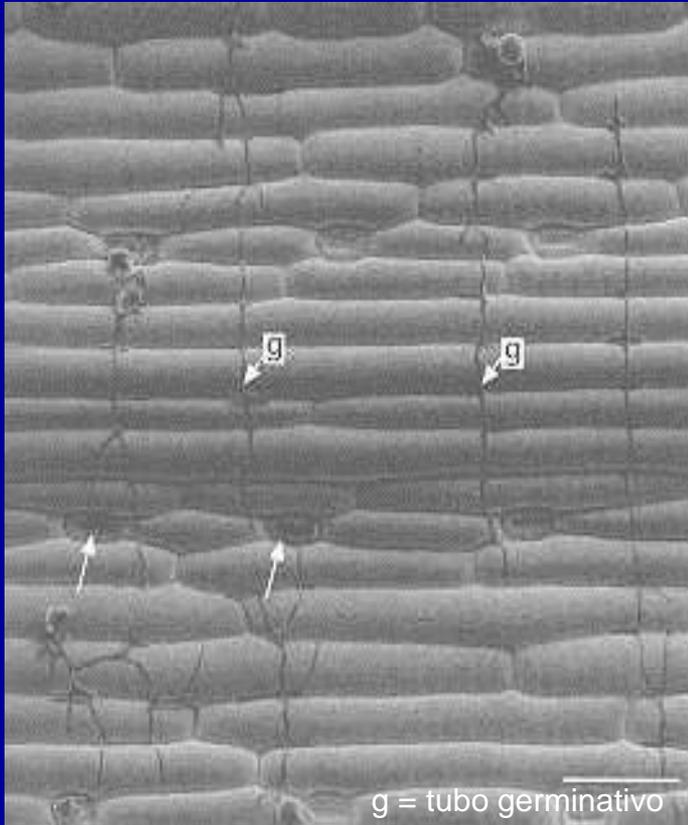
Raiz ferida (seta branca)

D - *Phytophthora* 0 min

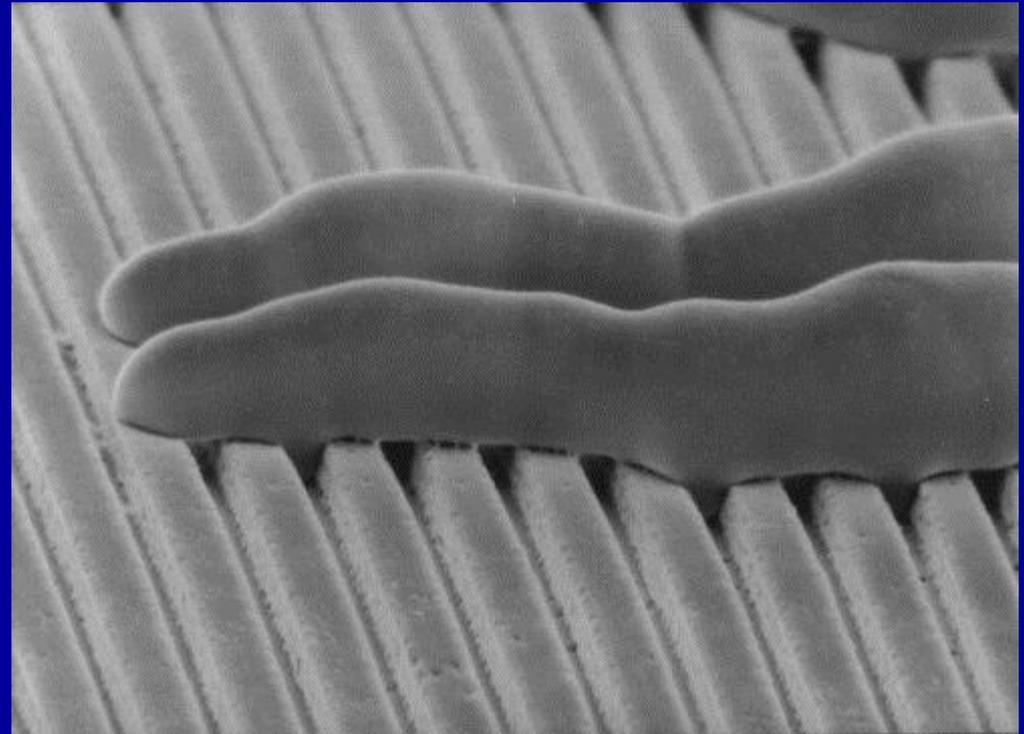
E - *Phytophthora* 10 min

F - *Pythium* 10 min

Pré-penetração - tigmotropismo



Tigmotropismo de hifas de *Puccinia hordei* em cevada

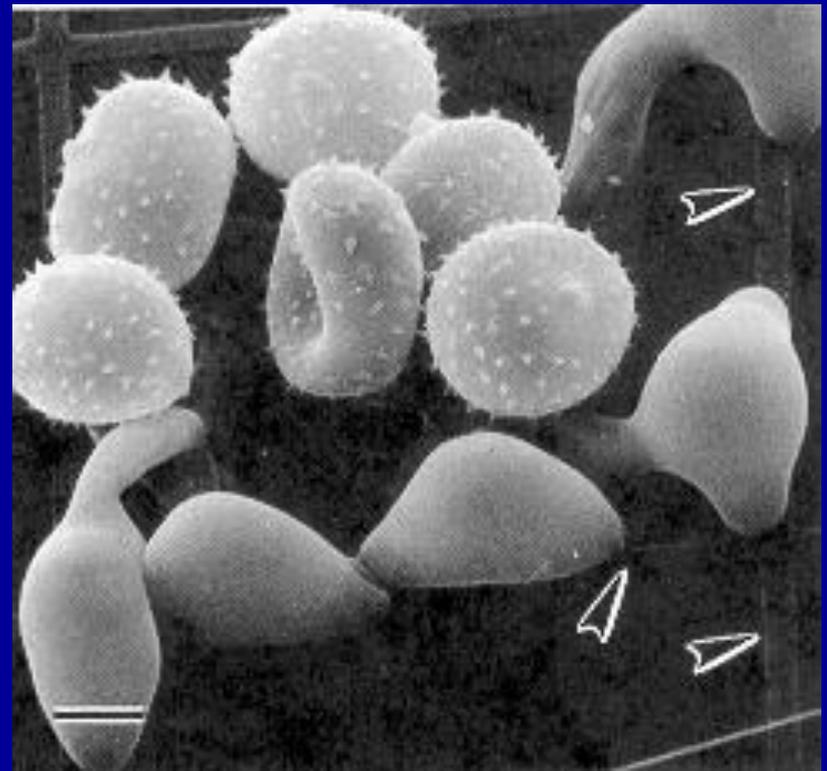
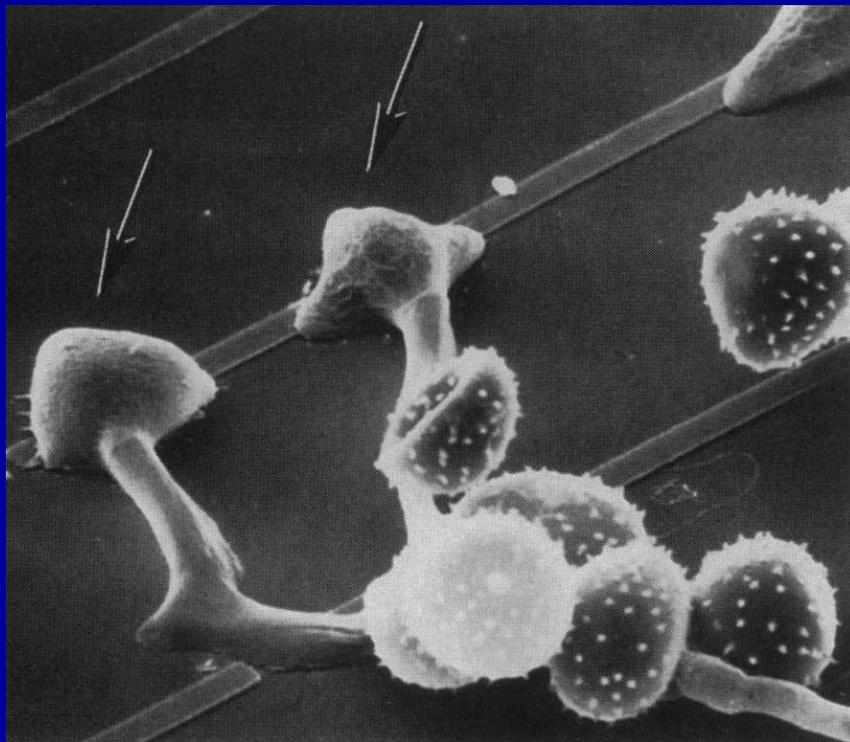


Fotografia em microscópio eletrônico de varredura de dois tubos germinativos de *Puccinia* sobre uma superfície artificial (poliestireno + silicone). As ranhuras e saliências induzem o crescimento direcional de tubos germinativos que também têm uma aparência "nose-down" mostrando que eles percebem e respondem à superfície sobre a qual crescem (Read *et al.*, 1992).

Pré-penetração

Uromyces appendiculatus

Formação de apressórios sobre ranhuras
(setas) de uma superfície artificial



Pré-penetração e penetração bacteriana

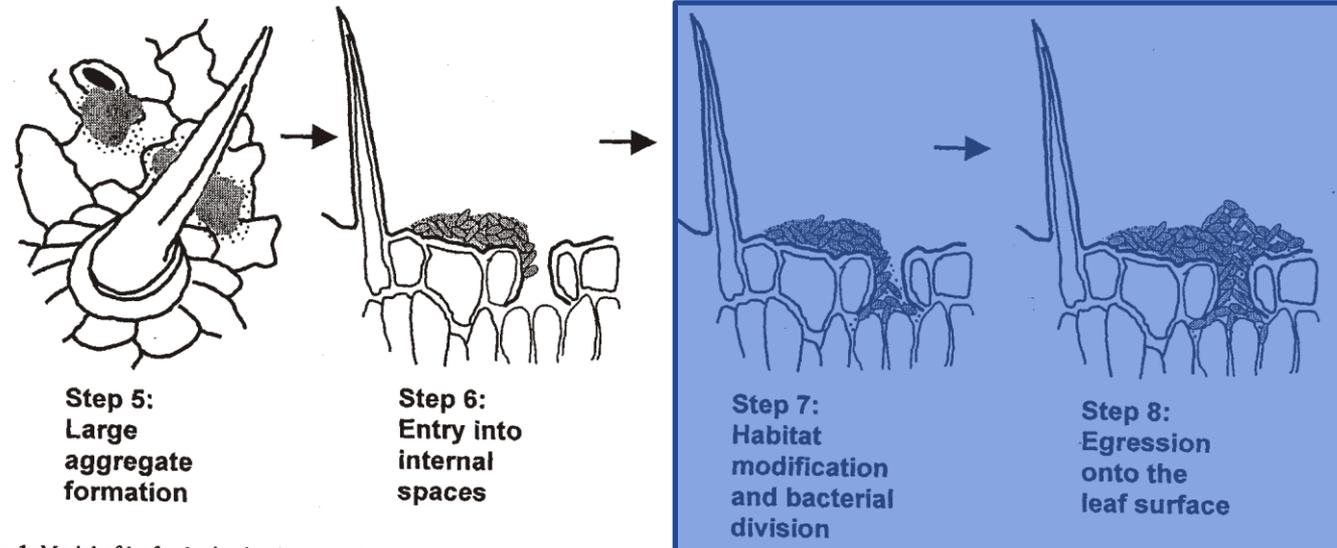
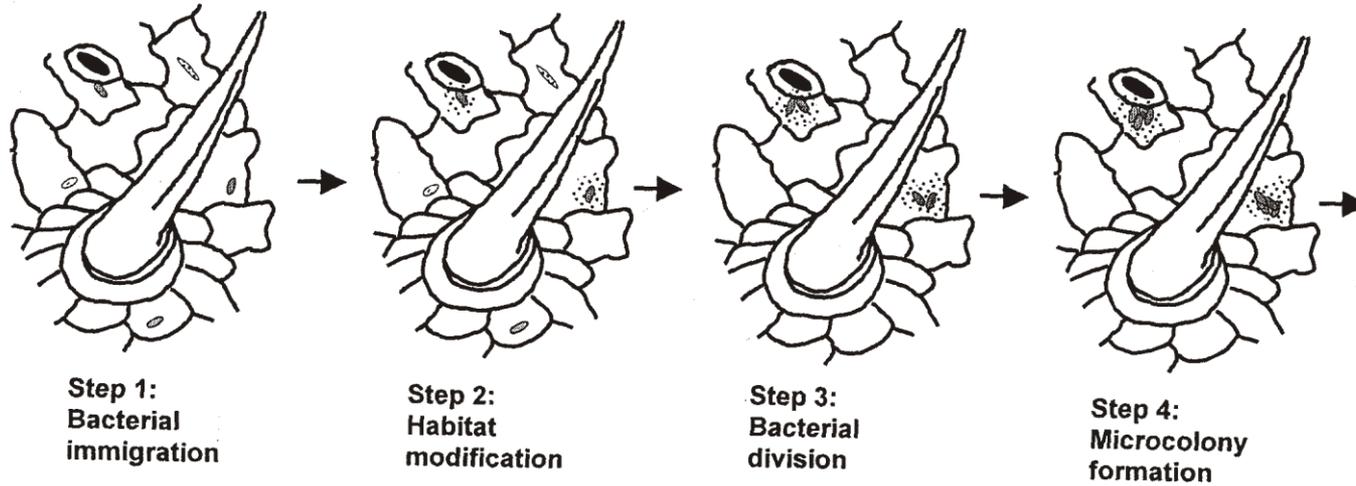


Fig. 1. Model of leaf colonization by phyllobacteria.

Pré-penetração e penetração bacteriana

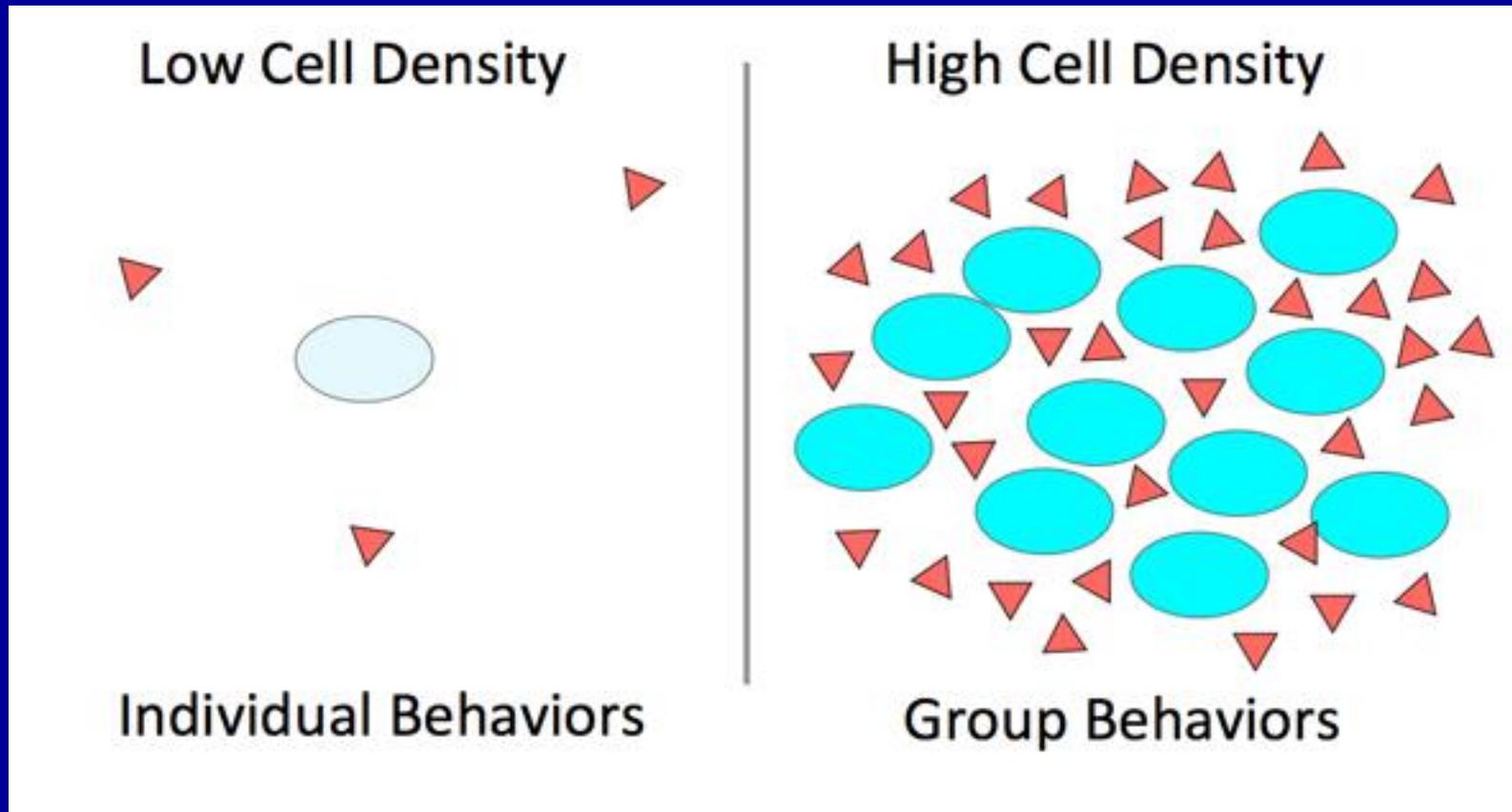
"QUORUM -SENSING"

Comunicação entre bactérias pela excreção de moléculas de baixo peso

Permite que a bactéria perceba a densidade populacional

Permite que as bactérias ajam em grupo ("seres sociais")

Sobrevivência, infecção e colonização

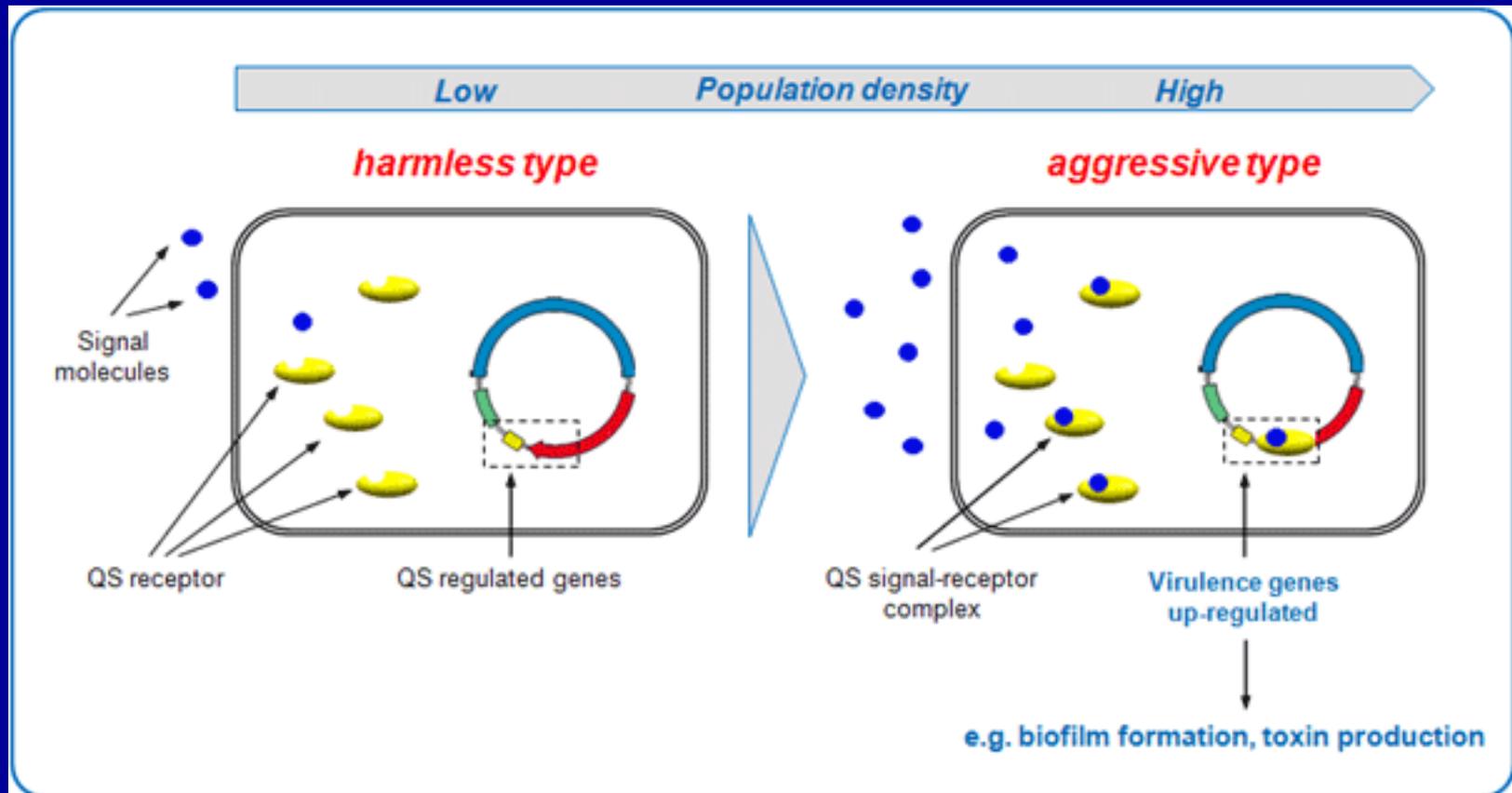


Pré-penetração e penetração bacteriana

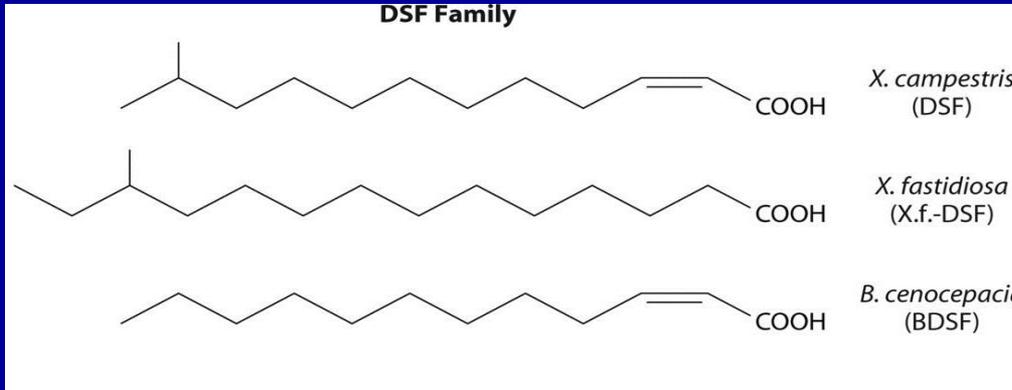
"QUORUM -SENSING"

Comunicação entre bactérias para regular coordenadamente a expressão de genes

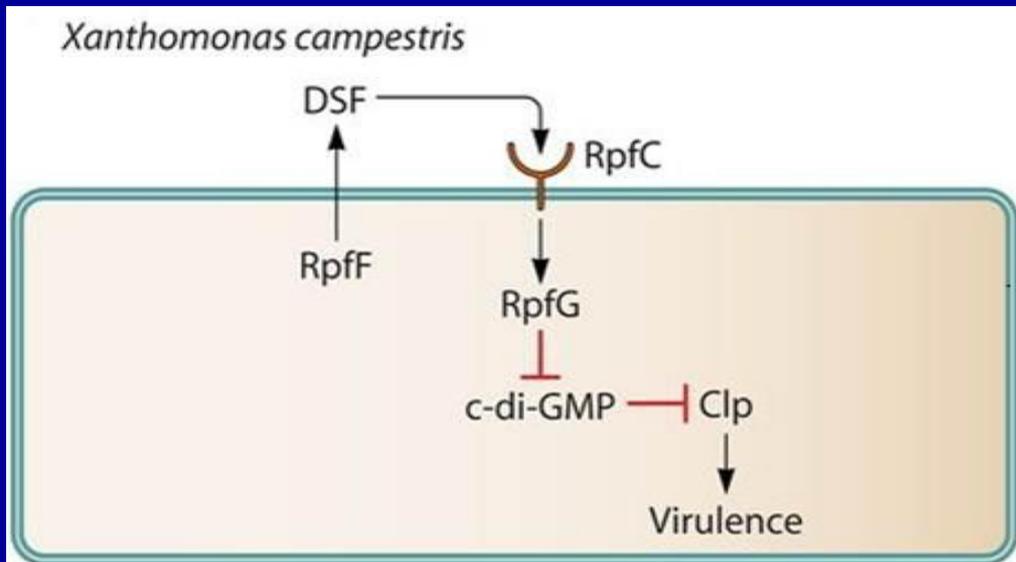
Presente em *Agrobacterium*, *Erwinia*, *Xanthomonas*, *Pseudomonas*, *Ralstonia*...



Moléculas sinalizadoras e receptoras de quorum sensing

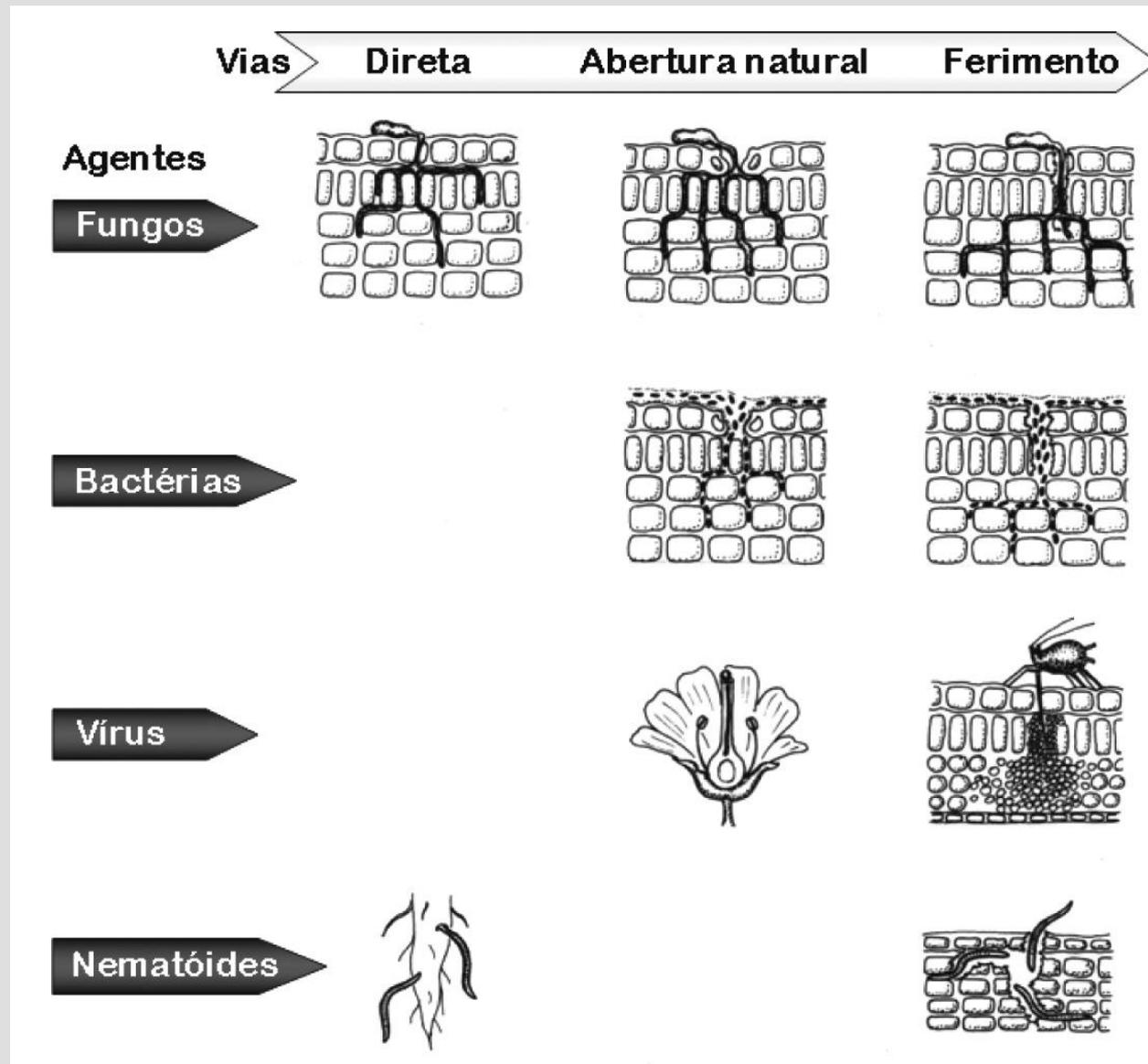


Efeito de inibidores de DSF (Difusible signal factor) em *Erwinia*: acima inoculação com presença de inibidores, abaixo inoculação sem a presença de inibidores



Inibição de quorum sensing é conhecida por “quorum quenching”

Penetração



Penetração direta

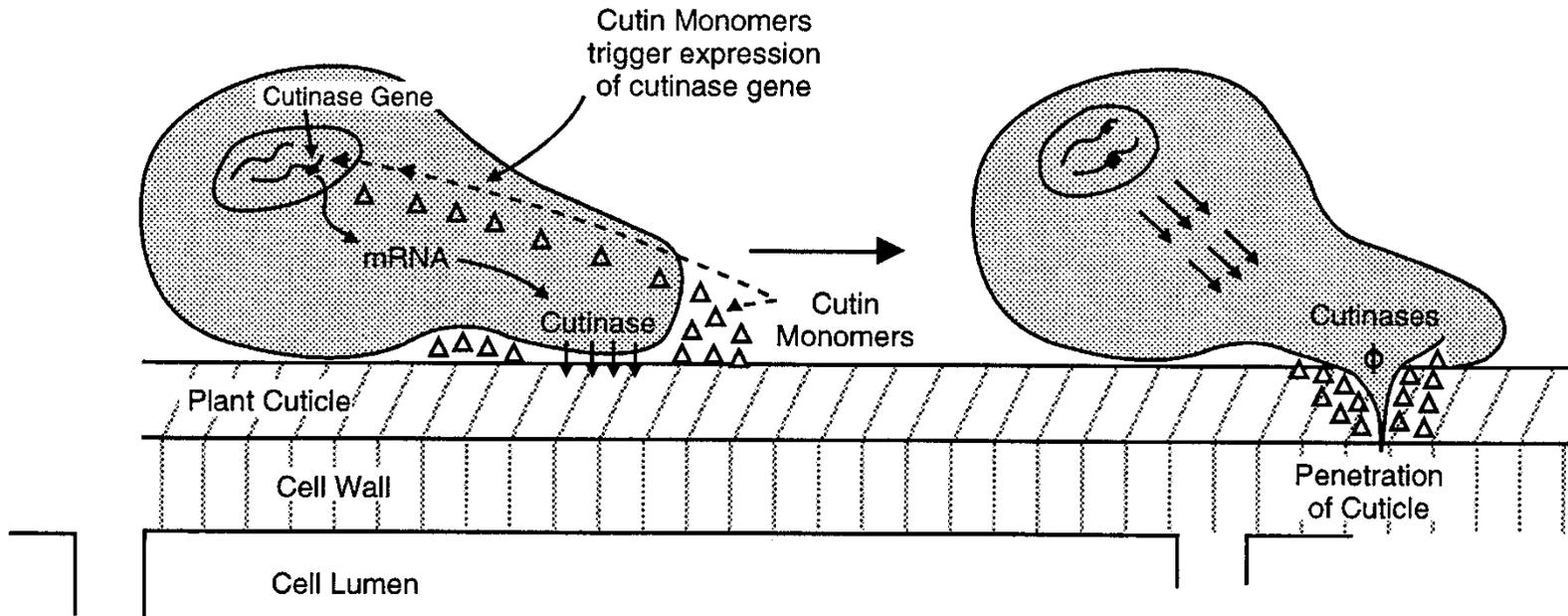


FIGURE 3-2 Diagrammatic representation of cuticle penetration by a germinating fungus spore. Constitutive cutinase releases a few cutin monomers from the plant cuticle. These trigger expression of the cutinase gene of the fungus, leading to production of more cutinase(s) which macerates the cuticle and allows penetration by the fungus.

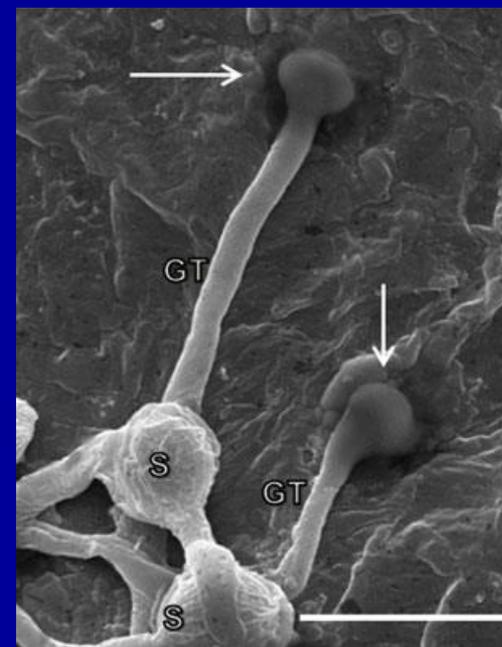
Penetração por ferimentos

Patógenos do Grupo I de Mc New: *Penicillium*,
Rhizopus, *Mucor*, etc

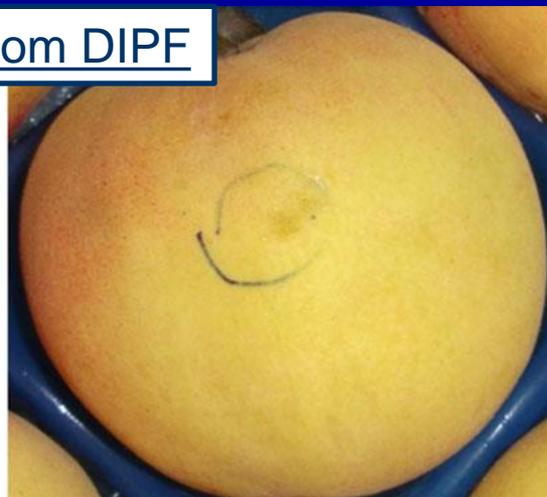
água



solução nutritiva



Tratados com DIPF



Penetração por aberturas naturais

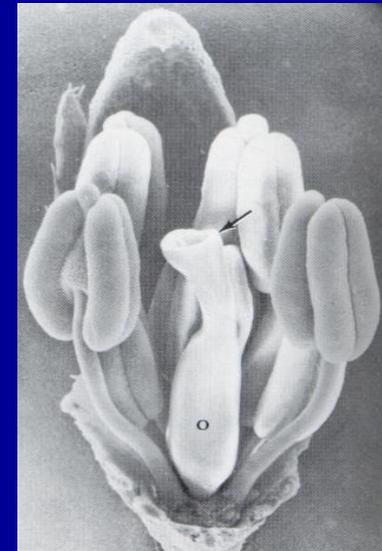
Lenticelas



1. ovário
2. estilete
3. estigma (*Ustilago*)
4. filamentos
5. anteras



3. nectário



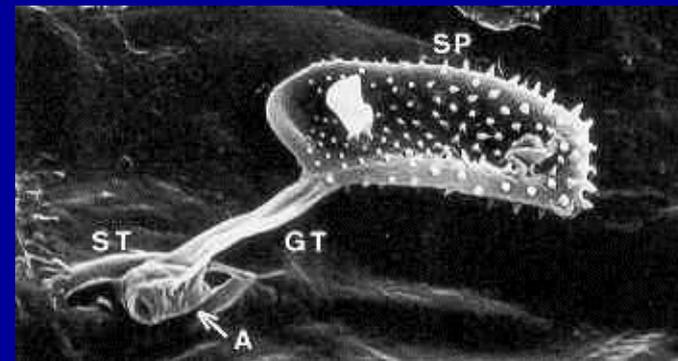
Penetração por abertura natural



Zoósporo encistado e
apressório de
Pseudoperonospora humili

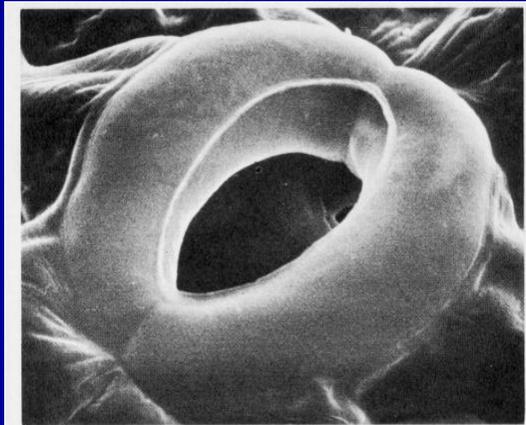


Urediniósporo de *Melampsora*



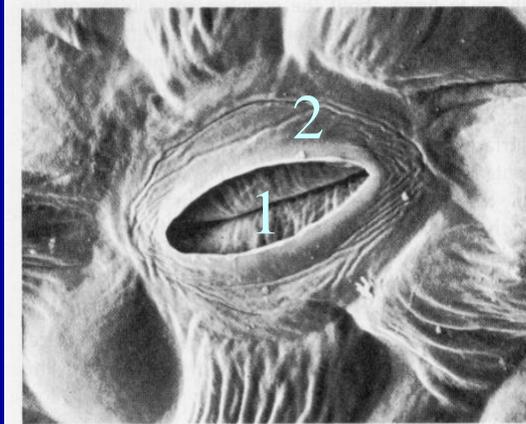
SP = esporo, GT = tubo germinativo
A = apressório, ST = estômato

Penetração por aberturas naturais e ferimentos



(a)

5 μm



(b)

5 μm

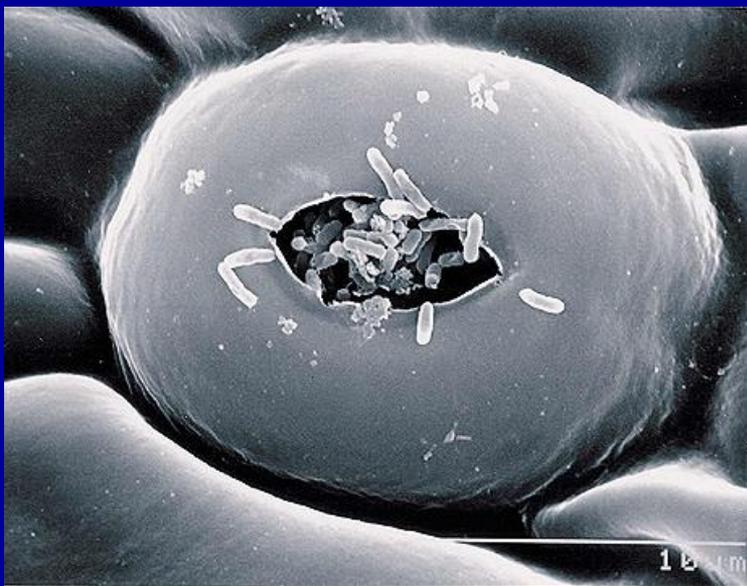
1. Células
-guarda

2. células subsidiárias



Citros com galeria
de larva minadora

Vias de penetração aberturas naturais & ferimentos



Xanthomonas axonopodis pv. *citri* em câmara subestomática de folhas de grapefruit (Gottwald et al., 2002)



Sintomas de cancro cítrico relacionados a ferimentos por espinhos (alto) e galeria da larva minadora.

Penetração por aberturas naturais e ferimentos

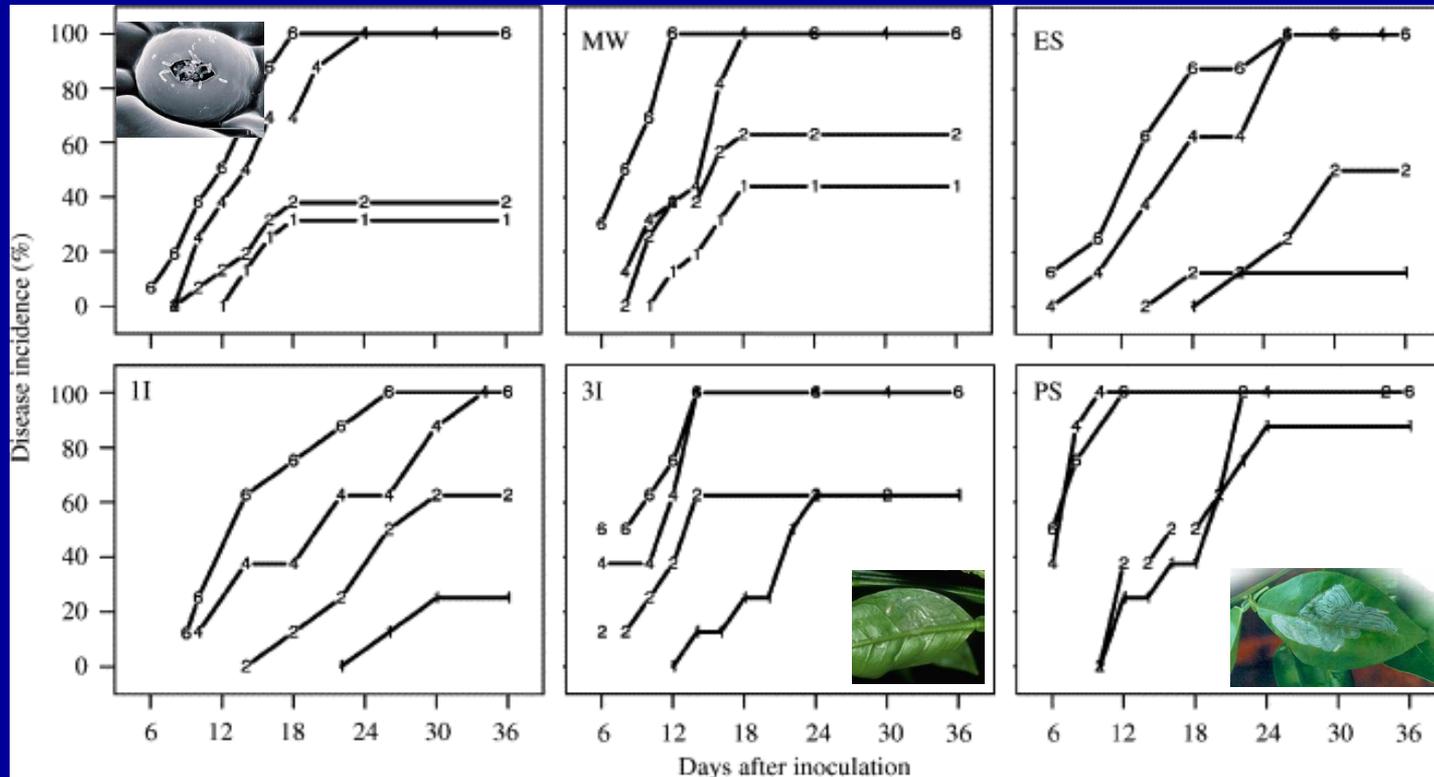
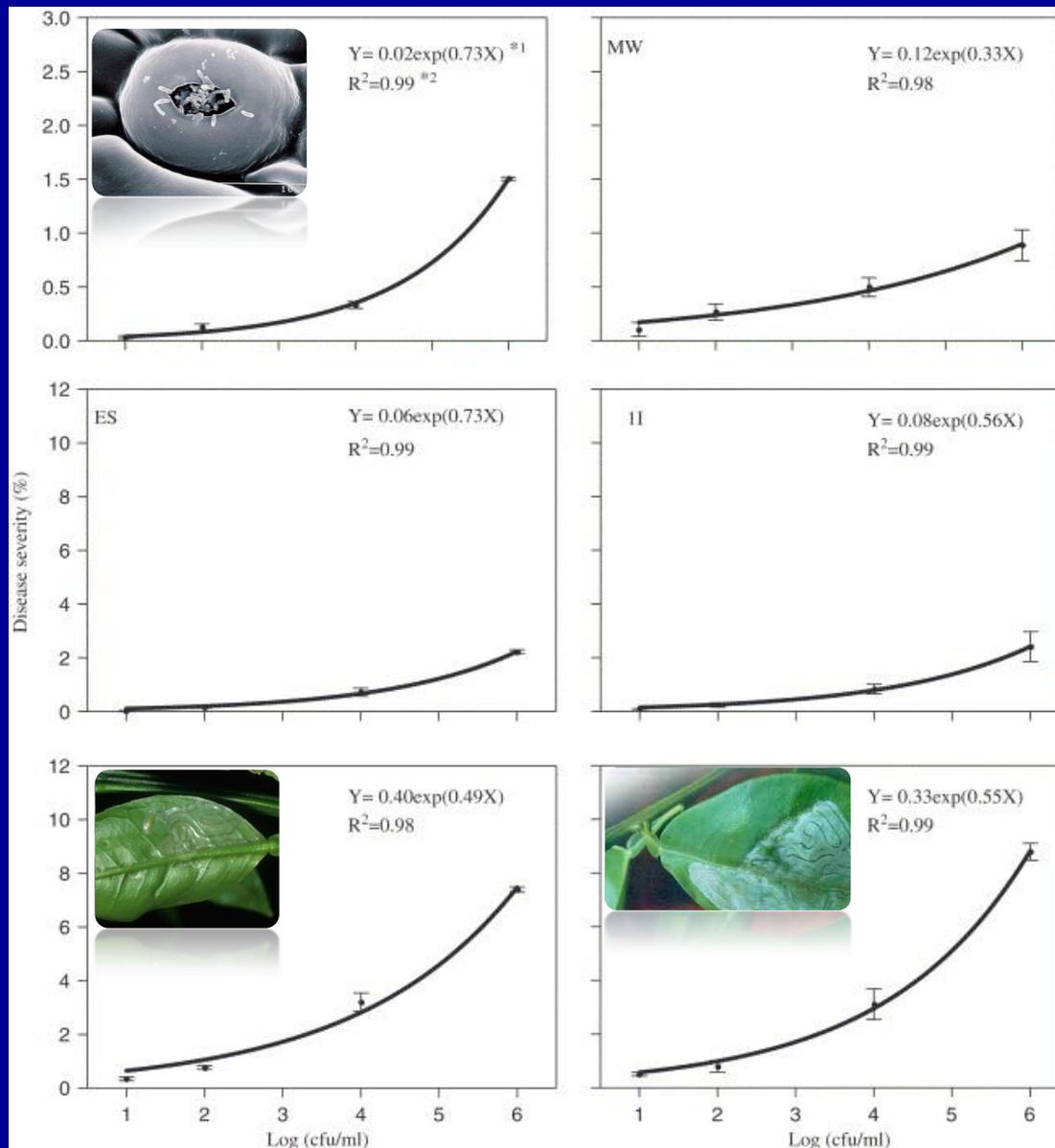


Fig. 1. Effect of leaf treatment and inoculum concentration on the progress of disease incidence (% diseased plants) of Asiatic citrus canker in Tahiti lime. Leaf treatments: intact leaf (IL), mechanically wounded (MW) and infested with various stages of citrus leaf miner: egg stage (ES), first instar (1I), third instar (3I) and pupal stages (PS). Inoculum concentrations of *Xanthomonas axonopodis* pv *citri* are: 10¹ (1), 10² (2), 10⁴ (4) and 10⁶ (6) cfu ml⁻¹. (Christiano et al., 2007)

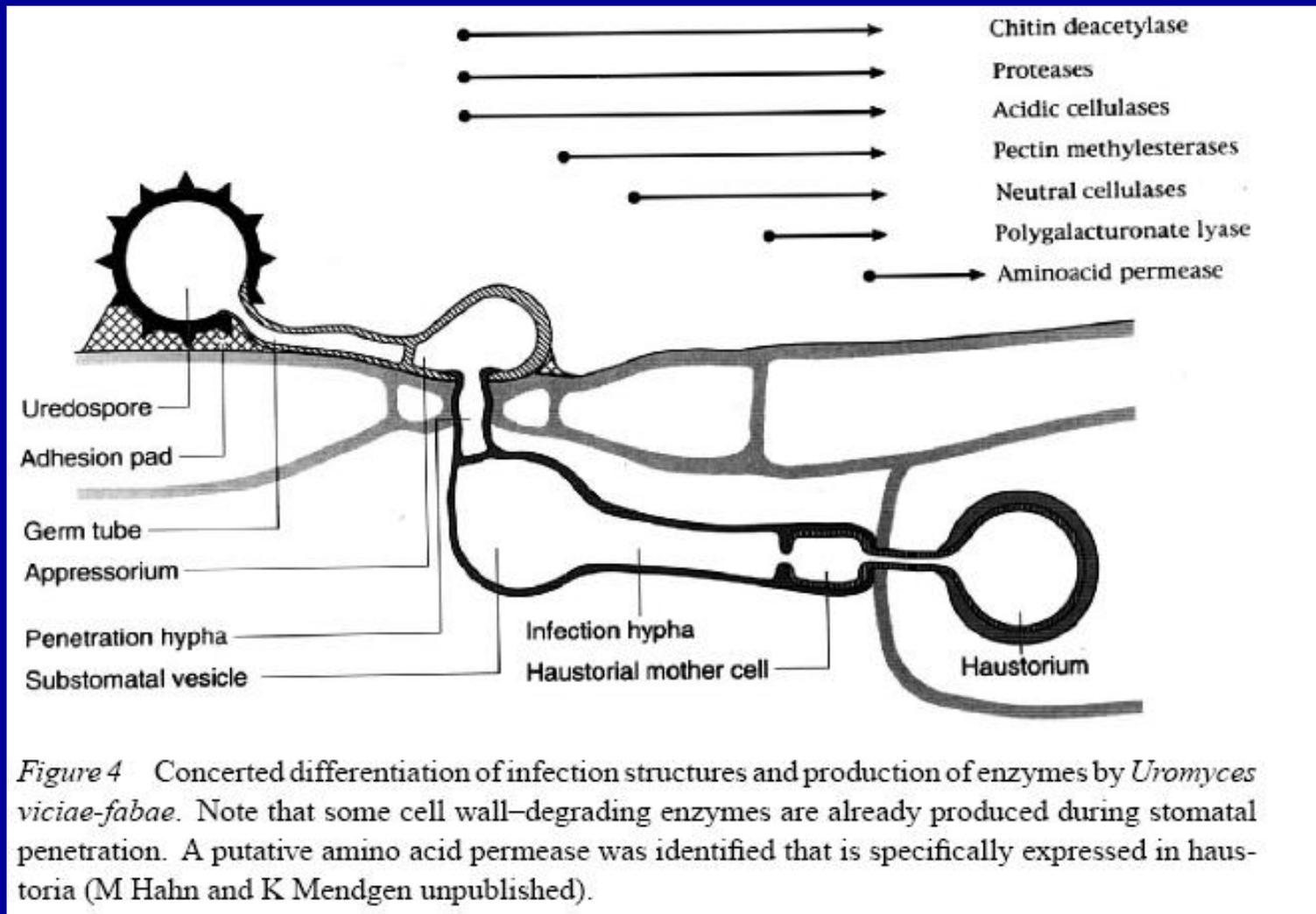
Vias de penetração aberturas naturais & ferimentos

Fig. 3. The effect of leaf treatment and inoculum concentration on the severity (% leaf area infected) of Asiatic citrus canker in leaves of Tahiti lime. Leaf treatments are: intact leaf (IL), mechanically wounded (MW) and infested with various stages of citrus leaf miner: egg stage (ES), first instar (1I), third instar (3I) and pupal stages (PS).

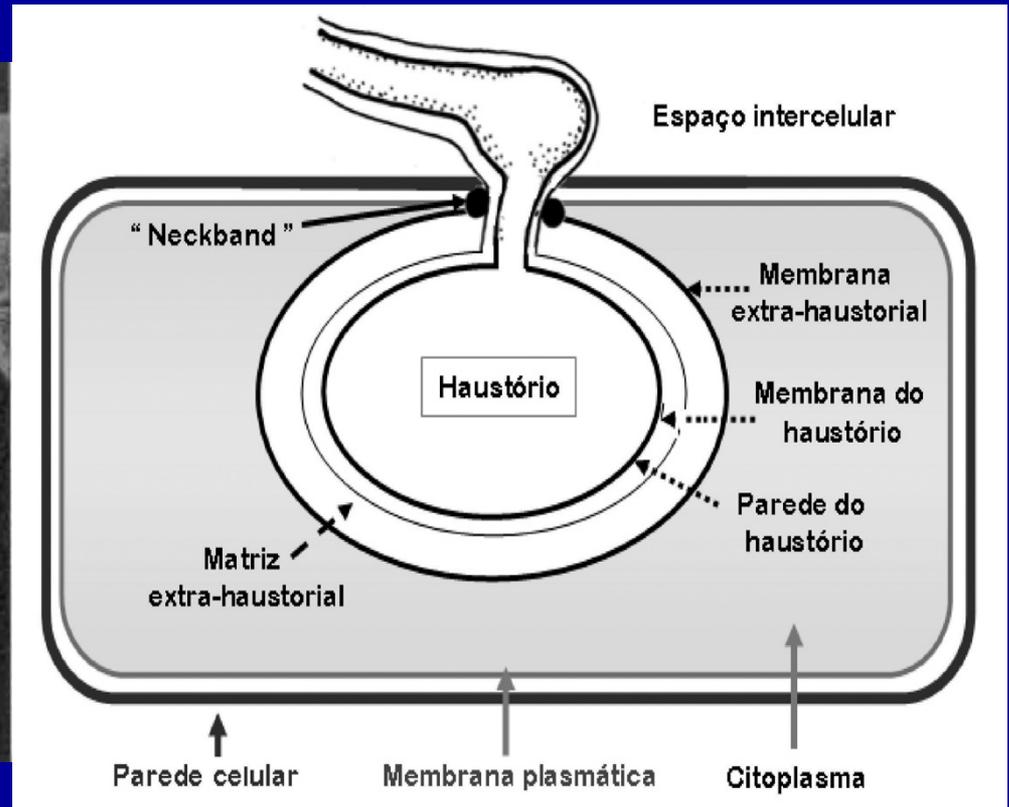
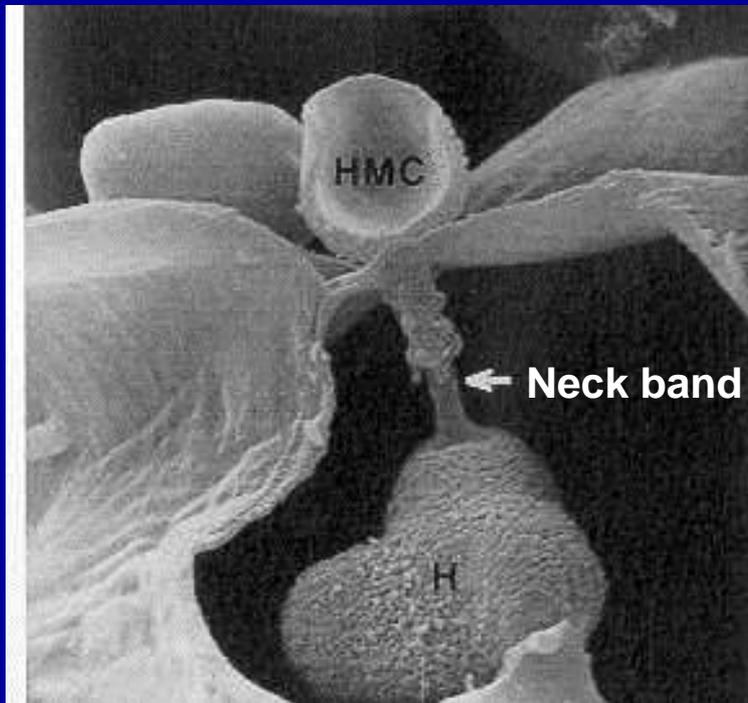
(Christiano et al., 2007)



Relações parasitárias estáveis



Relações parasitárias estáveis



HMC = CÉLULA MÃE DO HAUSTÓRIO
H = HAUSTÓRIO DE *Hemileia vastatrix*

Neck band - contenção da matrix
extrahaustório

Relações parasitárias estáveis

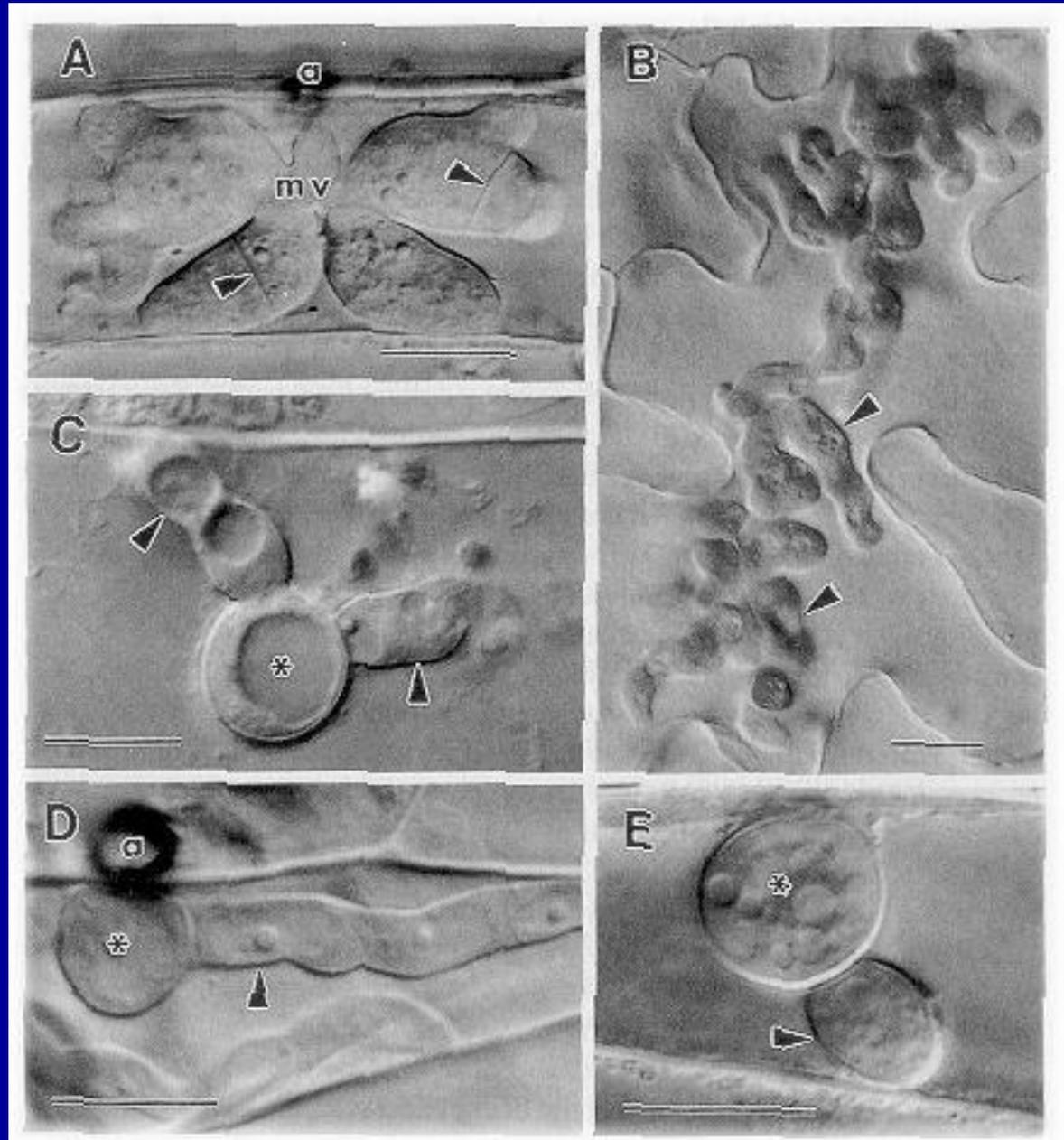
A = *Colletotrichum destructivum* - caupi (mv = vesícula multilobulada)

B = *C. truncatum* - ervilha (hifa primária na célula epidérmica)

C = *C. orbiculare* - pepino (hifas primárias se originando da vesícula)

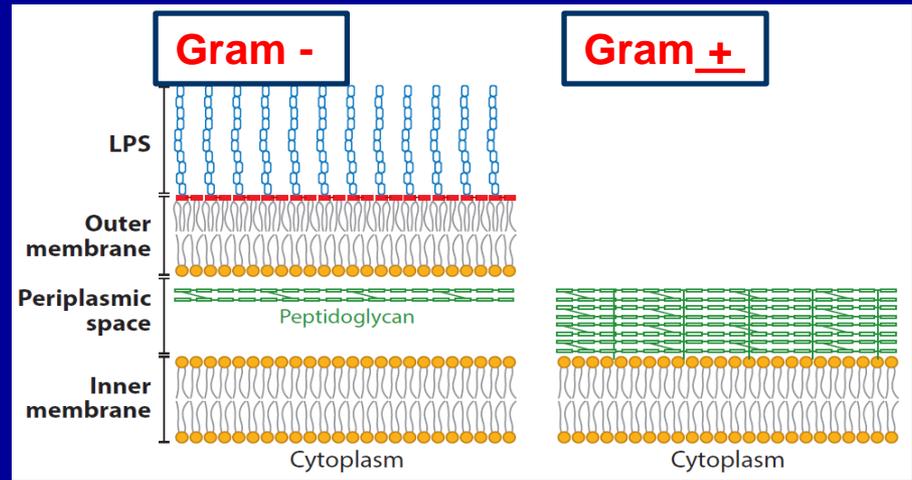
D = *C. malvarum* - *Sida spinosa* (hifa primária se originando da vesícula)

E = *C. lindemuthianum* - feijão (hifa primária se originando da vesícula)



Relações parasitárias estáveis

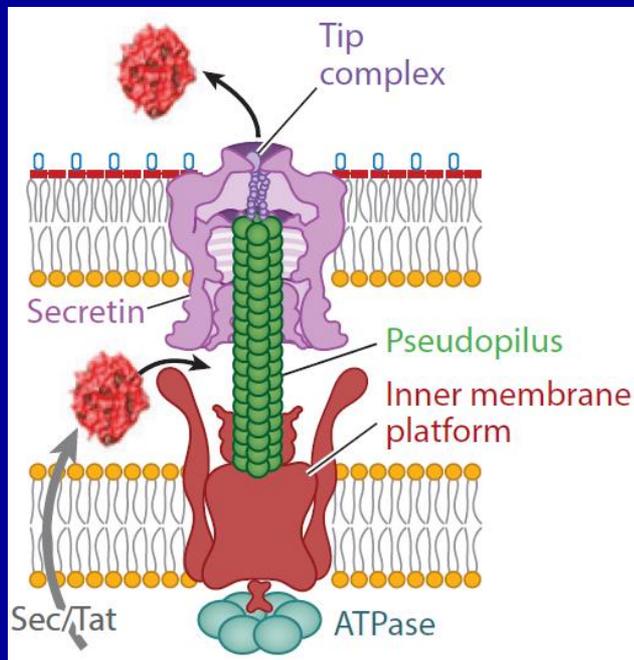
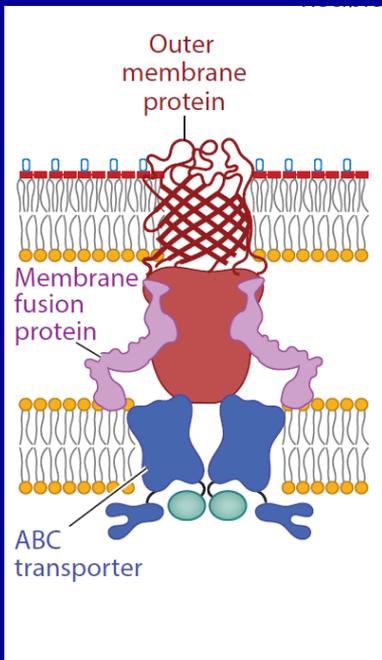
Sistemas de secreção bacterianos
 I a VI Gram –
 Tipo IV todas as bactérias
 Tipo VII Gram +



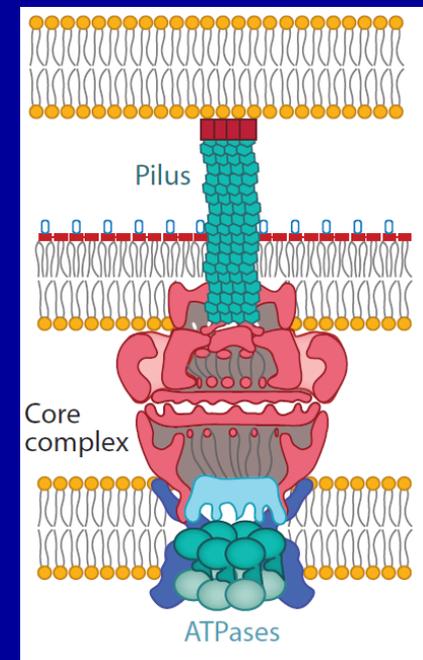
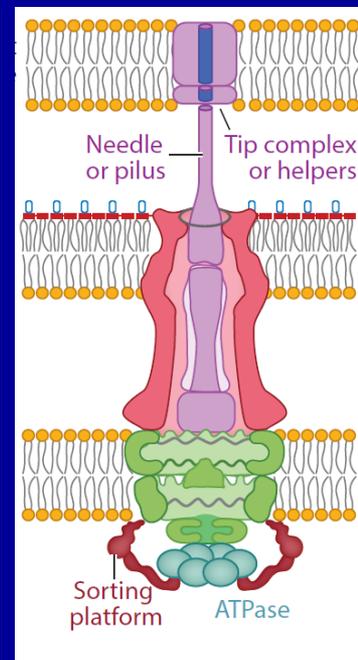
Secreção para o meio

Tipo I (TISS)

Tipo II (TIISS)



→ Secreção para dentro da célula
 Tipo III (TIISS) Tipo IV (TIVSS)



Sistema de secreção tipo III (needle – principalmente células animais)

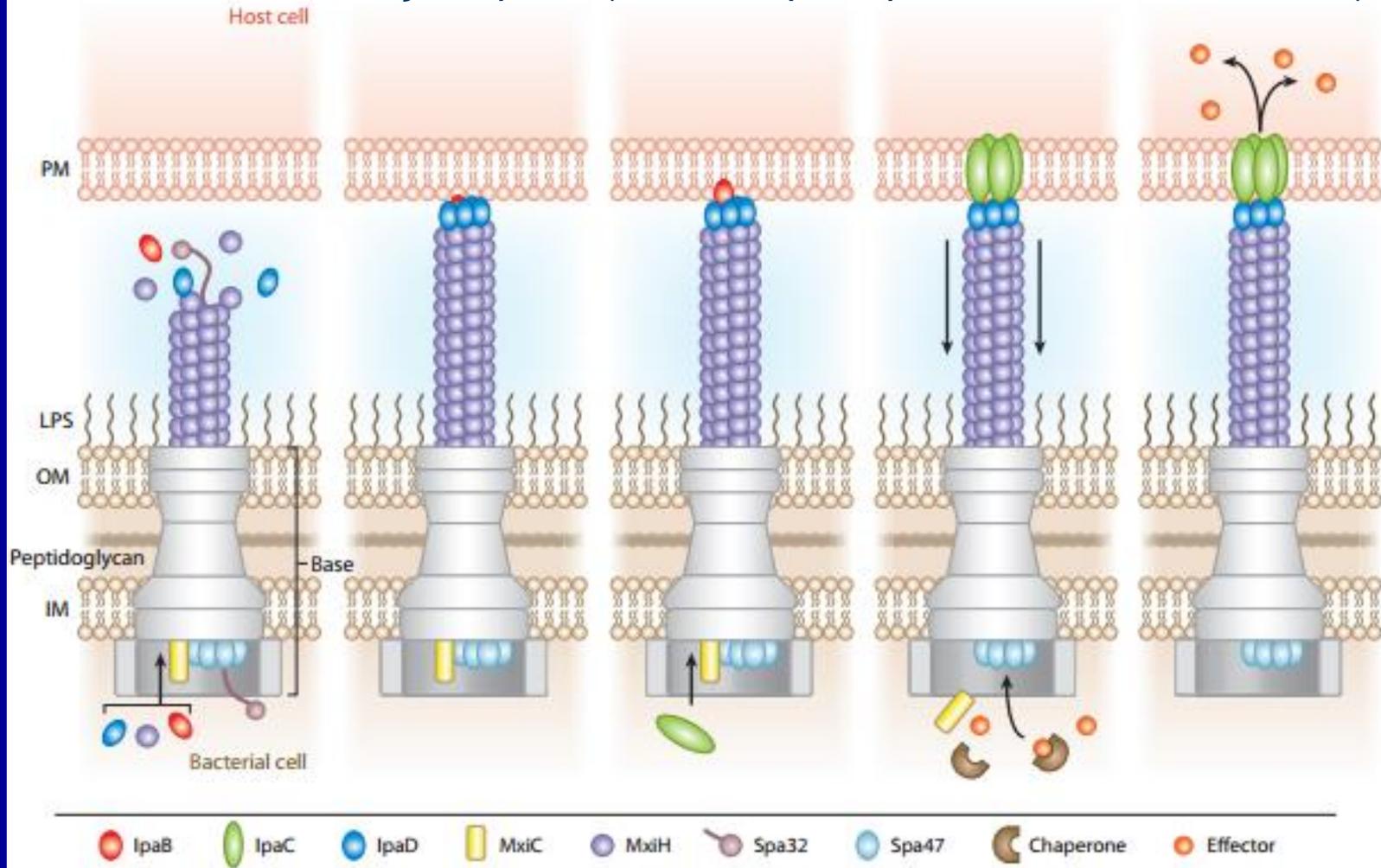


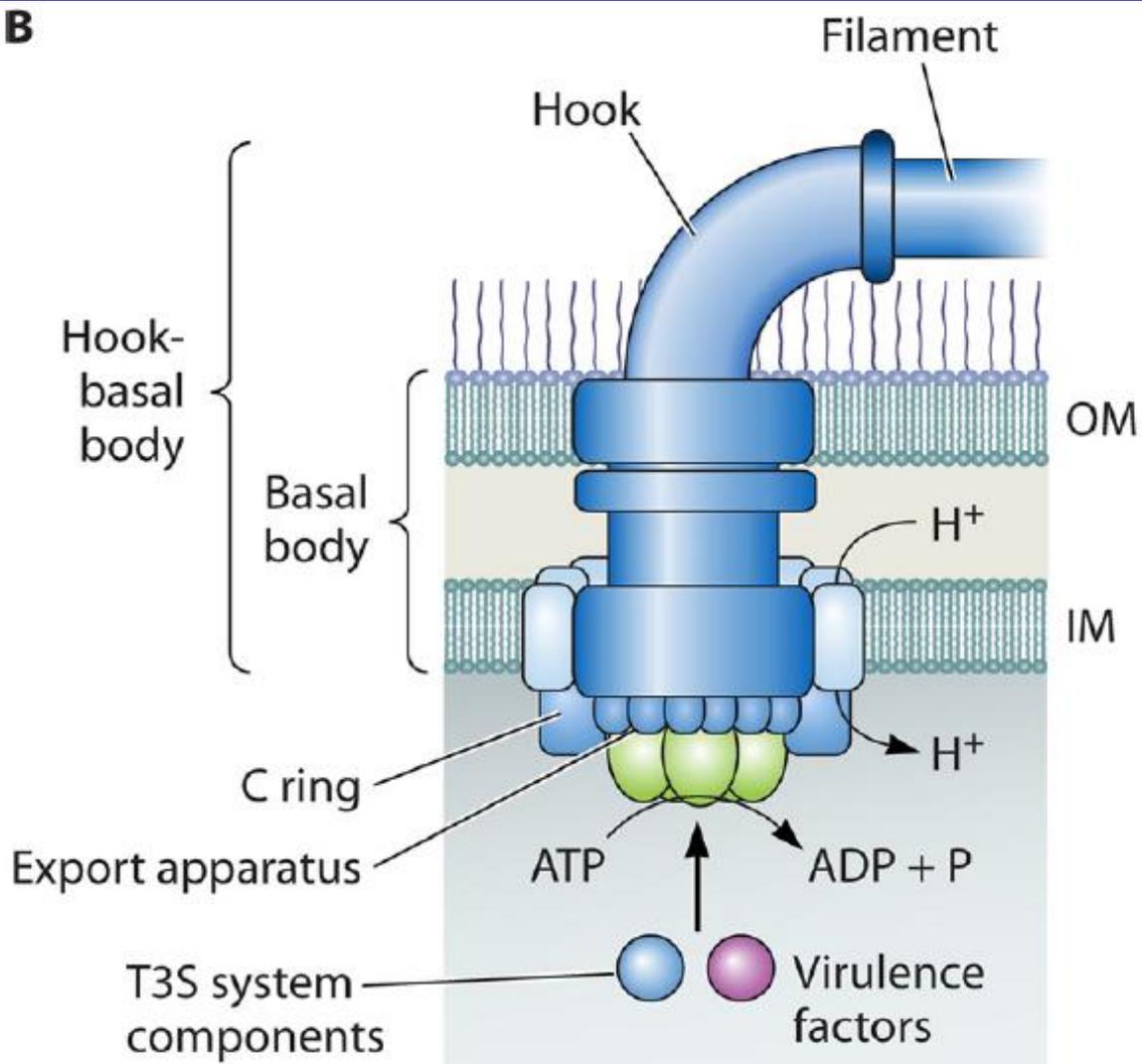
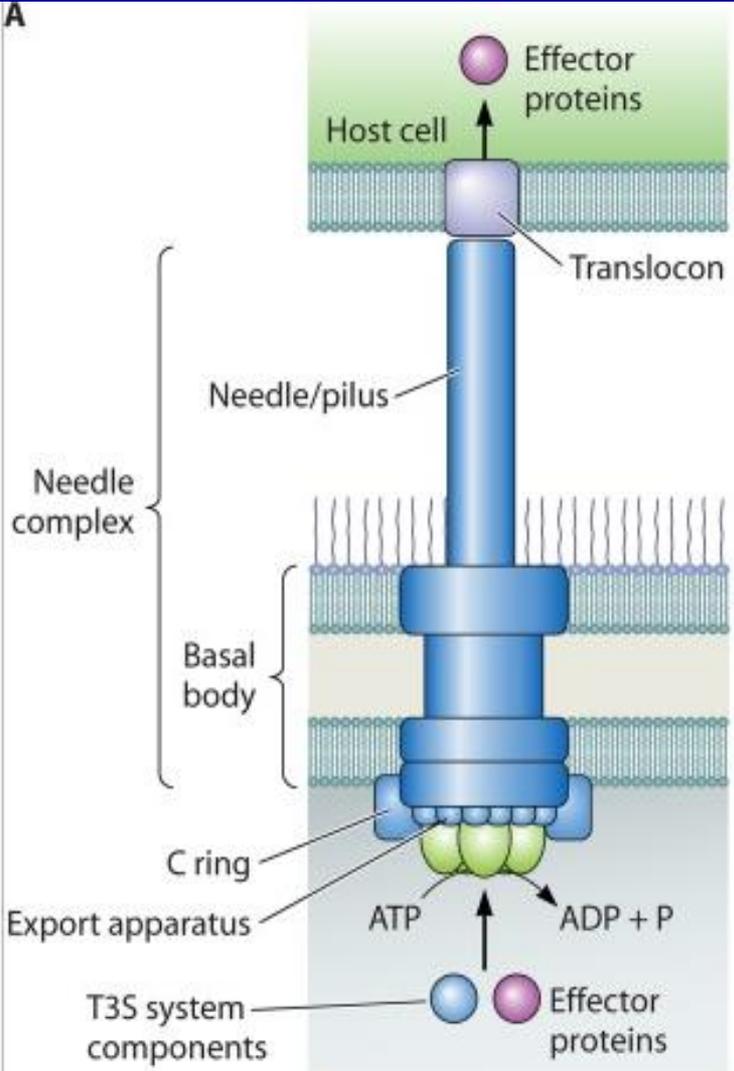
Figure 1

Proposed model for Type III secretion in *Shigella flexneri*. From left to right, MxiC inhibits the activity of ATPase Spa47, preventing effector secretion while allowing transport of needle proteins MxiH, IpaB, IpaC, and IpaD, and molecular ruler protein Spa32. Contact with the host triggers insertion of IpaB into the plasma membrane, resulting in IpaC transport and pore formation. Transmission of a signal from the needle tip to the base of the injectisome causes activation of Spa47 and effector secretion into the host cell. Abbreviations: IM, inner membrane; LPS, lipopolysaccharide; OM, outer membrane; PM, plasma membrane.

Sistema de Secreção tipo III – comum para transporte de efetores

Principalmente em animal – needle 80 nm

Vegetal – pilus ou flagelo 2 μm



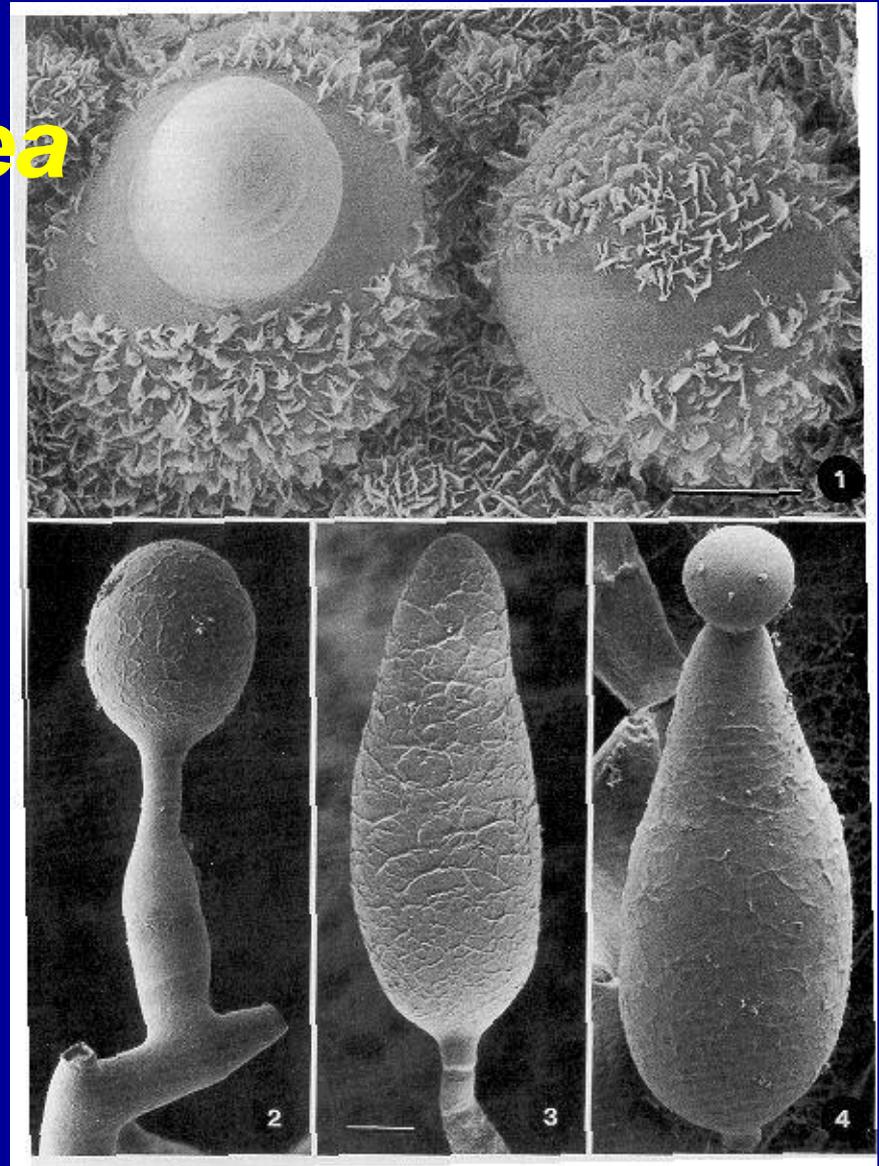
Infecção de *Magnaporthe grisea*

1 Início da formação do conidióforo na superfície da folha de arroz

2 Início da formação do conídio

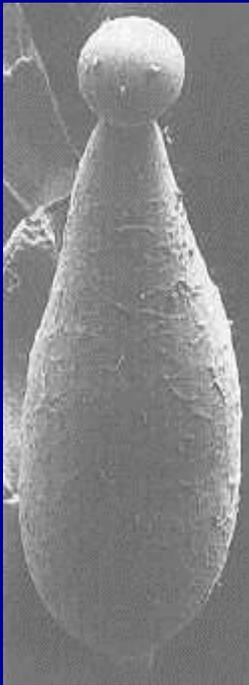
3 Conídio em estágio intermediário, em fase de expansão

4 Conídio completamente expandido com gota mucilaginosa apical, ainda preso ao conidióforo



Infecção de *Magnaporthe grisea*

Pré-penetração

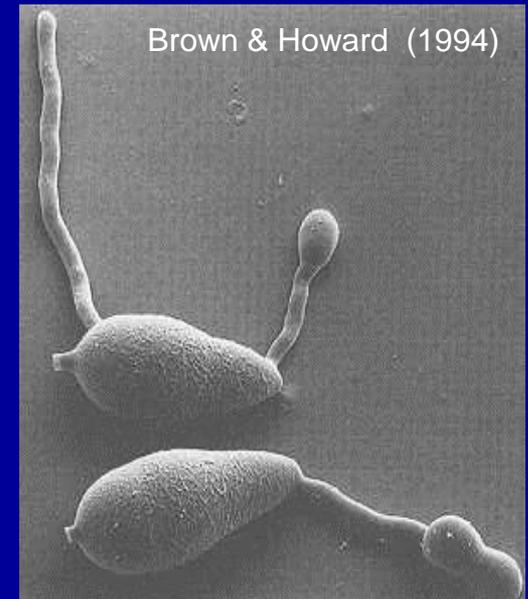


Conídio + gota apical
de mucilagem



Conídio preso ao
substrato por
adesão da mucilagem

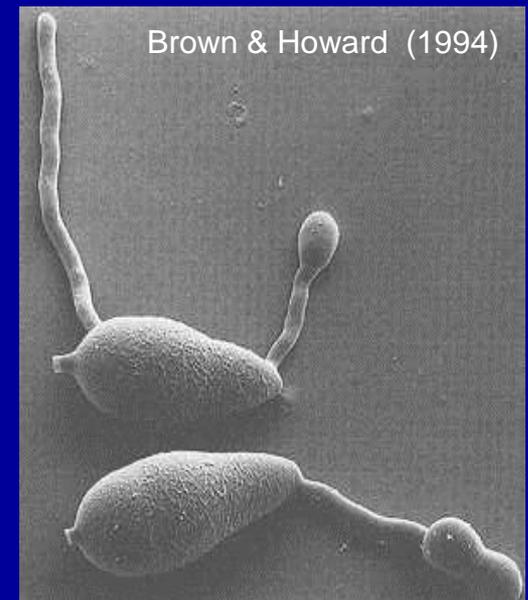
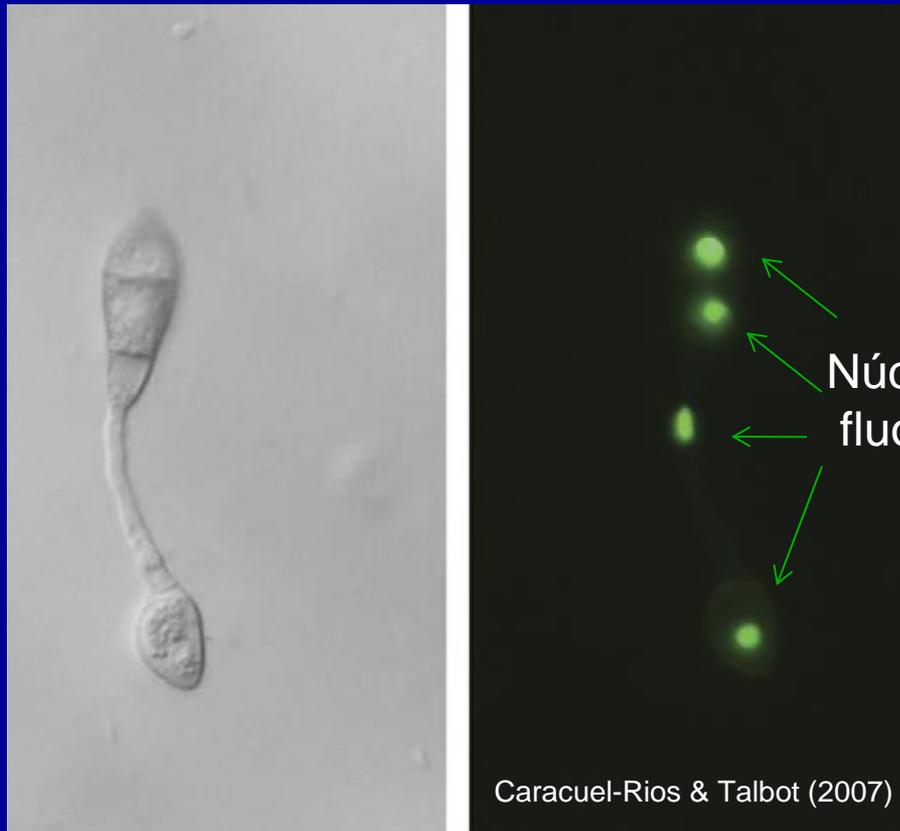
Germinação de conídios
e estágios iniciais de
formação de apressórios



Infecção de *Magnaporthe grisea*

Pré-penetração

Germinação de conídios e estágios iniciais de formação de apressórios

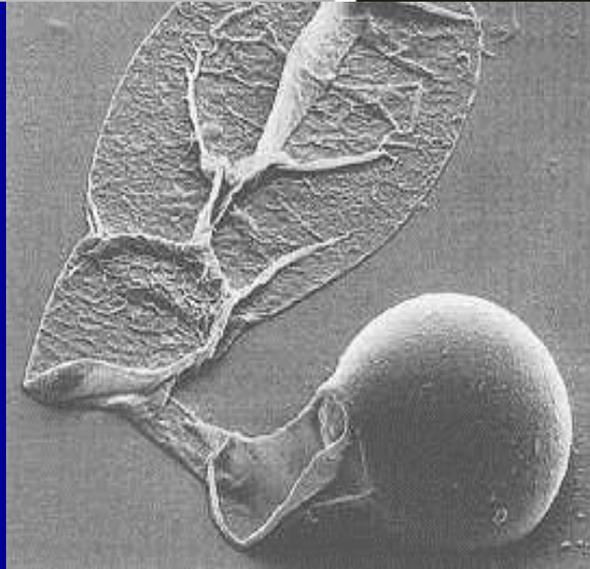


Infecção de *Magnaporthe grisea*



Degeneração
de núcleos do
conídio

Penetração

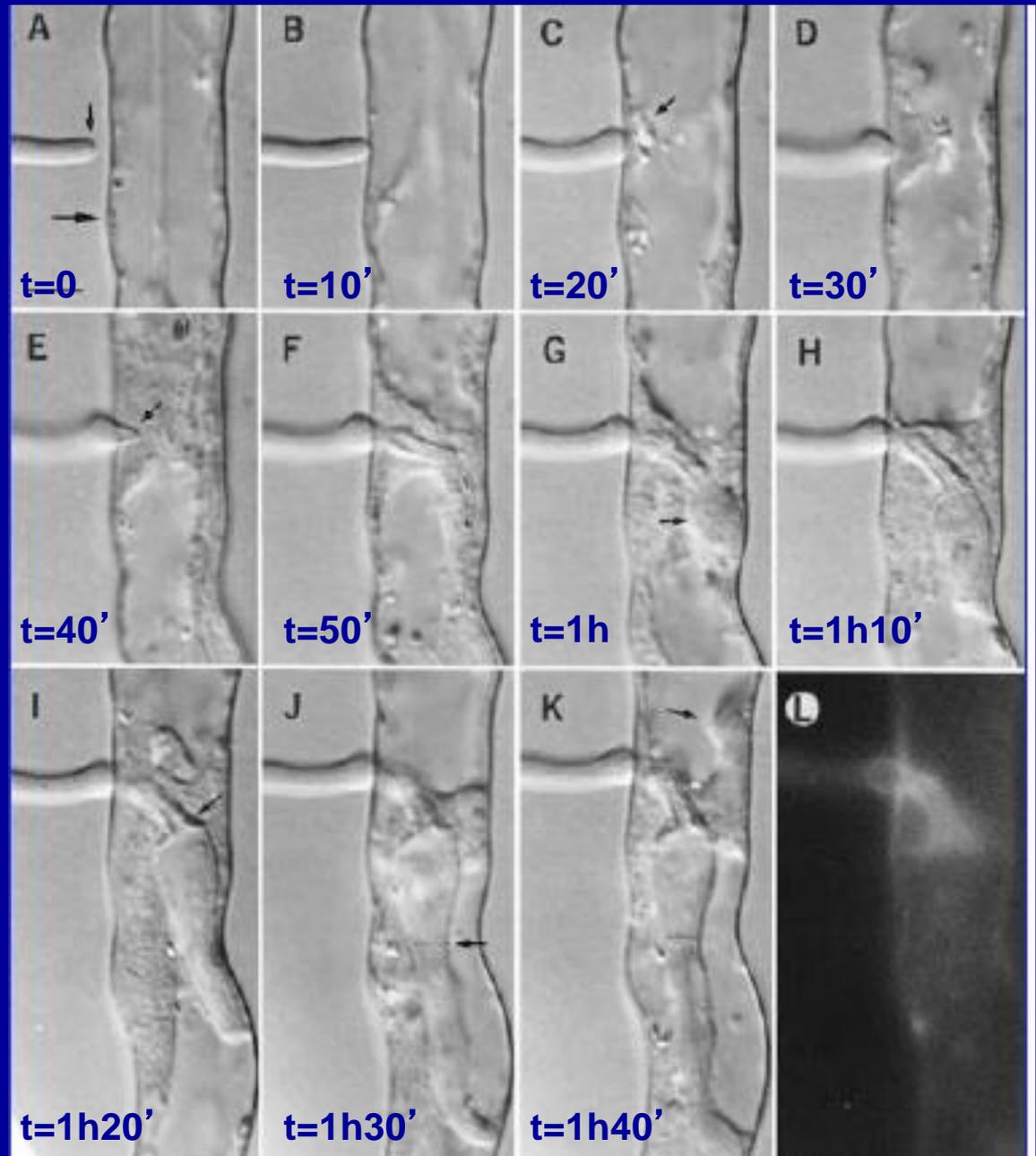


Apressório maduro, globoso,
preso ao conídio em colapso

Efeito do peg de penetração em superfície
de polietileno, após retirada do apressório

Infecção de *Thielaviopsis basicola*

Infecção de pelos radiculares de fumo por *T. basicola* (Hood & Shew, 1997)

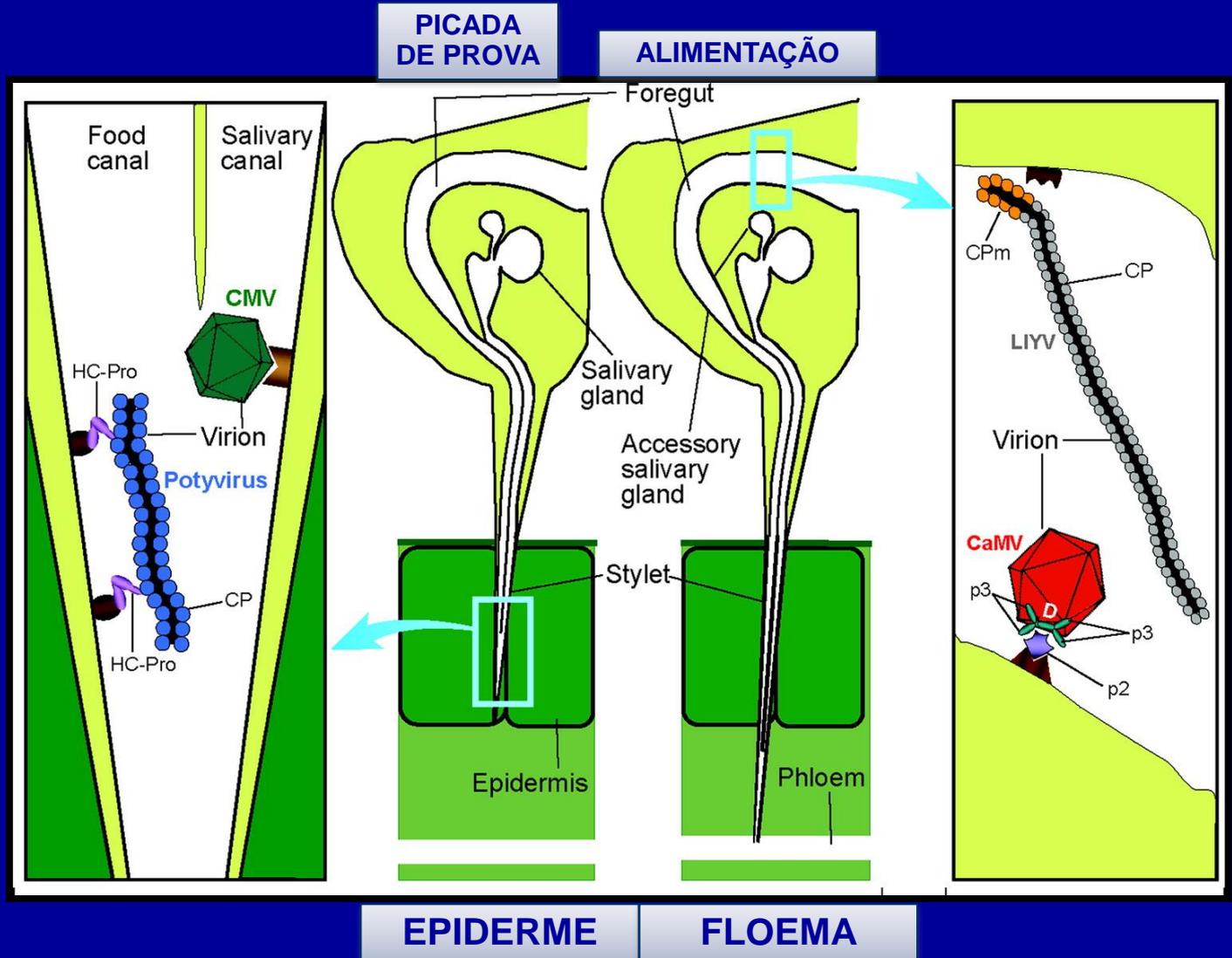


Vírus

TRANSMISSÃO NÃO-PERSISTENTE

TEMPOS DE

AQUISIÇÃO
INOCULAÇÃO
LATÊNCIA
RETENÇÃO



Vírus e Bactérias

TRANSMISSÃO PERSISTENTE

TEMPOS DE

AQUISIÇÃO
INOCULAÇÃO
LATÊNCIA
RETENÇÃO

