Monoxenic production of the entomopathogenic nematode *Steinernema carpocapsae* in an internal-loop airlift bioreactor: Evolution of the hydrodynamic conditions.

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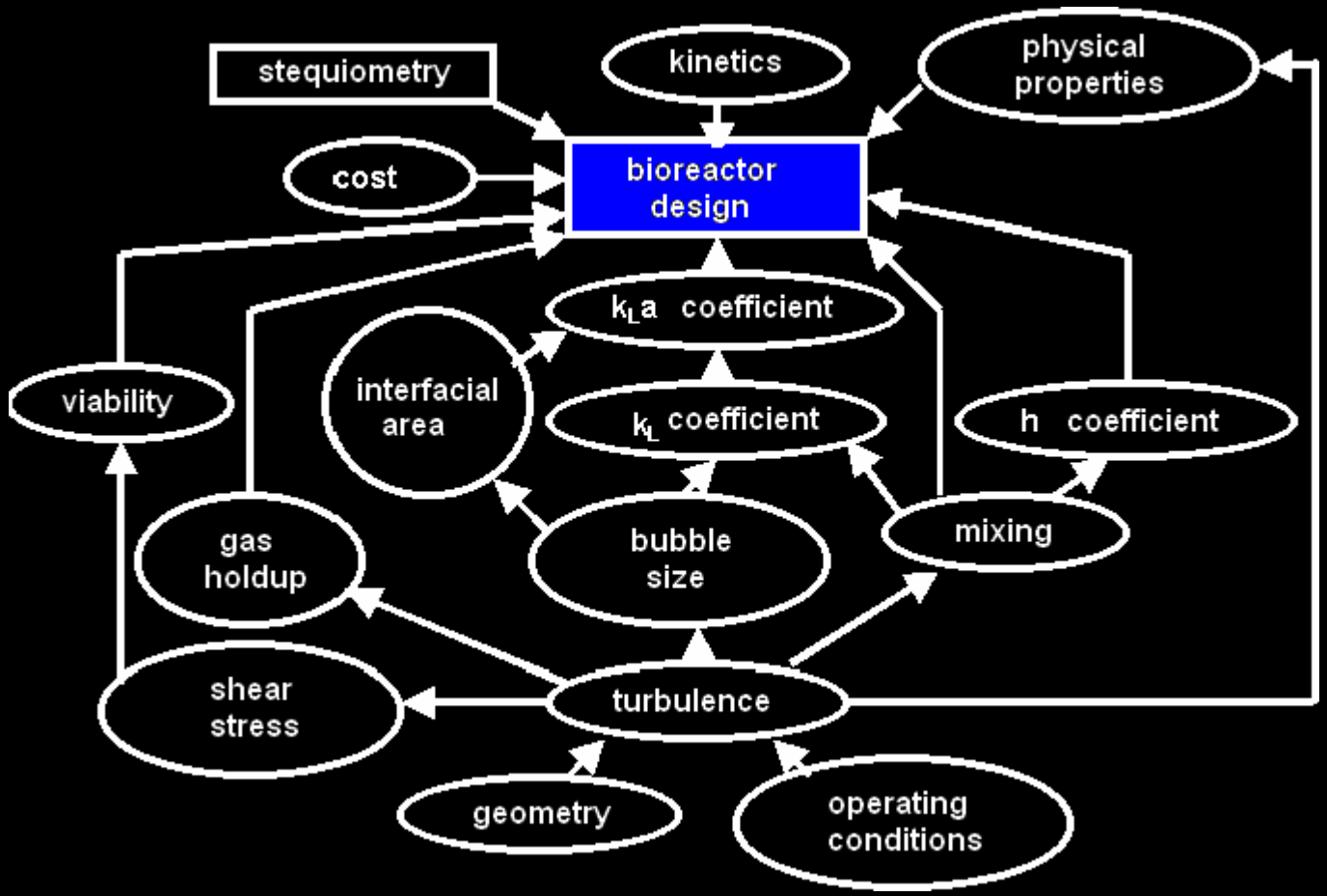
Food Science and Technology Research Centre, Autonomous University of Hidalgo State. MEXICO.

Biocontrol Symbiosis/Entomopathogen
Nematodes and Symbiotic Bacter

INTRODUCTION

Biological control of insect pests (Mass production of IJ stages of entomopathogenic nematodes, *Steinernema* spp.; *Heterorhabditis* spp.).

INTRODUCTION



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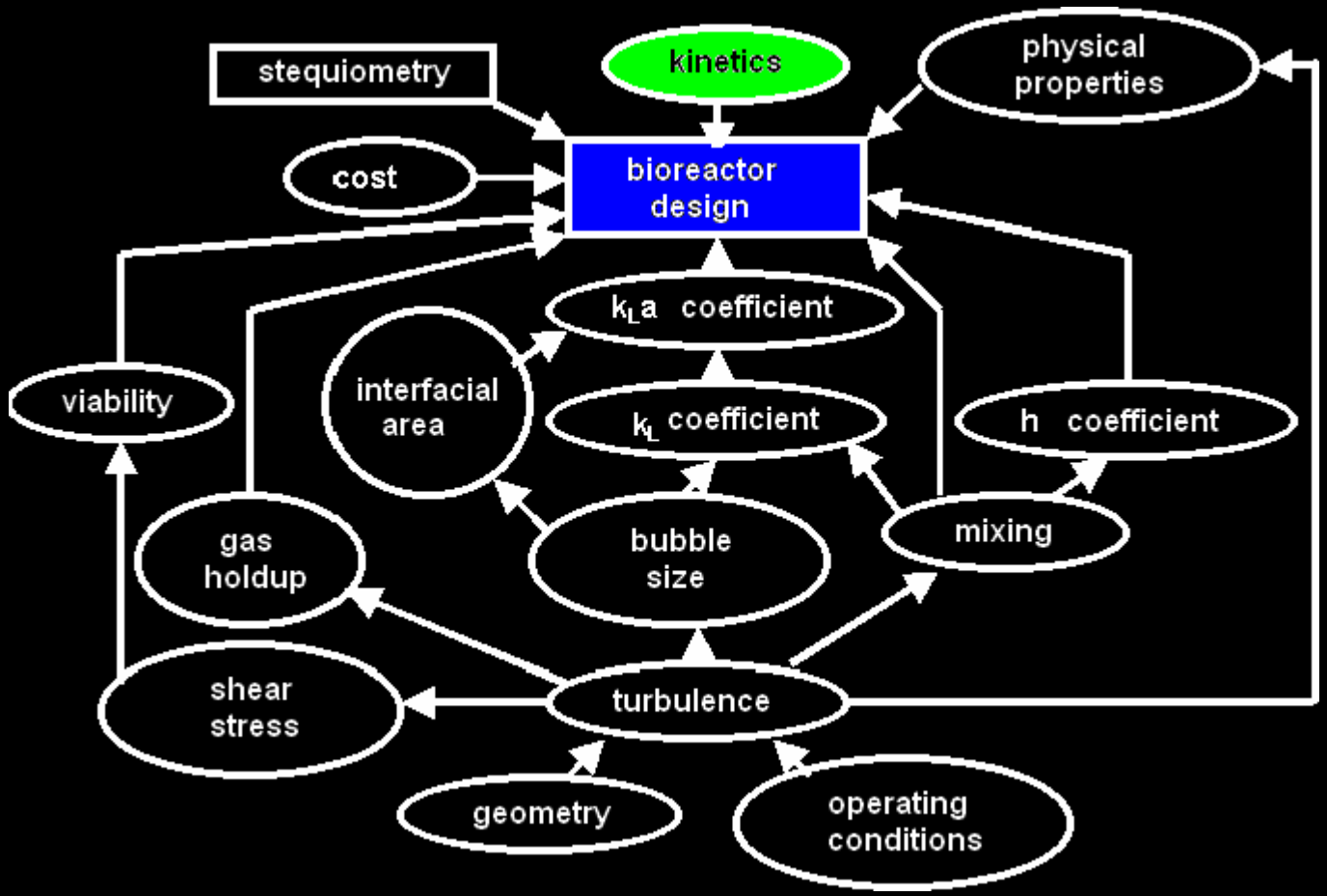

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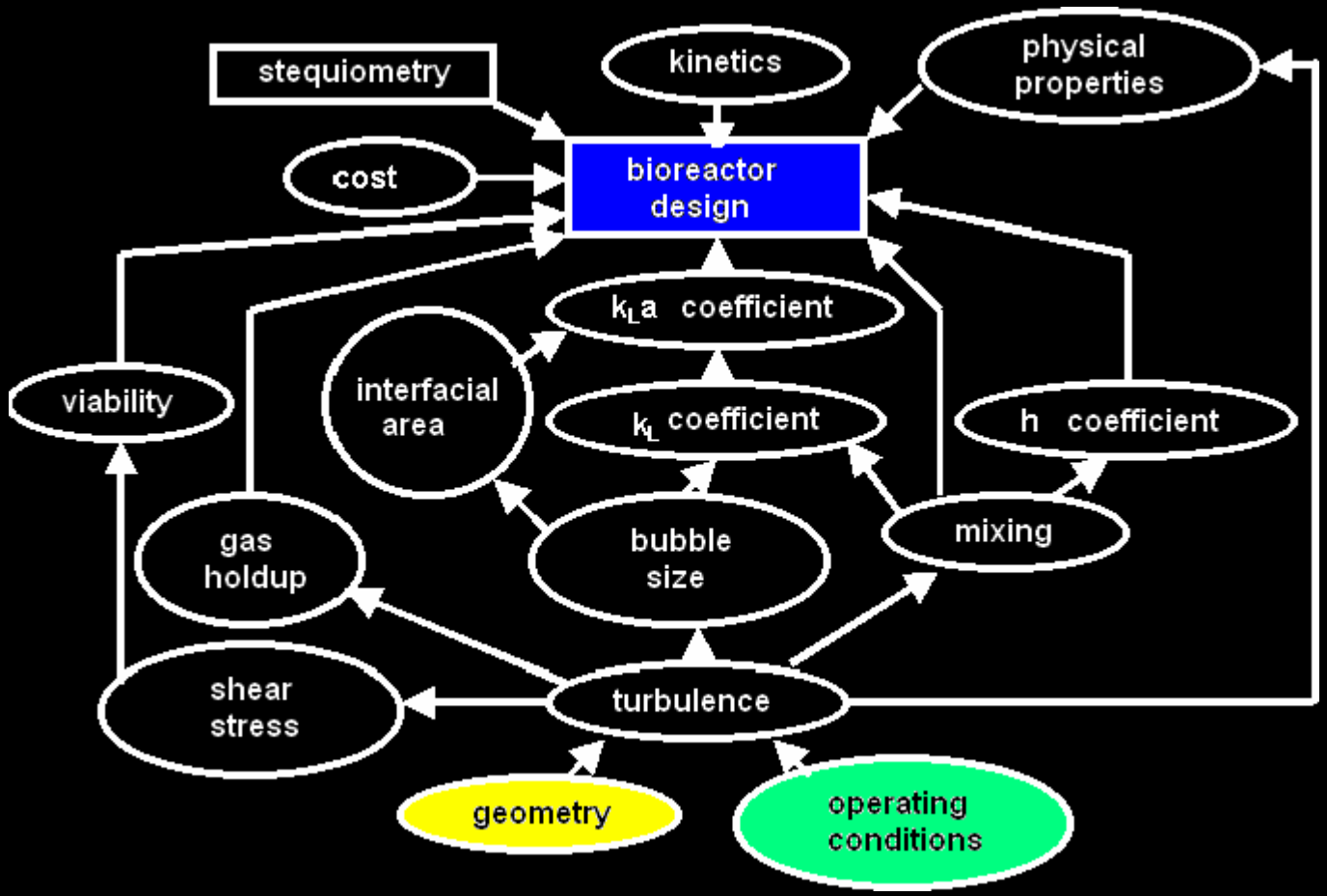

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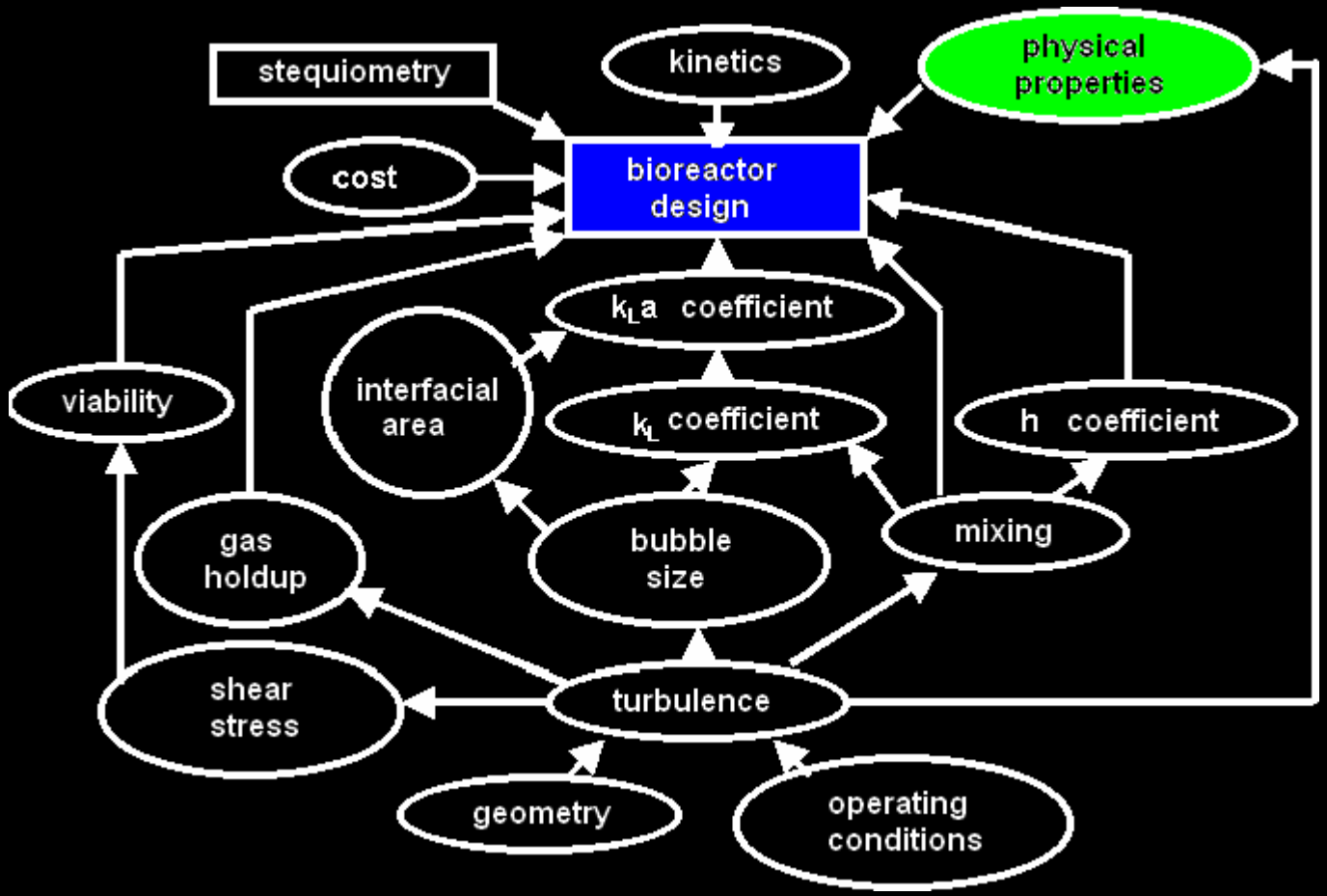
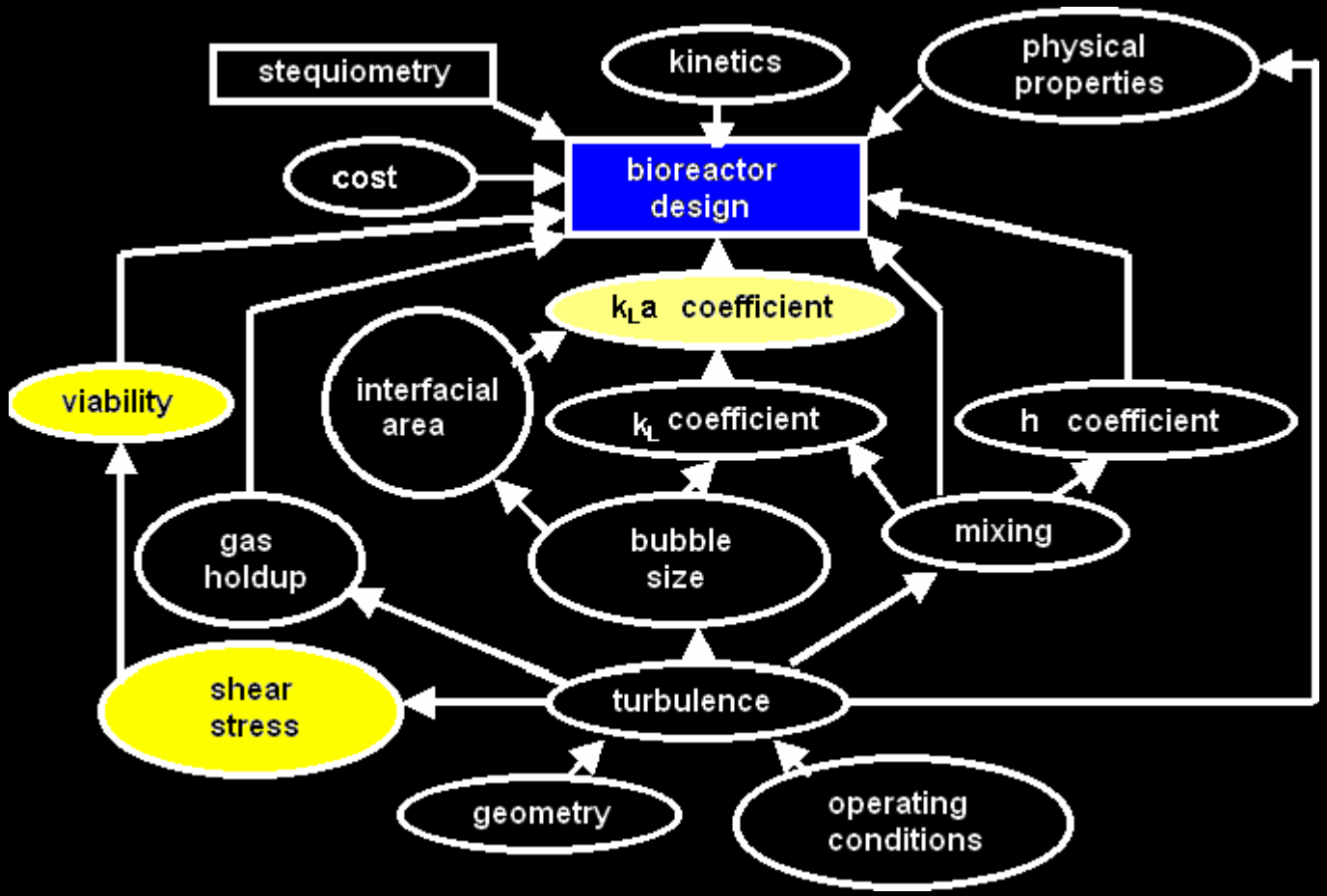
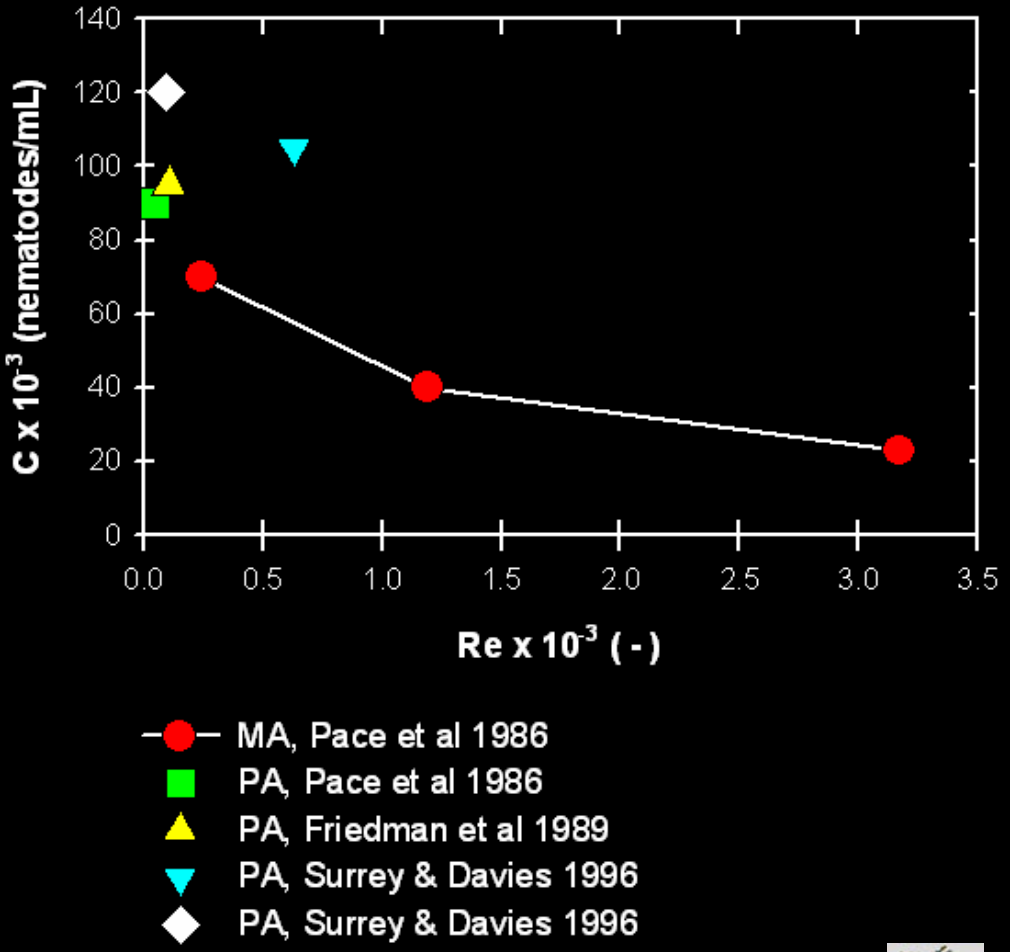
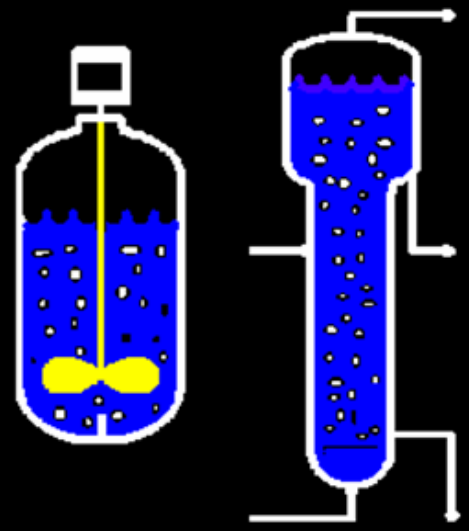


INTRODUCTION



- Using data of *K* and *n* (Chavarría-Hernández et al 2003)
- Using equations -Reynolds number-, Re = f (N, D,...)





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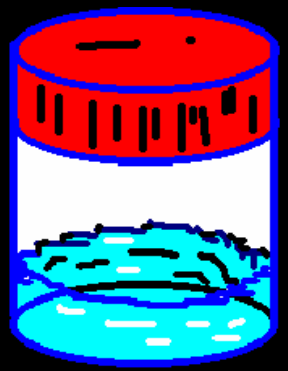
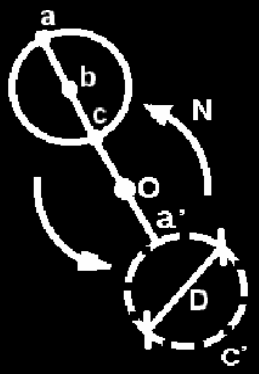
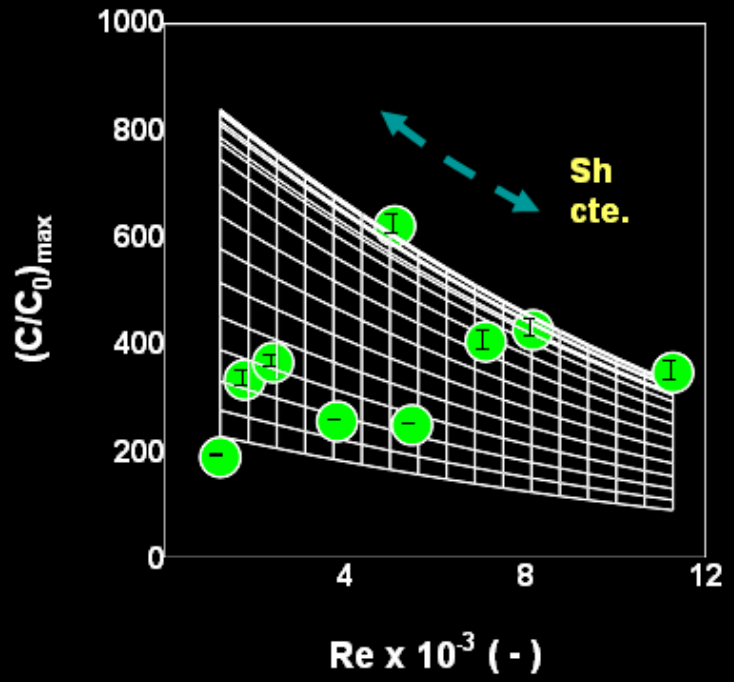
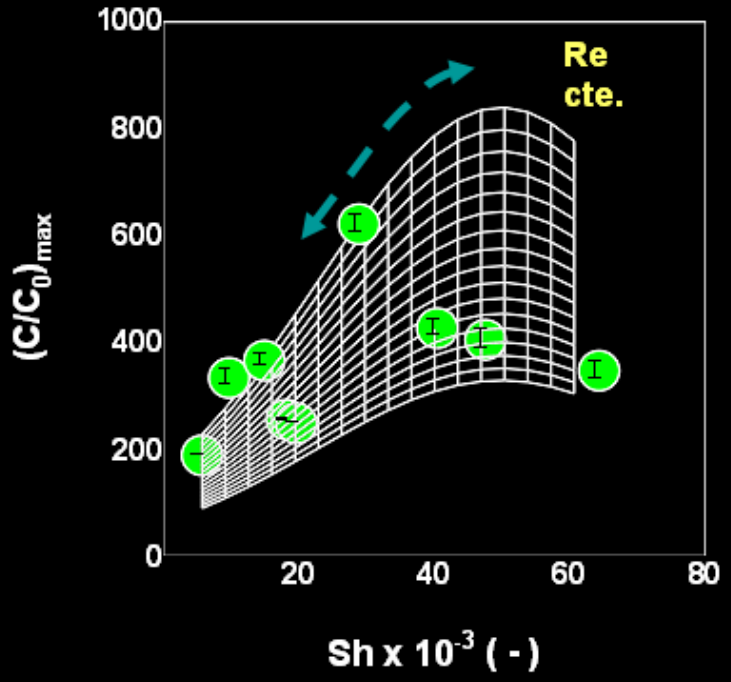


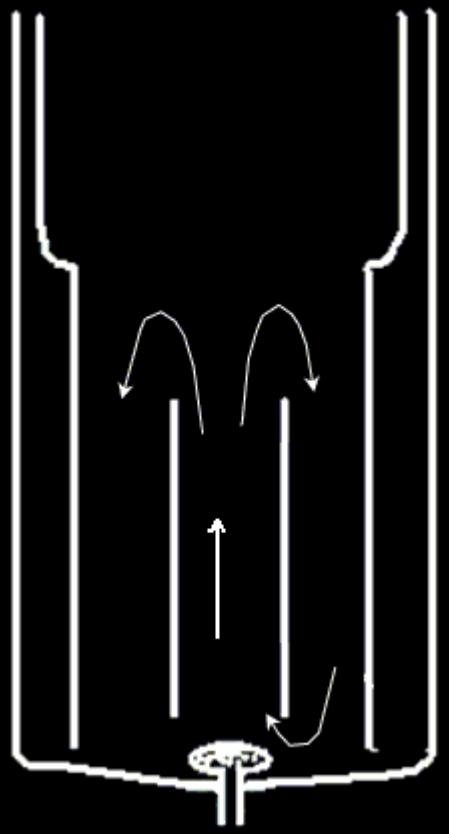


- Re; hydrodynamics.
- Sherwood number –Sh-, oxygen transfer conditions.

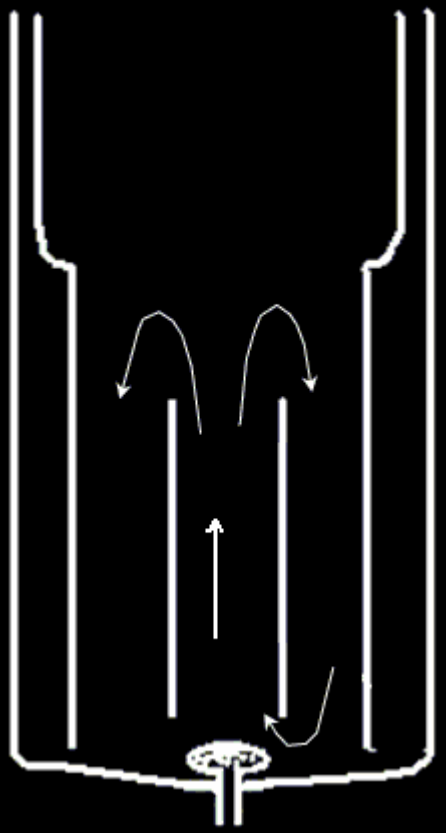


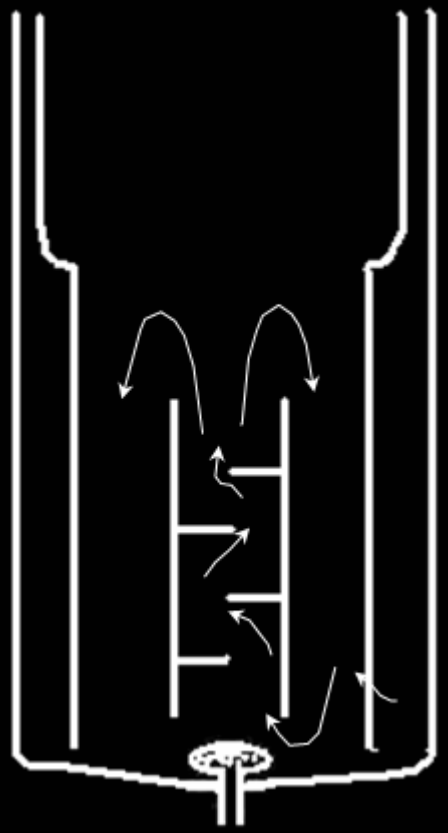


Internal loop airlift bioreactor



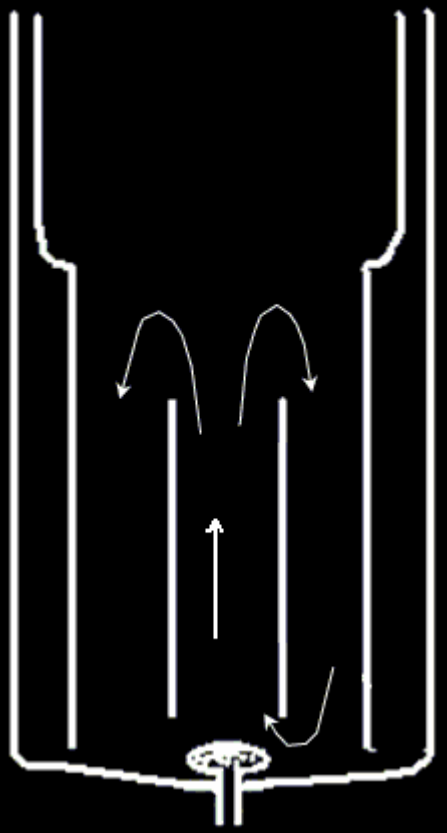
Internal loop airlift bioreactor

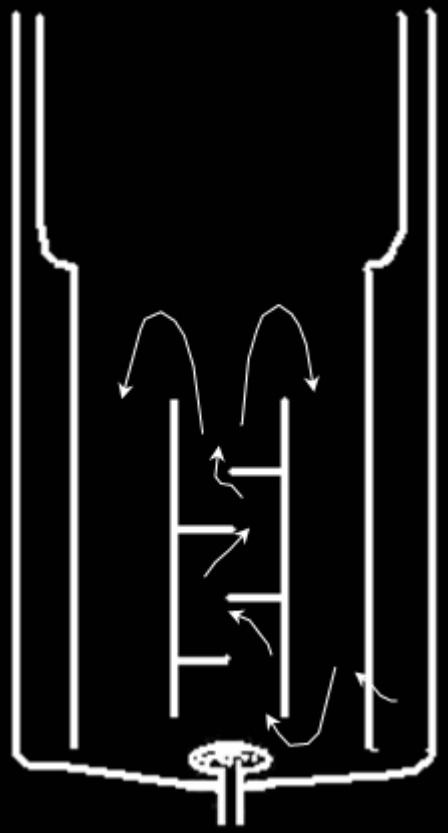




Static mixer

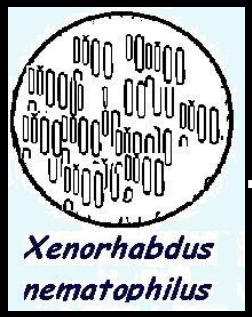
Internal loop airlift bioreactor





Static mixer

Cultures of *Steinernema carpocapsae* and *Xenorhabdus nematophila*



25 % v/v
glycerol

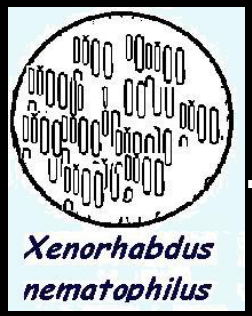
T= -80°C

Bacterium
culture

150 rpm,
30°C, 24 h

MATERIALS & METHODS

Cultures of *Steinernema carpocapsae* and *Xenorhabdus nematophila*



Bacterium culture

150 rpm, 30°C, 24 h

5 % (v/v)

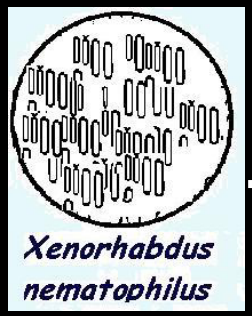
Production medium

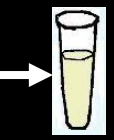
Q=1 vvm, 30°C, 48h

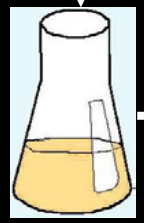
MP1. 50% v/v whey, 2% w/v yeast extract, 1% w/v dried egg yolk, 0.3% w/v NaCl, 3.7% v/v corn oil (Chavarría-Hernández et al 2006).

MP2. 8.2% v/v aguamiel (sap from *Agave* spp.), 1.2% w/v dried egg yolk, 0.4% w/v yeast extract, 0.5% w/v NaCl, 2.5% v/v corn oil (Islas-López et al 2005).

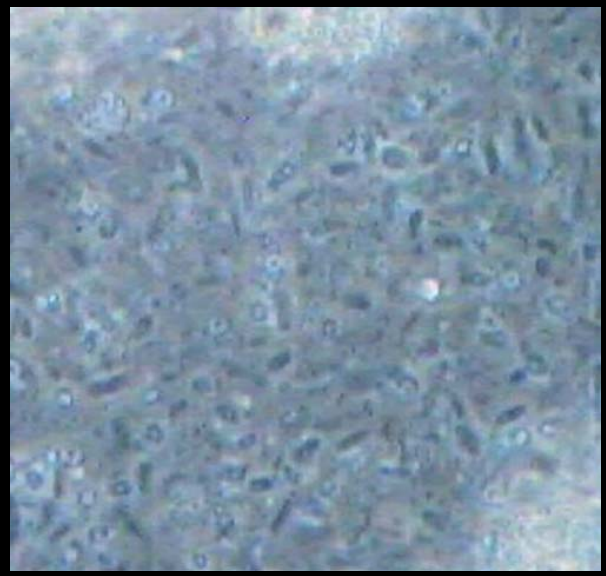
Cultures of *Steinernema carpocapsae* and *Xenorhabdus nematophila*



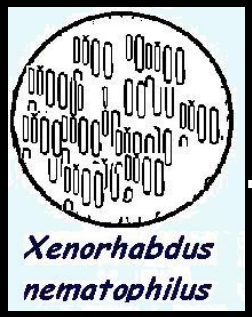


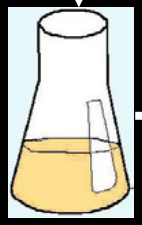


5 %
(v/v)



Cultures of *Steinernema carpocapsae* and *Xenorhabdus nematophila*

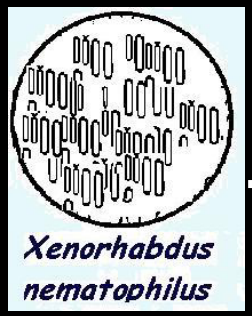


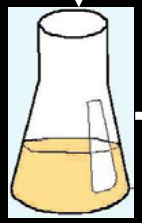


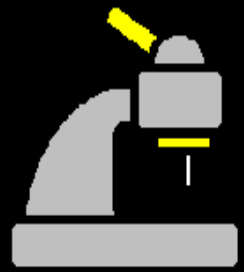
IJ stages in sterilised distilled water, 4°C

C_0=1,000 IJ/mL, approximately

Cultures of *Steinernema carpocapsae* and *Xenorhabdus nematophila*





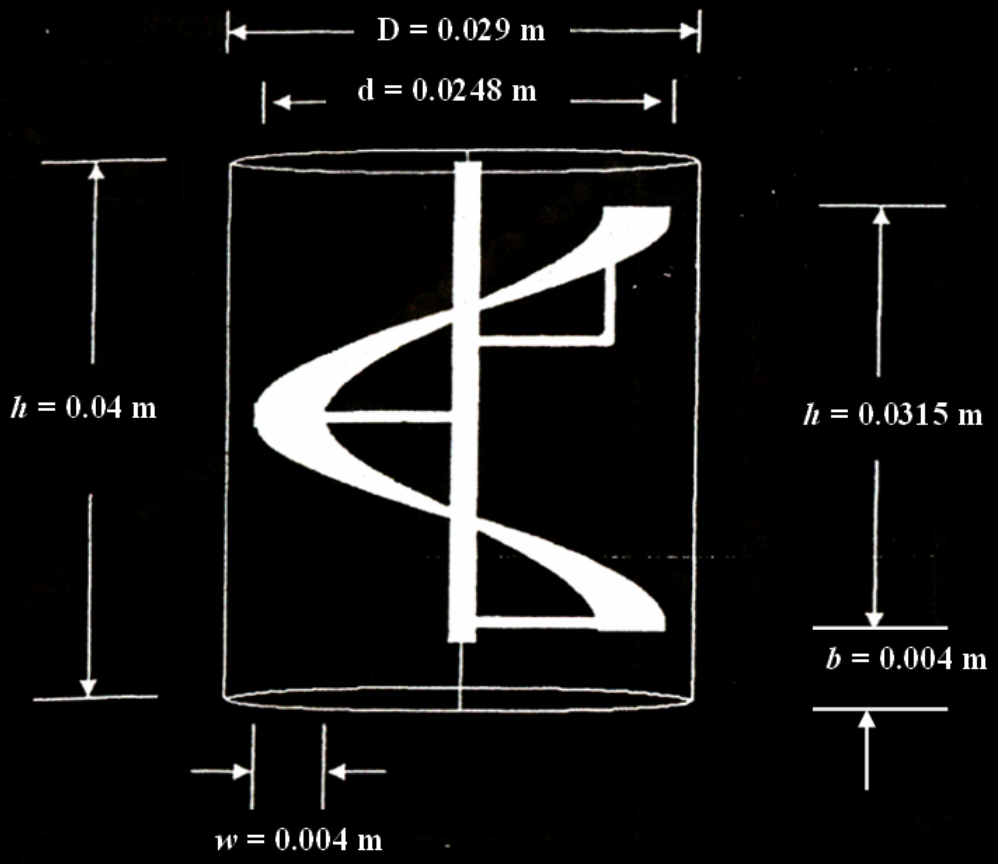


- Nematode concentration (viable count, C; and *X*)
- Process viscosity, η (mixing viscometry)

Q variable, **22°C**, 24 d

Taking samples every 2 - 4 d

Process viscosity, η (mixing viscometry)



$$\log \Gamma = A_n + n \cdot \log N$$

$$\mathbf{Kp} = (162.5454) \cdot \left(24.64^{(n-1)}\right) \cdot \left(0.91^{\left[\frac{n-1}{n}\right]}\right)$$

$$K = \frac{2 \cdot \pi \cdot A_n}{Kp \cdot (0.0248^3)}$$

$$\eta = K \dot{\gamma}_a^{(n-1)}$$

Hydrodynamics analysis. Culture broth rheological properties were evaluated through the Ostwald-de Waele model - $\tau = K (\gamma)^{n}$ -.

Riser section: $\gamma = 5000\ v_{sg}$ (Nishikawa et al 1977) with superficial gas velocity (v_{sg}=Q/A).

Downcomer section:

$$\gamma_w = 3.36(1 - U_{GR})^{-32.56} \left(1 + \frac{A_D}{A_R}\right)^{0.89} (H_D)^{0.44}$$

(Al-Masry et al 2005)

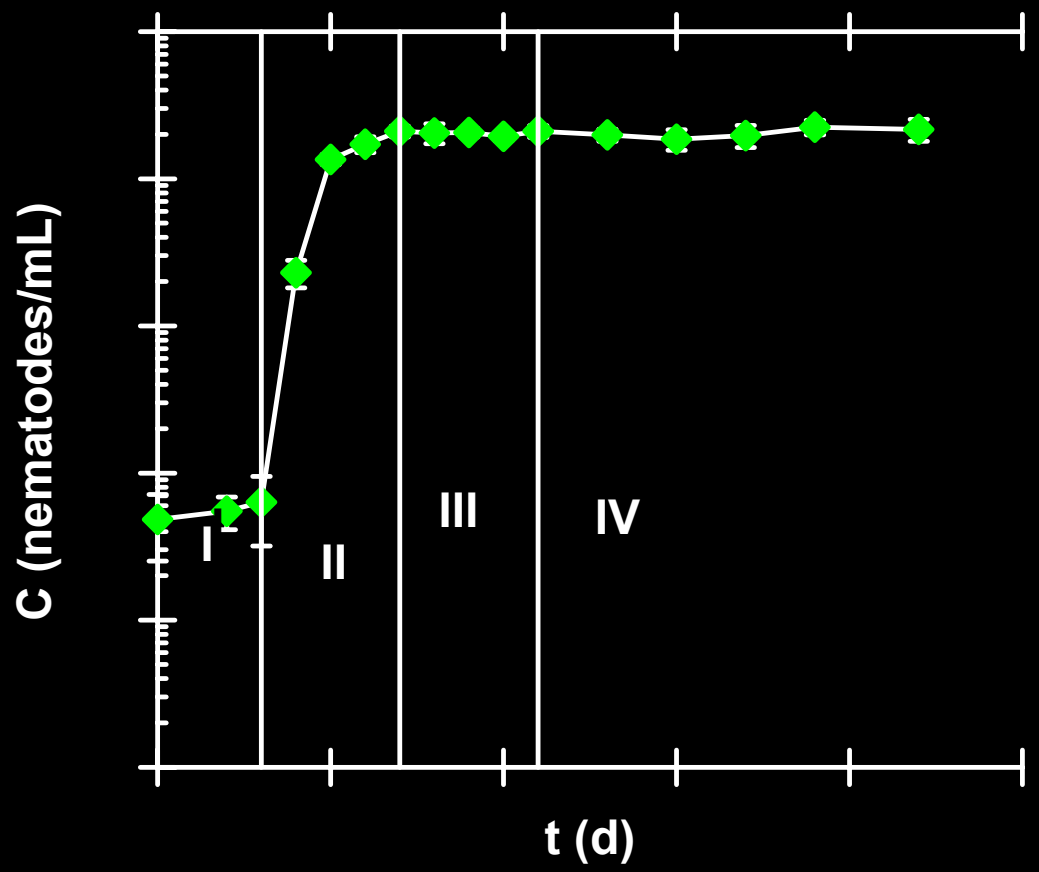
Actual flow regime was calculated using (considering the gas expansion):

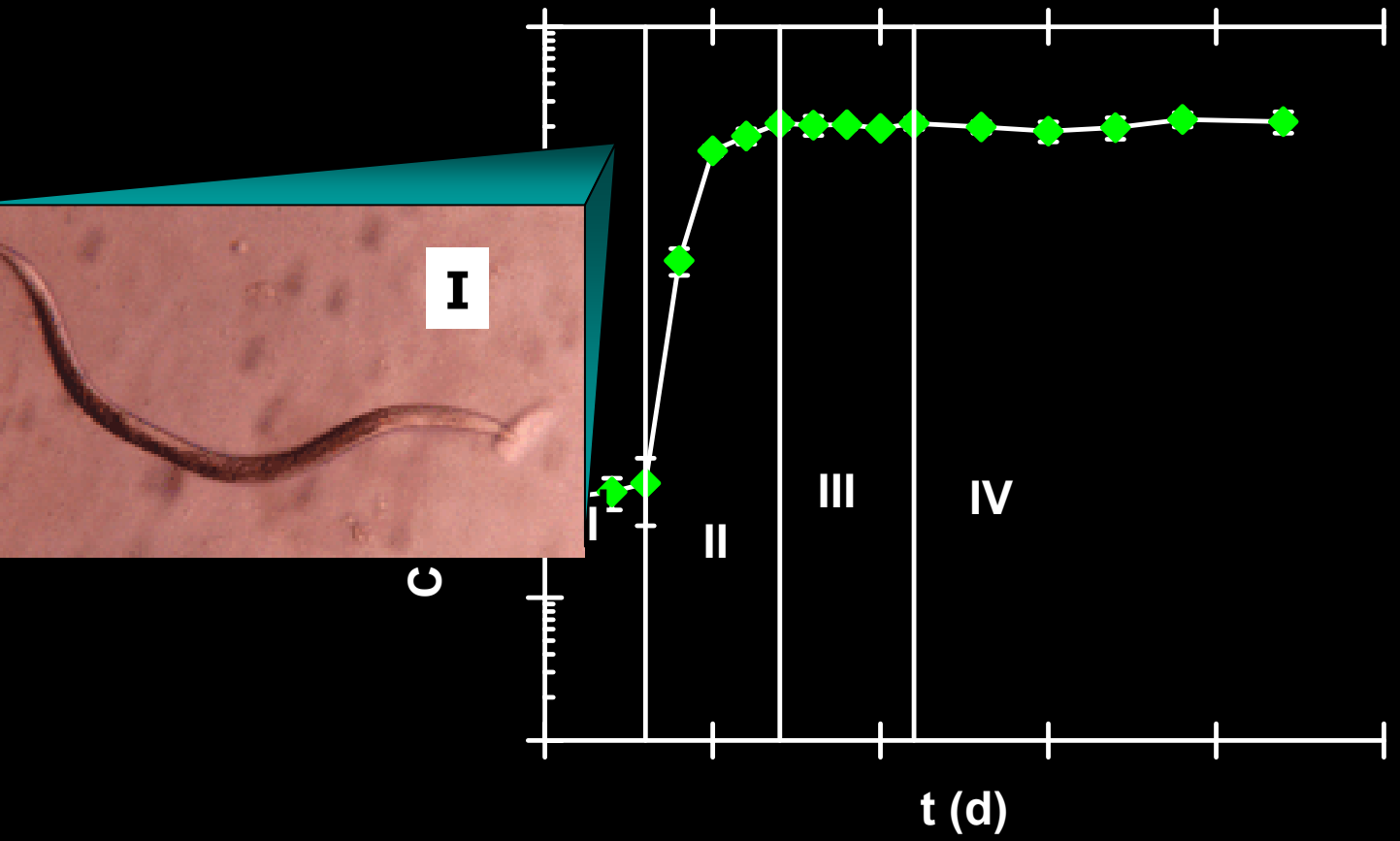
$Re=(\rho D_H v)/\eta$

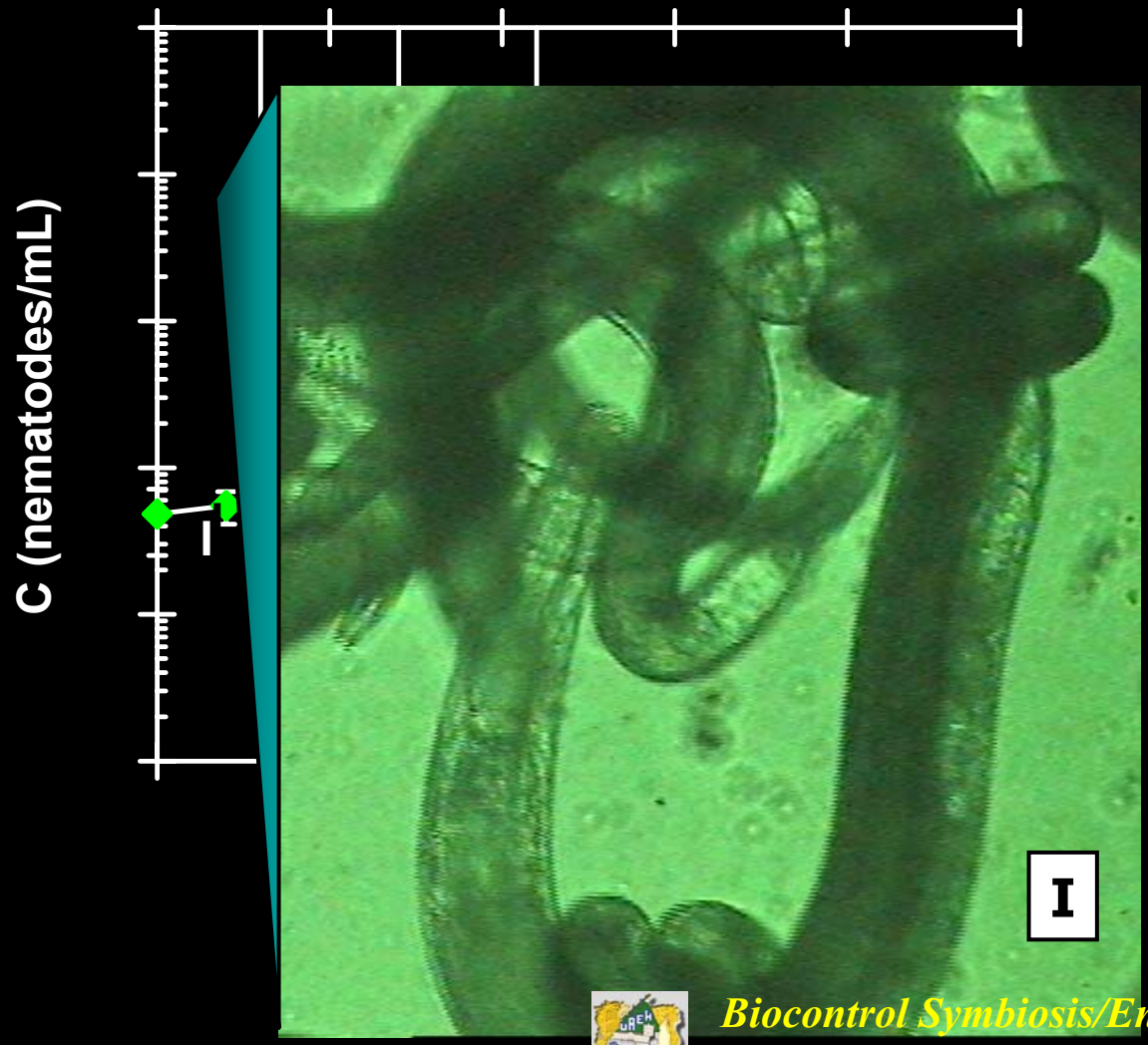
The gas-liquid mass transfer efficiency was calculated using:

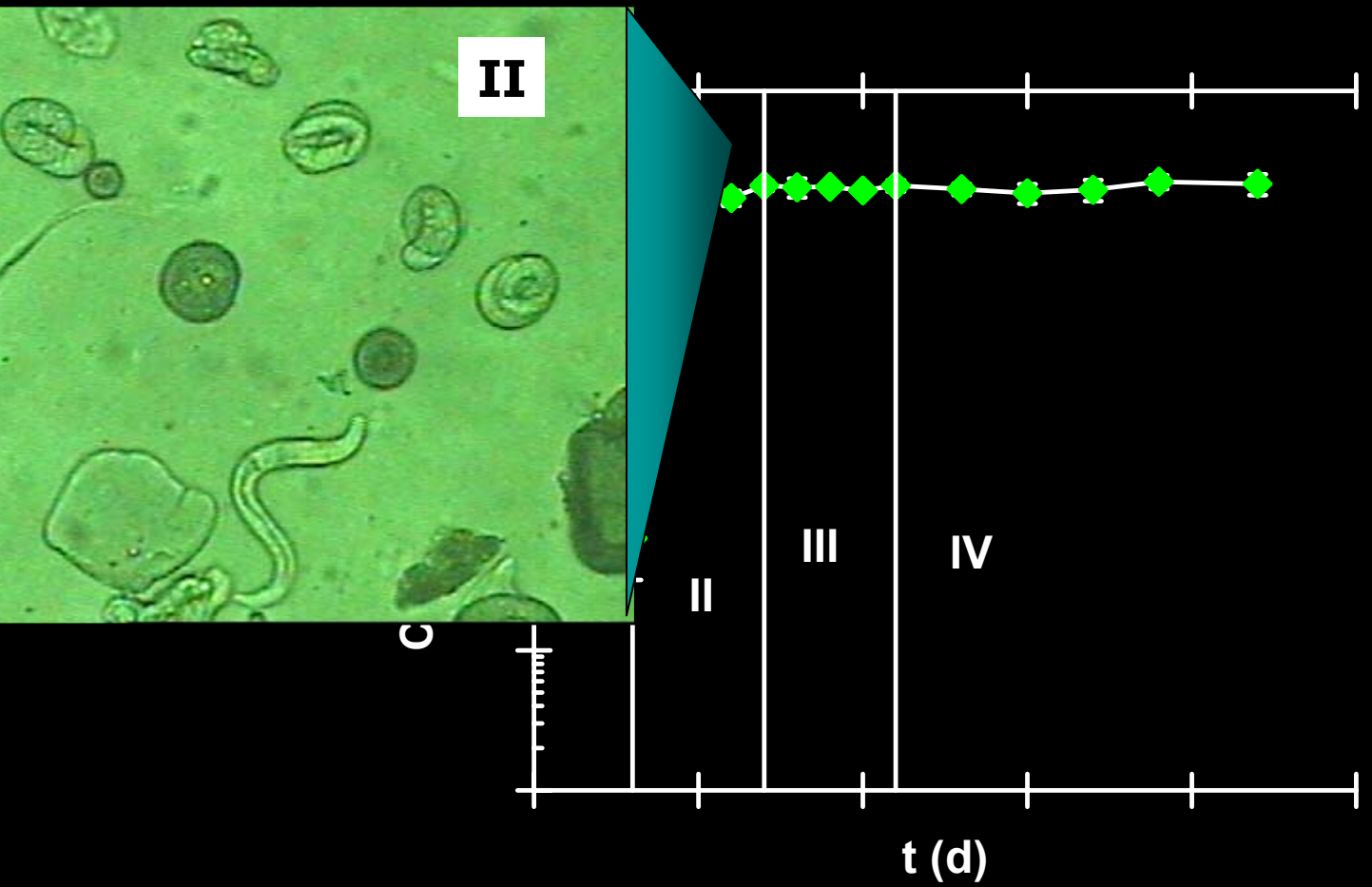
$$\frac{k_L a D}{V_{sg}} = 2.95\times10^{-3}\left(\frac{DV_{sg}\delta}{\eta_a}\right)^{0.37}\left(\frac{V_{sg}^2 D\delta}{\sigma}\right)^{-0.13}\left(\frac{V_{sg}}{\sqrt{gD}}\right)^{-0.31}\left(\frac{H}{D}\right)^{-0.50}$$

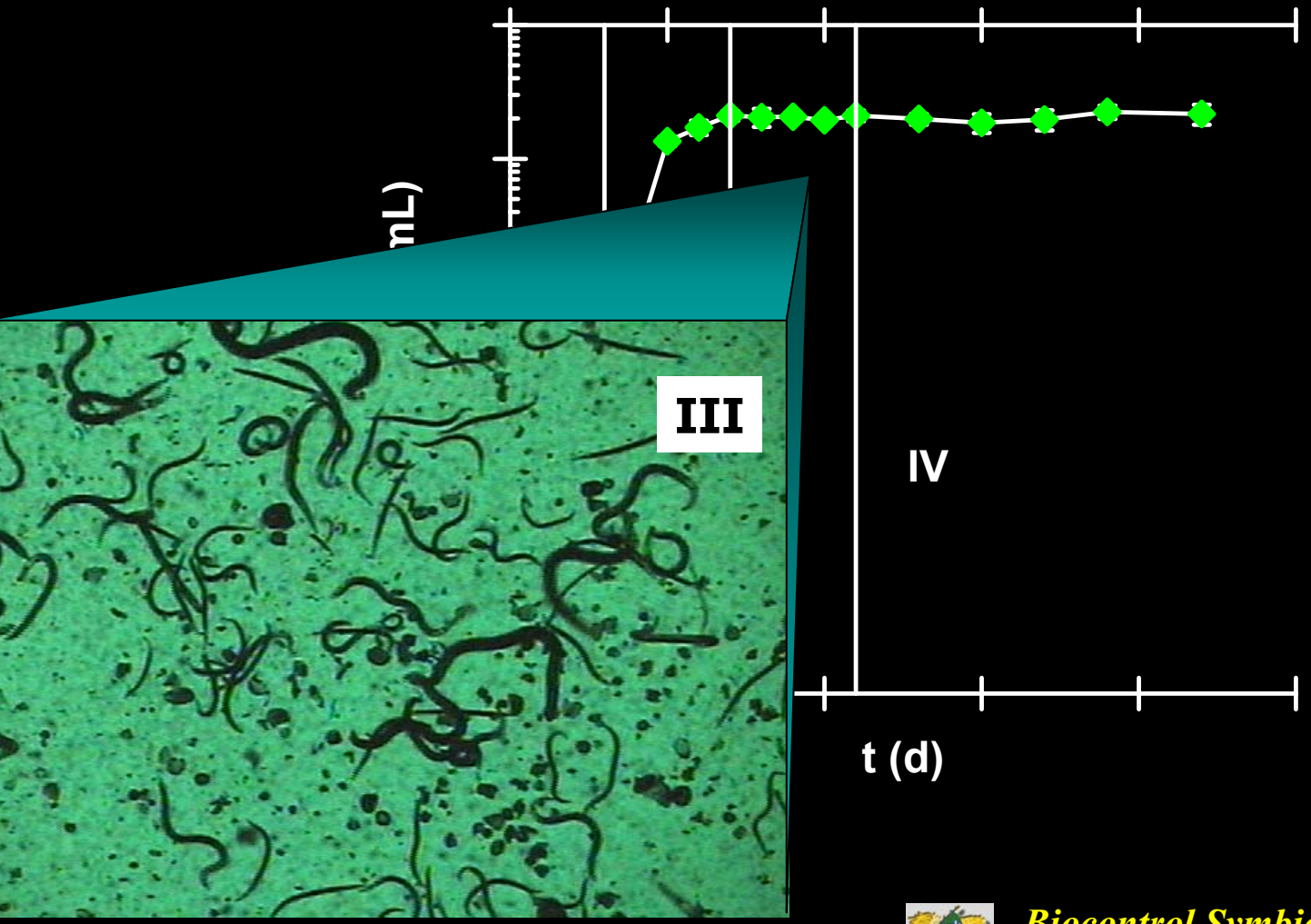
(Zhao et al 1994)

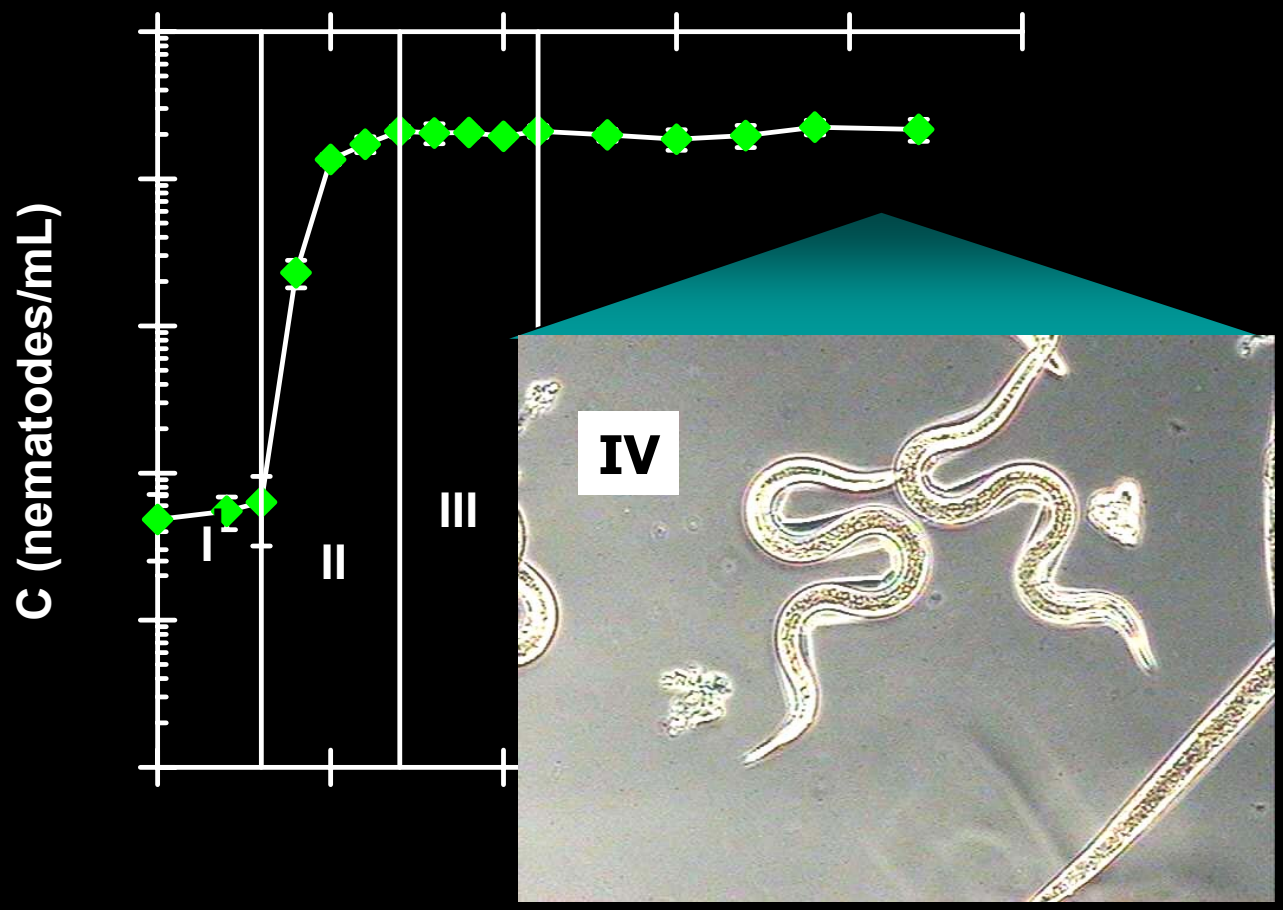

C (nematodes/mL)
t (d)
I
II
III
IV

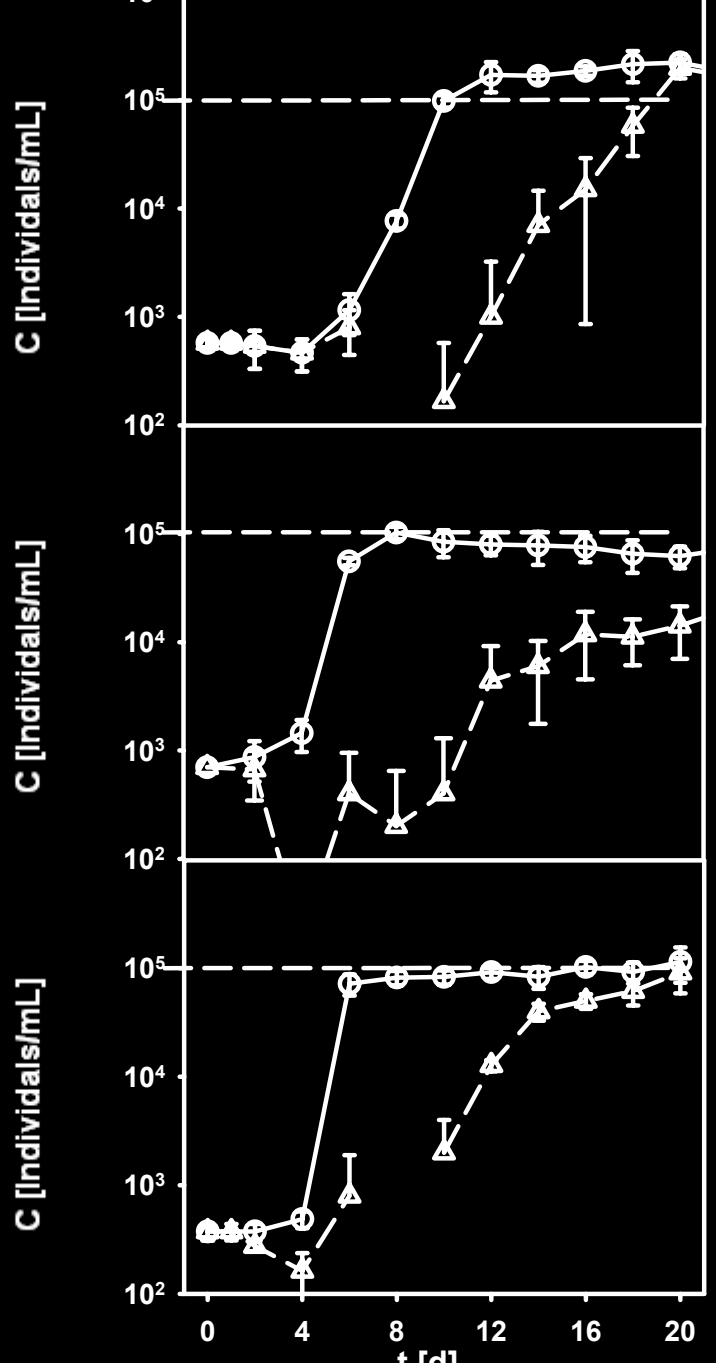

I
II
III
IV
t (d)


C (nematodes/mL)
I


II
II
III
IV
t (d)


III
IV
t (d)


C (nematodes/mL)
I
II
III
IV



RESULTS

SDT-MP1

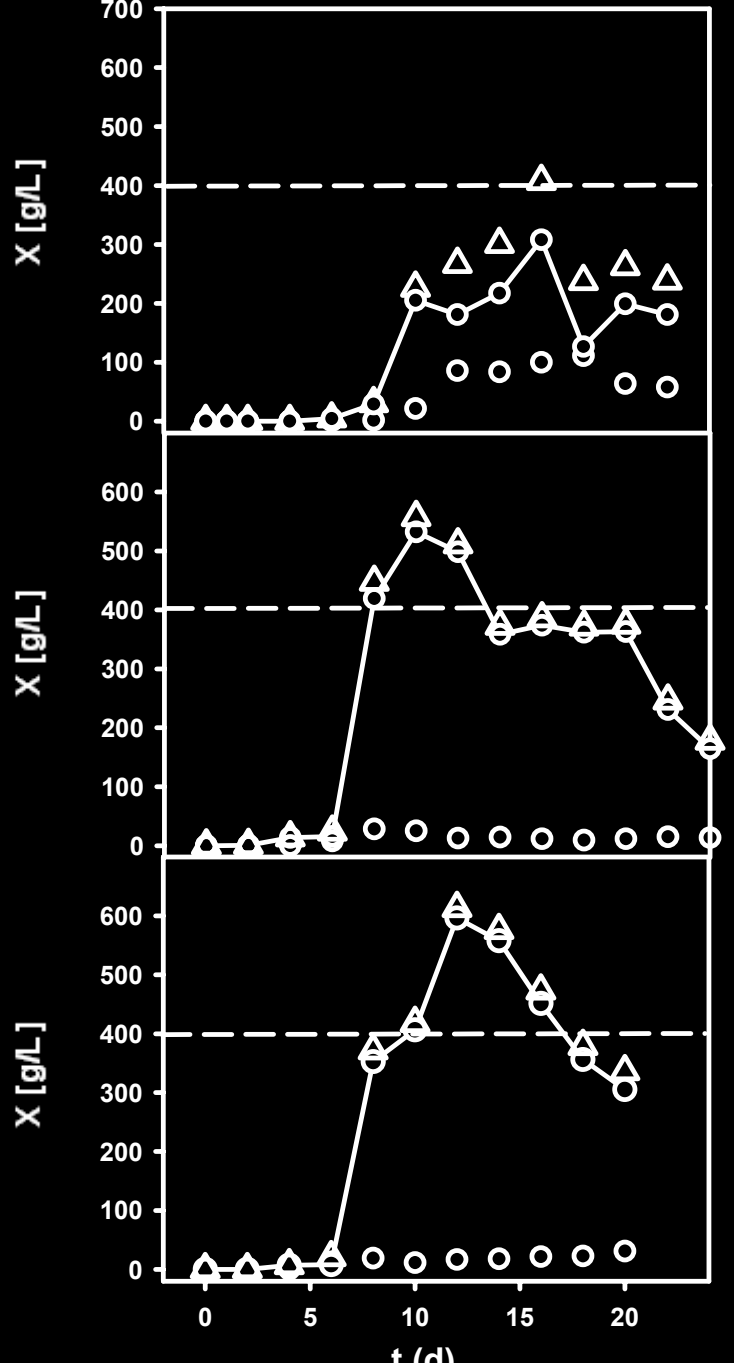
SM-MP1

SM-MP2

RESULTS

Fermentation	Maximum nematode concentration (individuals/mL) $(\sigma)^2$	% of IJ stages	Maximum IJ concentration [individuals/mL]	Maximum nematode biomass concentration (X, [g/L])
SDT-MP1	222,000 (41,923)	87.12	193,406	407.7 (t = 16 d)
SM-MP1	62,200 (14,131)	22.83	14,200	557.2 (t = 10 d)
SM-MP2	114,600 (41,058)	79.58	91,199	613.1 (t = 12 d)

RESULTS


X [g/L]
700
600
500
400
300
200
100
0
SDT-MP1
X [g/L]
600
500
400
300
200
100
0
SM-MP1
X [g/L]
600
500
400
300
200
100
0
SM-MP2
0
5
10
15
20
t (d)

RESULTS

Fermentation	Maximum nematode concentration (individuals/mL) $(\sigma)^2$	% of IJ stages	Maximum IJ concentration [individuals/mL]	Maximum nematode biomass concentration (X, [g/L])
SDT-MP1	222,000 (41,923)	87.12	193,406	407.7 (t = 16 d)
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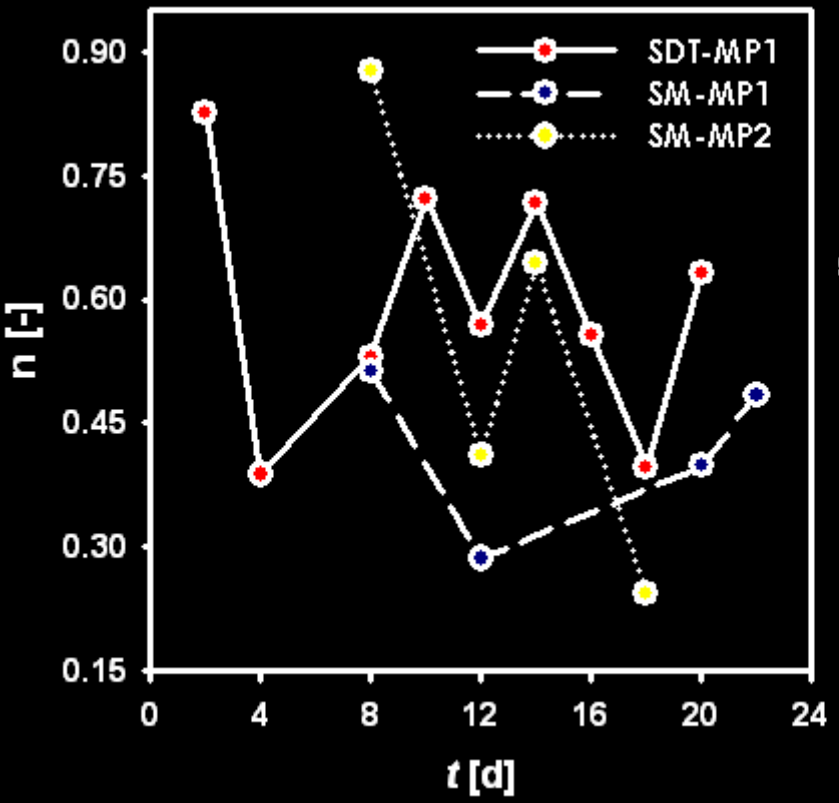
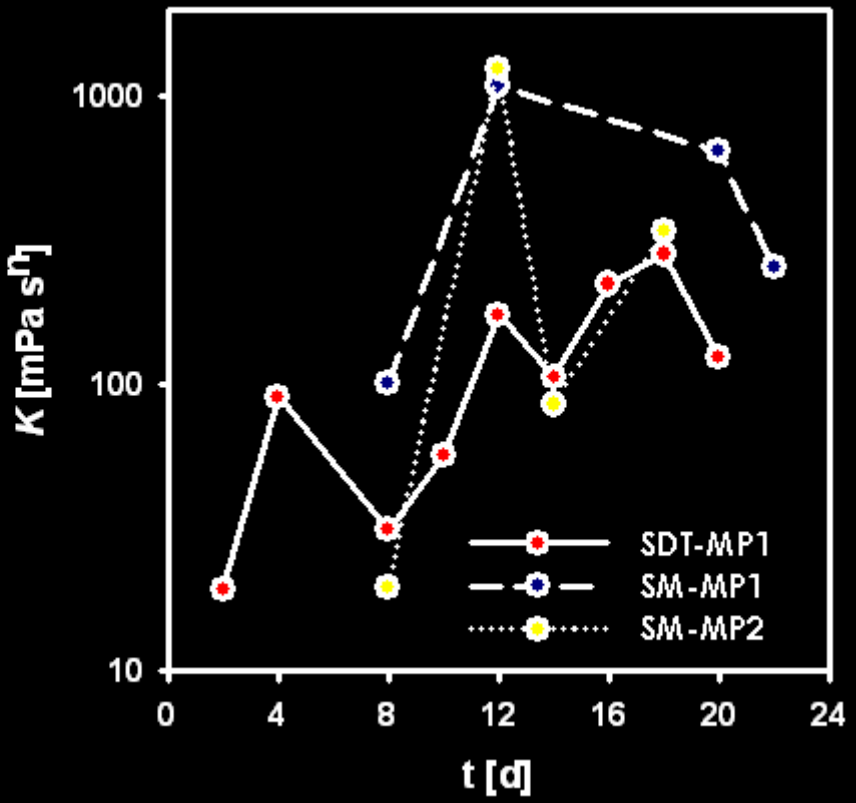
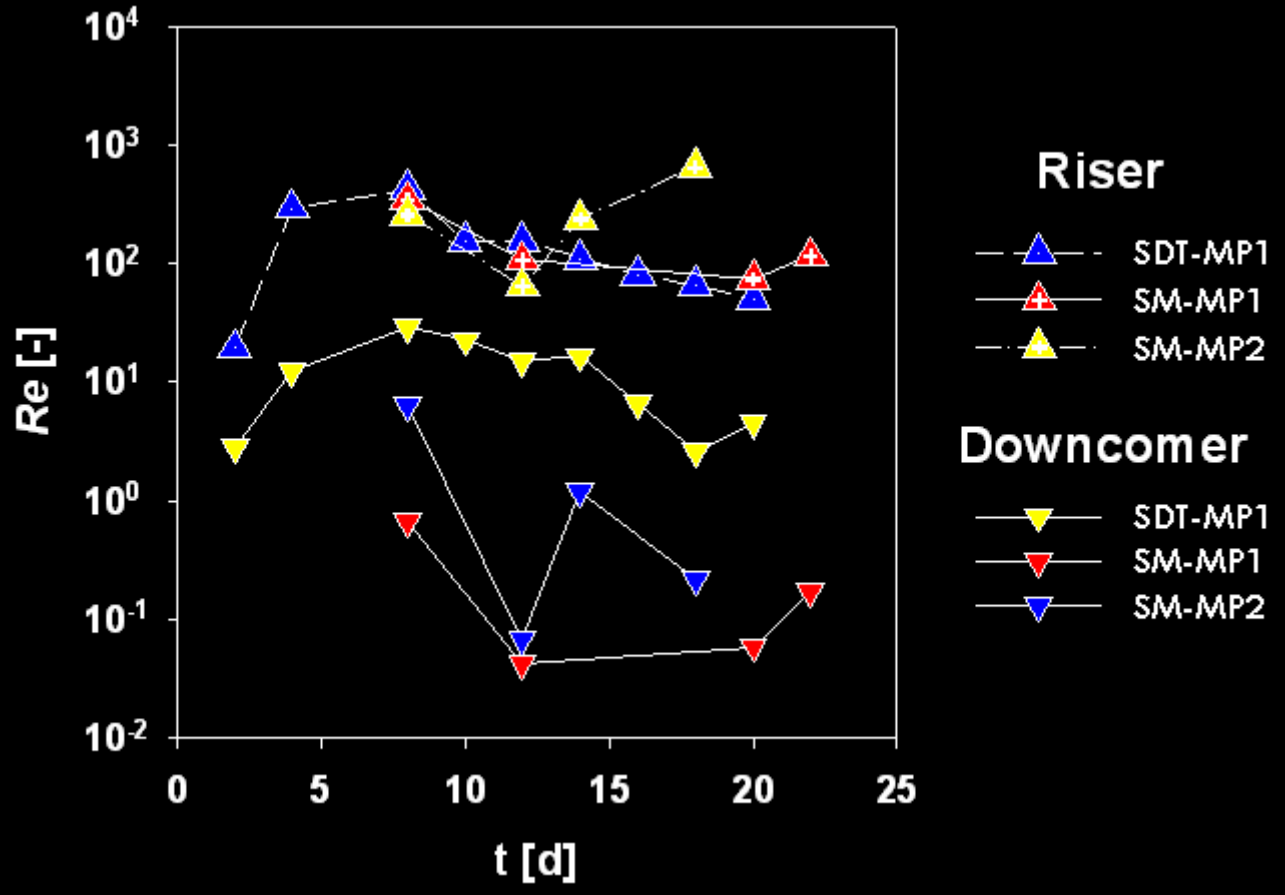
Rheological properties evolution

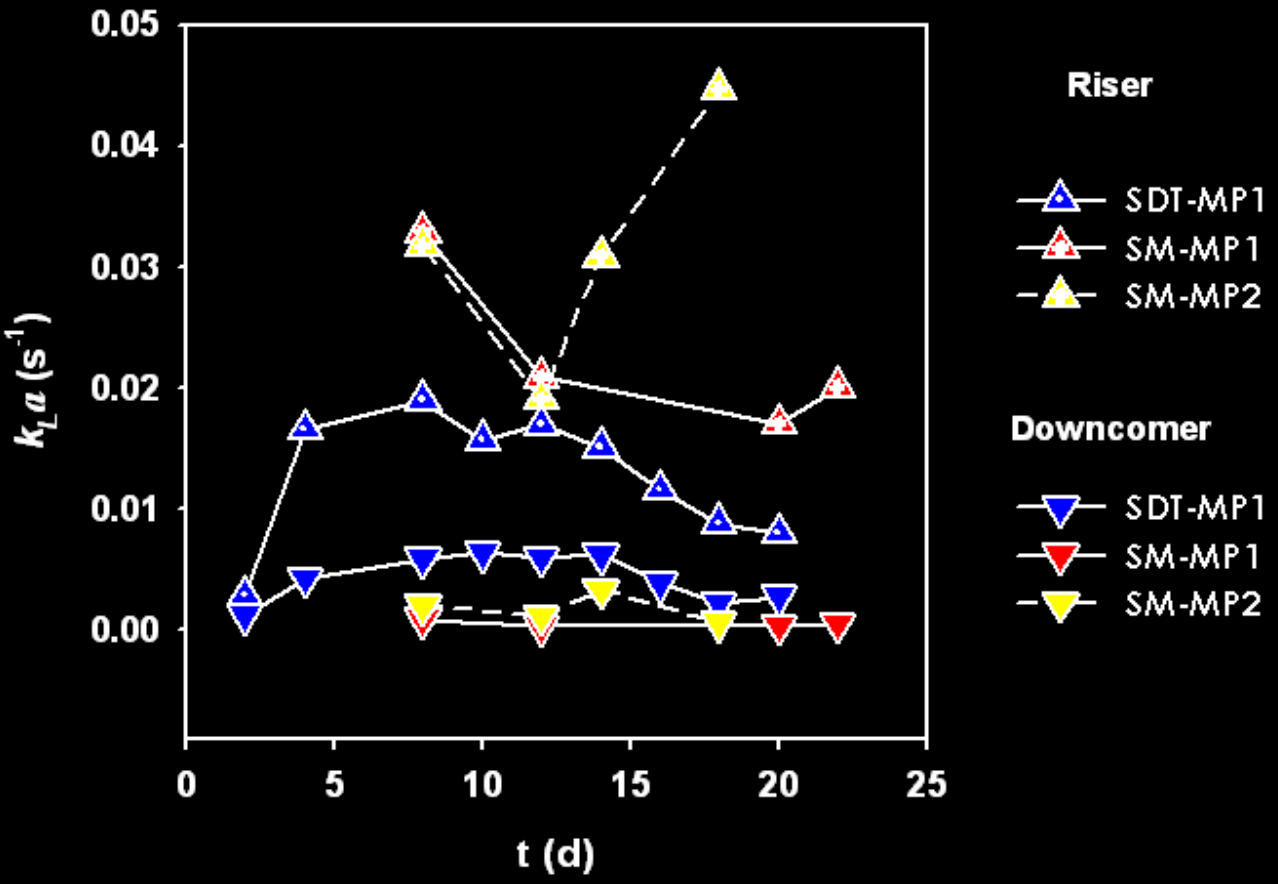




Hydrodynamics evolution



Evolution of the gas-liquid mass transfer efficiency



Conclusions

-High nematode concentrations of *Steinernema carpocapsae* were achieved using an internal-loop airlift bioreactor (up to 190,000 IJ/mL; MP1 medium).

-Using the estimated rheological properties, it is first presented the evolution of both the hydrodynamics and the gas-liquid mass transfer conditions along the fermentations (i.e., Re & Sh).

-SDT geometry rendered the best IJ productivities. Nonetheless, SM geometry appears to provide better conditions for the nematode population development.

-Finally, medium MP2 –that contains lower nutriments concentrations than MP1- rendered higher IJ productivities.

Acknowledgements

• Team: René, Gabriela, Marco, Uriel, Eduardo.

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•Grant 200201-9206, FOMIX-CONACYT-Gob. Hgo.

Biocontrol Symbiosis/Entomopathogen
Nematodes and Symbiotic Bacter