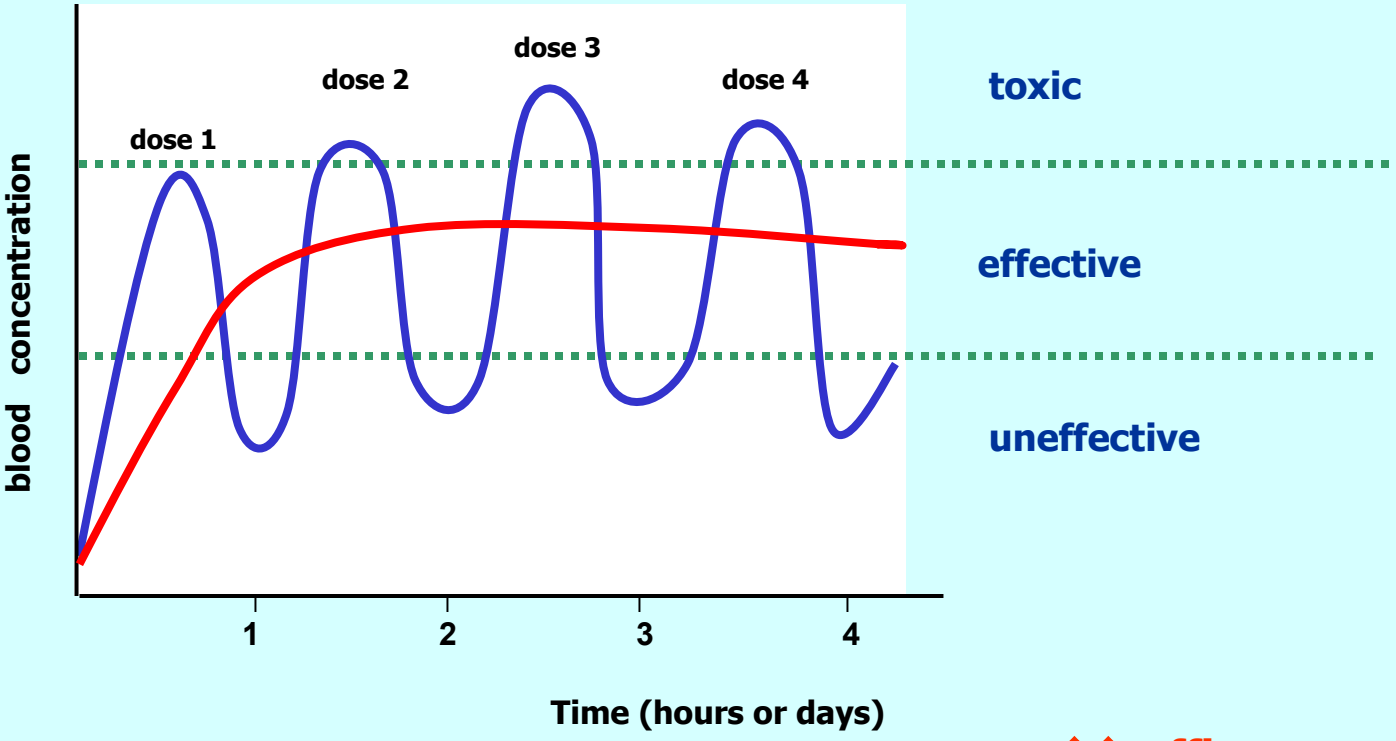
FEDERAL UNIVERSITY OF RIO DE JANEIRO - UFRJ
Institute of Biophysics
Laboratory of Immunopharmacology

NANOPARTICLES FOR DRUG AND VACCINE DELIVERY :

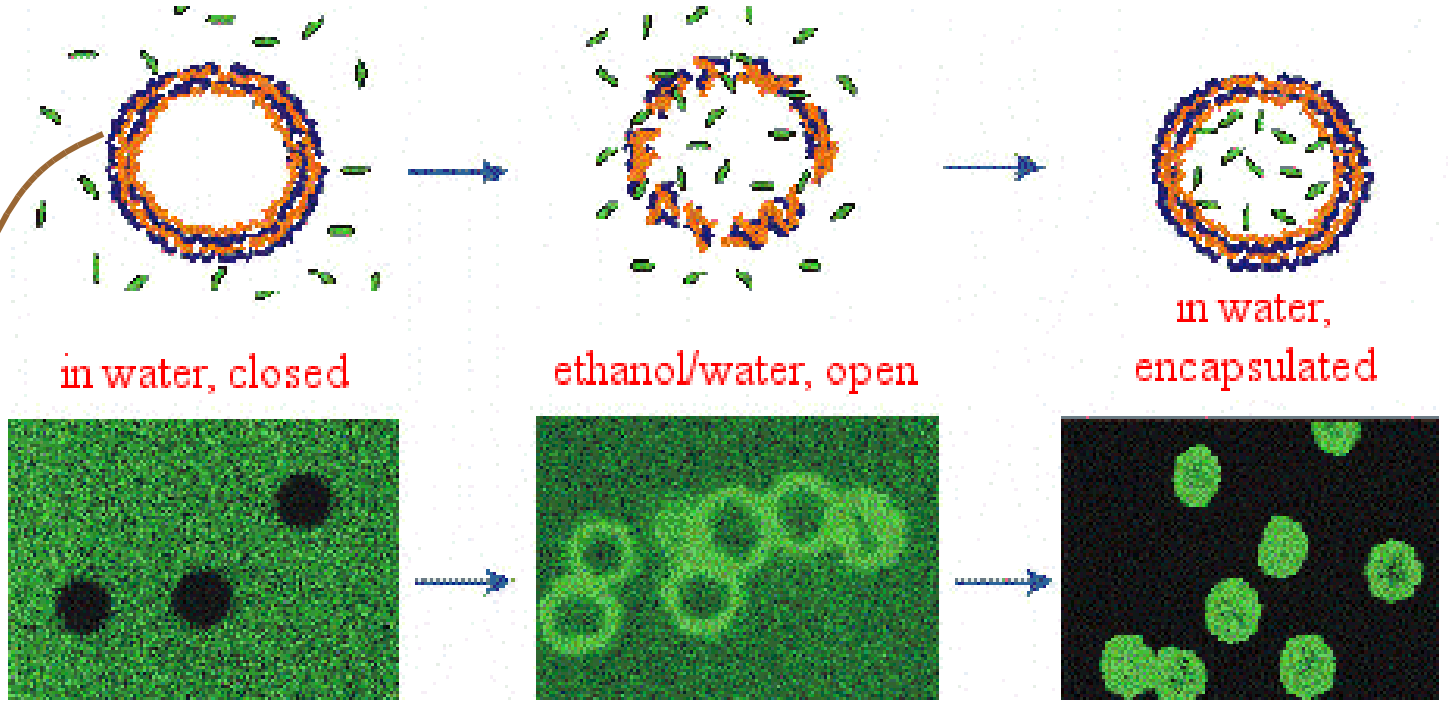
Leishmaniasis as a disease model

Bartira Rossi Bergmann

CONTROLLED RELEASE



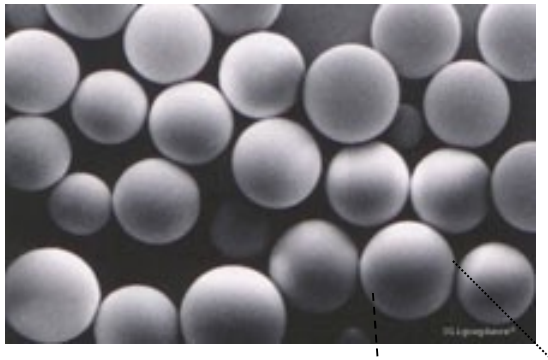
↑↑ efficacy
↓↓ toxicity
↓↓ doses



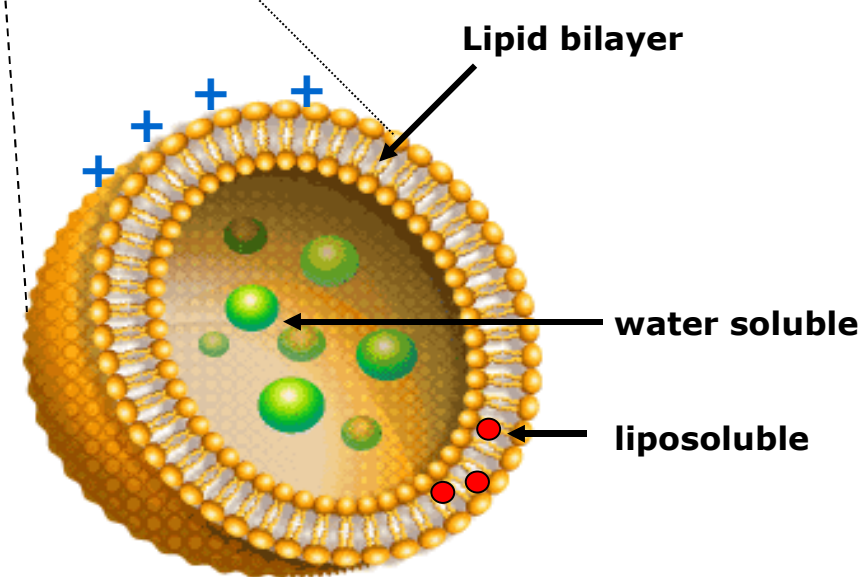
BIODEGRADABLE AND BIOCOMPATIBLE

- **LIPOSOMES**
- **POLYMERS (PLGA, chitosan,…)**
- **CYCLODEXTRINS**
- **DENDRIMERS**

LIPOSOMES

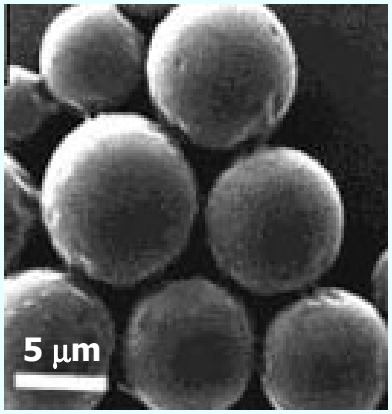


50 nm

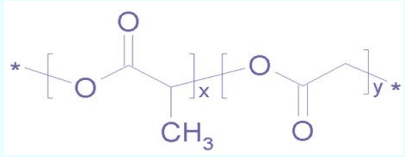


Totally biocompatible
(phosphatidylcholine expensive)

POLYMERIC PARTICLES



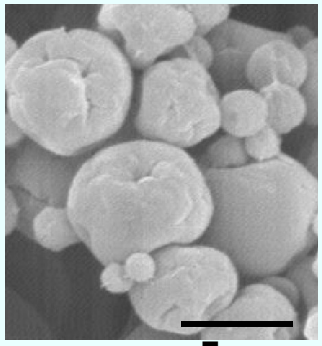
Poly (lactide-co-glicolide) = PLGA



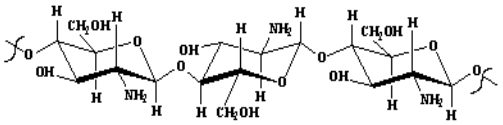
133 days in water

US$ 3000 / Kg

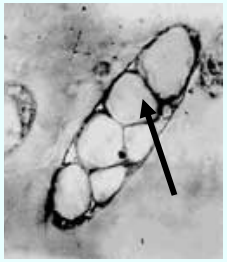
chitin



Chitosan
(biopolymer)



saccharosis
+
Alcalígenes_s.p



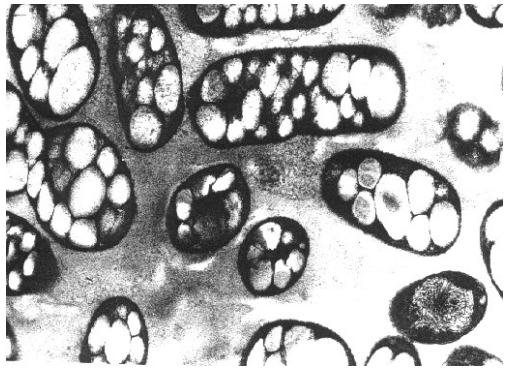
Polyhydroxyalkanoates
(biopolymer
100% Brazilian)

Pharmaceutically
suitable ??

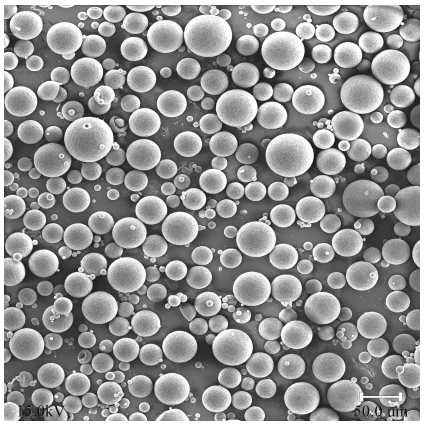
U$ 5 / Kg

Biosynthetic Polyhydroxyalkanoates and their Potential in Drug delivery

bacterial storage polyesters accumulated as carbon and energy reserve granules.

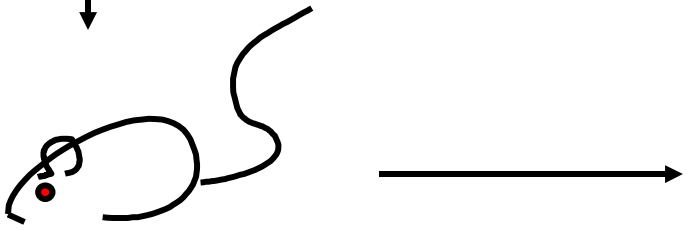


PHA - Biopolymers (polyhydroxyalkanoates)



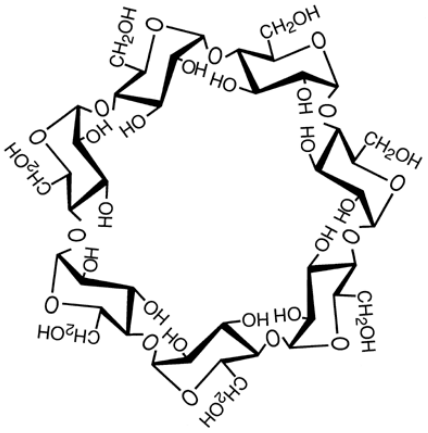
PHA micro and nanospheres prepared from an emulsion-evaporation solvent process

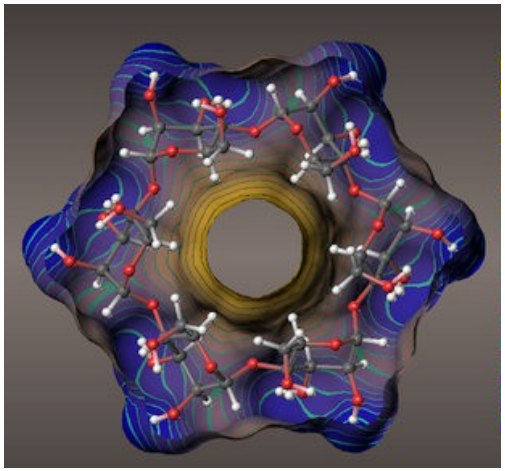
1 mg (s.c)
or
5 mg (i.p.)

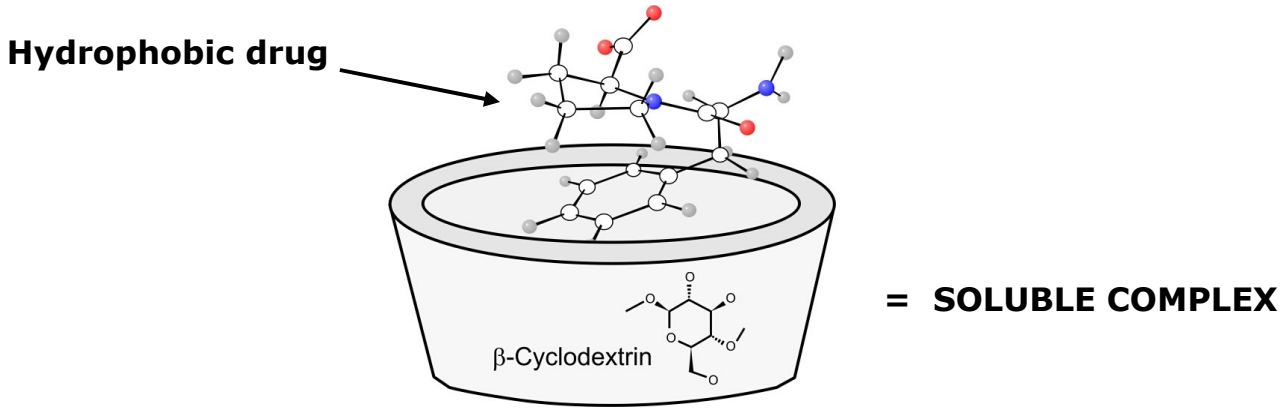


- **No detectable allergic reaction**
- **No detectabe inflammatory reaction**

BETA-CYCLODEXTRIN

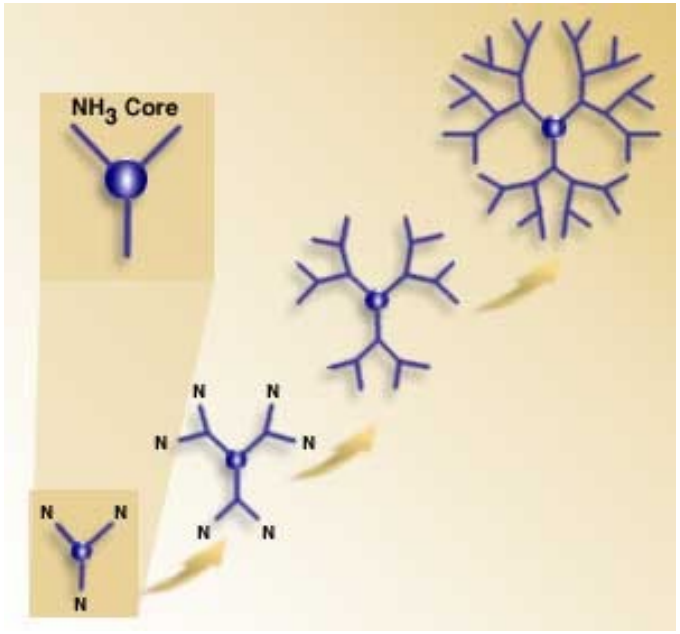


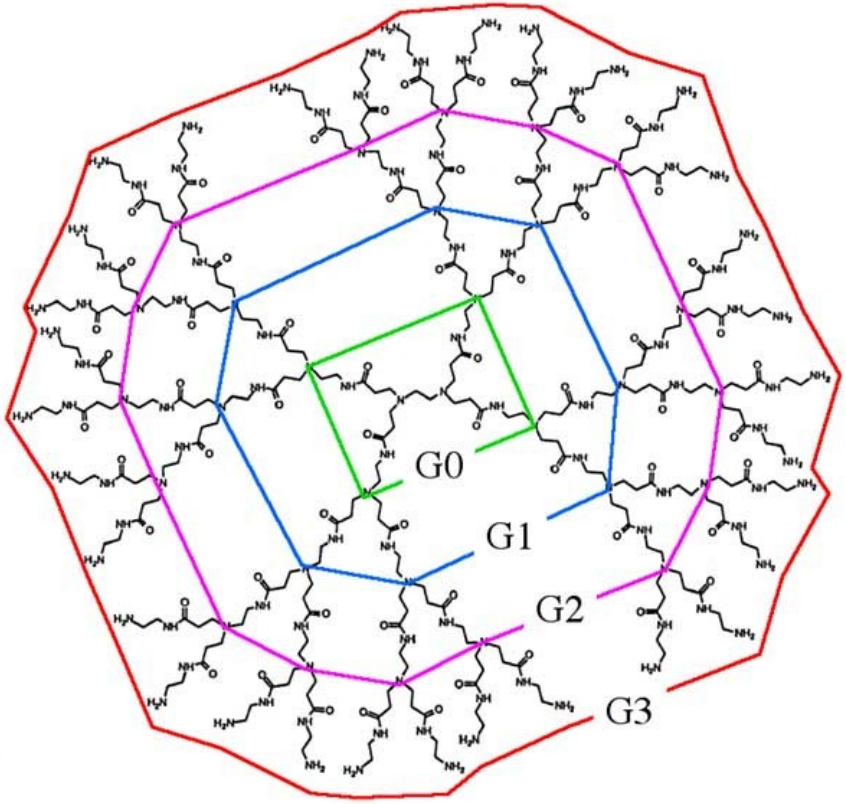




= SOLUBLE COMPLEX

DENDRIMERS

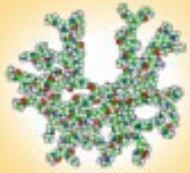




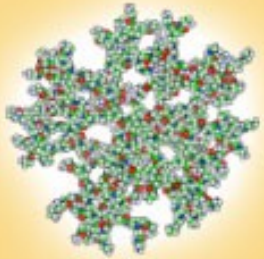
PAMAM: POLY (AMIDOAMINE)

Dendrimer Size Comparison

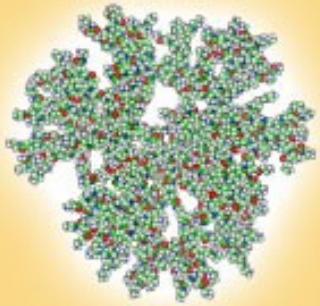
G3 Dendrimer



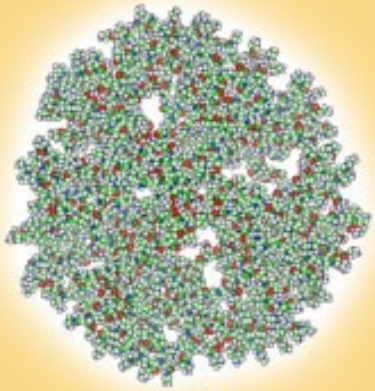
G4 Dendrimer



G5 Dendrimer



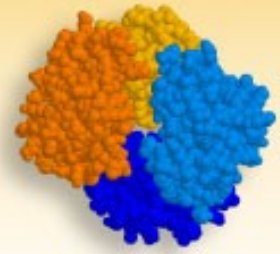
G6 Dendrimer



G7 Dendrimer

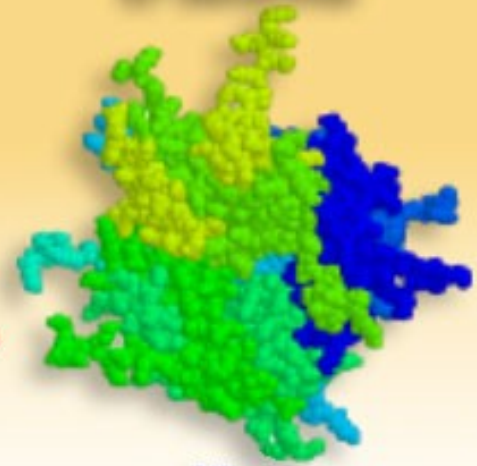
Insulin

Cytochrome C



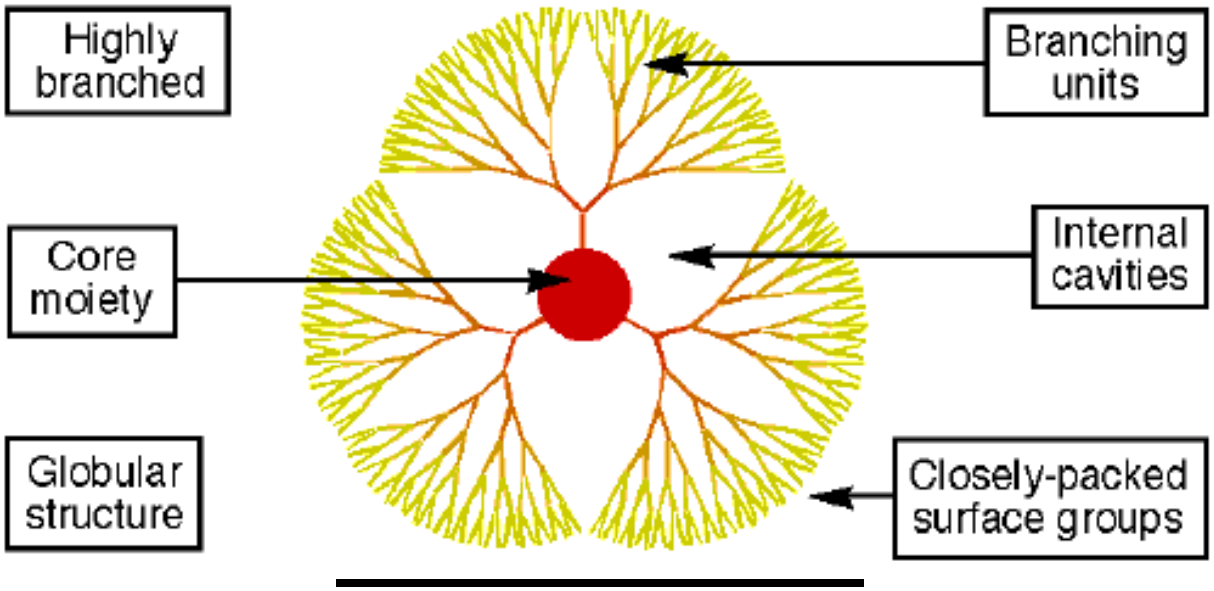
Hemoglobin

Transthyretin



Histone

The Dendritic Structure

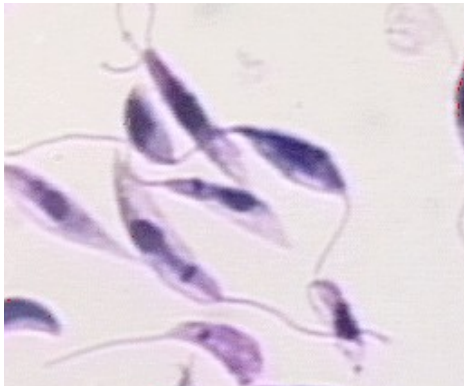


Delivery of:

- **Nucleic acids**
- **Encapsulated drugs**
- **Covalently linked drugs**

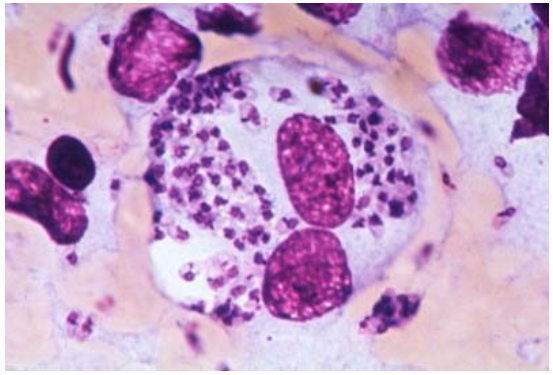
NANOPARTiCLES FOR THE TREATMENT AND VACCINATION OF LEISHMANIASIS

LEISHMANIA LIFE CYCLE



FLAGELATE PARASITES

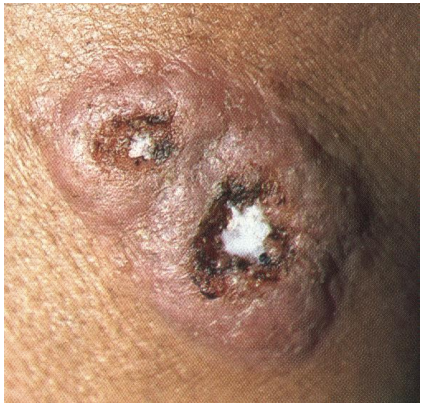
MAN



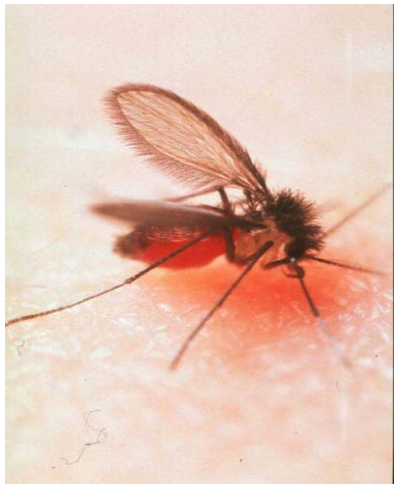
PARASITES
INSIDE
MACROPHAGES

VISCERAL
LEISHMANIASIS

LIVER, SPLEEN,
BONE MARROW

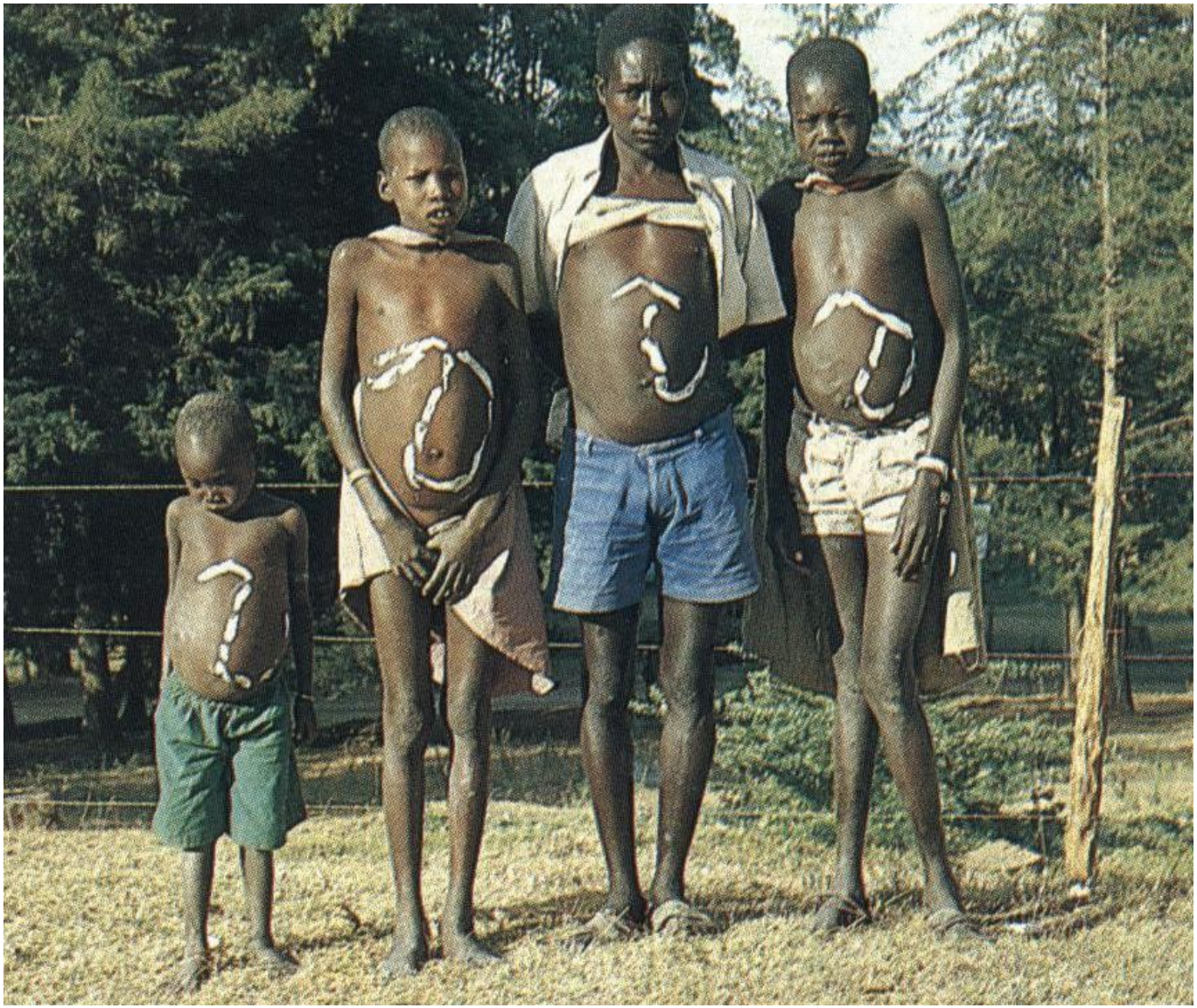


CUTANEOUS
LEISHMANIASIS



SANDFLY

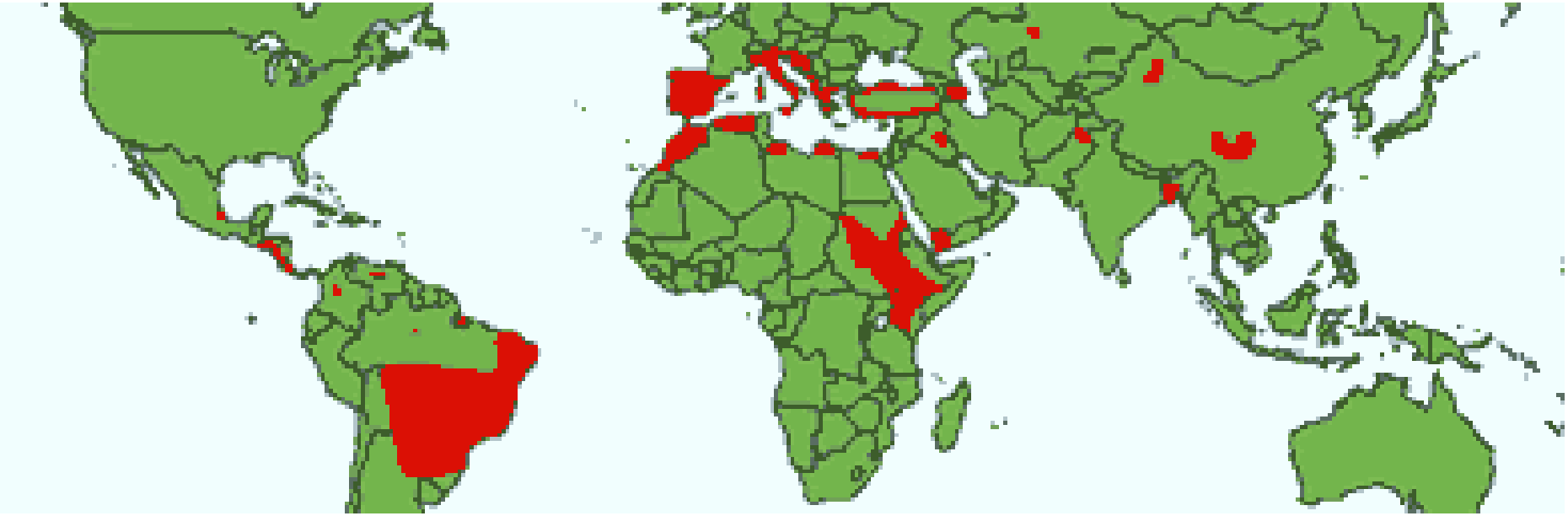
VISCERAL LEISHMANIASIS



Leishmania donovani

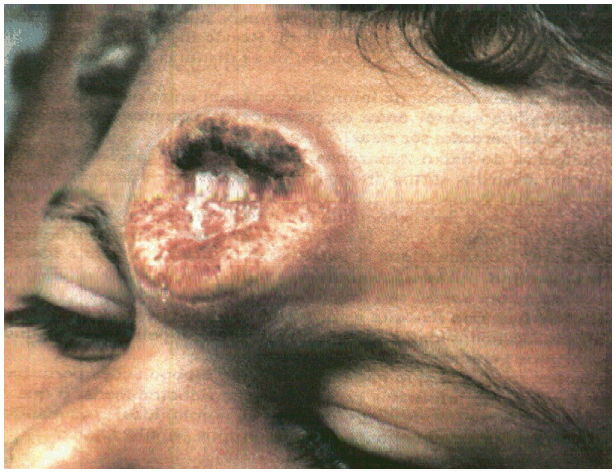
Visceral Leishmaniasis (Kala-Azar)

Europe = Spain, France and Italy

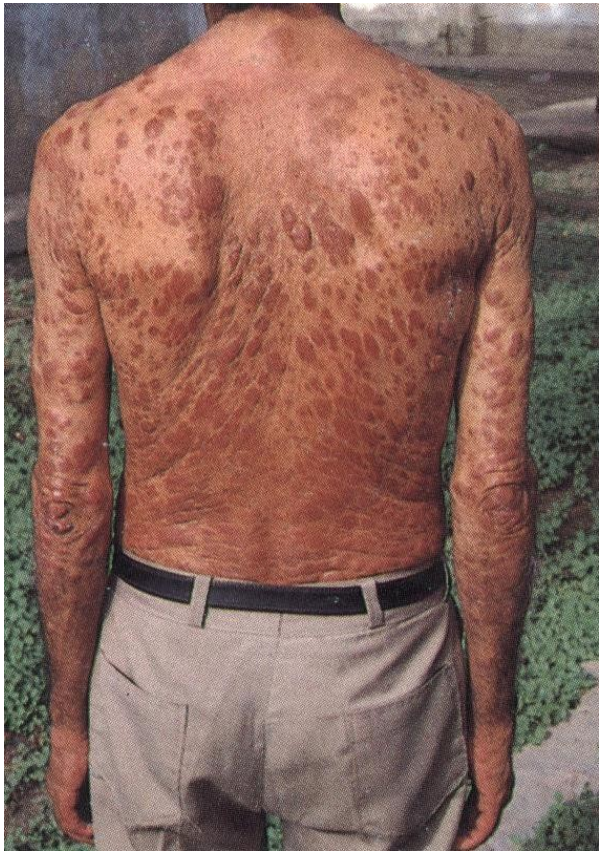


= Endemic areas

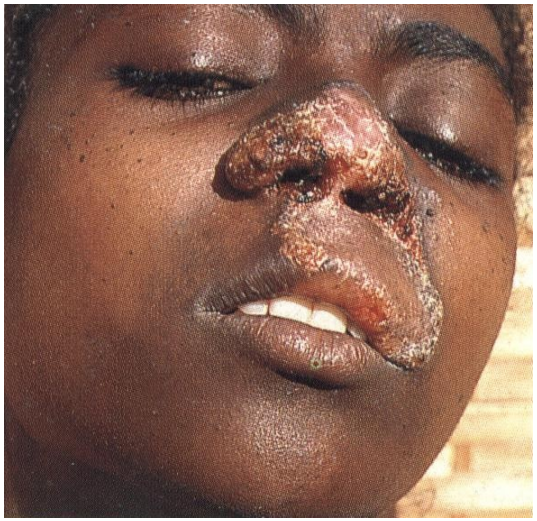
CUTANEOUS LEISHMANIASIS



***L. major* + outras**



L. amazonensis



L. braziliensis


Cutaneous Leishmaniasis
Highly Endemic Countries (90% of cases)
Highly Endemic Countries
WHO/CTD, 1996

CONVENTIONAL TREATMENT :

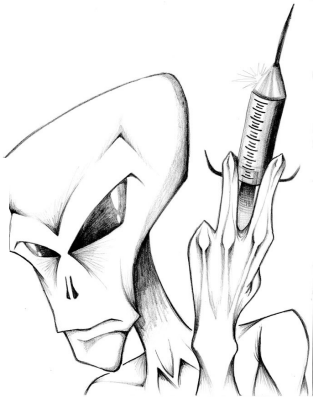
1 intramuscular injection a day / 20 days
with 5-10 ml pentavalent antimonial
(Glucantime or Pentostan)

↓ **if no cure**

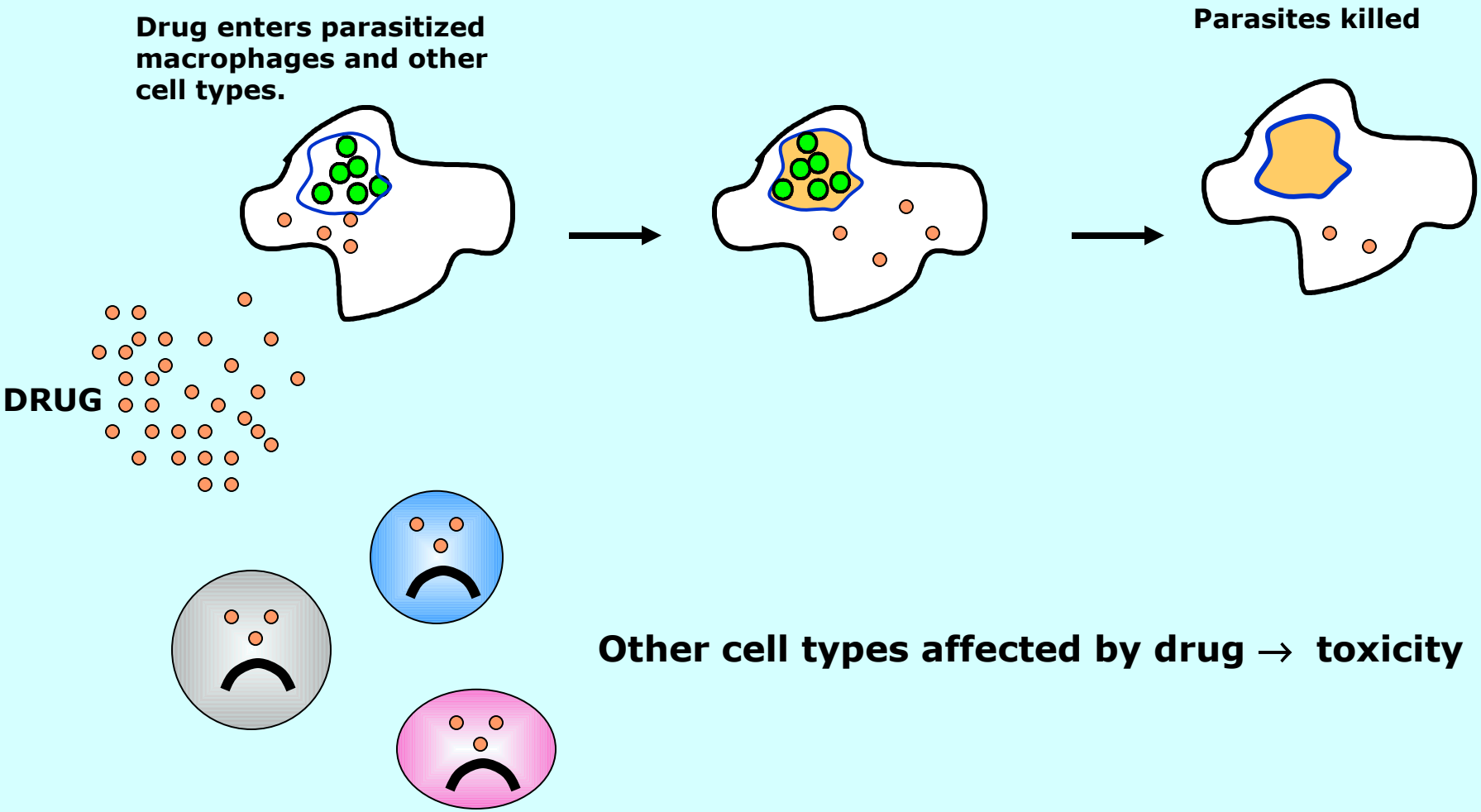
repeat (i.v.)

⇣

heart toxicity
muscular and bone pain

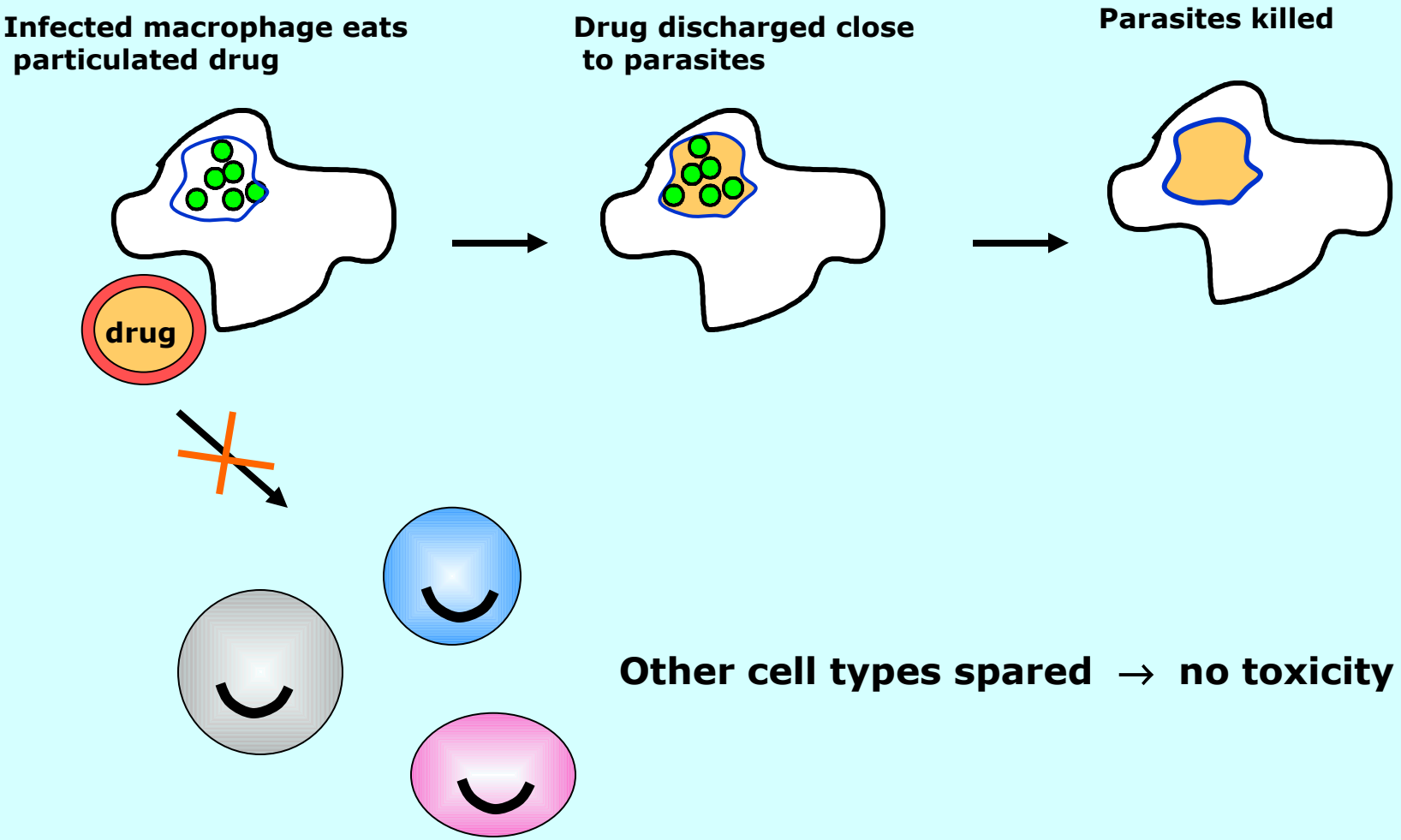


TREATMENT OF LEISHMANIASIS WITH SOLUBLE DRUGS



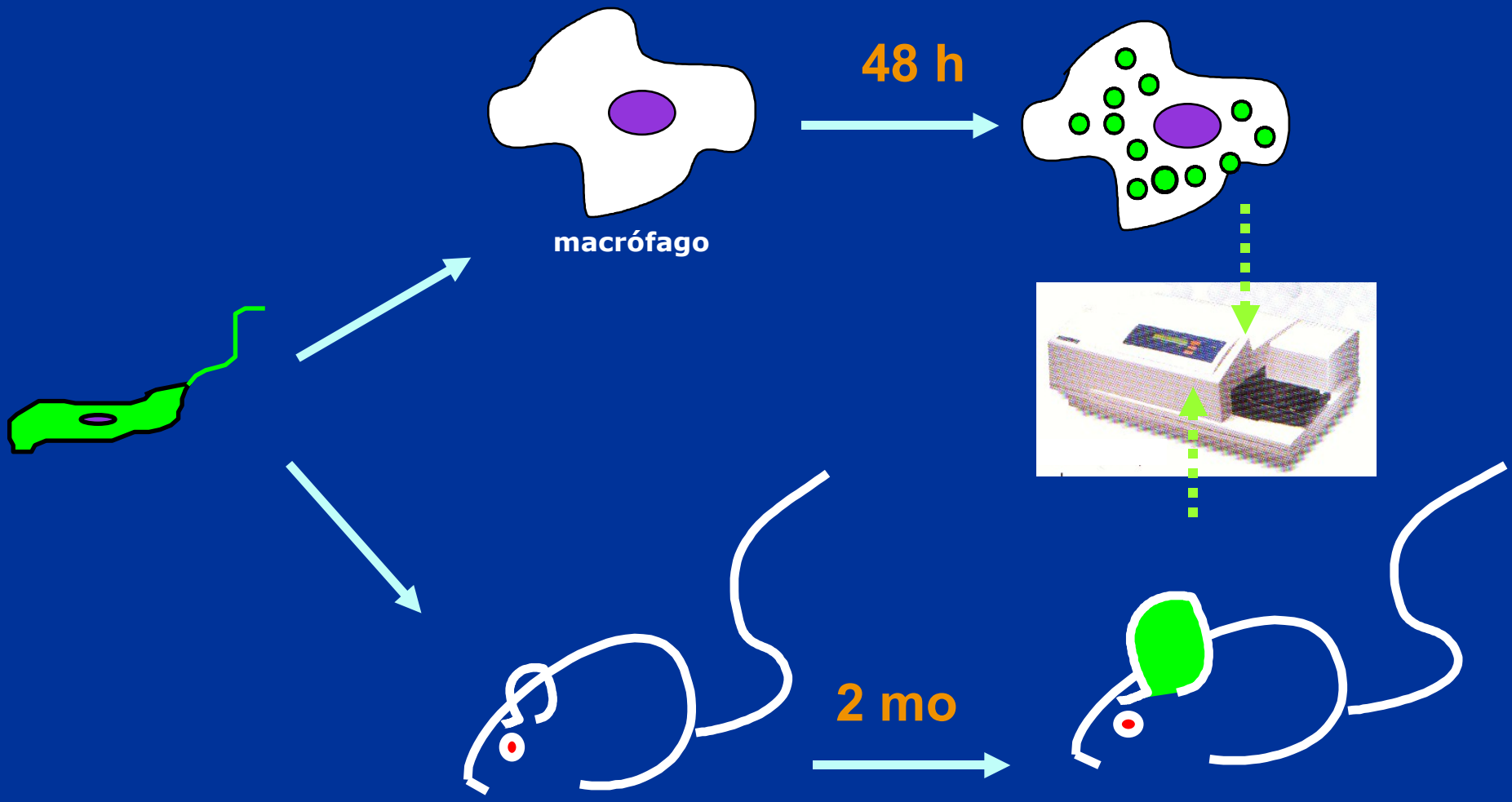
Other cell types affected by drug → toxicity

TREATMENT OF LEISHMANIASIS WITH PARTICULATED DRUGS
(leishmania only infects the macrophage, a phagocytic cell)



Other cell types spared → no toxicity

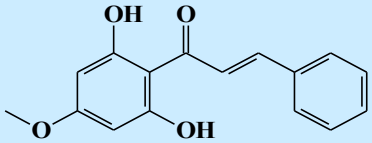
A RAPID AND SENSITIVE METHOD TO QUANTIFY PARASITES IN LESIONS USING GFP-TRANSFECTED FLUORESCENT LEISHMANIA



Rossi-Bergmann et al, submitted

Piper aduncum

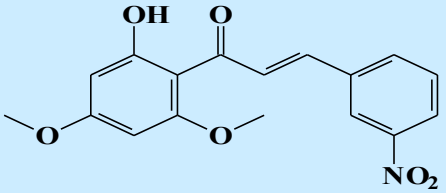
active natural chalcone



CH

Torres-Santos *et al*(1999) *Antim. Ag .Chemother* 43:1453

synthesis (21 analogues)

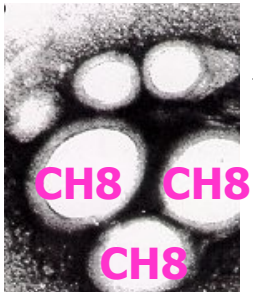


CH8

Patent PI 0204079-4

polymeric:

- **PLA**
- **Chitosan**
- **PHA**

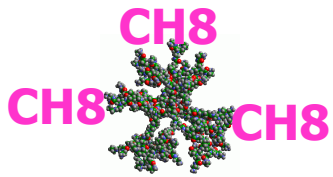


oral , topical, i.v.

Visceral **Cutaneous**

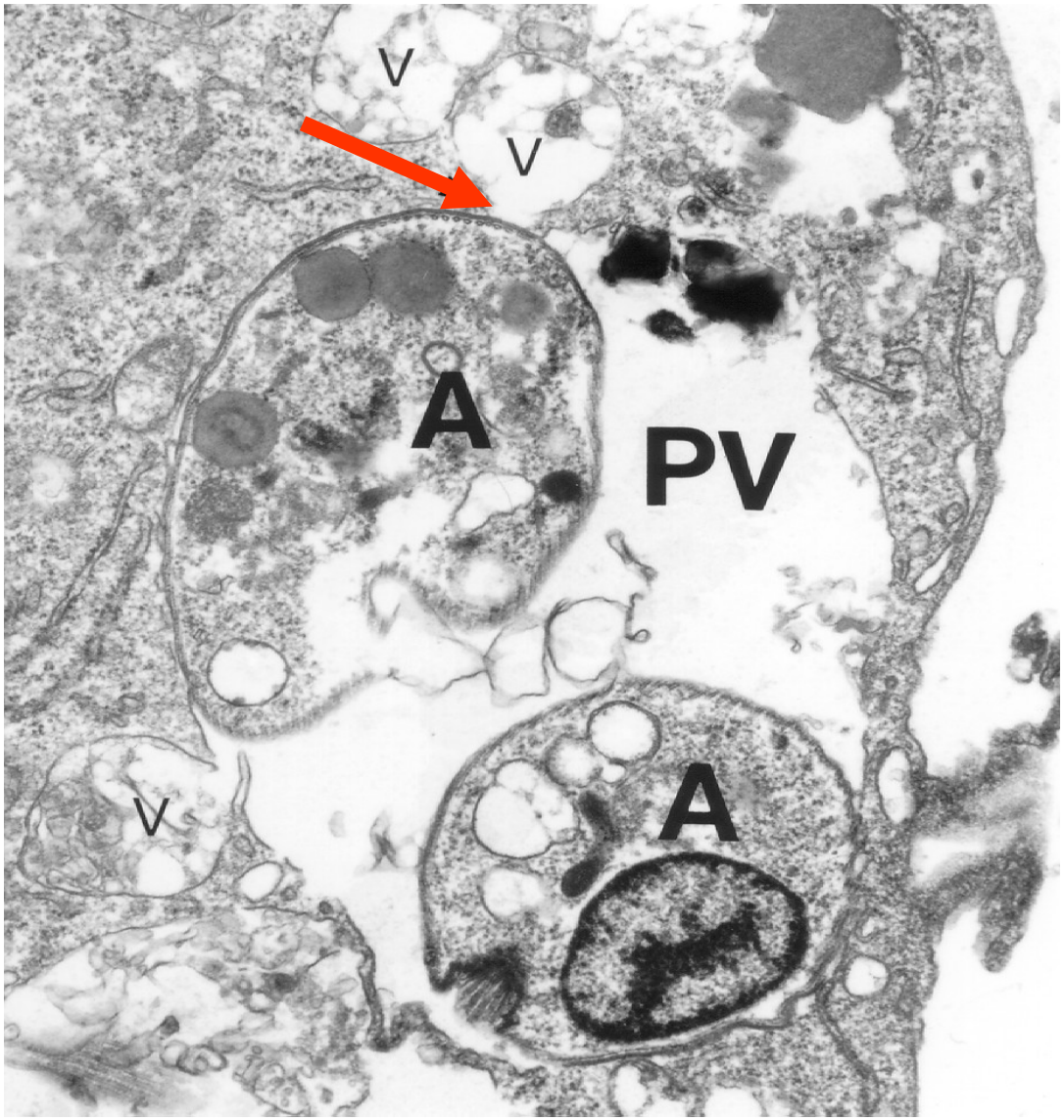
liposomes

β-ciclodextrin

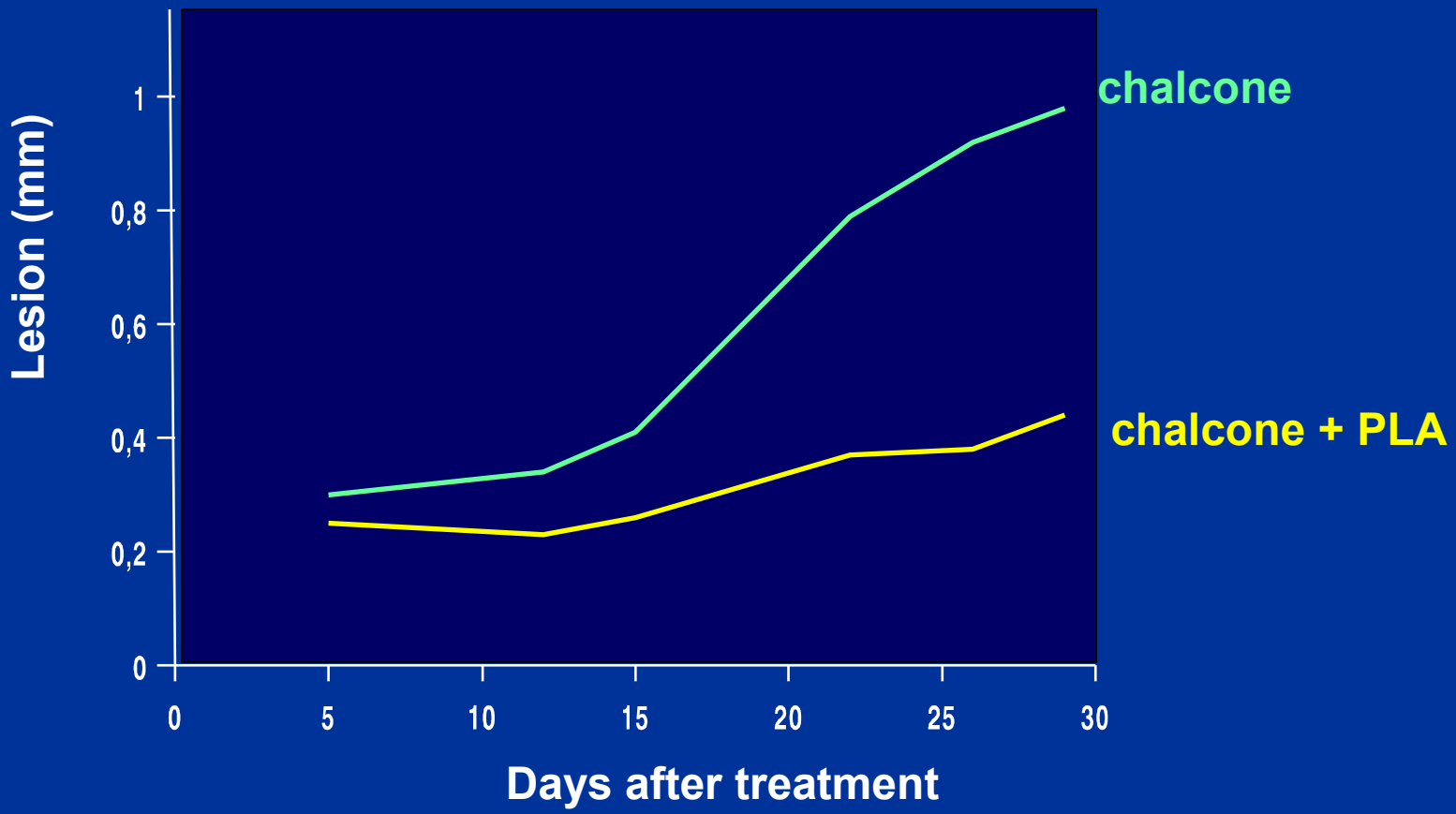


PAMAM dendrimer

Poly(D,L-lactide) nanospheres inside infected macrophages, near the parasites



CHALCONE ENCAPSULATED IN PLA NANOSPHERES
(cutaneous leishmaniasis), 20 ug s.c., days 40 and 45



Torres-Santos, E.C *et al*.(1999 b) *Antim. Ag .Chemother* 43:1776-

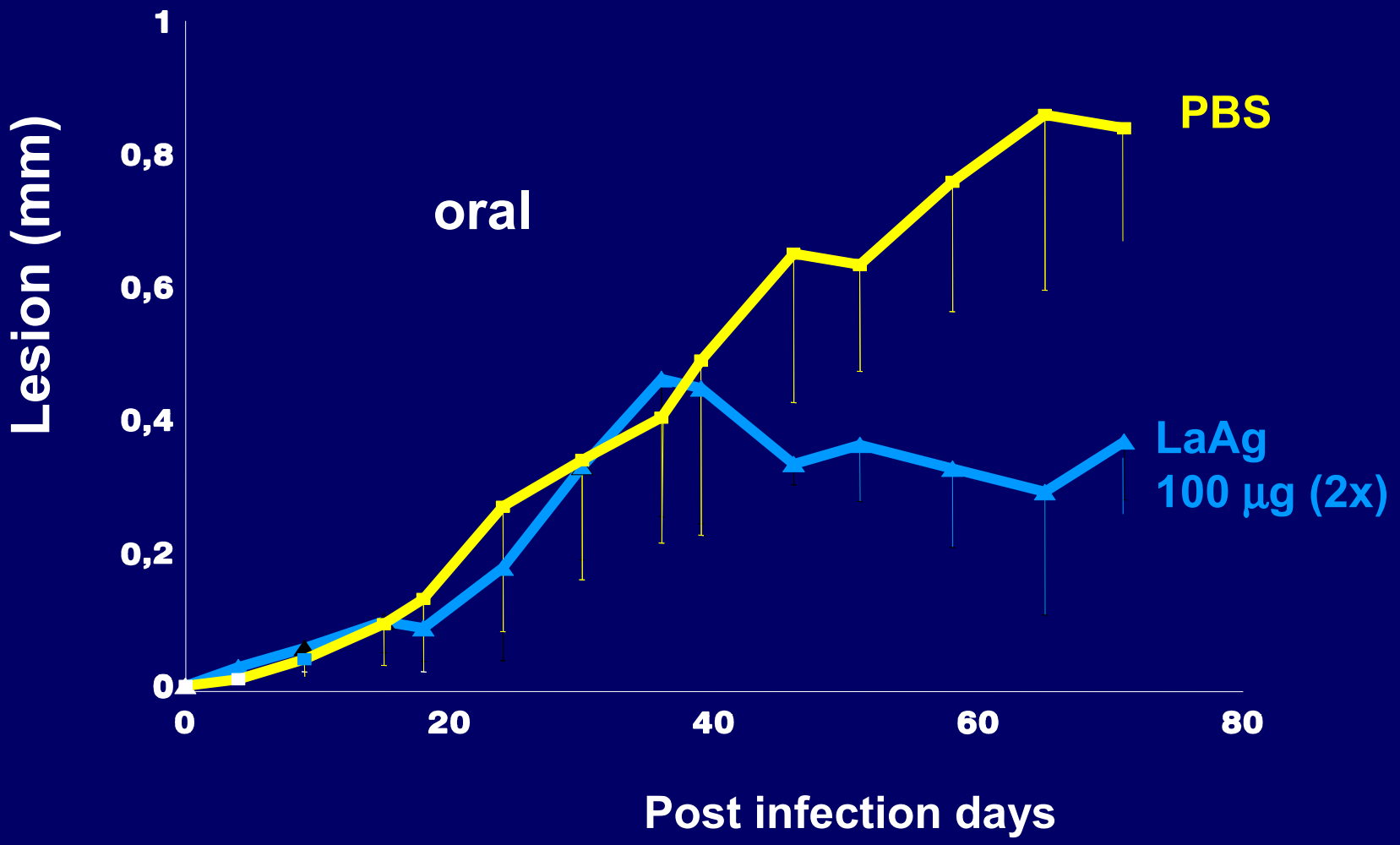
PARTICULATED VACCINES FOR MUCOSAL ADMINISTRATION

LaAg
(Total antigen)

FREE VACCINE √

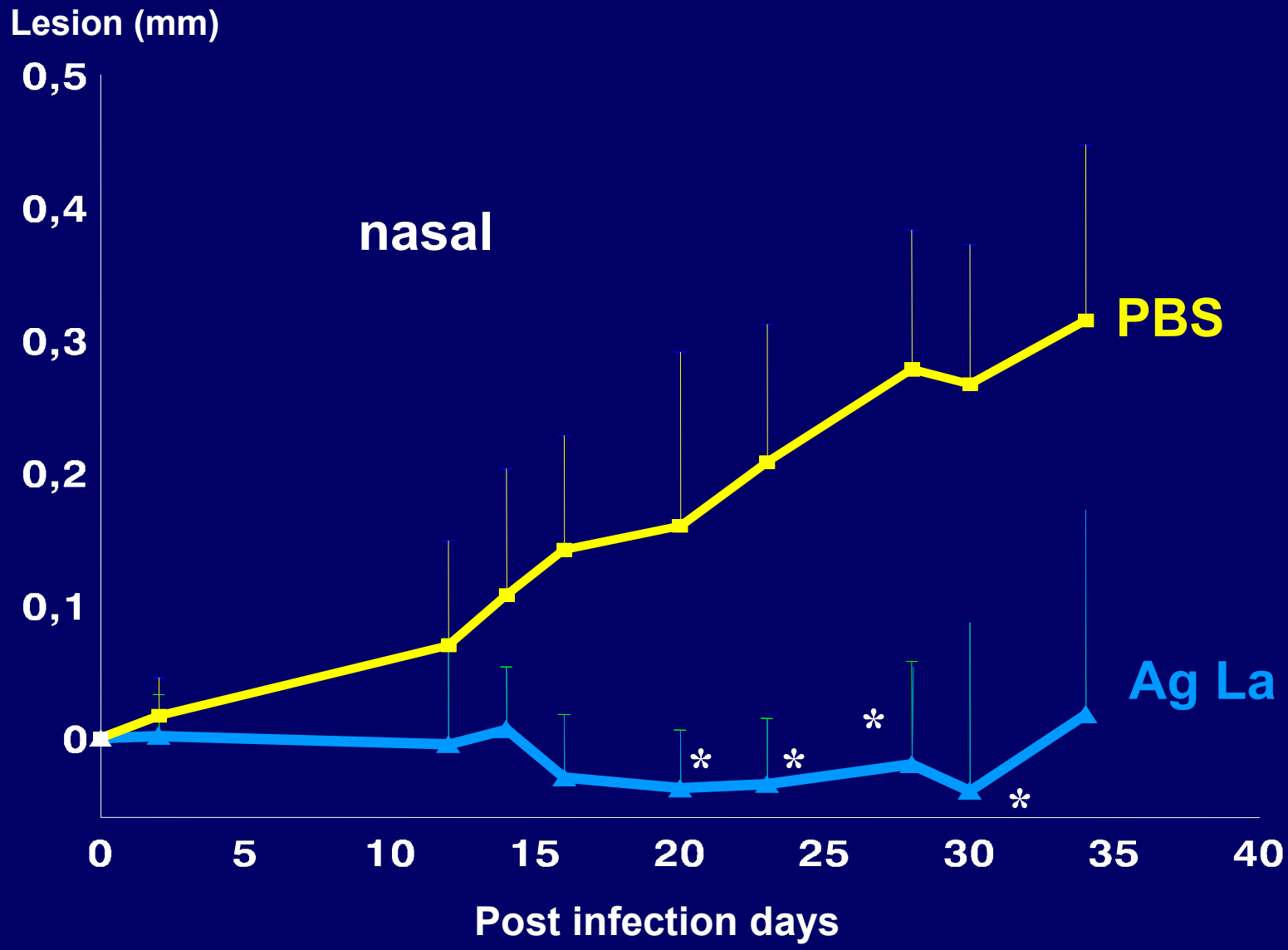
Oral: Pinto E.F. et al (2003) Vaccine
Nasal: Pinto E.F. et al (2004) Inf. Immun

Oral La Ag : protection against infection with *L.amazonensis*



Pinto E.F. et al (2003) Vaccine

Nasal La Ag : protection against infection with *L.amazonensis*



Pinto E.F. et al (2004) Inf. Immun

PARTICULATED VACCINES FOR MUCOSAL ADMINISTRATION

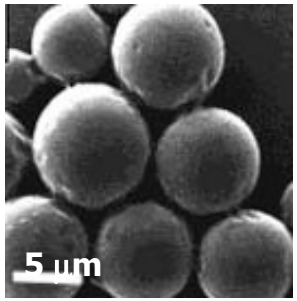
LaAg
(Total antigen)

→ **FREE VACCINE** √

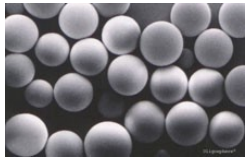
Oral: Pinto E.F. st al (2003) Vaccine
Nasal: Pinto E.F. et al (2004) Inf. Immun

S2A (soluble)
or LaAg

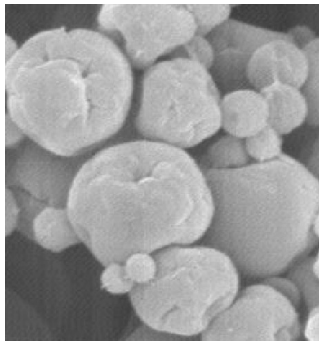
PLGA PARTICLES



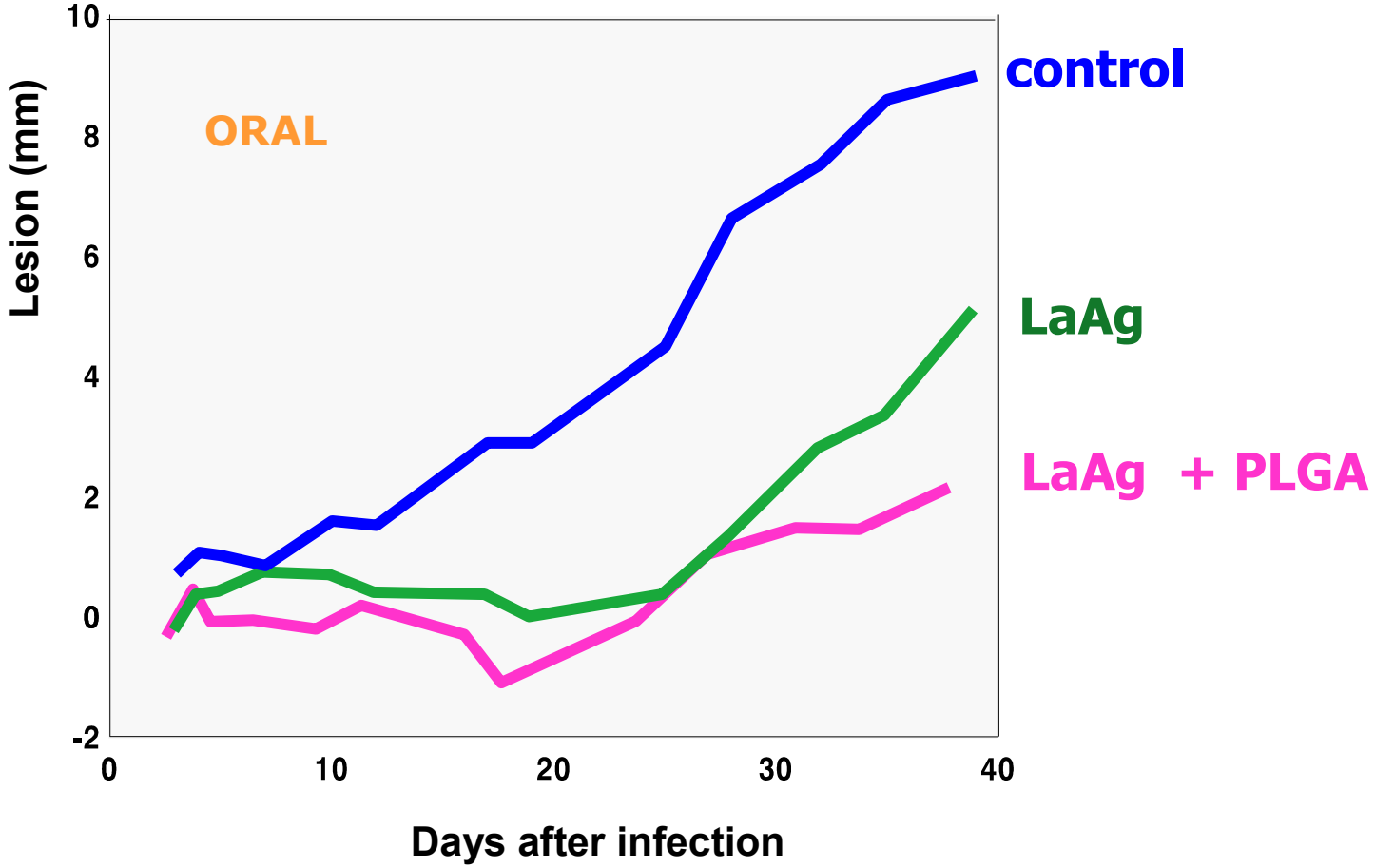
LIPOSOMES



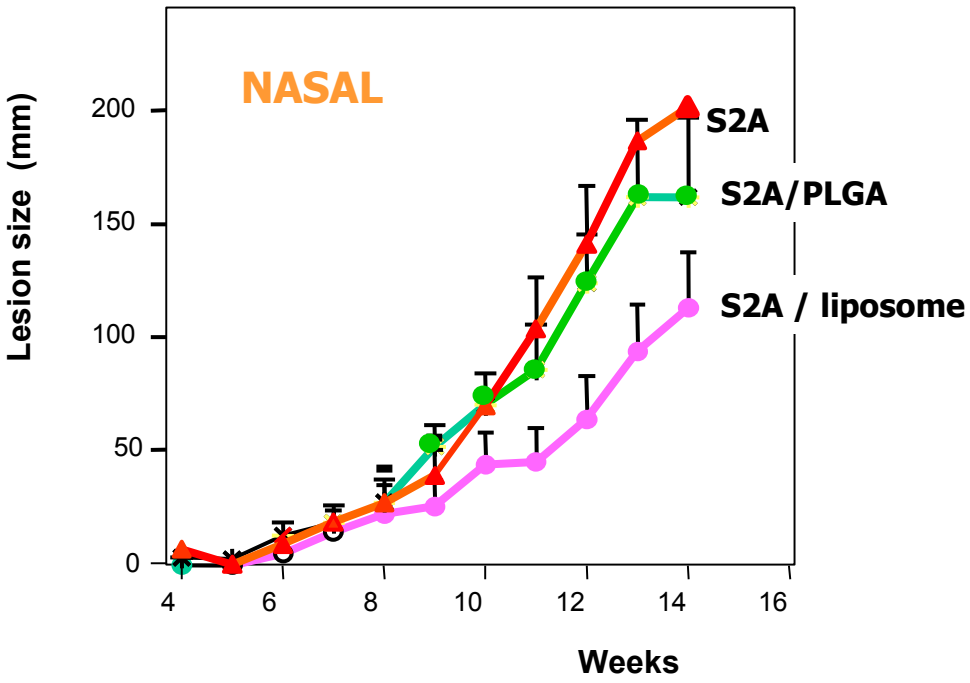
CHITOSAN PARTICLES



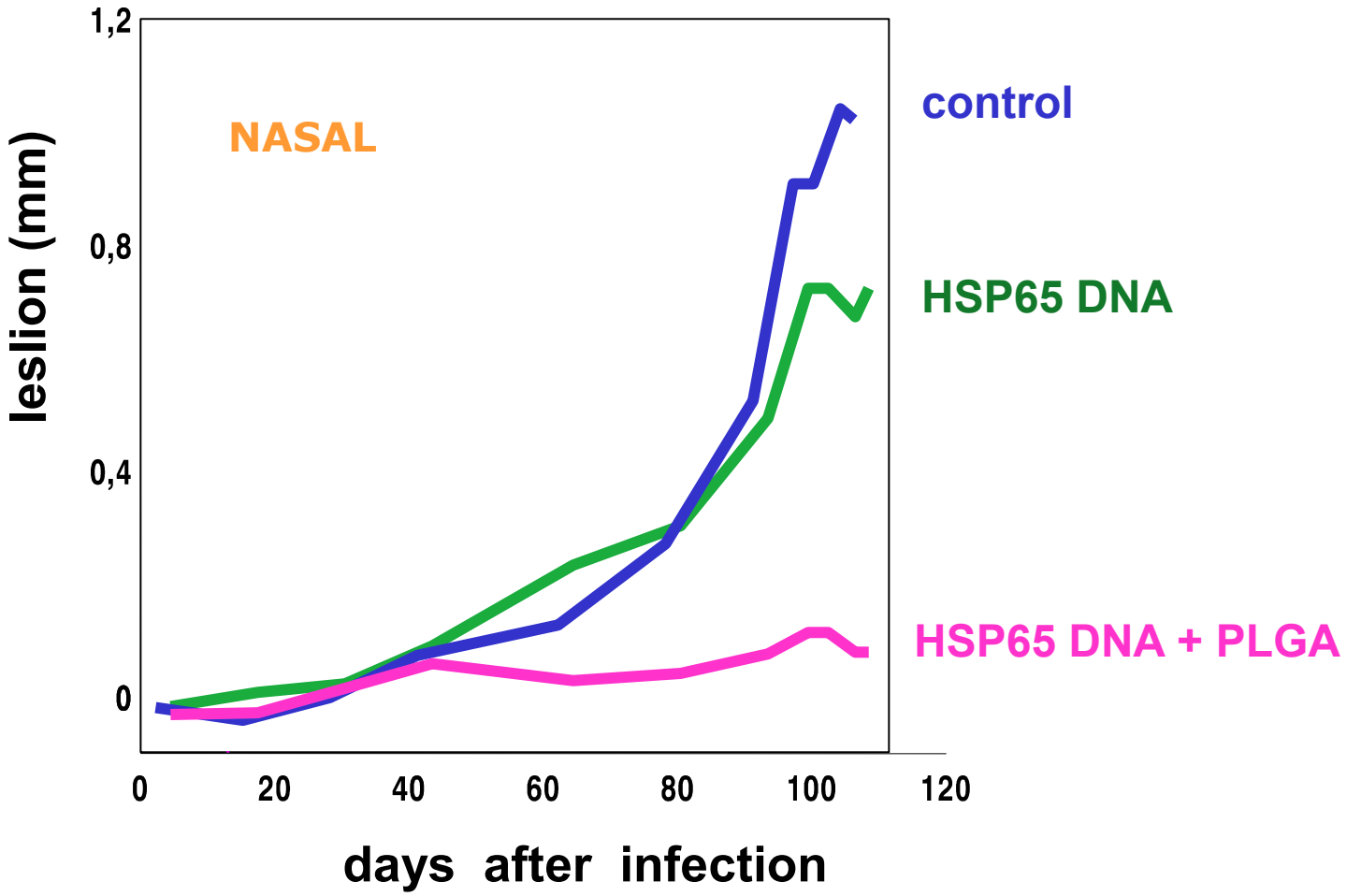
LaAg in PLGA PARTICLES - ORAL



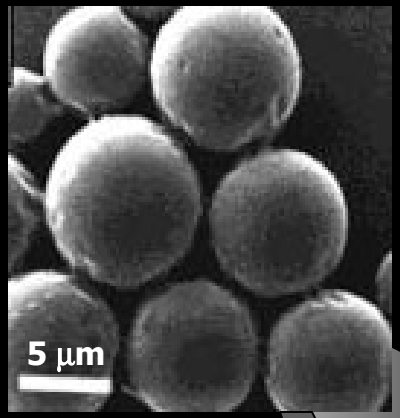
S2A in LIPOSOMES - NASAL

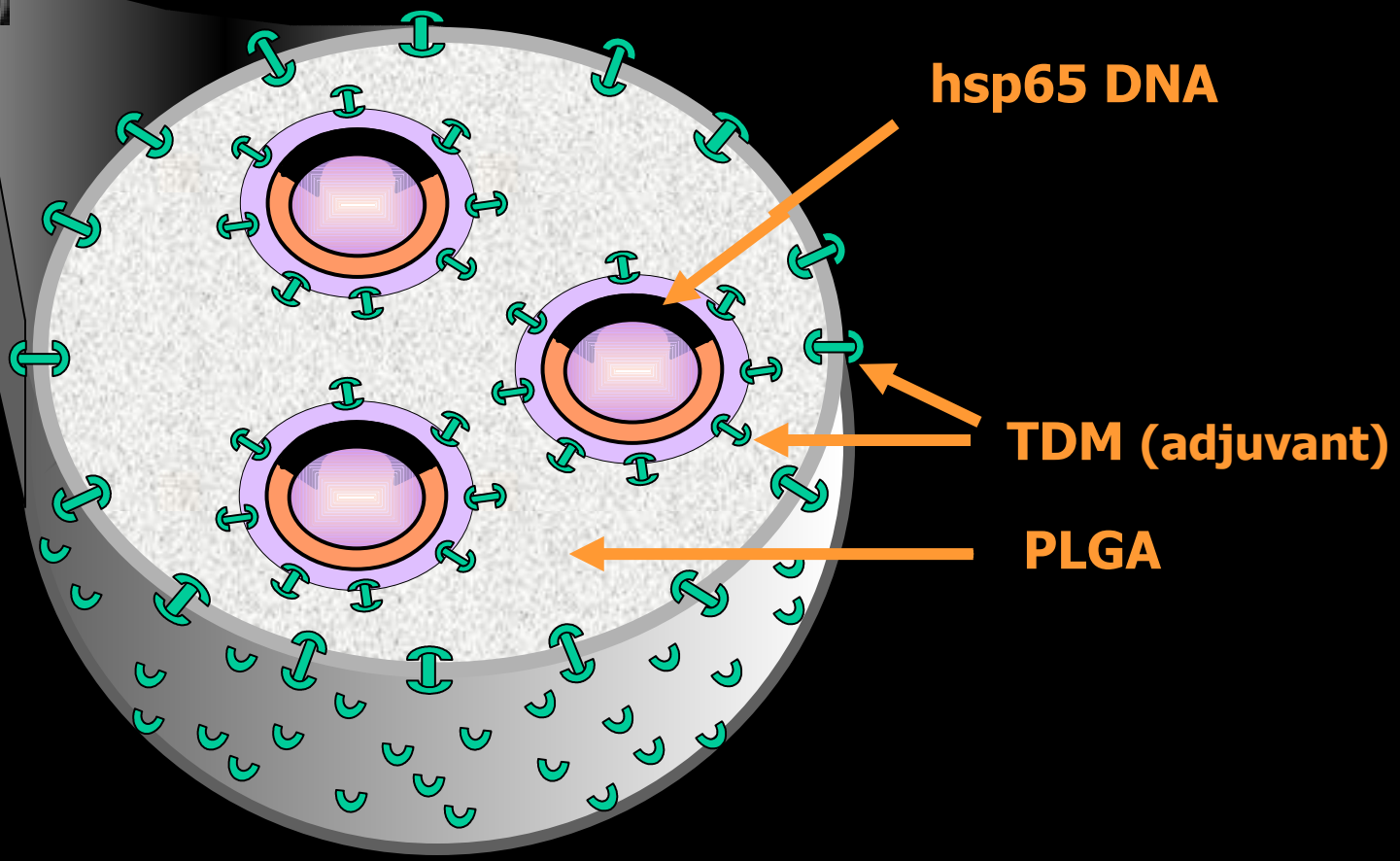


DNA VACCINE IN PLGA PARTICLES - NASAL

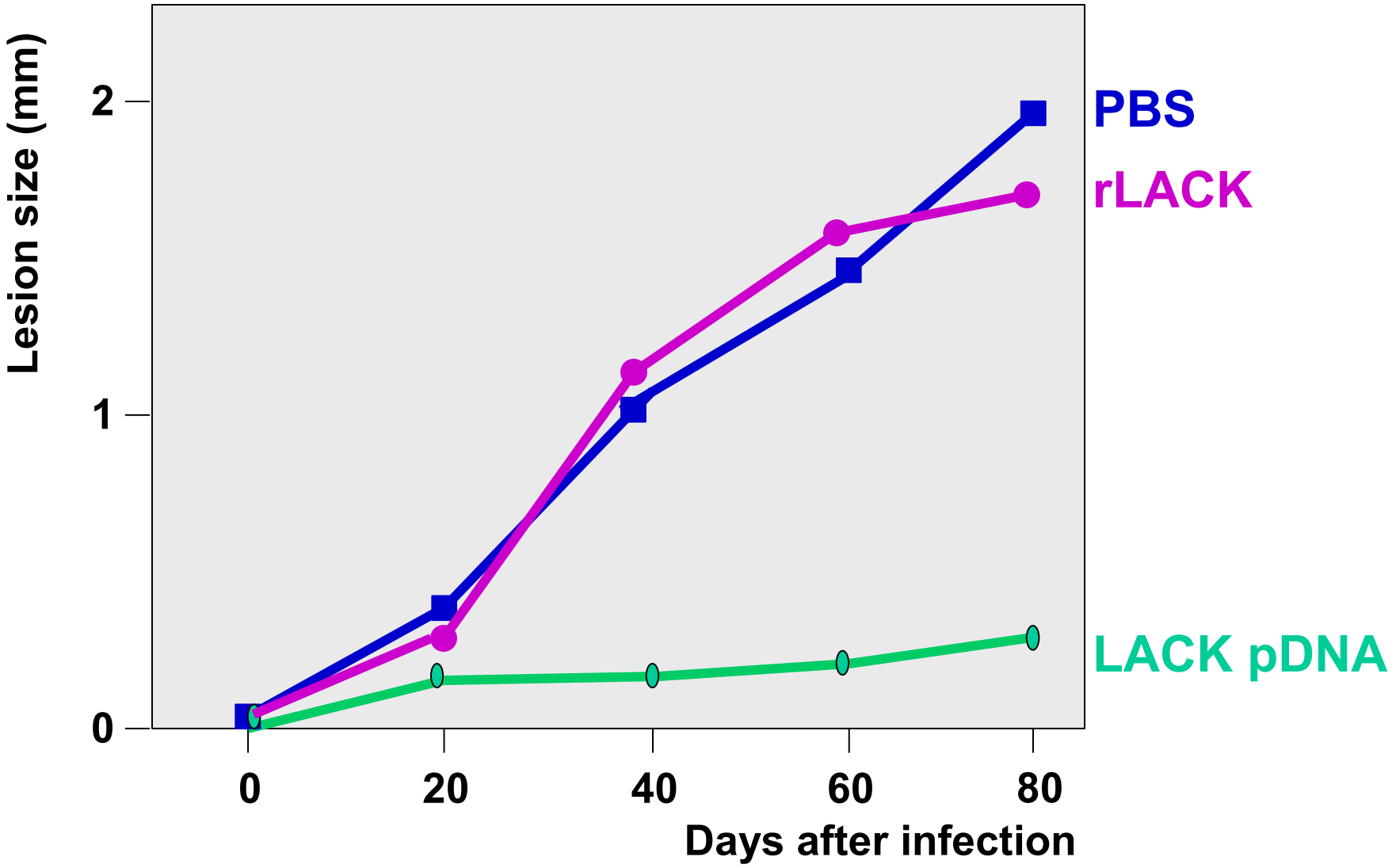


hsp65 DNA vaccine + TDM (trehalose dimicolate) loaded in PLGA microspheres





PROTECTIVE EFFECT OF NASAL IMMUNIZATION WITH LACK DNA



LEISHMANIASIS AS A DISEASE MODEL

- Endemic in Brazil (also in Europe).
  - No vaccine available.
  - Current drugs are toxic.

- Leishmania are the only parasites that replicate exclusively inside a phagocyte → Good model to test particulated drugs.

- We have a novel fluorimetric method to more rapidly assess drug and vaccine efficacy.

- New drug (synthetic chalcone) and nasal vaccine to be used as prototypes for evaluation of nanoparticle formulations.

- Cheaper nanoformulations under development:
  - Chitosan particles: topical and mucosal administration.
  - Brazilian biodegradable PHA polymer →

REDE DE NANOBIOTEC (CNPq / MCT)

Leader : Nelson Durán Cabalero (UNICAMP)

Participants of this project:

Maria Inês Ré (IPT)	**Nano and Microspheres of Chitosan** **Nano and Microspheres of PHAs**
José Rodrigues Júnior (Nanocore).....	**Nano and Microspheres of PLA e PLGA**
Frederic Frèzard (UFMG).....	**β-cyclodextrin** **Liposomes**
Vanessa Furtado (UFRJ).....	**Dendrimers**
Bartira Rossi Bergmann (UFRJ).....	**Drug (chalcone)and Mucosal Vaccine**