Borrelia Attack Models

University of New Haven Symposium

May 19, 2007

Alan B. MacDonald MD
Staff Pathologist
St Catherine of Siena Medical Center
Smithtown , New York

Some Parasitic Diseases

Actually

Require Long Term

Antiparasitic Therapy

Spirochetes are Parasites

Therapy for Spirochetal parasites should be

<u>Customized</u> by the Physician to

<u>Eliminate</u>

<u>The Parasitic Spirochetal Infestation</u>

Module #1
Examples of Spirochetes
INSIDE
of Living cells

Preface

"Members of the Genus ***Borrelia*** display one of the most unusual genome structures in the bacterial world, if not in creation"

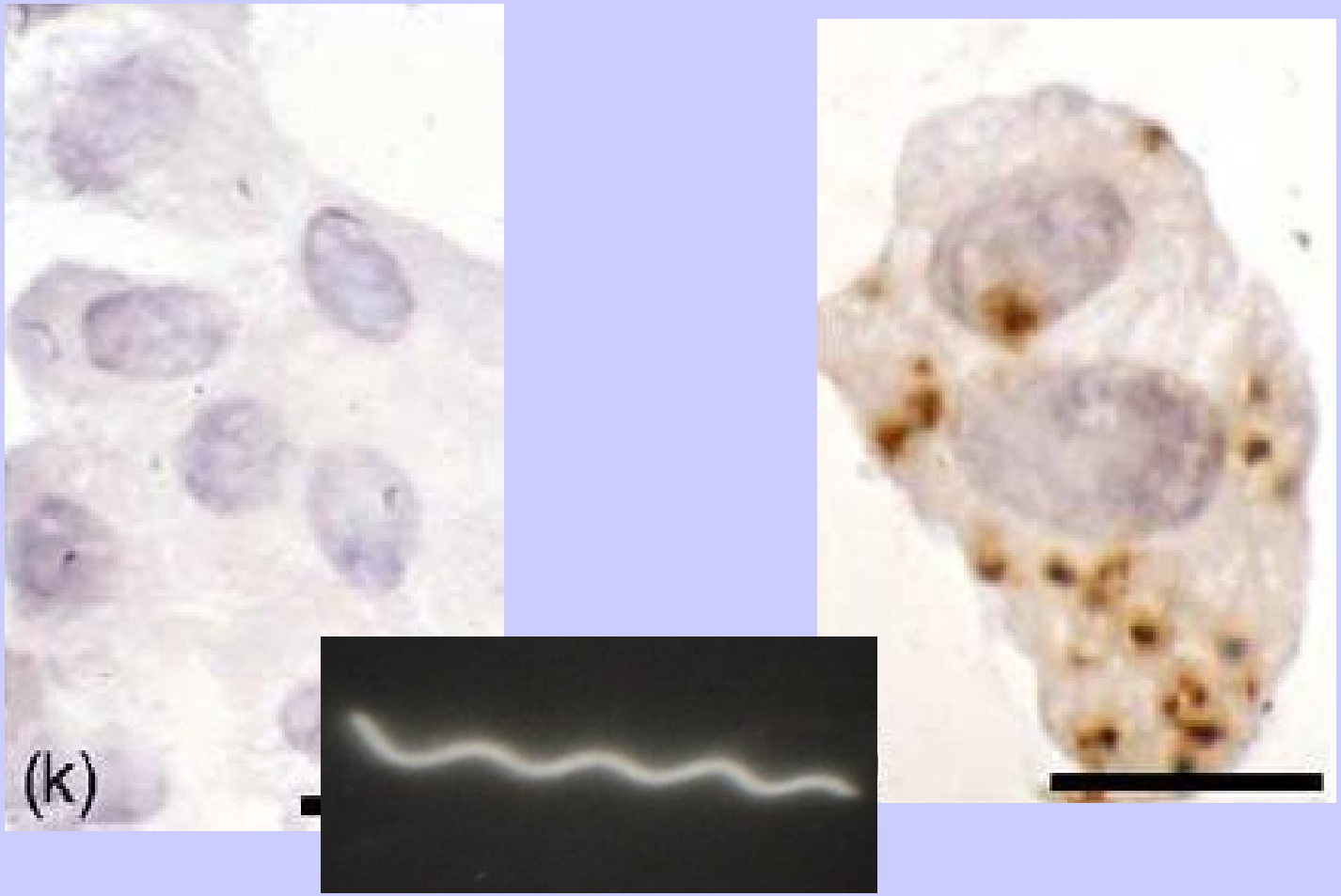
Dr George Chaconas

Professor and Canadian Research Chair in the Molecular Biology of Lyme Disease

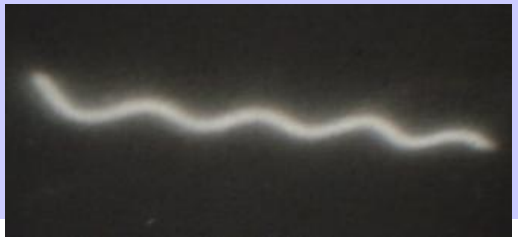
University of Calgary, Canada

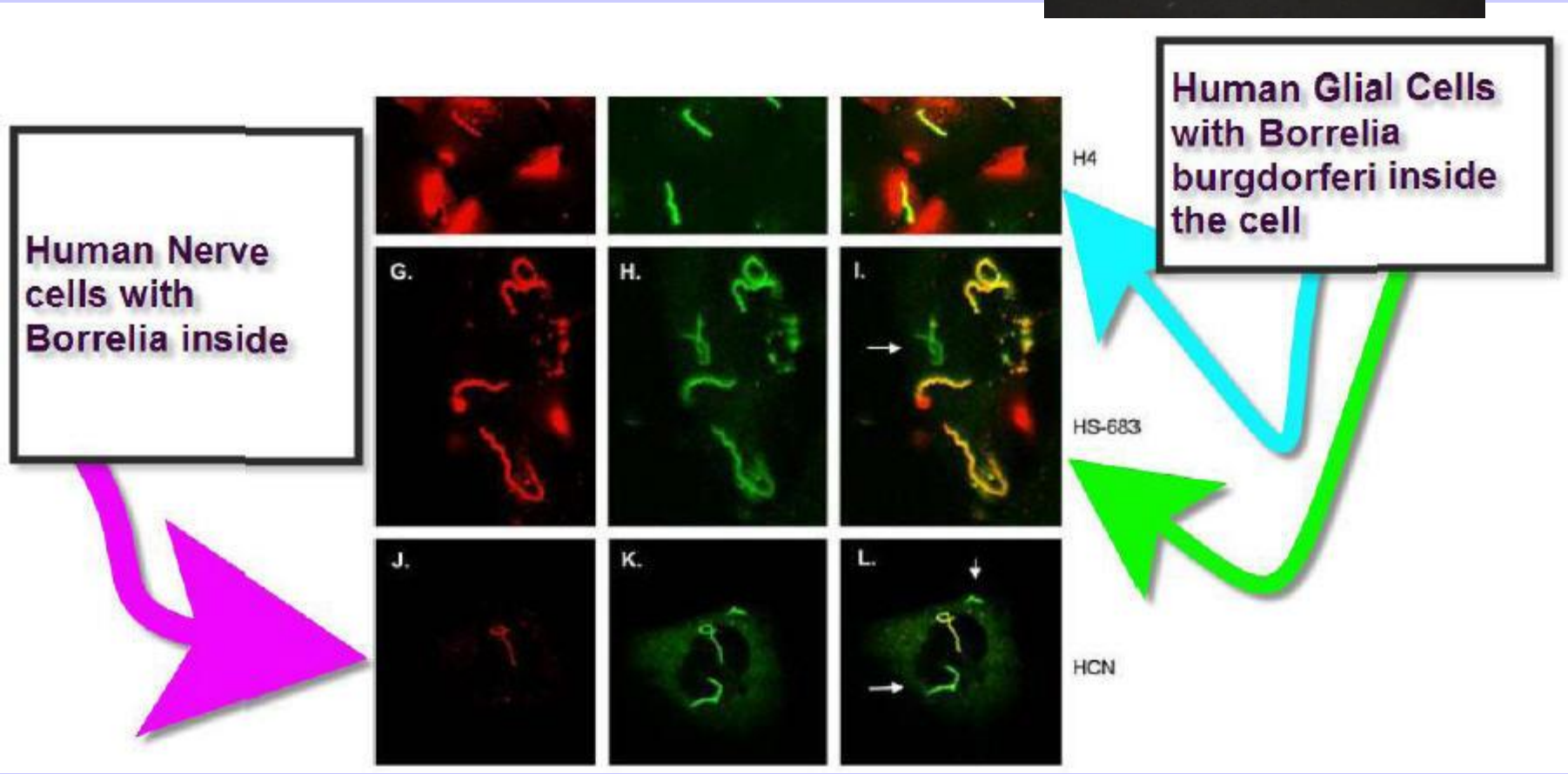
Miklossy Rat Neuron

Miklossy Rat Neuron

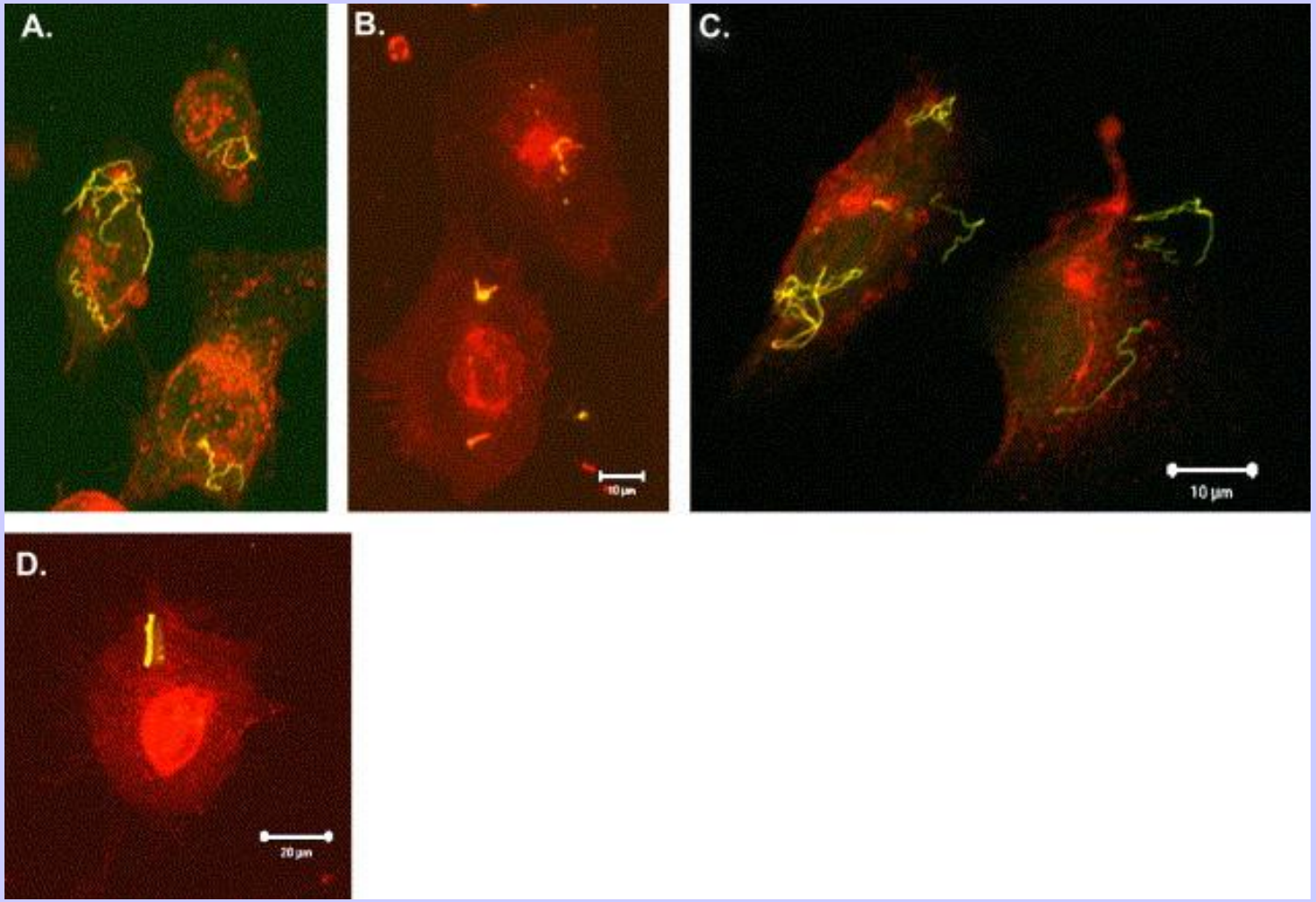


Livengood and Gilmore
Human Nerve cells

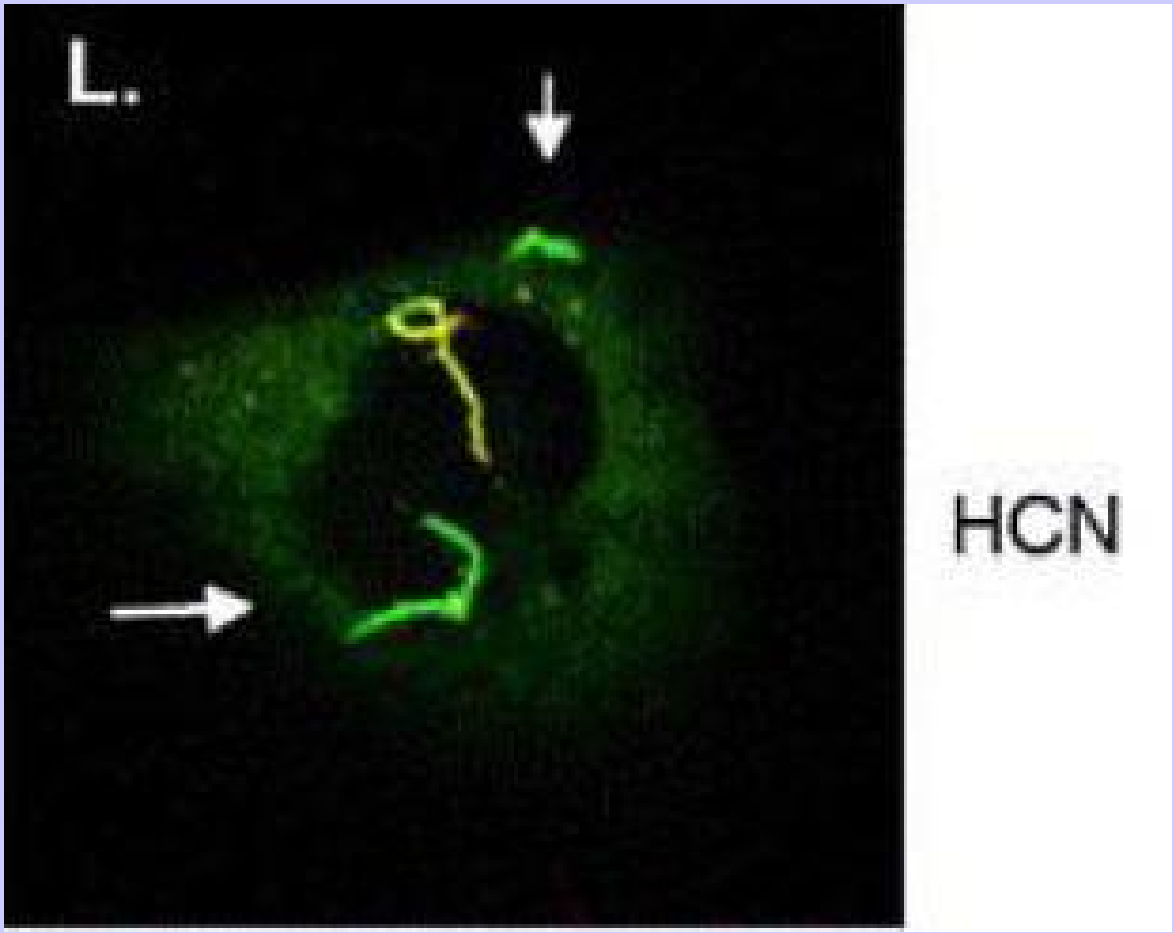




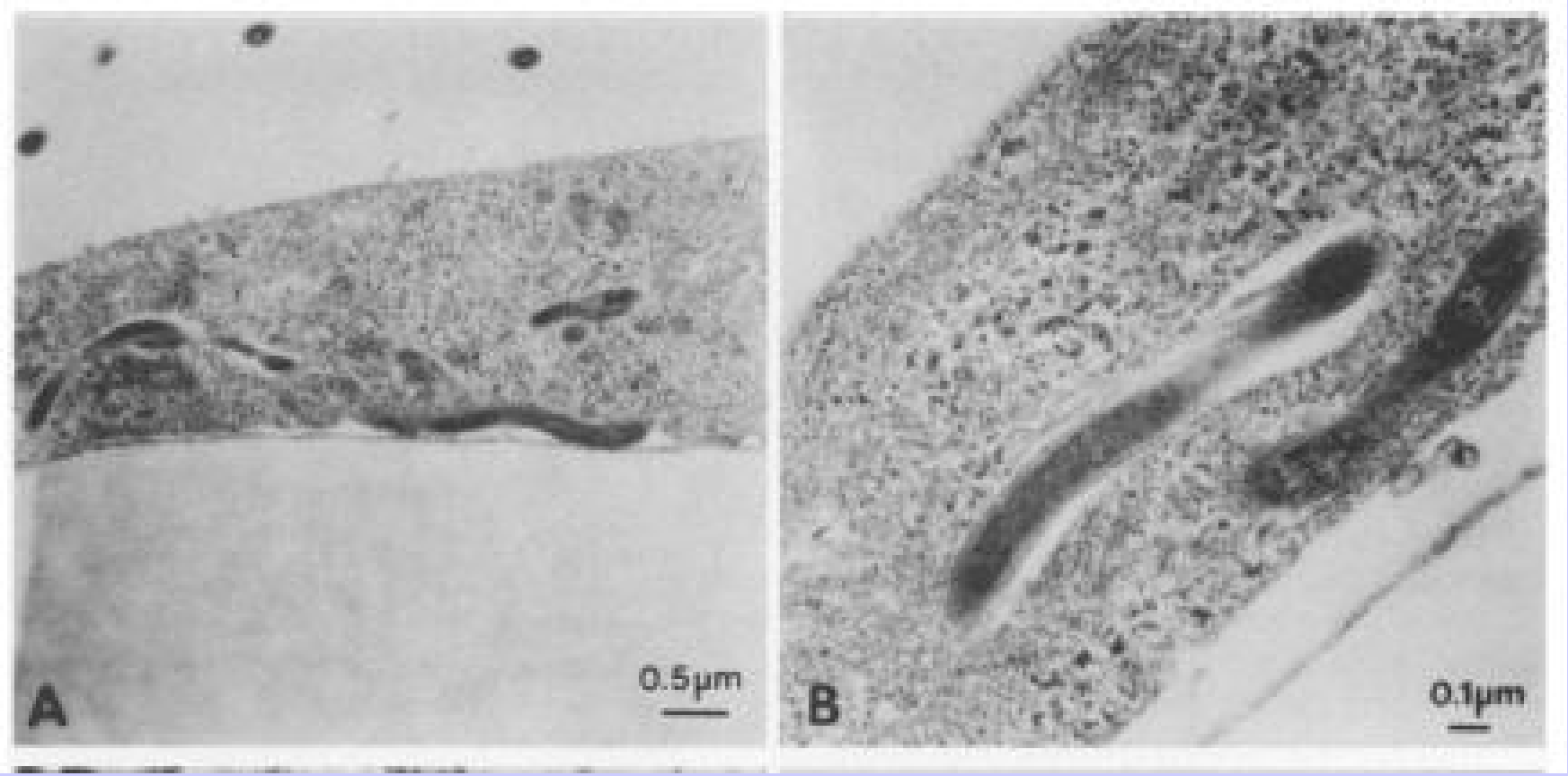
Livengood and Gilmore Human Nerve cells



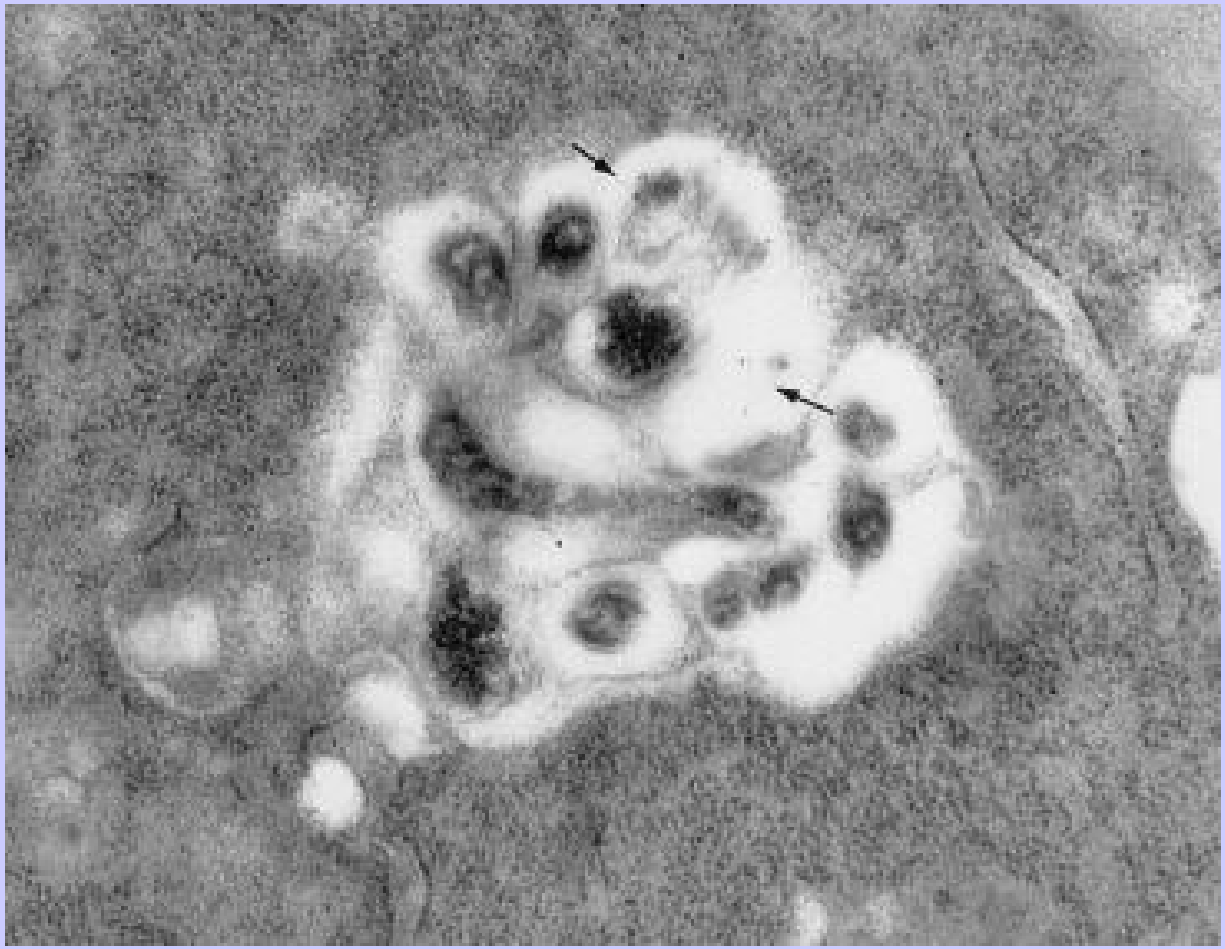
Livengood and Gilmore Human Nerve cells



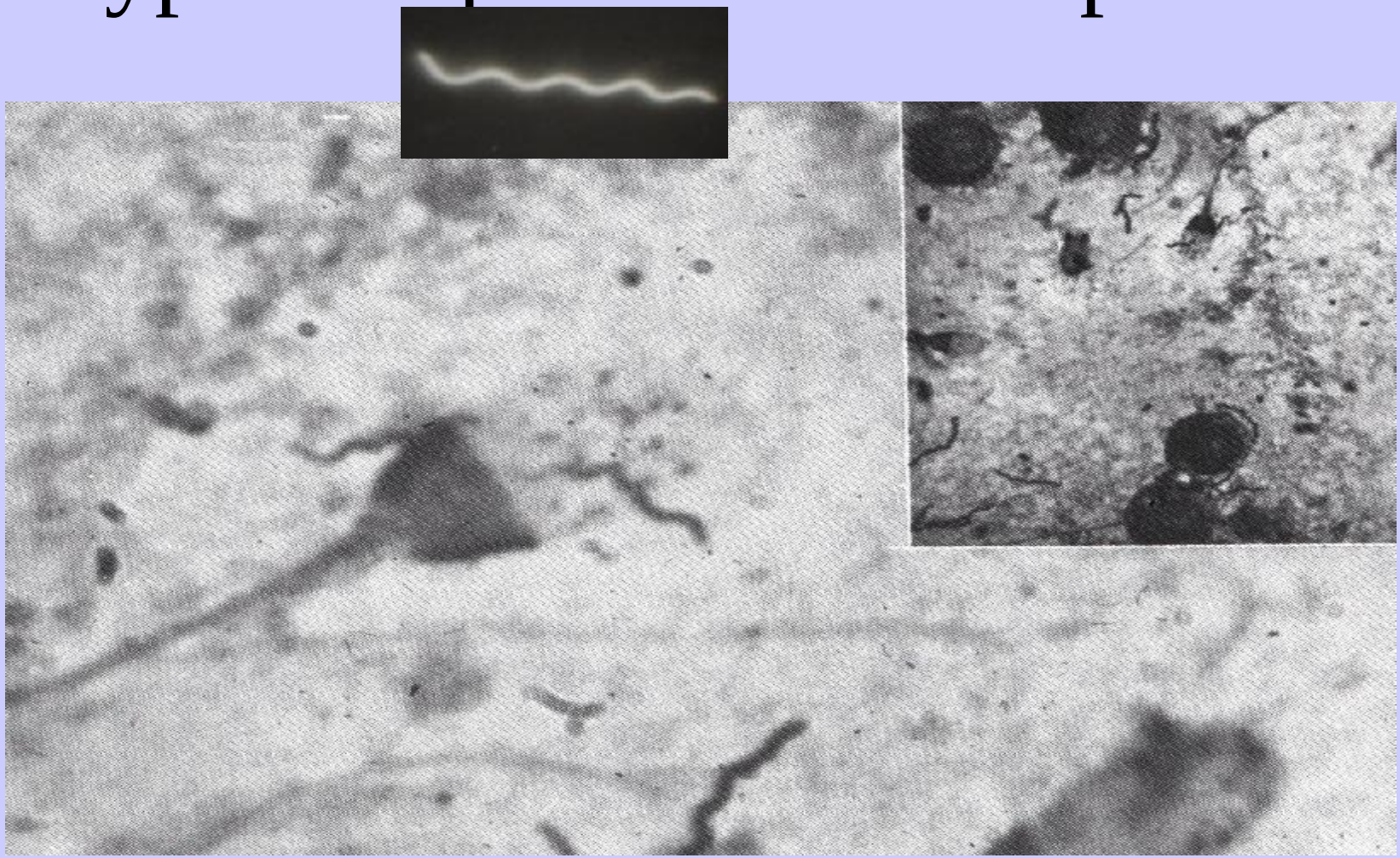
Thomas and Comstock
Human Endothelial cells
Invaded by Borrelia



Montgomery – Macrophages Invaded by Borrelia

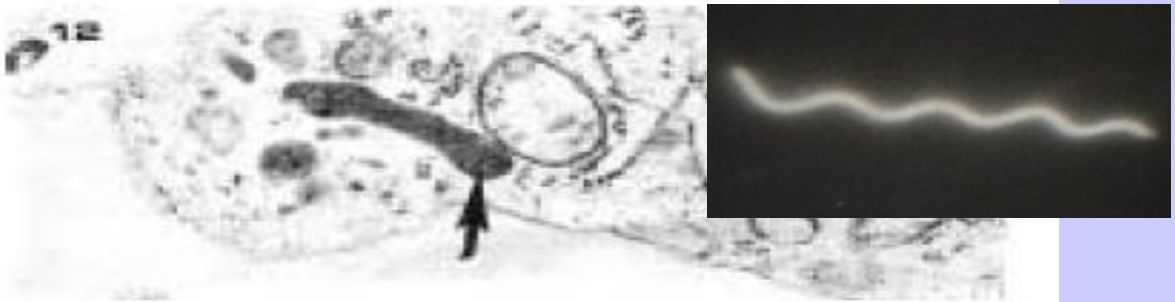


Syphilis Spirochete and Sperm



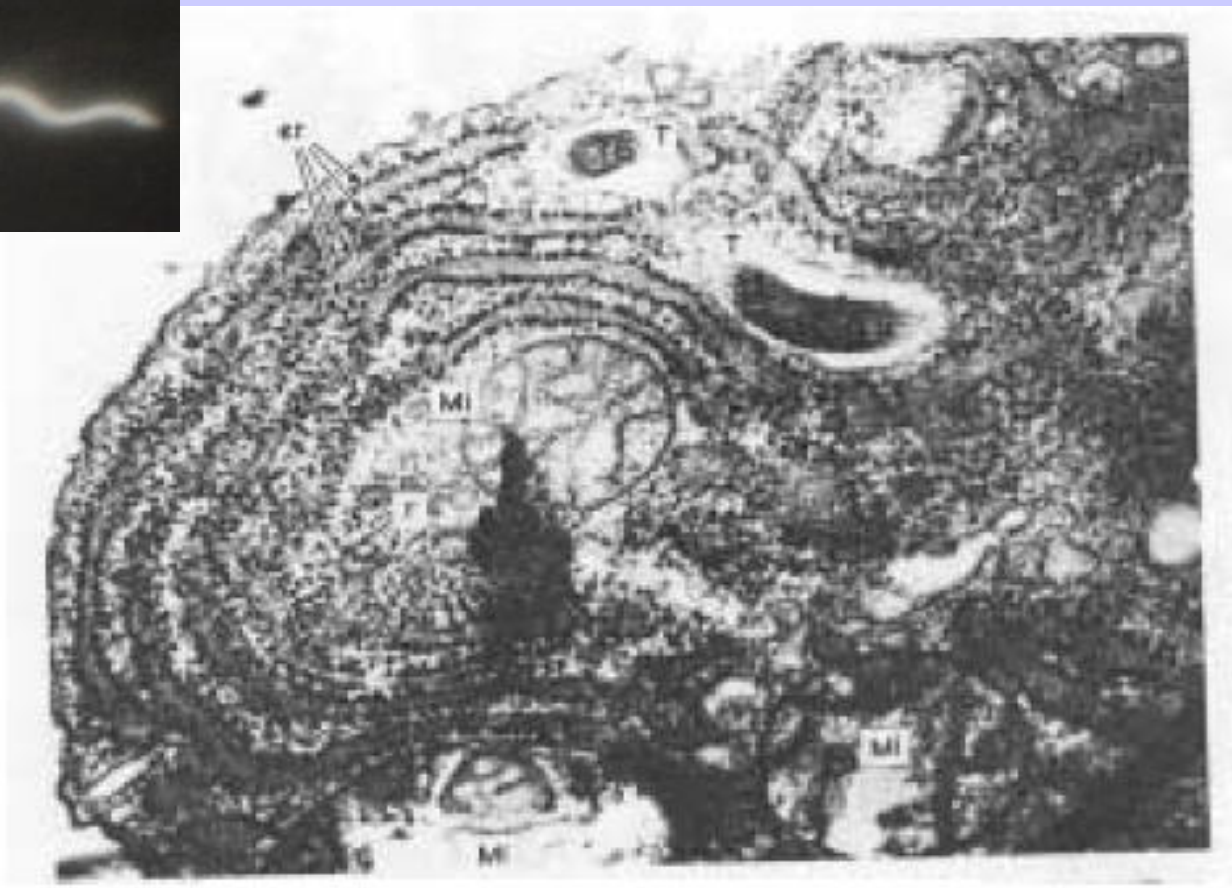
Spirochetes Invade Cells Because Spirochetes are Parasites

T. pallidum. Inside a fibroblast.
Lauderdale V; Goldman JN. 1972.



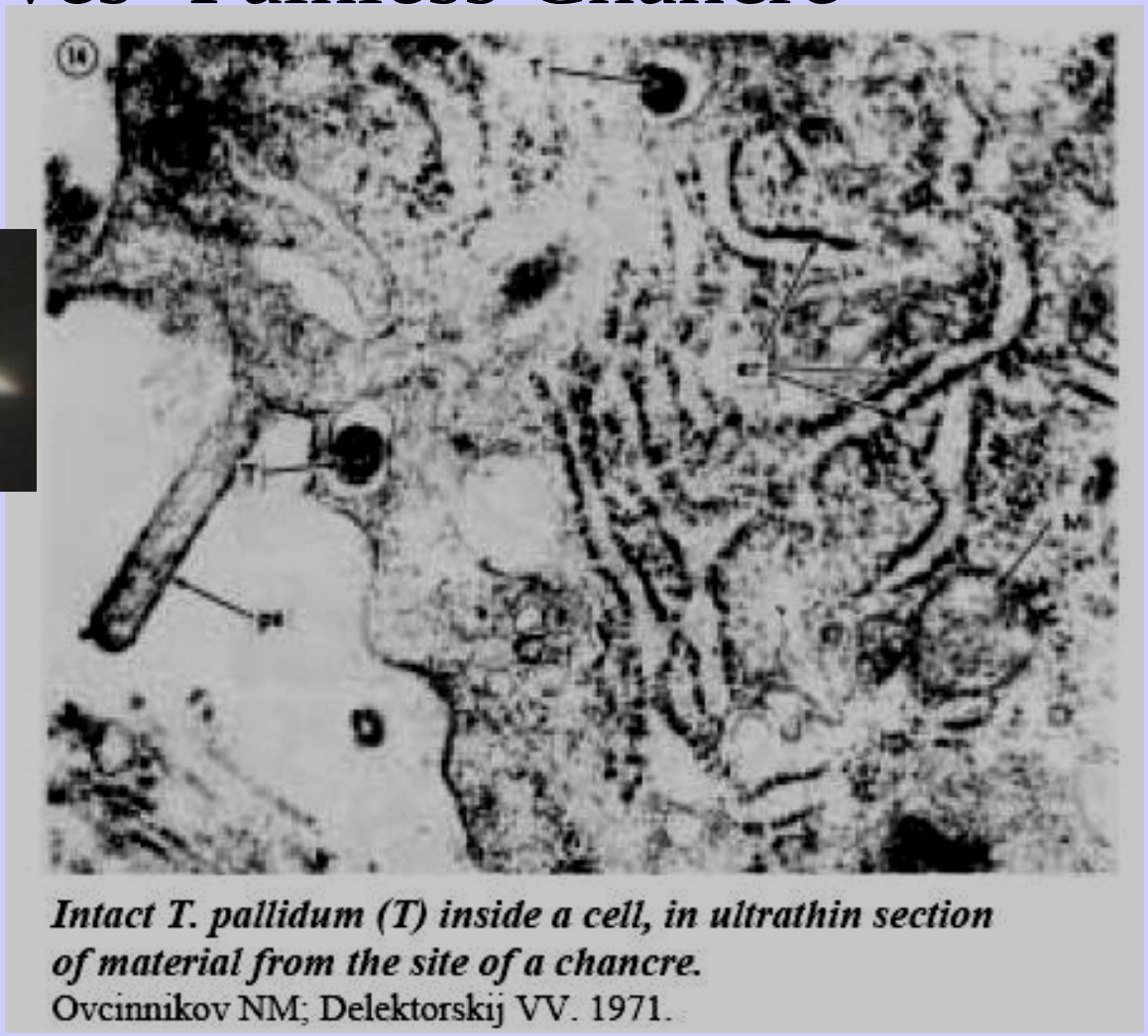
T. pallidum. Inside a Leydig cell.
Lauderdale V; Goldman JN. 1972.

Syphilis Spirochetes Invade Cells



T. pallidum (T) inside plasma cell.
Ovcinnikov NM; Delektorskij VV. 1971.

Syphilis Spirochetes
Block Nerves- Painless Chancre



Intact T. pallidum (T) inside a cell, in ultrathin section of material from the site of a chancre.
Ovcinnikov NM; Delektorskij VV. 1971.

Module #2
Examples of
Spirochetal
Life Cycles

Borrelia Life Cycle Concepts

A Life Cycle – Typical of Parasites

Multiple Forms of the Parasite

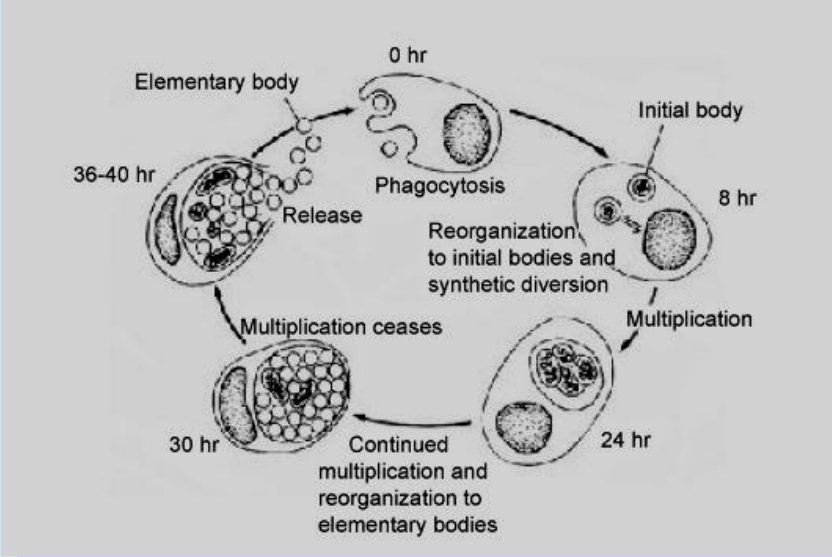
Blood Phase Forms

Tissue Phase Forms

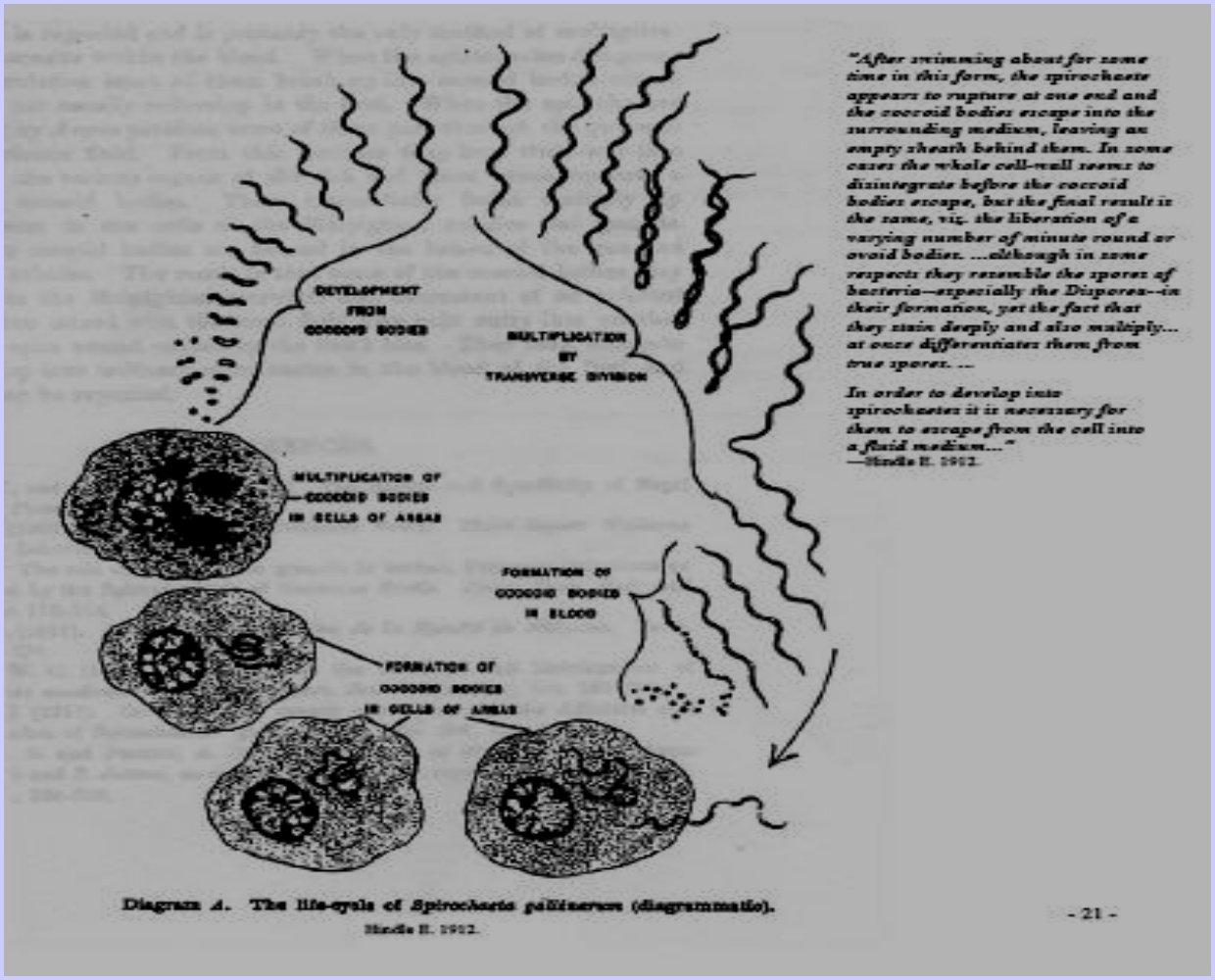
Insect Vector Forms

A circular Loop or Loops of Interconnections

between the Various Forms



Spirochetal Life Cycle – Hindle – Year 1912



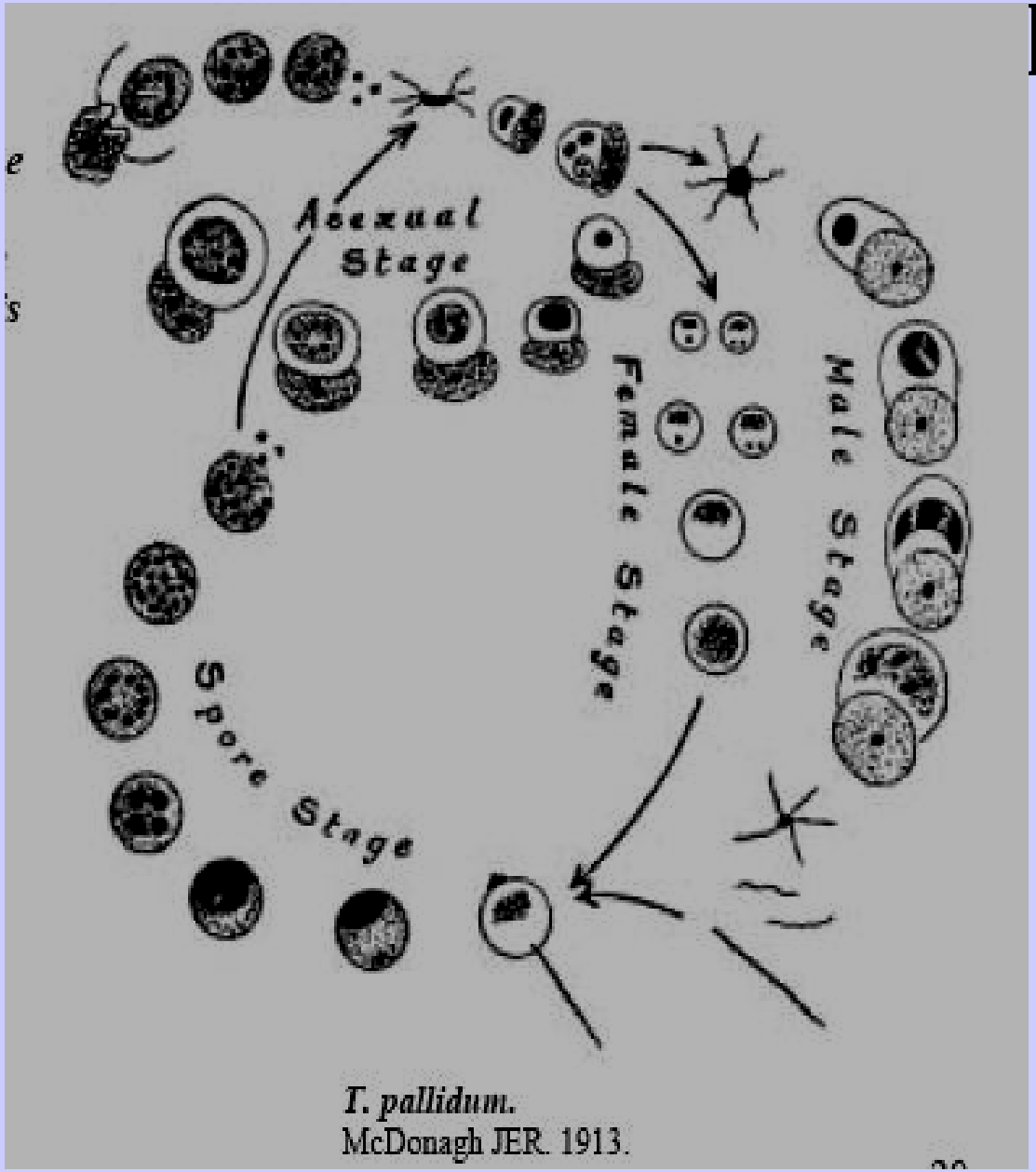
"After swimming about for some time in this form, the spirochaete appears to rupture at one end and the coccoid bodies escape into the surrounding medium, leaving an empty sheath behind them. In some cases the whole cell-wall seems to disintegrate before the coccoid bodies escape, but the final result is the same, viz. the liberation of a varying number of minute round or ovoid bodies. ...although in some respects they resemble the spores of bacteria—especially the Diaspores—in their formation, yet the fact that they stain deeply and also multiply... at once differentiates them from true spores. ...

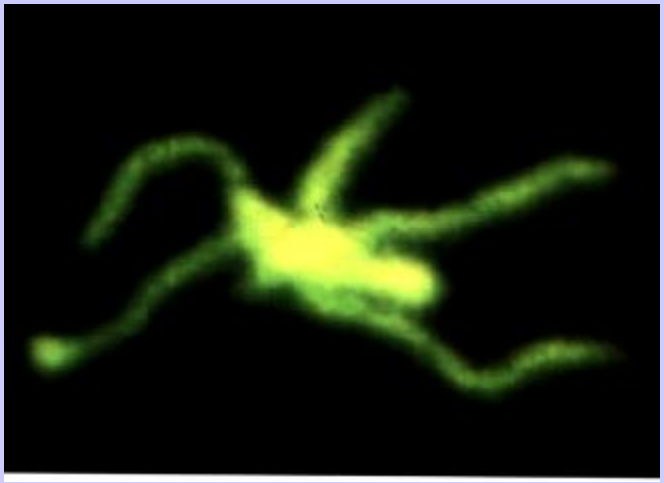
In order to develop into spirochaetes it is necessary for them to escape from the cell into a fluid medium..."
—Hindle E. 1912.

Diagram A. The life-cycle of *Spirochaeta gallinarum* (diagrammatic).
Hindle E. 1912.

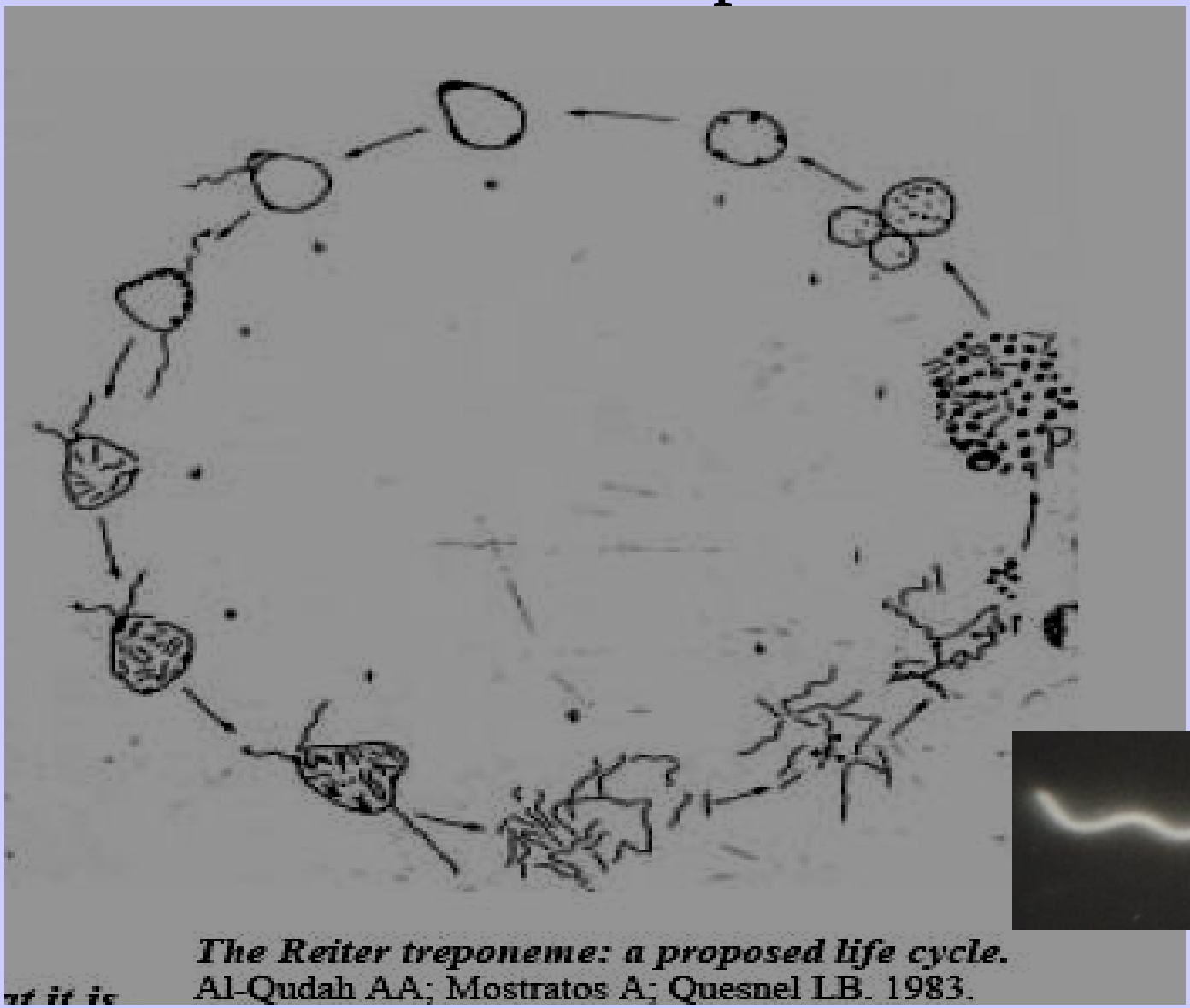
- 21 -

Spirochete Life Cycle McDonaugh Year 1913



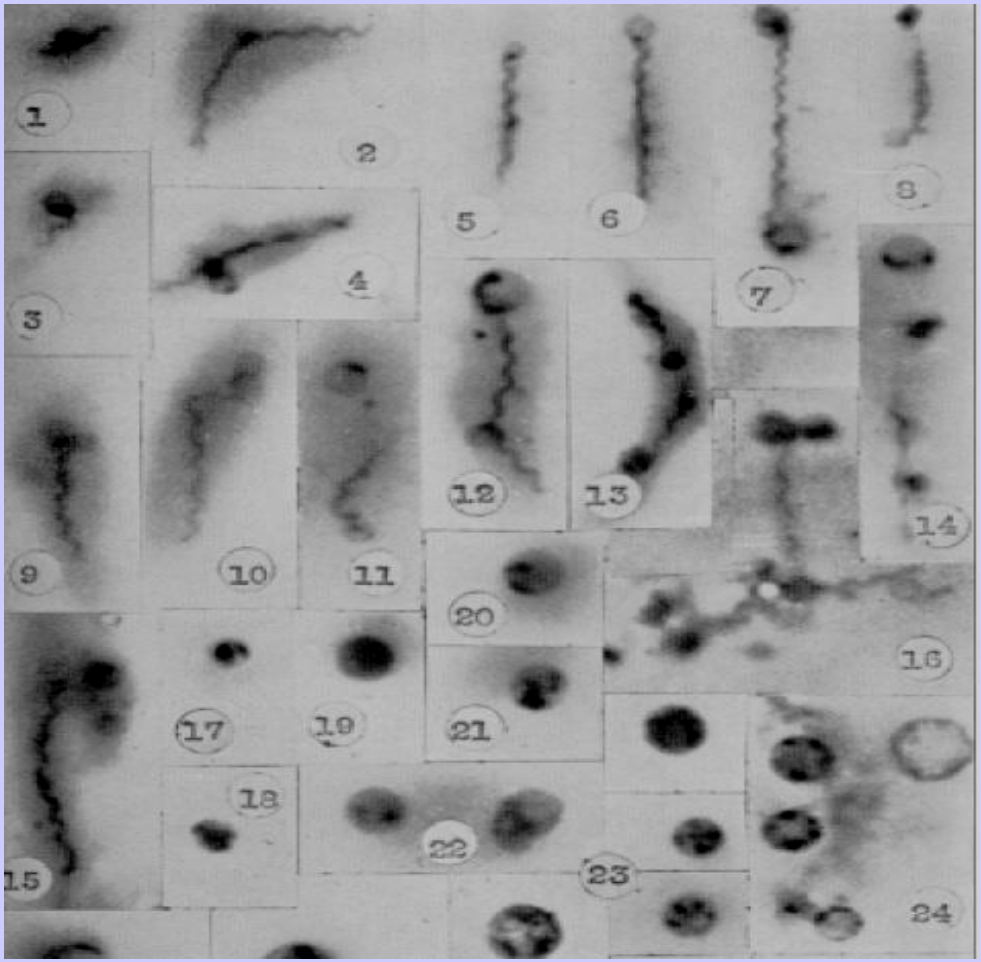


Spirochete Life Cycle



The Reiter treponeme: a proposed life cycle.
Al-Qudah AA; Mostratos A; Quesnel LB. 1983.

Life Cycle for Treponemes
Dr Delamater – Year 1950



Borrelia Life Cycle

ELSEVIER

medical hypotheses

http://intl.elsevierhealth.com/journals/mehy

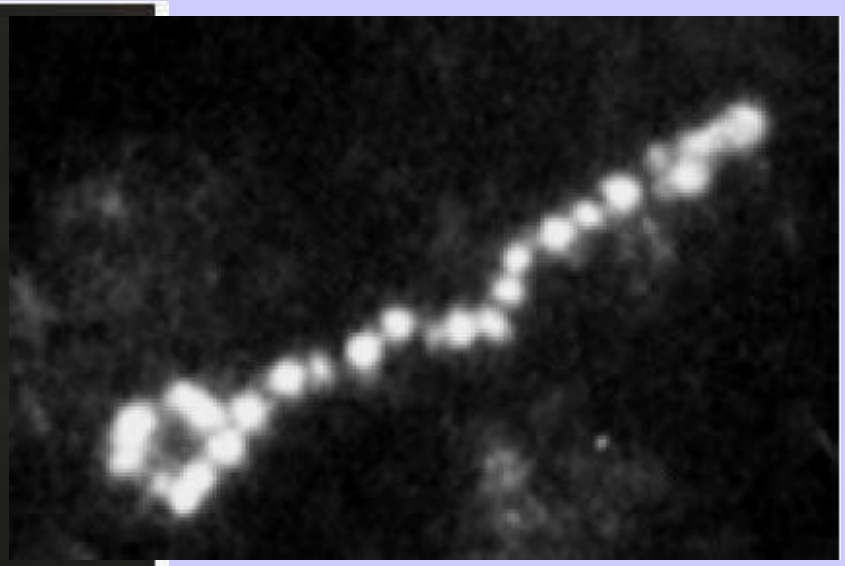
A life cycle for *Borrelia* spirochetes?

Alan B. MacDonald *,1

St. Catherine of Siena Medical Center, Department of Pathology, 50 Rte 25 A, Smithtown, NY 11787, USA

Received 20 March 2006; accepted 21 March 2006

MacDonald – Borrelia Life Cycle



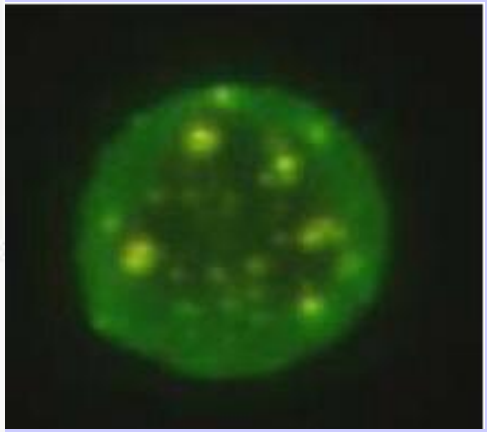
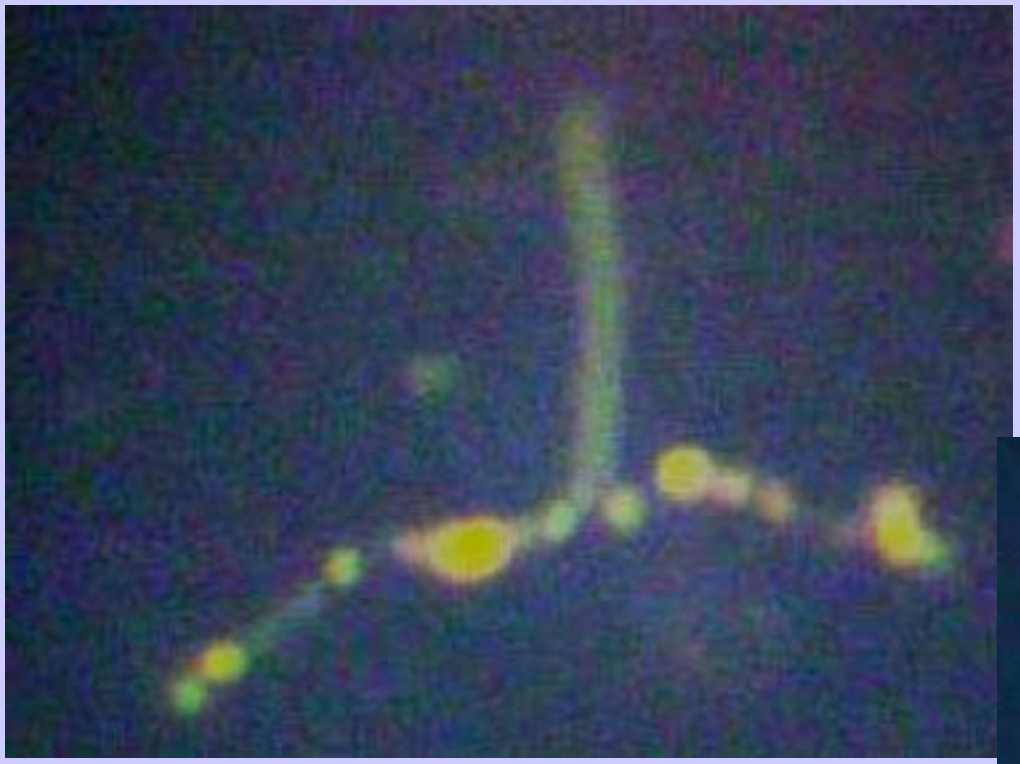
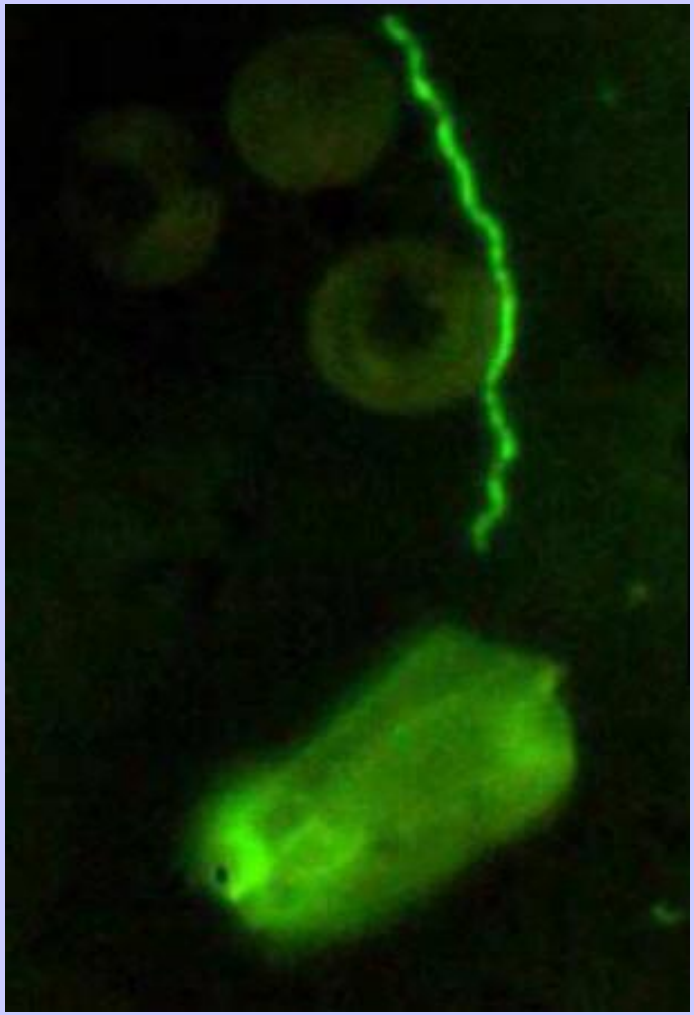


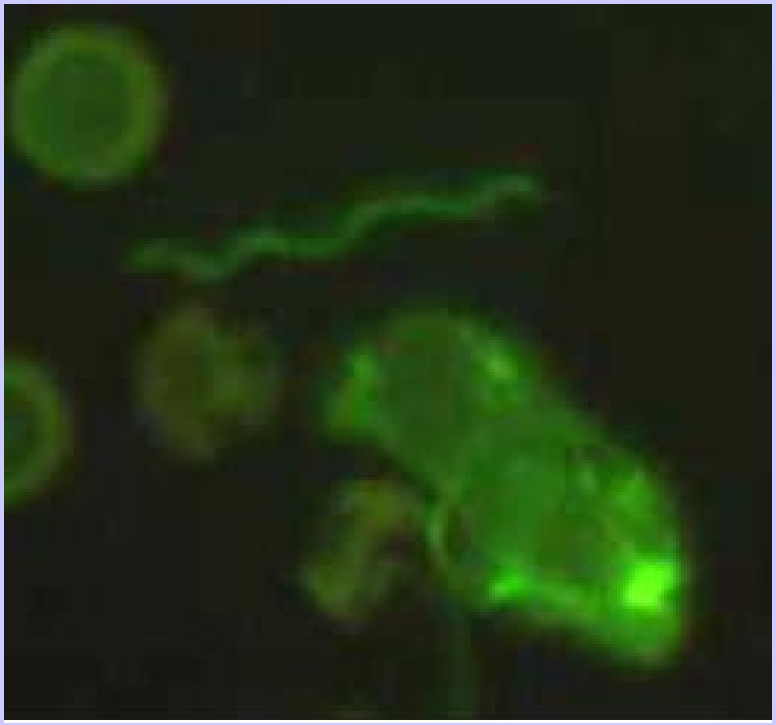
Figure 1 Cystic Forms of *Borrelia burgdorferi* (American Type Culture Collection 35210), darkfield image 1000x, original, Alan MacDonald, MD unpublished, Photograph date 1988

MacDonald – Borrelia Life Cycle



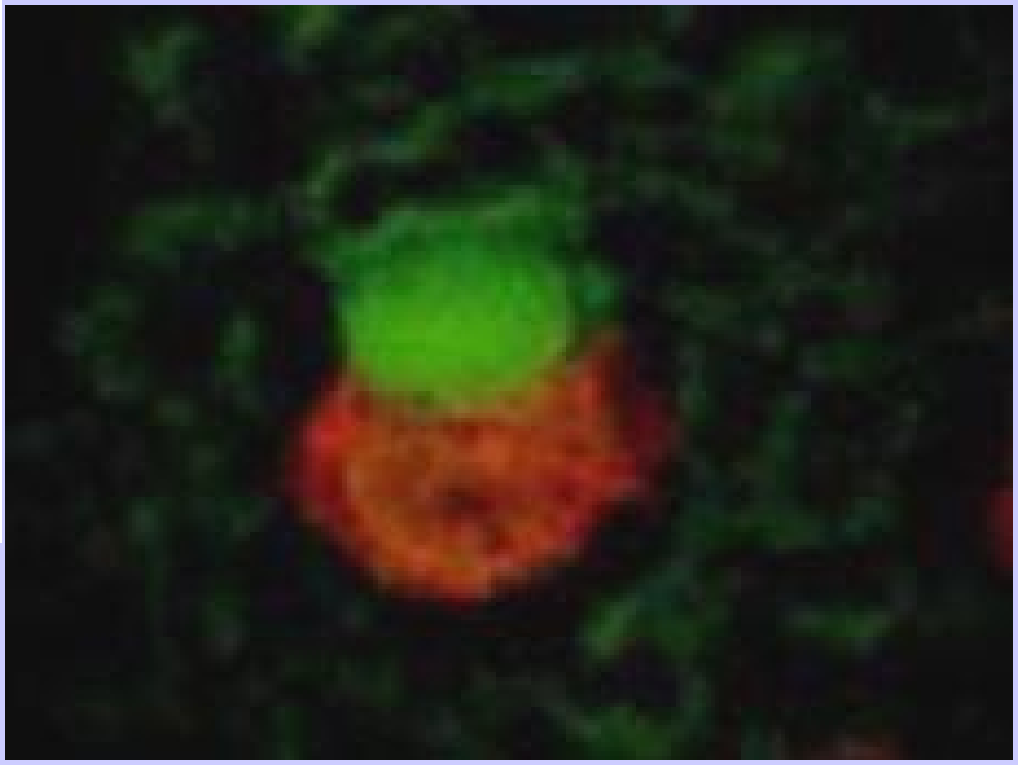
MacDonald – Borrelia Life Cycle



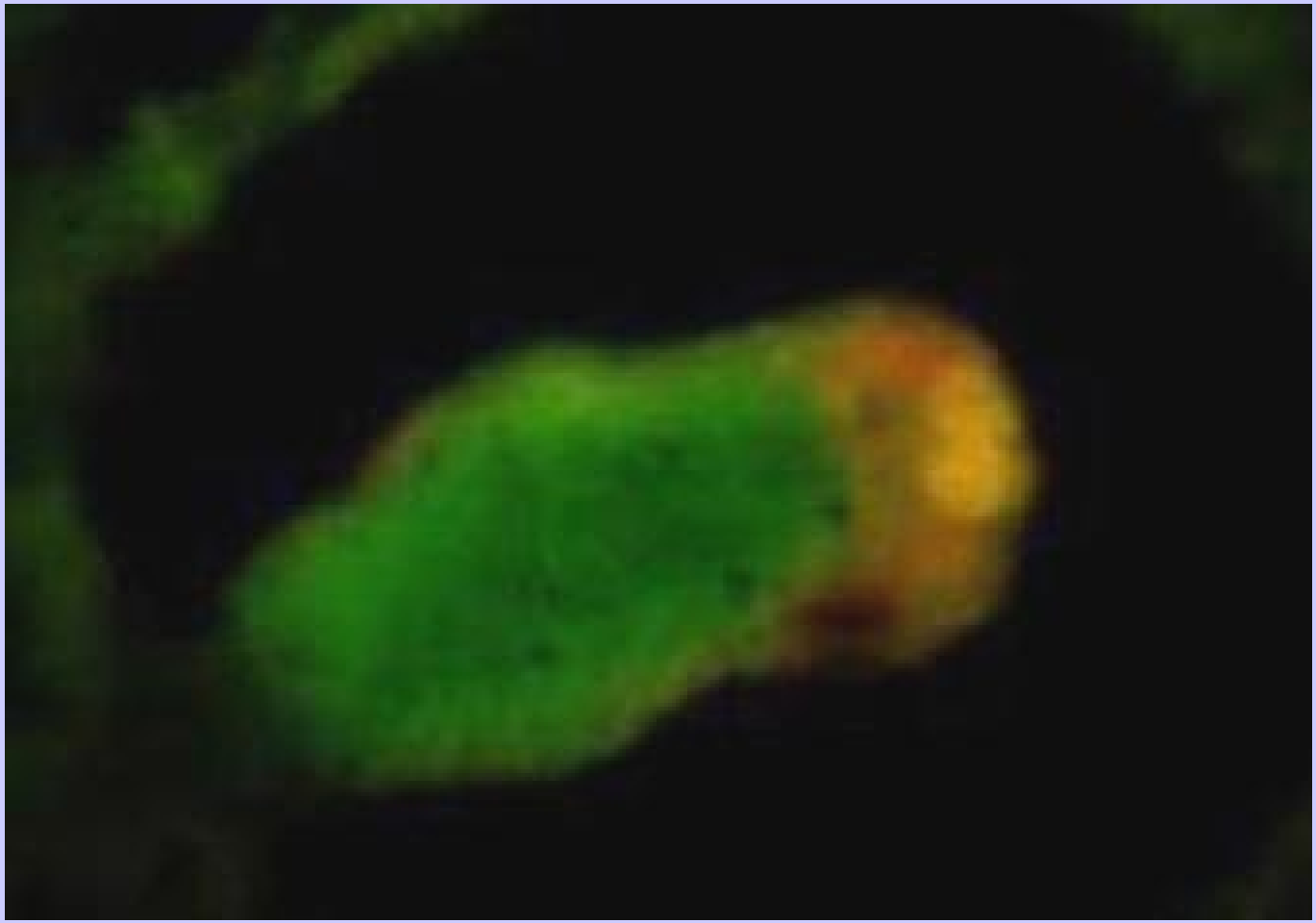


Module #3
Examples of
Borrelia
as Weapons

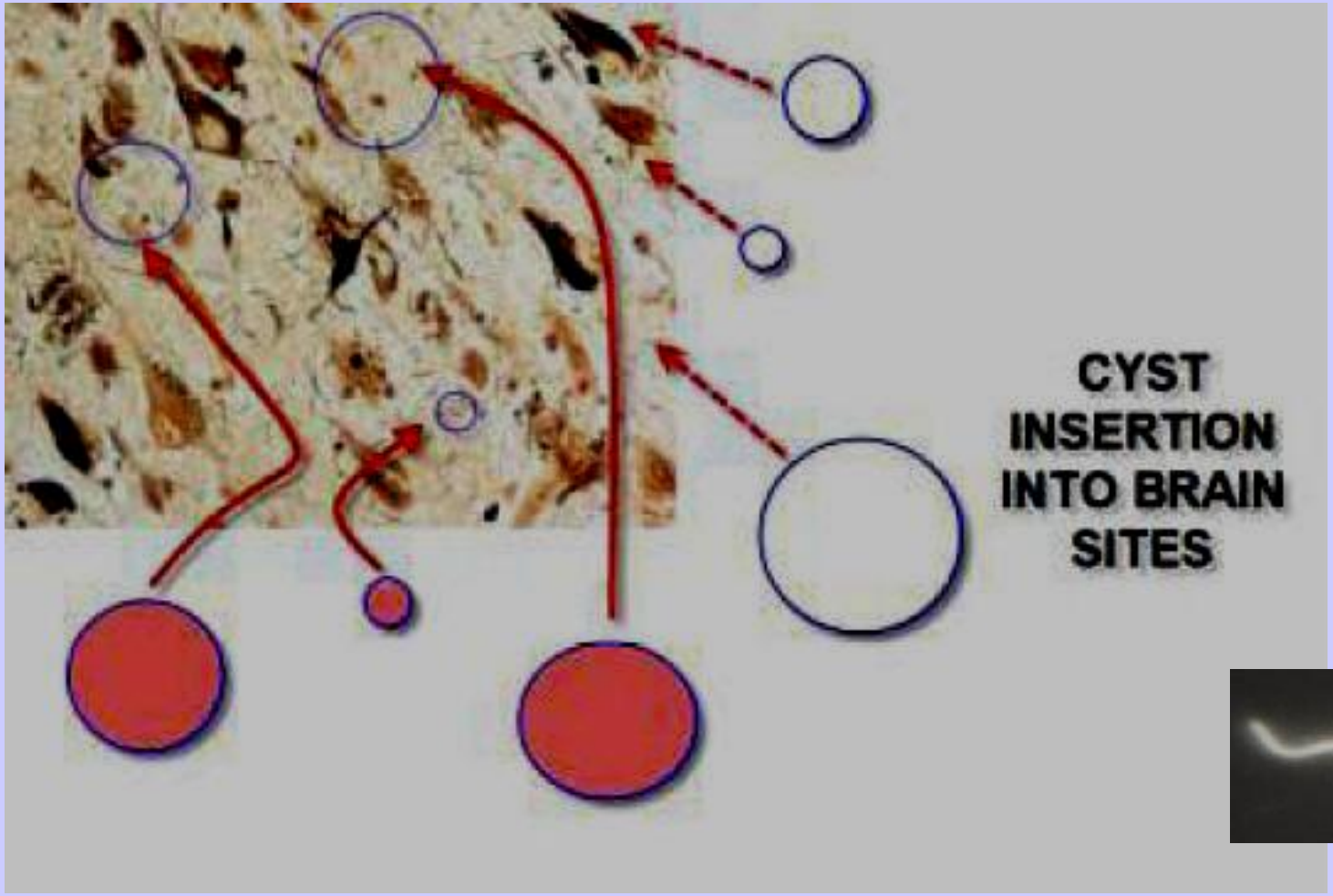
Borrelia Cysts as Weapons



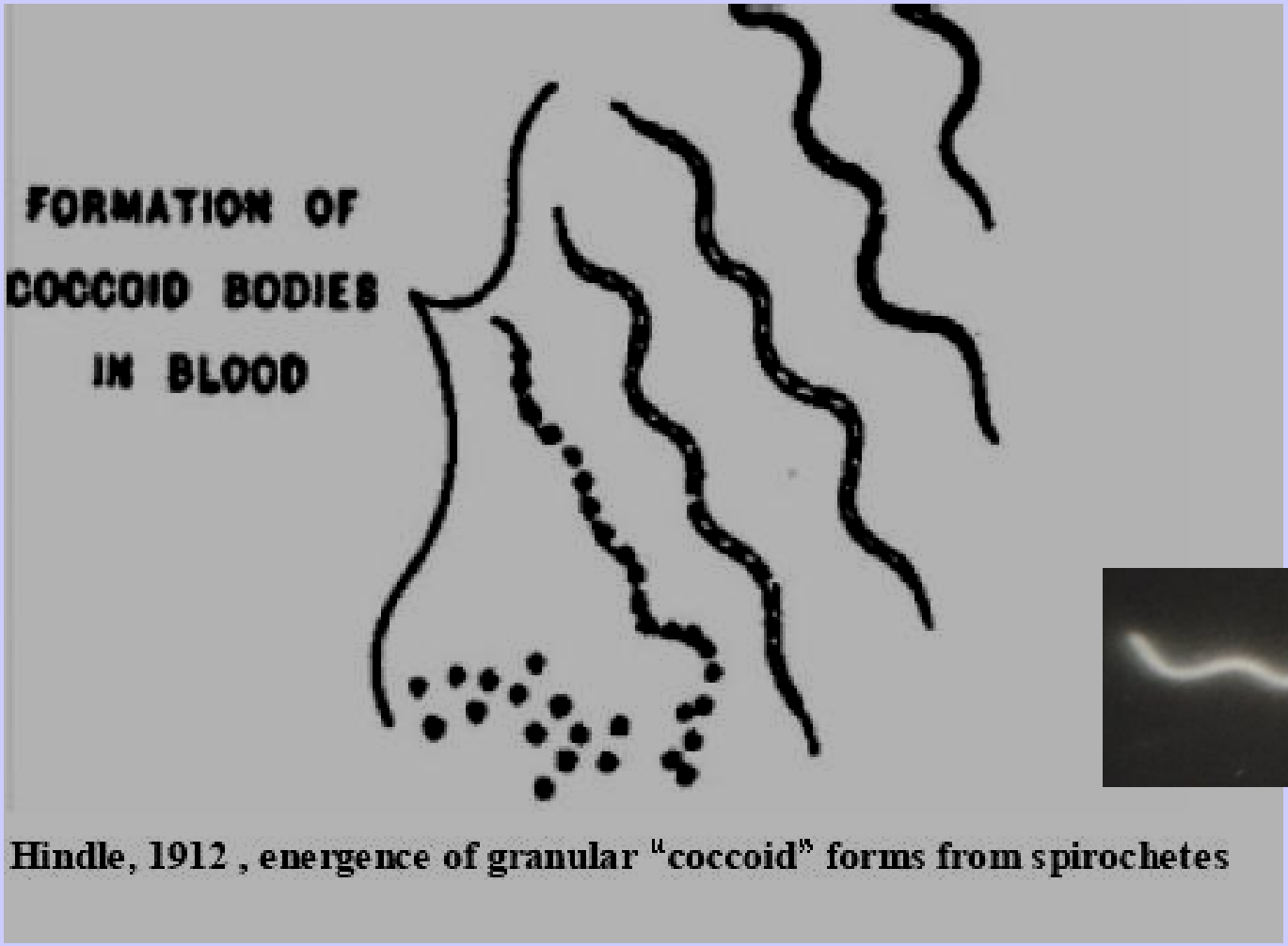
Borrelia Cysts as Weapons



Borrelia Cysts as Weapons

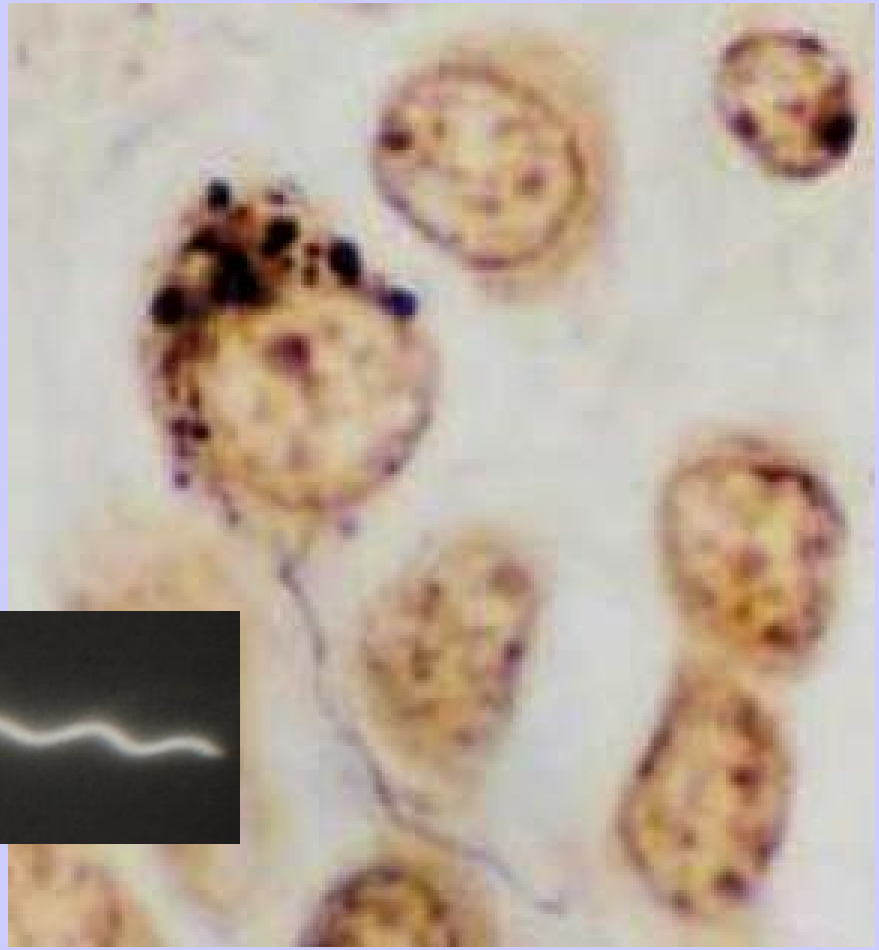


Borrelia Granules as Weapons

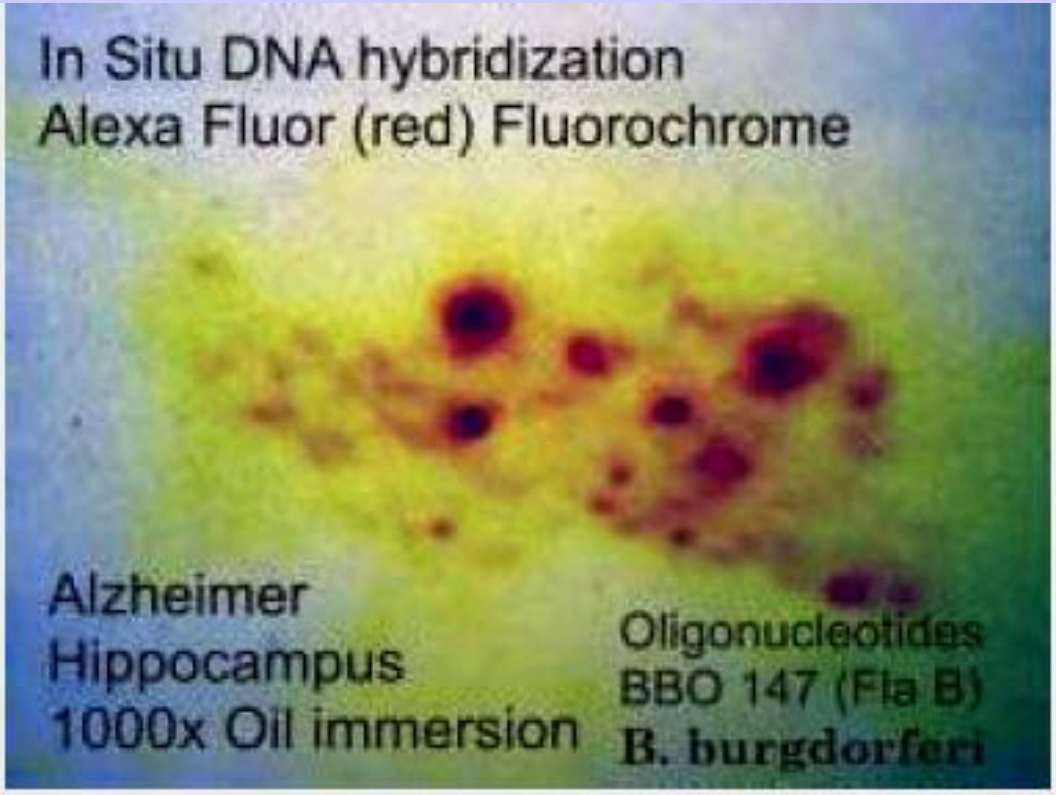


Hindle, 1912 , energence of granular "coccoid" forms from spirochetes

Borrelia Granules as Weapons

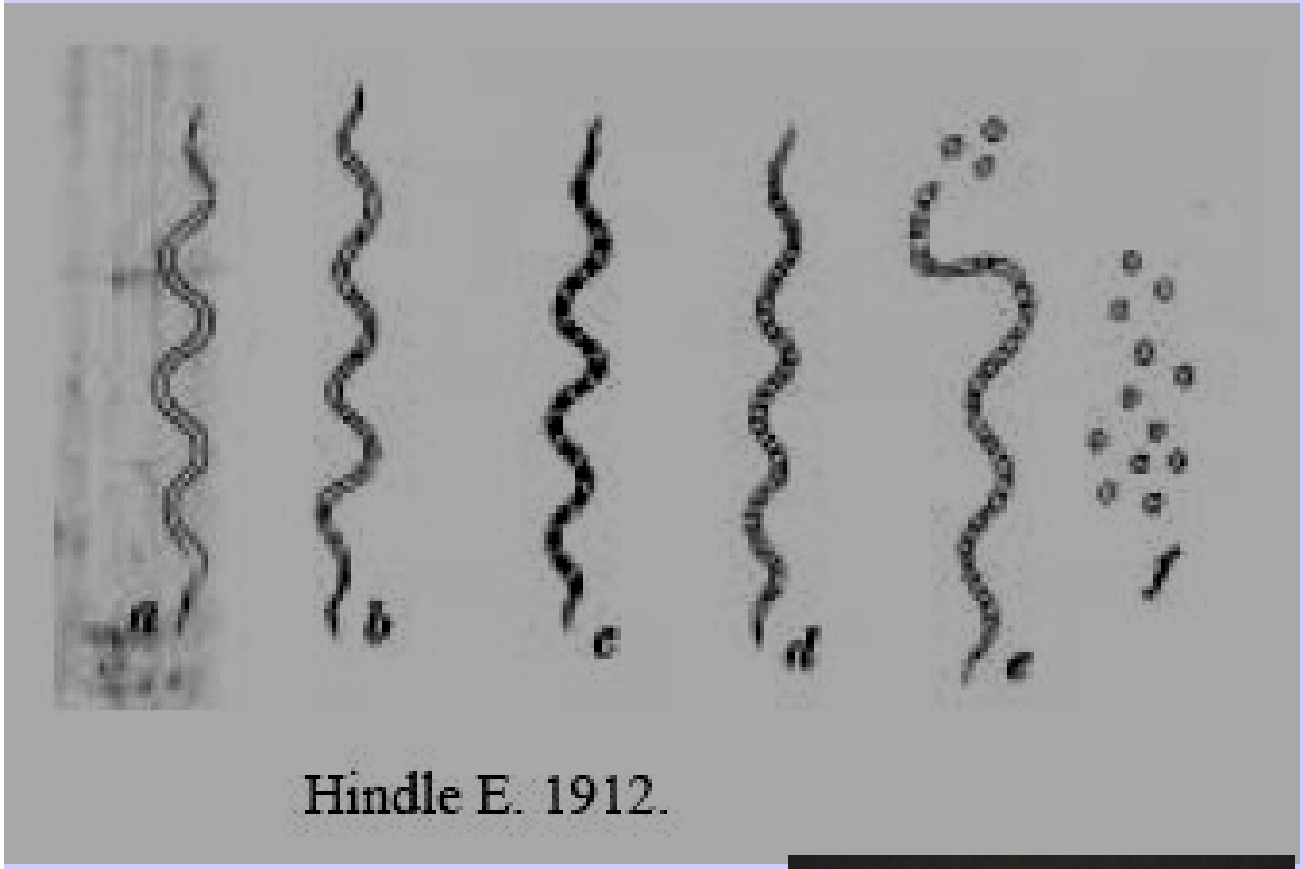


Borrelia Granules as Weapons

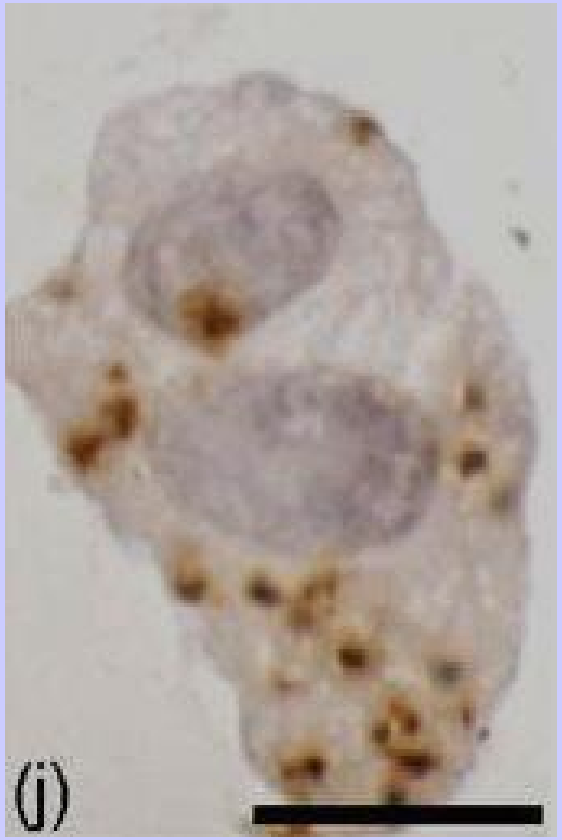


In situ DNA hybridization with flagellin DNA probes from *Borrelia burgdorferi*. Alzheimer hippocampus. Red signals within the cytoplasm of a nerve cell recapitulate the cytoplasmic profiles of granulovacuolar degeneration in Alzheimer's disease.

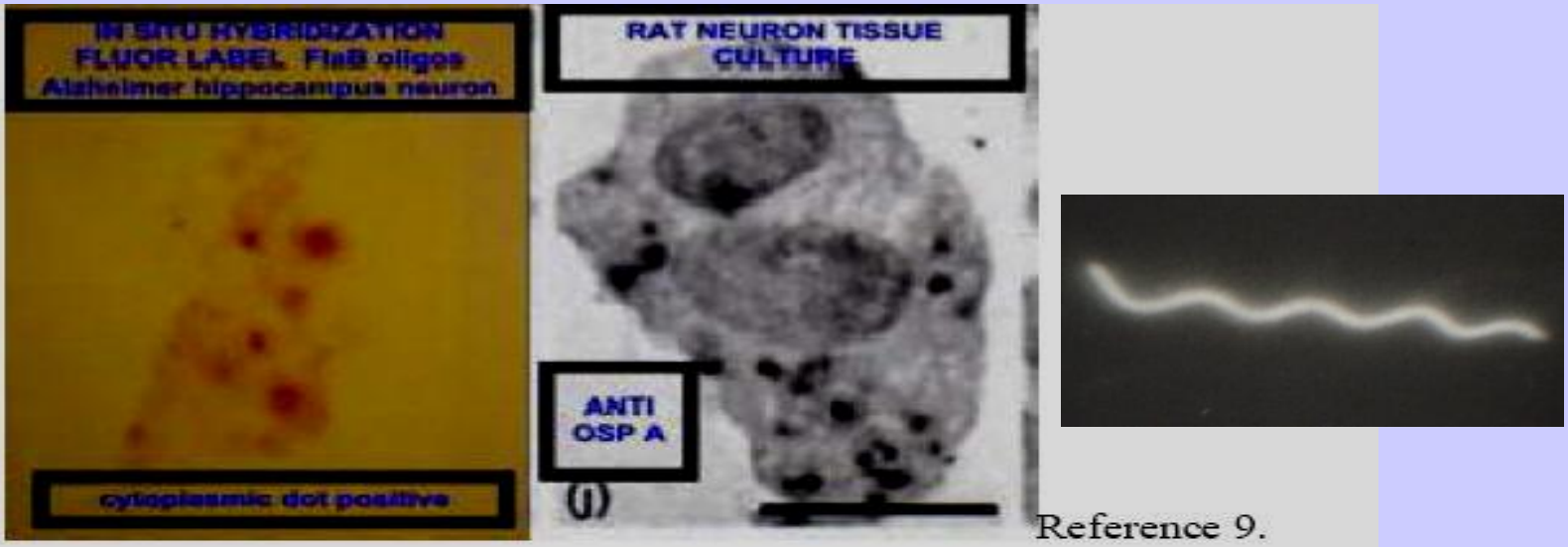
Borrelia Granules as Weapons



Hindle E. 1912.



Borrelia Granules as Weapons



Reference 9.

Comparison of intracellular *Borrelia* spirochetal profiles

Right panel shows rounded coccoid bodies within the cytoplasm of a rat nerve cell incubated in tissue culture with virulent Borrelia burgdorferi for several weeks to yield evidence of intracellular penetration of spirochete fragments (reactive with monoclonal antibody H5332, which uniquely recognizes an epitope of outer surface protein A (OspA) which defines Borrelia burgdorferi at the proteomic level. (Miklossy et al)
Left panel is a human Alzheimer nerve cell which shows positive signals in the cytoplasm in a granulovacuolar degeneration profile. (In situ DNA hybridization with flagellin B DNA of *Borrelia burgdorferi*.)

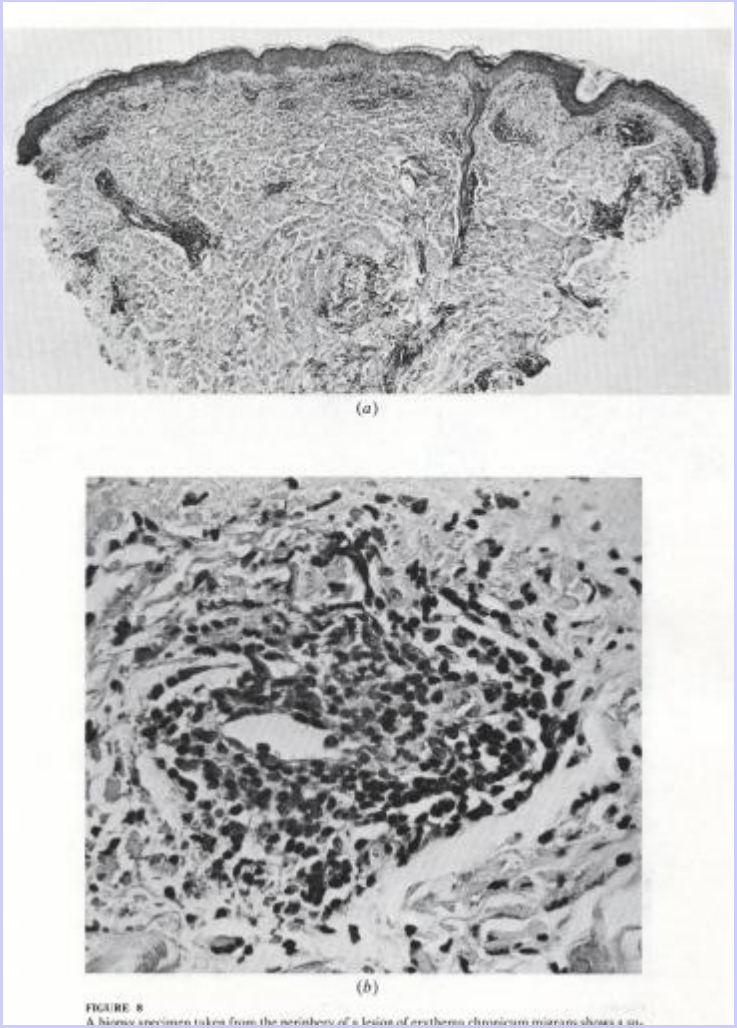
Module #4
Skin
as Target Site
for
Virulent
Borrelia

Erythema Migrans

GURE 1
tiny tick, *Ixodes dammini*, atop an early papule of erythema
ronicum migrans.

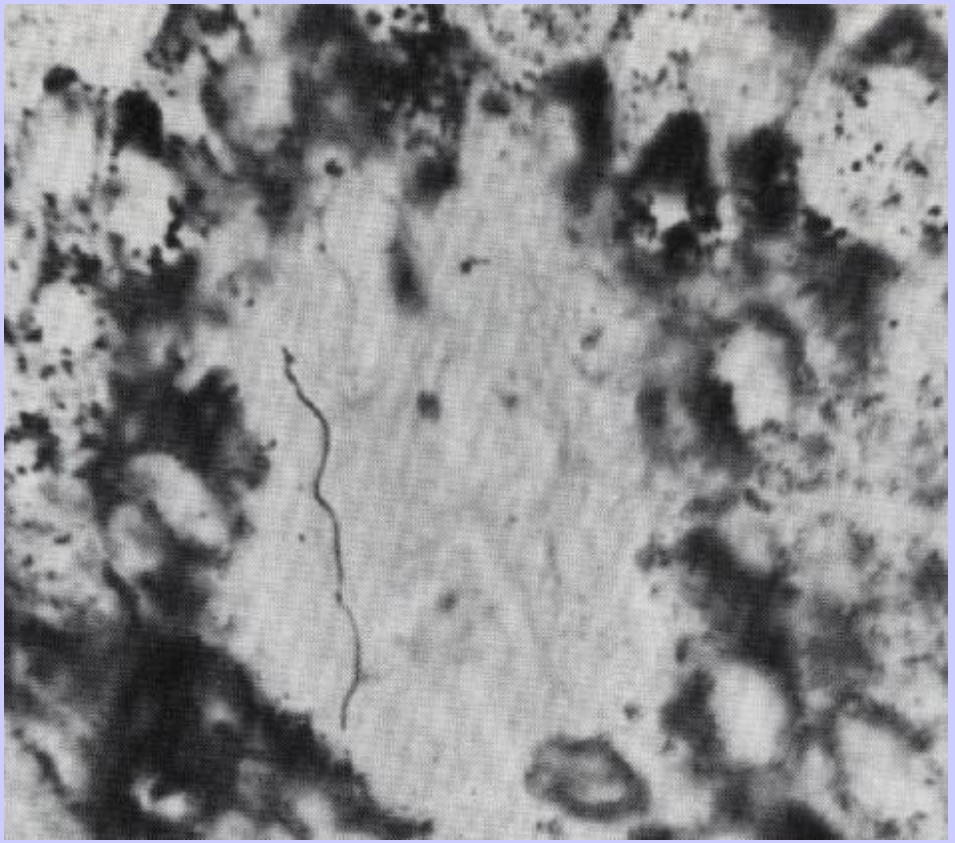
FIGURE 4
Two somewhat oval plaques of erythema chronicum migran

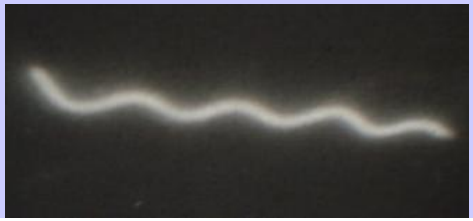
Borrelia in Erythema migrans


(a)

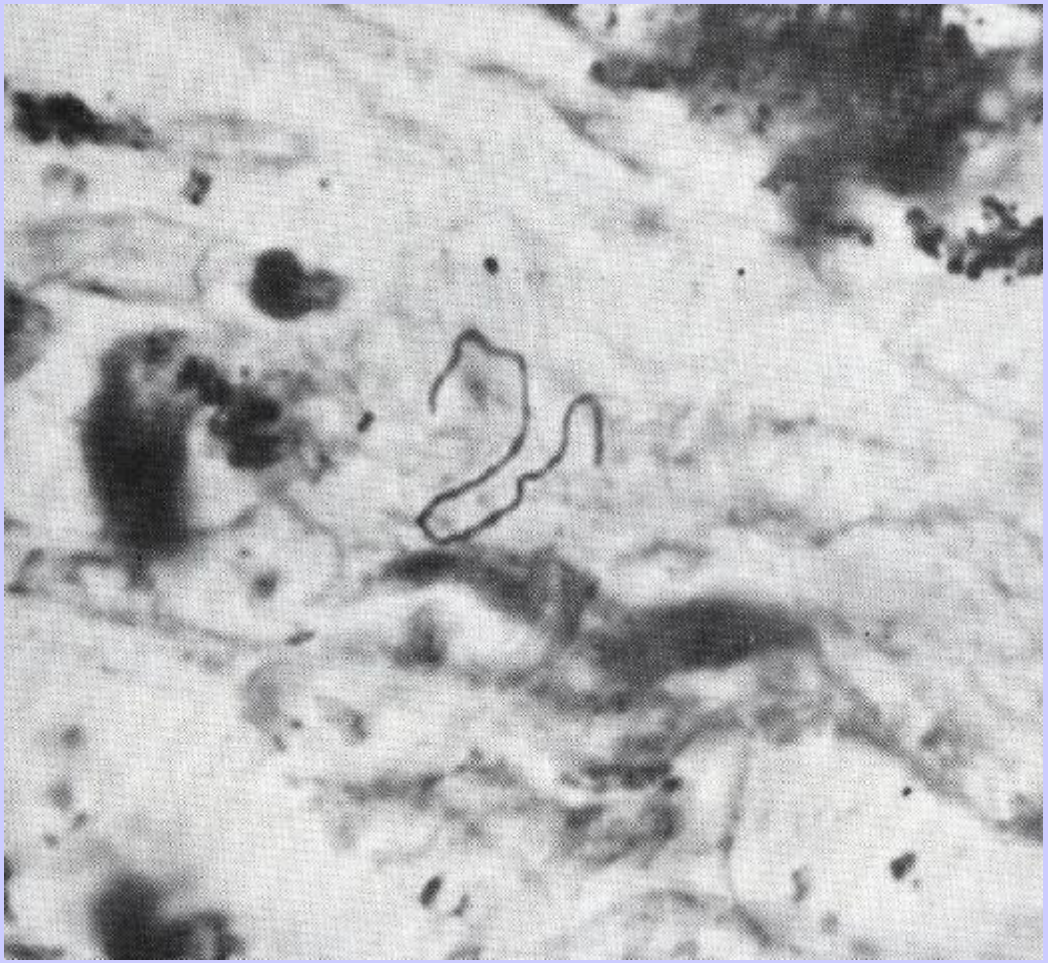
(b)

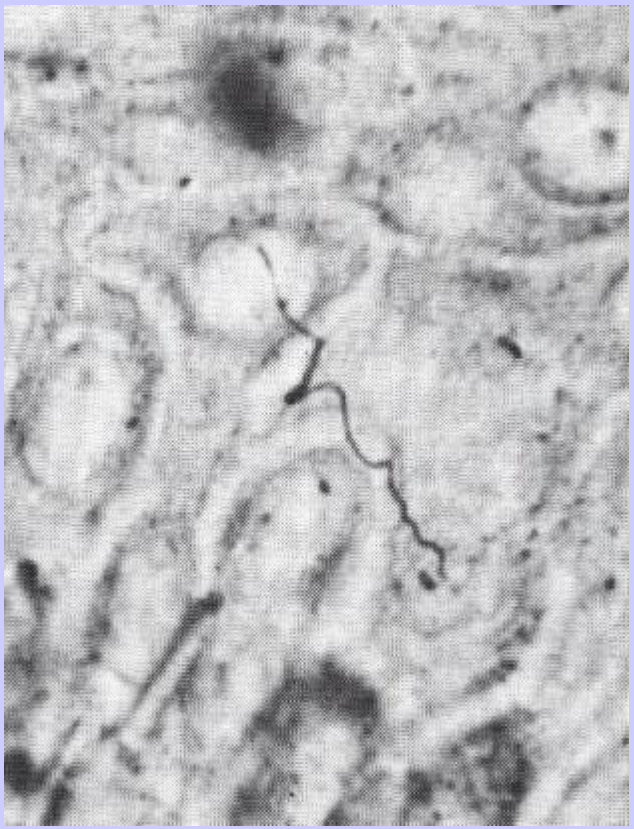
FIGURE 8

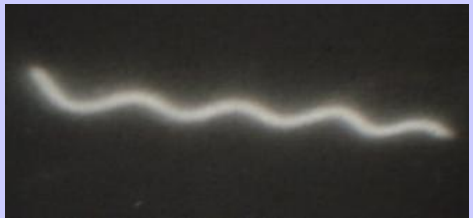




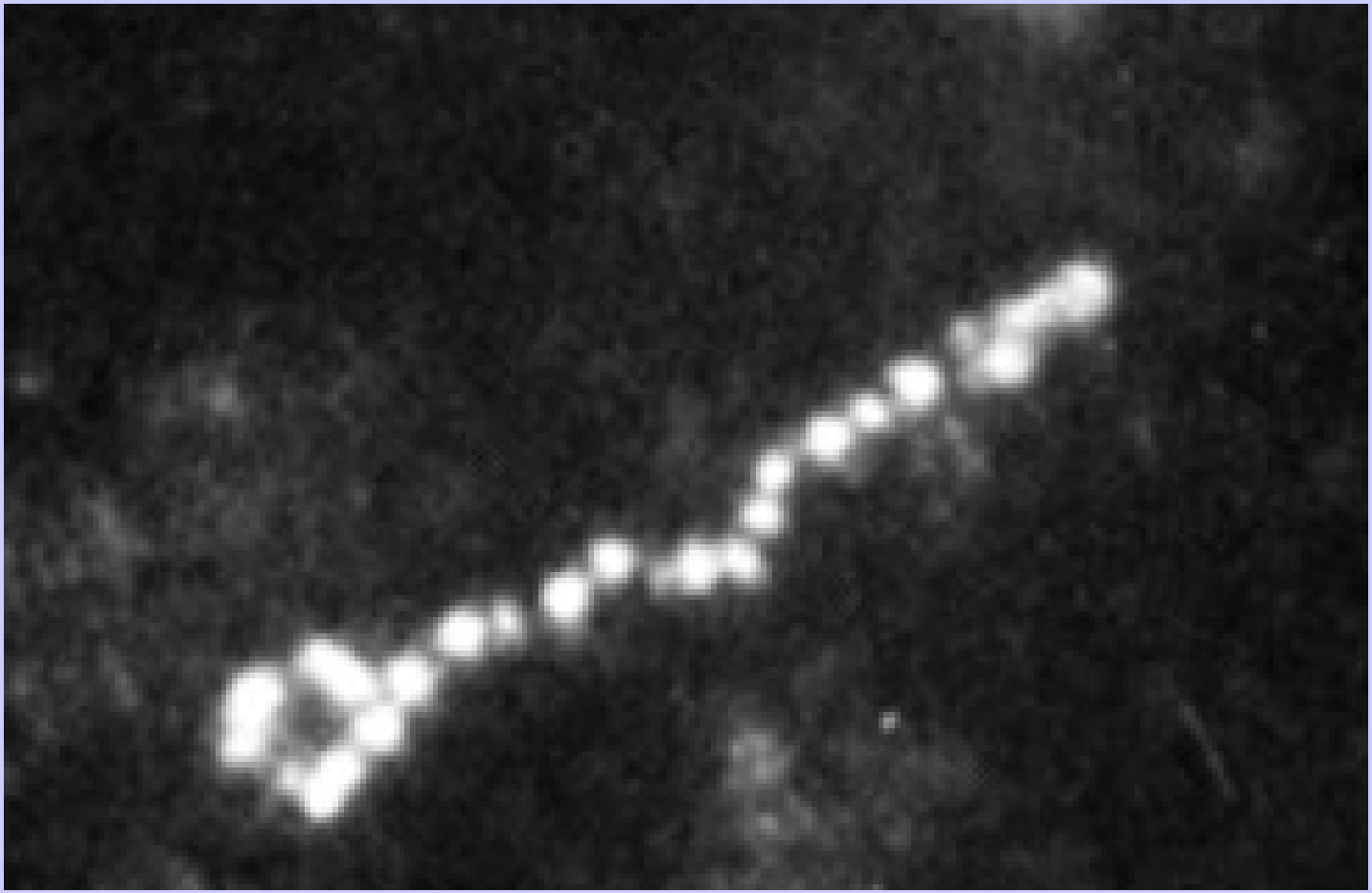
Borrelia in Erythema migrans



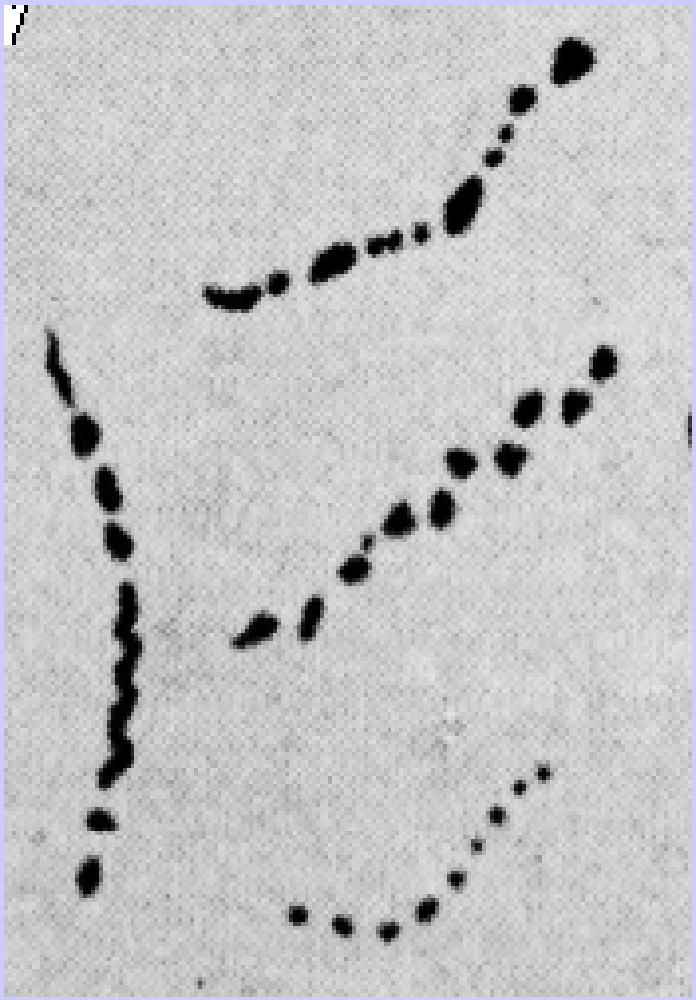


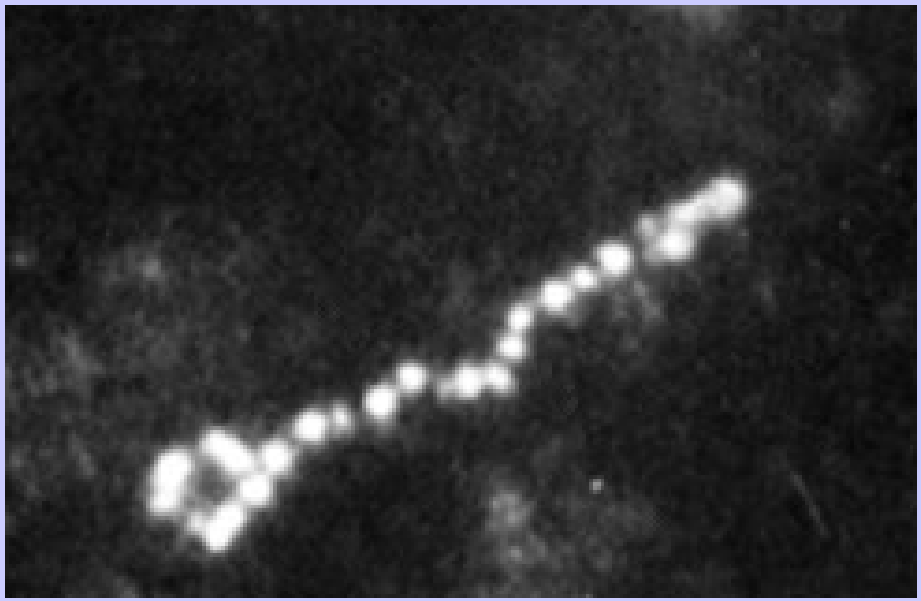


Is this a Borrelia spirochete?



Is this a Borrelia spirochete?



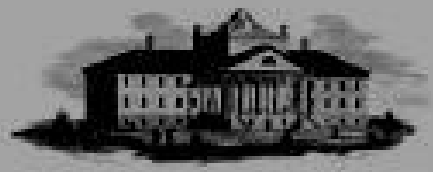


Module #5
Brain and Spinal Cord as Targets of Virulent Borrelia

Borrelia in the Brain

The NEW ENGLAND JOURNAL of MEDICINE

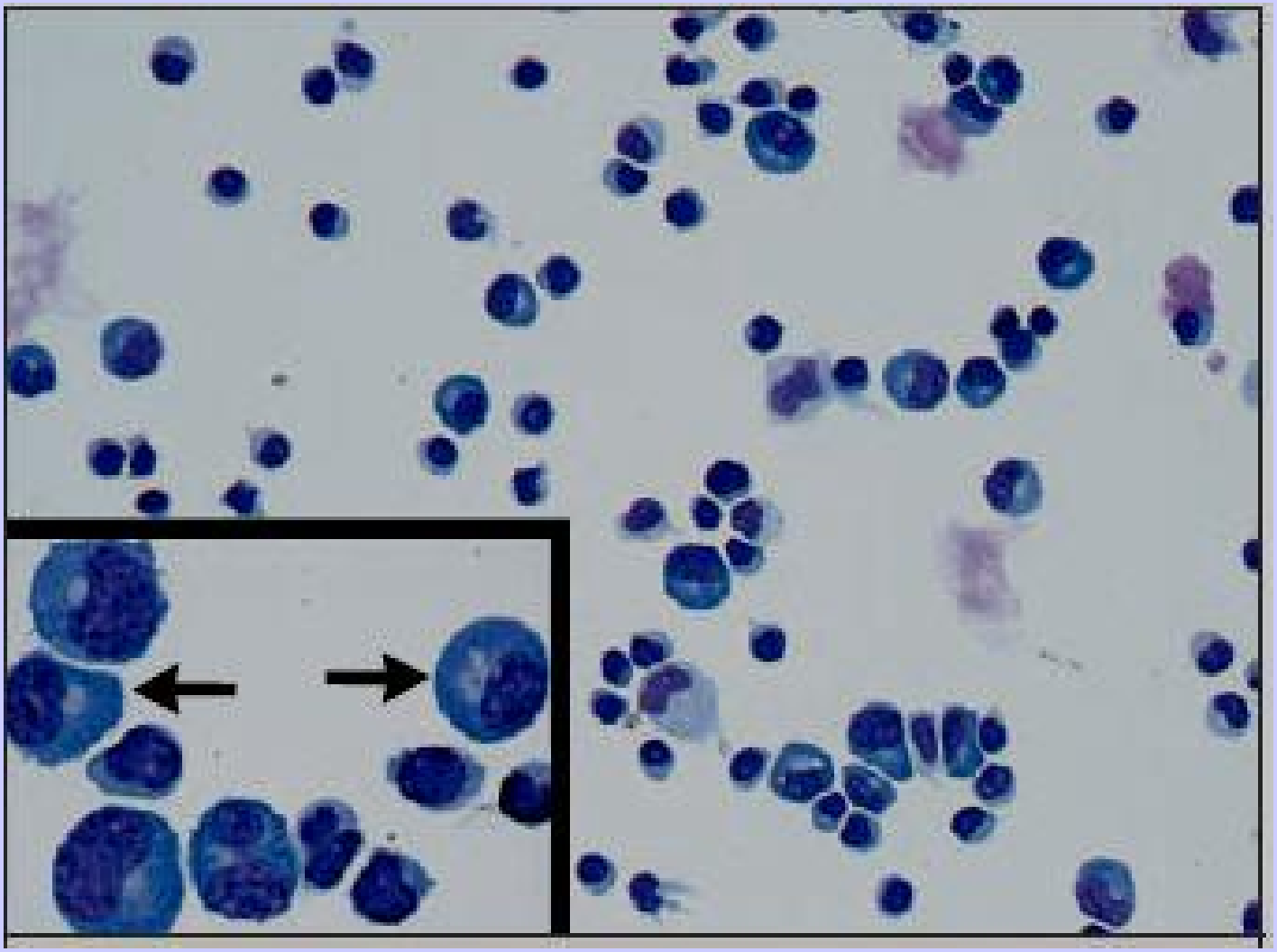
CASE RECORDS of the MASSACHUSETTS GENERAL HOSPITAL



Case 11-2007: A 59-Year-Old Man with Neck Pain, Weakness in the Arms, and Cranial-Nerve Palsies

Borrelia in the Brain

Borrelia in the Brain



Brain Monkey Model of Dr Fikrig

Skin – Hypodermic Needle Injection mimics Bite site

Autopsy Study of Monkey months later

BRAIN and Heart sites

Site Specific Molecular “signatures” of the Spirochetes

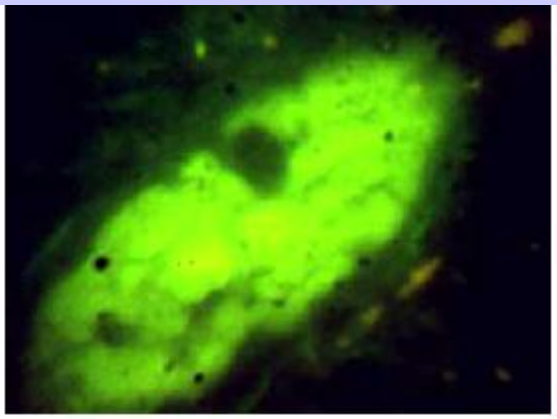
Brain signature differs from Cardiac signature

Brain Monkey Model of Dr Fikrig

Fig. 3. Confirmation of the expression profiles. (A) cDNA prepared from

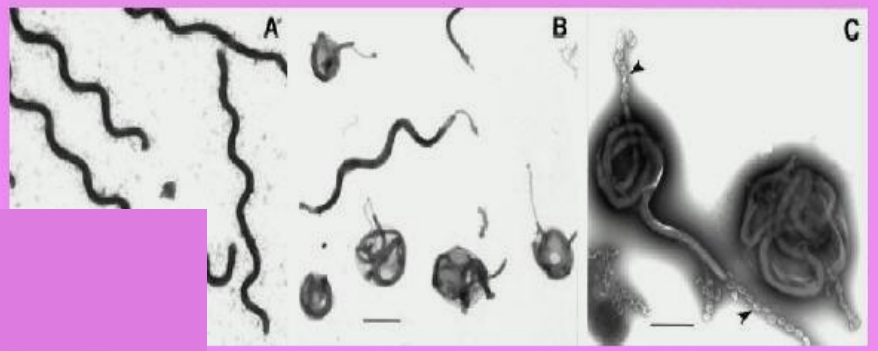
Fikrig's Gene probes for DNA of Bb

Brain Alzheimer Model of Dr MacDonald



Borrelia burgdorferi **Flagellin DNA . In situ hybridization. Large Plaque**
1000x original magnification

Nelson – Dynamic equilibrium of Borrelia burgdorferi cysts to vegetative forms

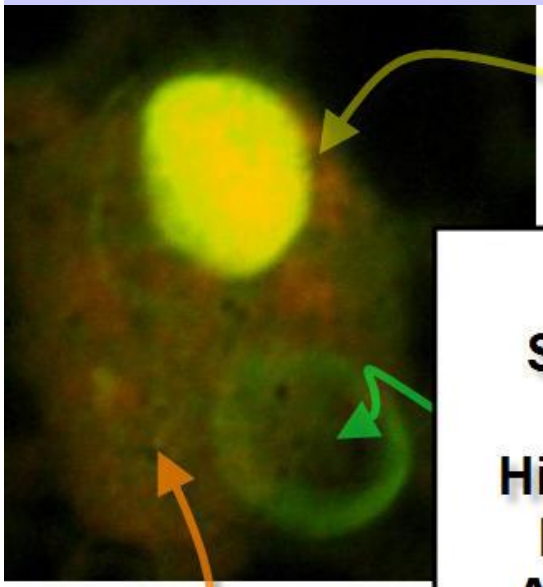


plets" - eligible to participate in a "splash"

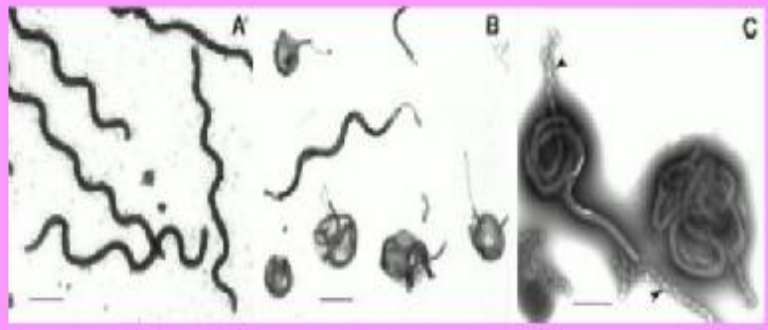
Nelson Cystic "stress altered" form

Brain Alzheimer Model of Dr MacDonald

Invade, Penetrate, Injure or Kill



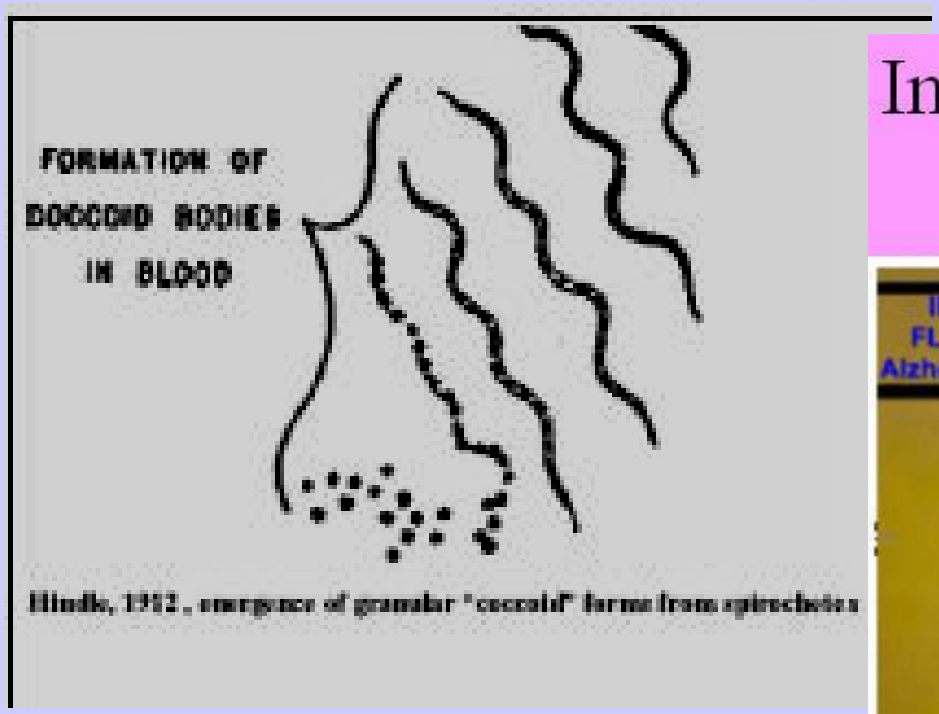
Nelson – Dynamic equilibrium of Borrelia burgdorferi cysts to vegetative forms



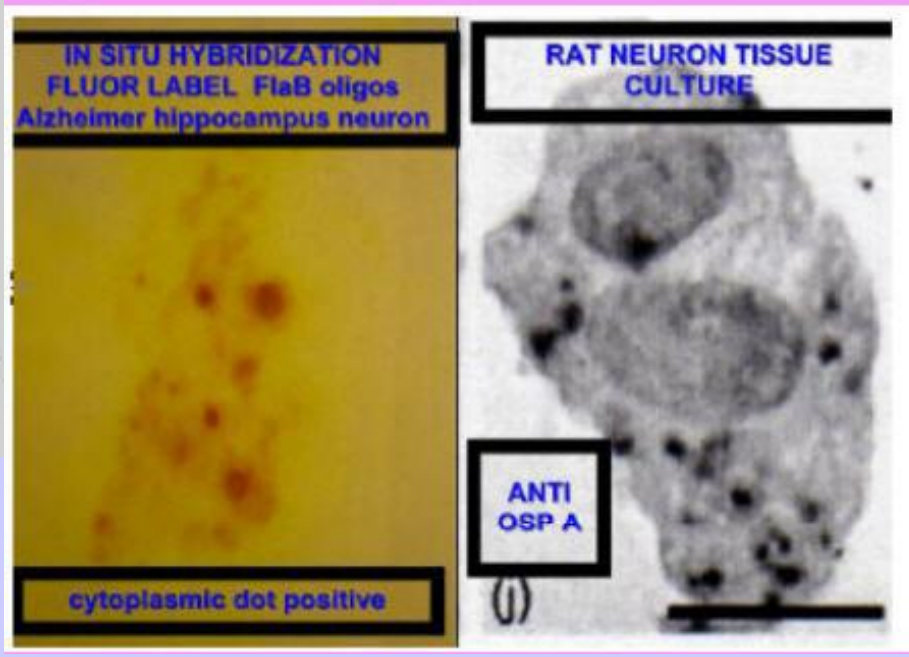
Cysts as "droplets" - eligible to participate in a "splash"

Brain Alzheimer Model of Dr MacDonald

Invade, Penetrate, Injure or Kill



In Situ Dna and Rat Tissue culture



Module #6 Immunologic Diversity of Borrelia and its Genetic Basis

Antigenic Variation in Borrelia

" In Lyme disease Borrelia, antigenic variation involves segmental gene conversion…..Gene Conversion events involve replacement of random, variable length segments…As a result, mammals could harbour ***Thousands of different variants at any one time…*** confounding efforts by the immune system to keep up with the sequence variation."

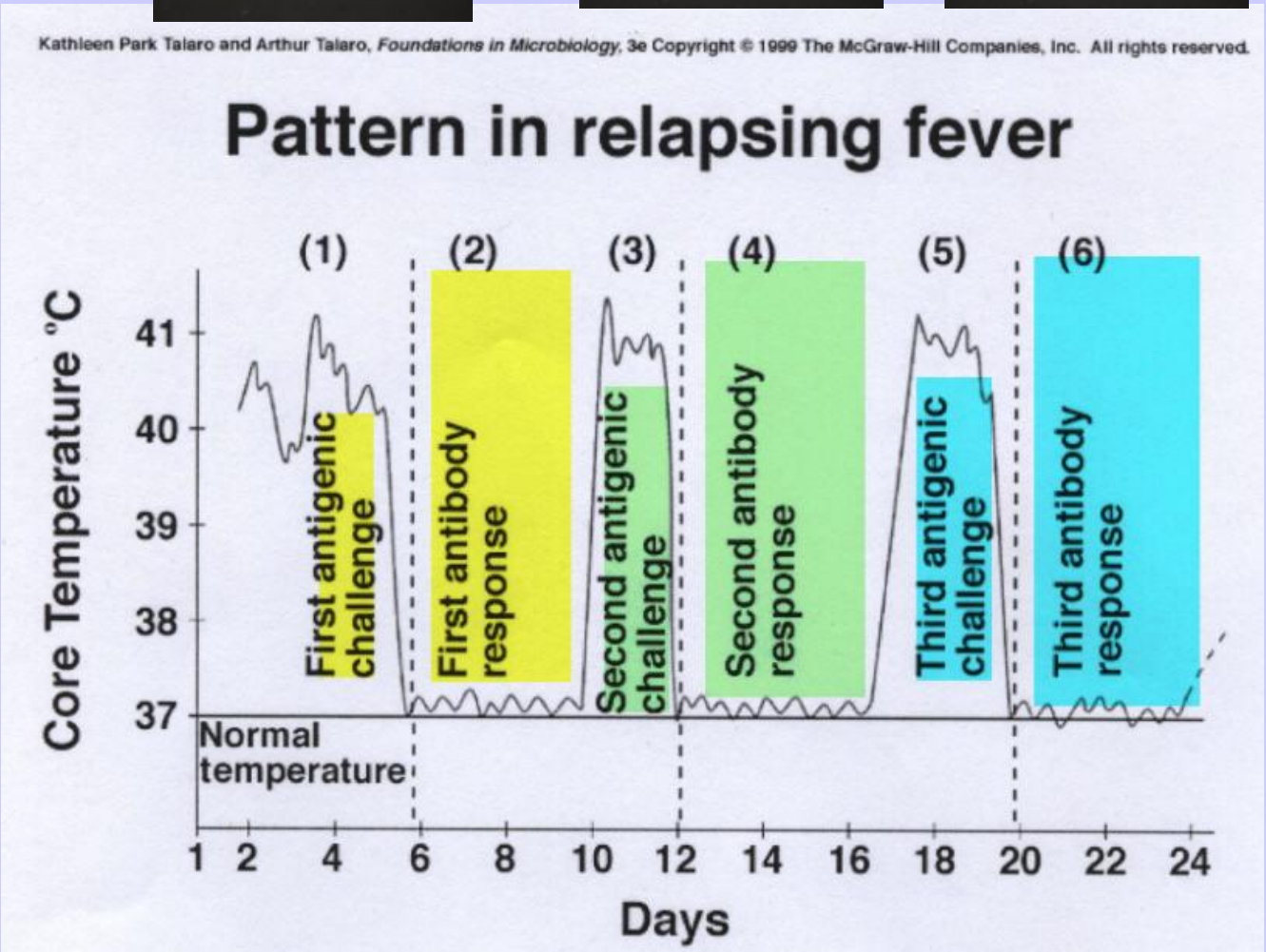
Dr Steven Norris

Professor, University of Texas Medical School at Houston

Molecular Microbiology, 2006

Antigenic Variation in Borrelia

Re  g f   Bo



Antigenic Variation in Trypanosome

SEQUENCE OF EVENTS

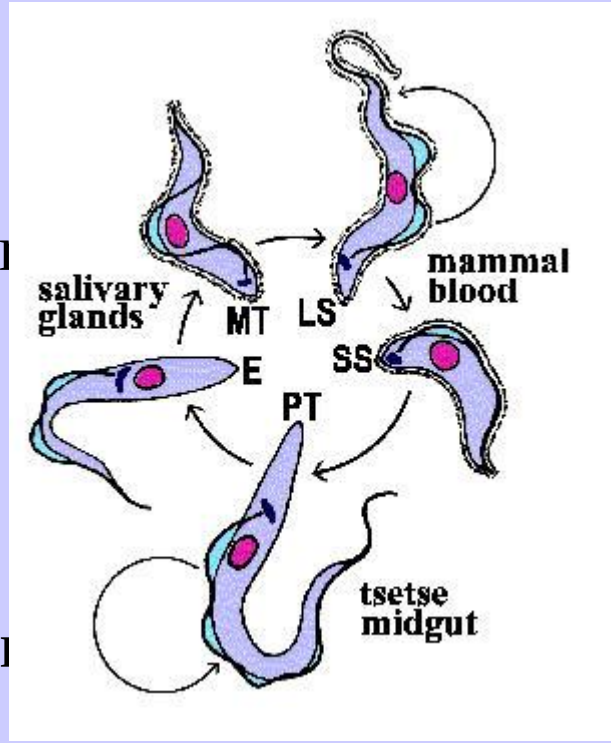
Skin Bite by Arthropod Vecto

Chancre

Blood Forms of Trypanosome

Relapses of Trypanosomes i

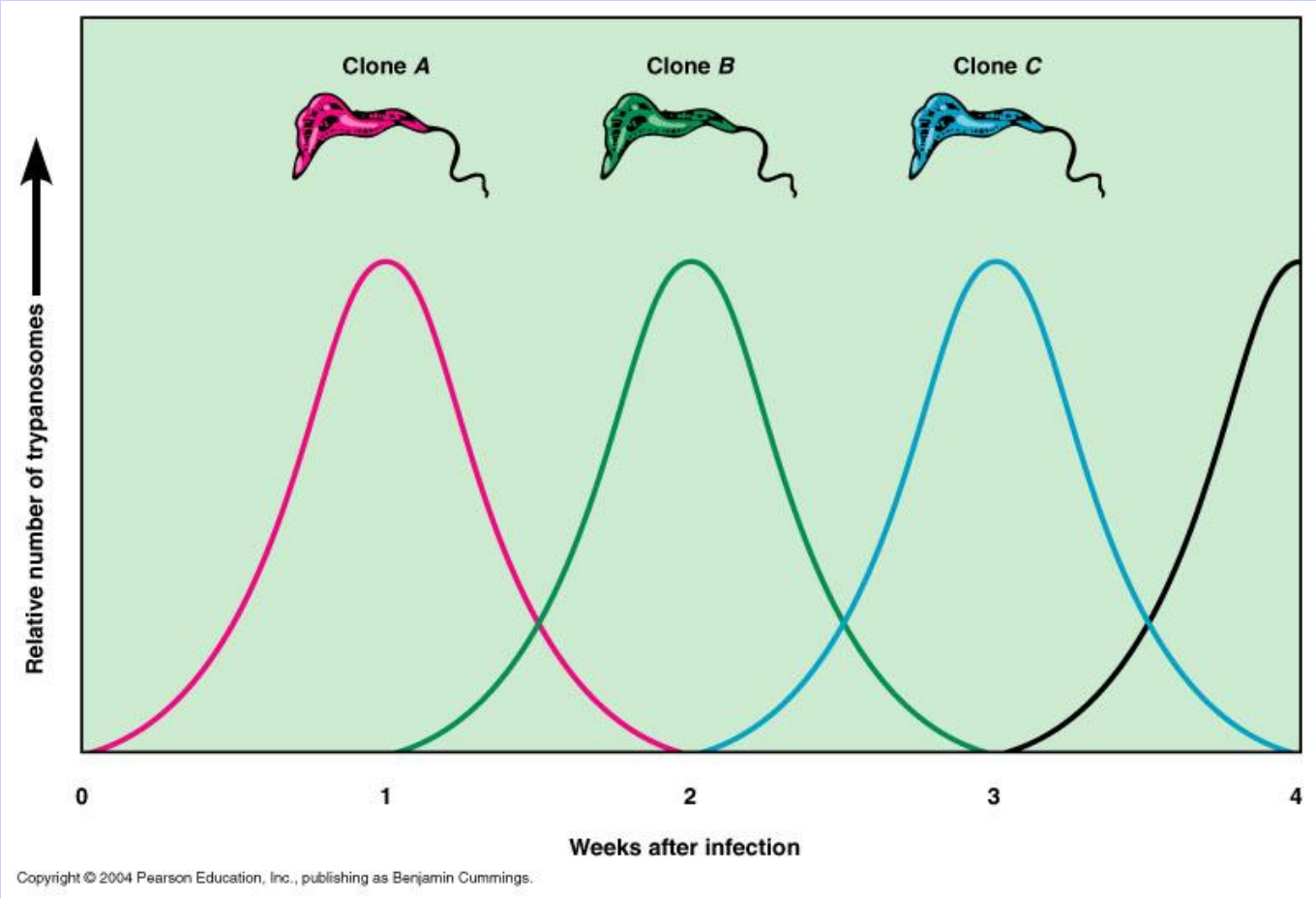
with Fever Spikes



Invisible Trypanosomes in Brain after Death from

Sleeping Sickness

Antigenic Variation in Trypanosome



Module #7
Borrelia
in the
Pregnant patient

Pregnancy Borrelia

Skin bite Site – (no Chancre) - Erythema Migrans Absent

Blood Phase – Spirochetes travel through Umbilical Cord to Reach the Placenta (No Barrier in Placenta)

Placental Villi – Oxygen, Carbon Dioxide, Spirochetes

Infection Sequelae in the Developing Fetus

Miscarriage, Malformation, Stillbirth, Sudden Death of Infant in 1st year,

Tertiary Lyme Borreliosis in Youth or Adolescence

Pregnancy Borrelia

Antibodies

From Mother

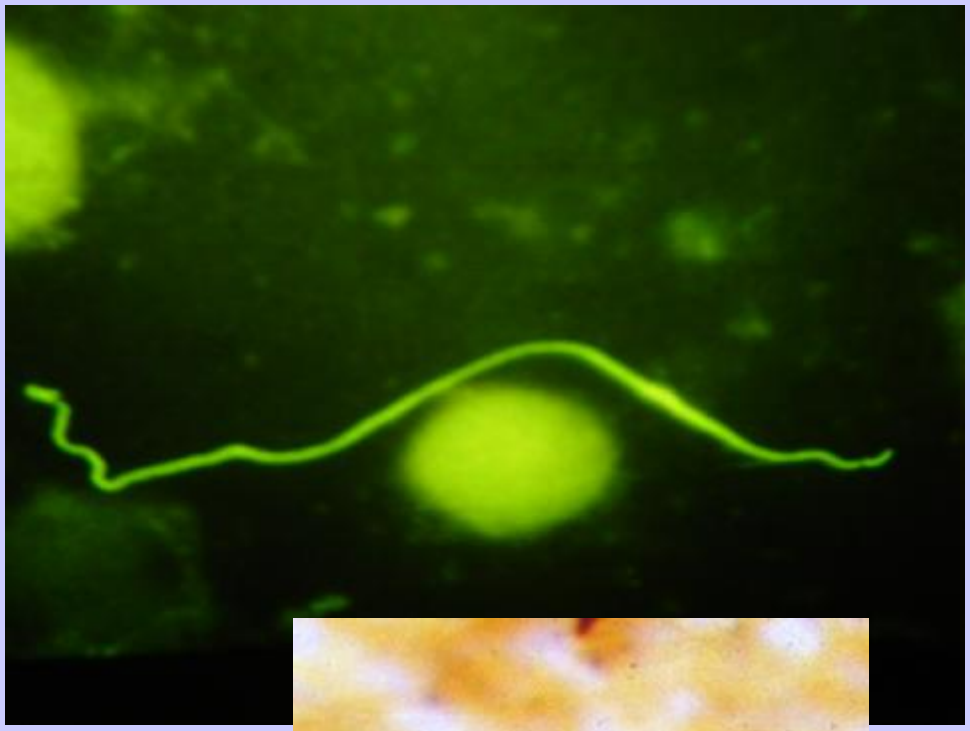
From Fetal Immune Response to intrauterine infection

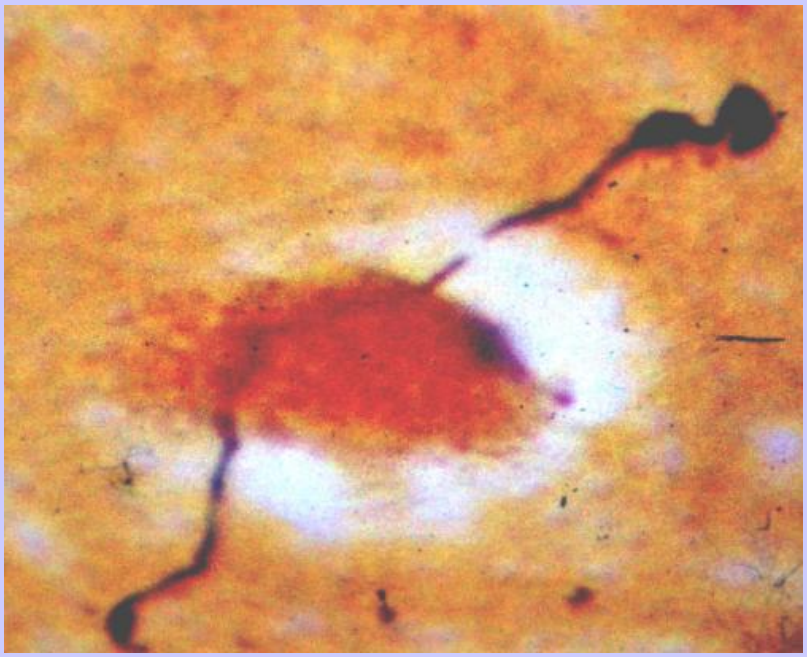
Pregnancy and the Maternal Immune System - Down Regulated

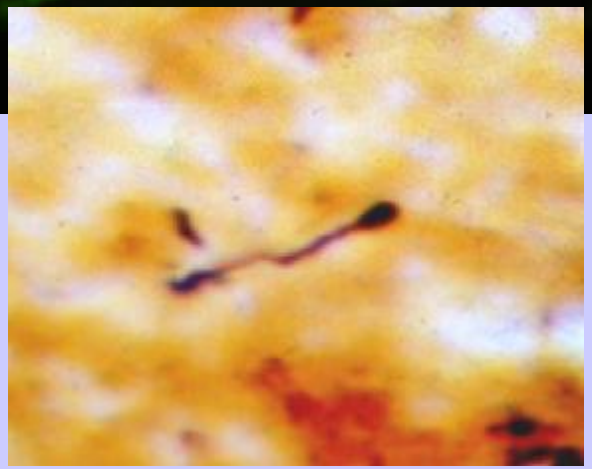
Teratogen Issues in the Developing Fetus –

Rubella infection model for
Intrauterine Infections

Pregnancy Borrelia



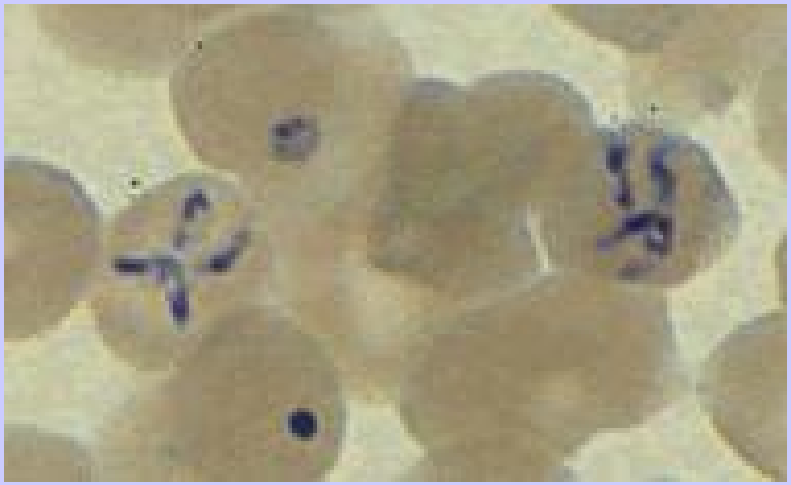




Module #8
Multiple Infections in the Borrelia Attack Model

Multiple Infections Borrelia

Co - Infections



Babesia

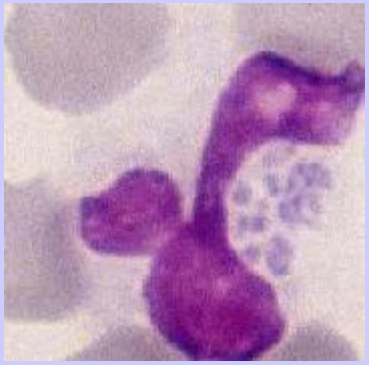
Ehrlichia

Bartonella

Anaplasma

Theileria

Mycoplasma



Multiple Infections Borrelia

Repeat ARTHROPOD ASSAULT

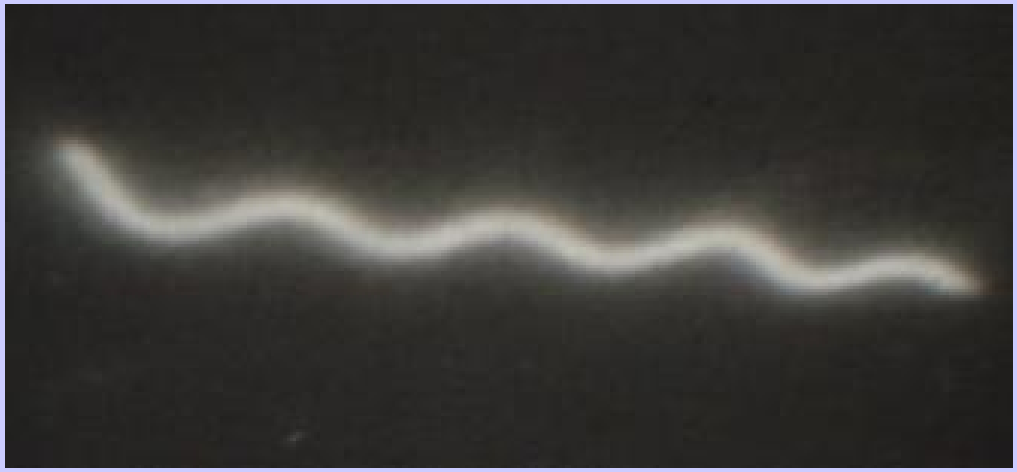
- Mosquito
- Biting Flies
- Ticks

Blood Transfusion

Organ Transplantation

??Gastrointestinal Routes

Venereal routes



Module #9
Latency
in
Borrelia
Infections

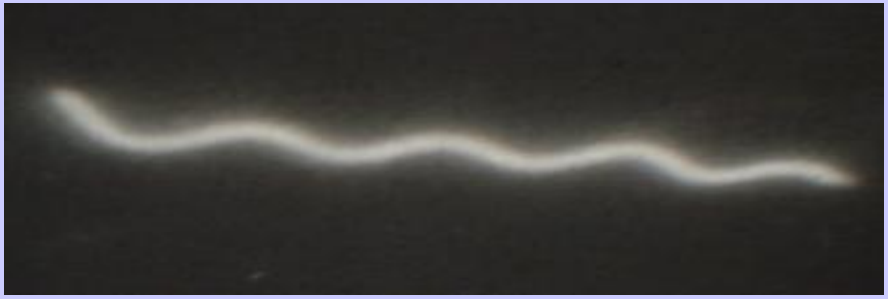
Latency Model for Borrelia

Spirochete – (Borrelia burgdorferi) – Resting State

Months

Years

Decades



“Bad Blood” -- No Blood Donation??????

??Issue of the eligibility for blood donation in Borreliosis patients??

Latency Model for Syphilis

Spirochete – (Treponema Pallidum) – Resting State

Months

Years

Decades

“Bad Blood” -- No Blood Donation ever if you have had syphilis

Latency – Other Microbes

Whipple's Disease

Tuberculosis

Chicken Pox – Varicella Zoster

Herpes Simplex (Cold Sore)

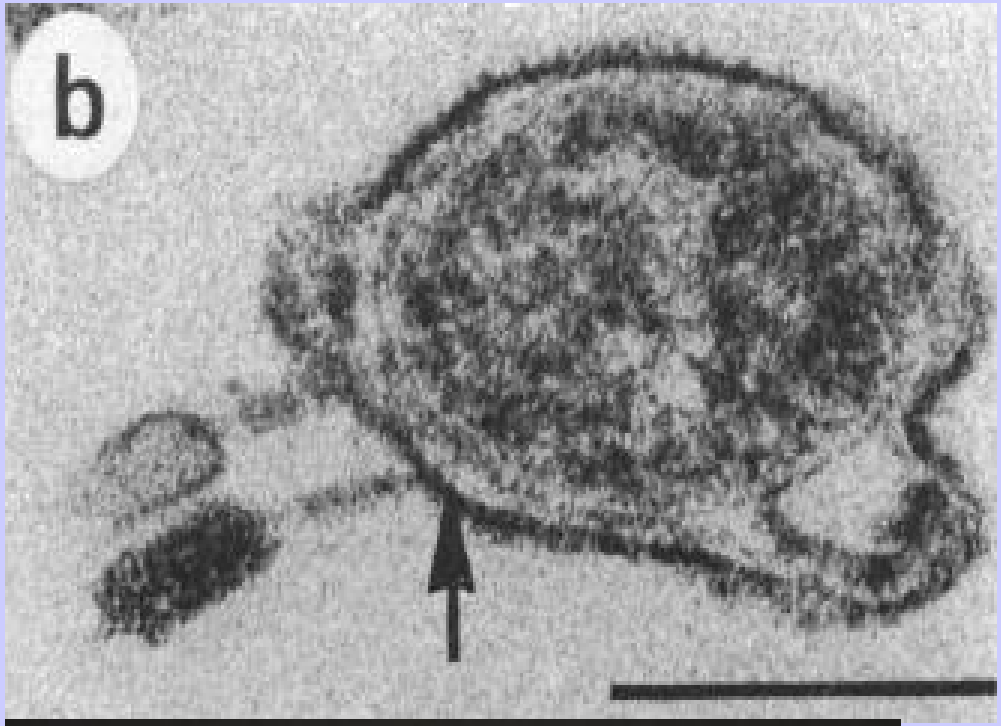
Borrelia Burgdorferi

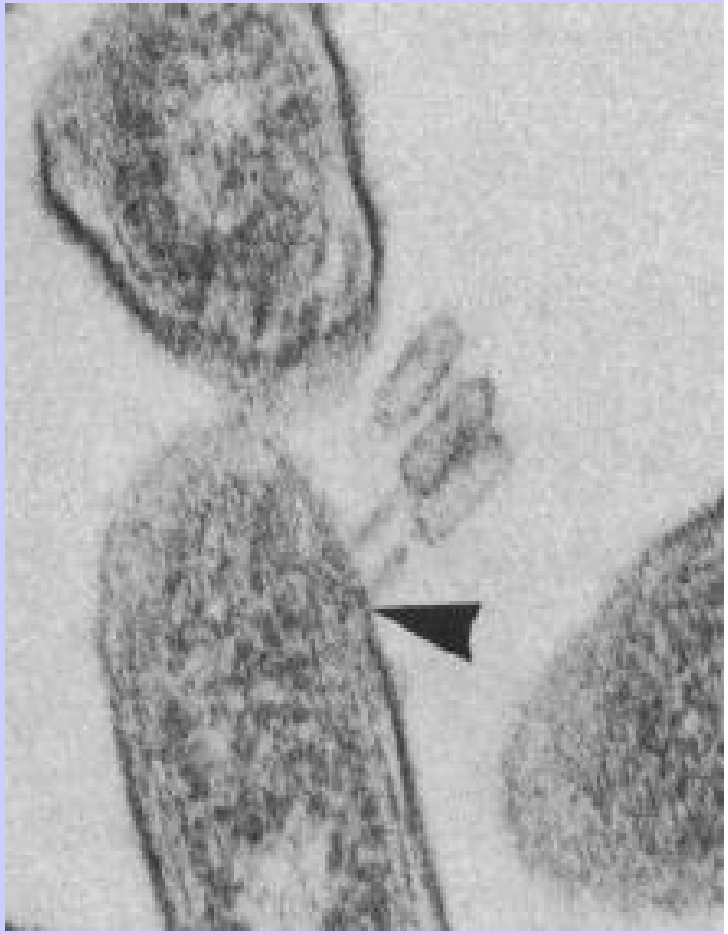
Bugs in your body which lie in a dormant "Resting" state

and which are capable of

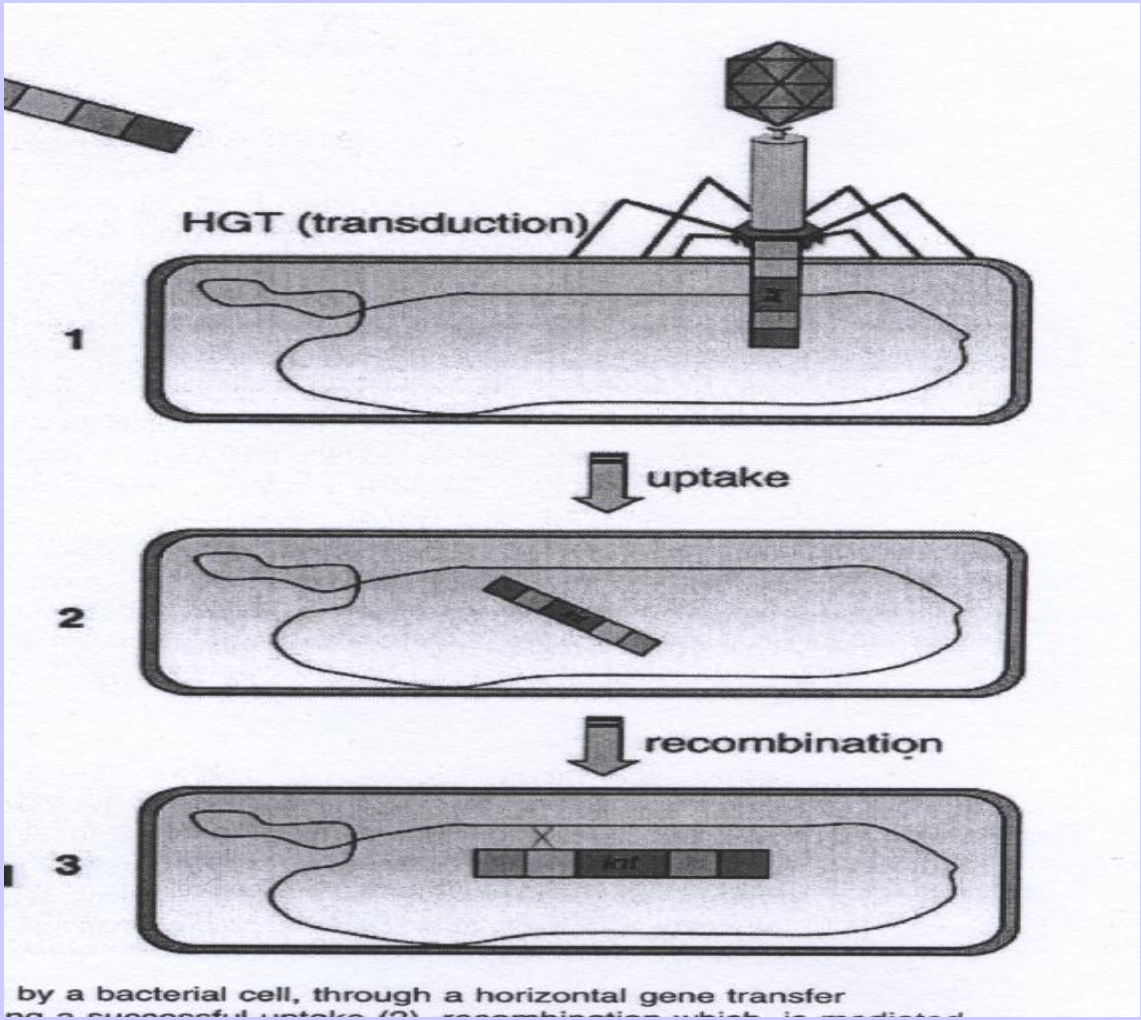
"Reactivation"

Module #10
Borrelia Genomes in Flux



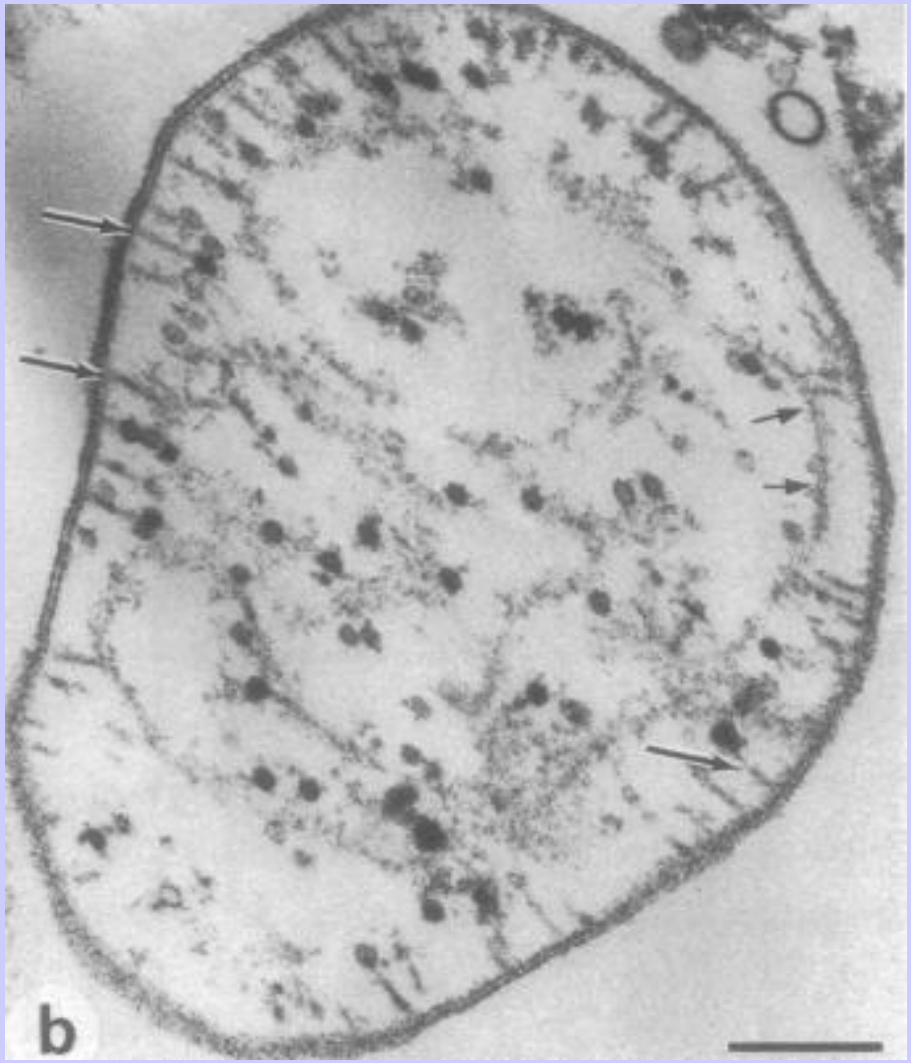


Borrelia Genomes in Flux

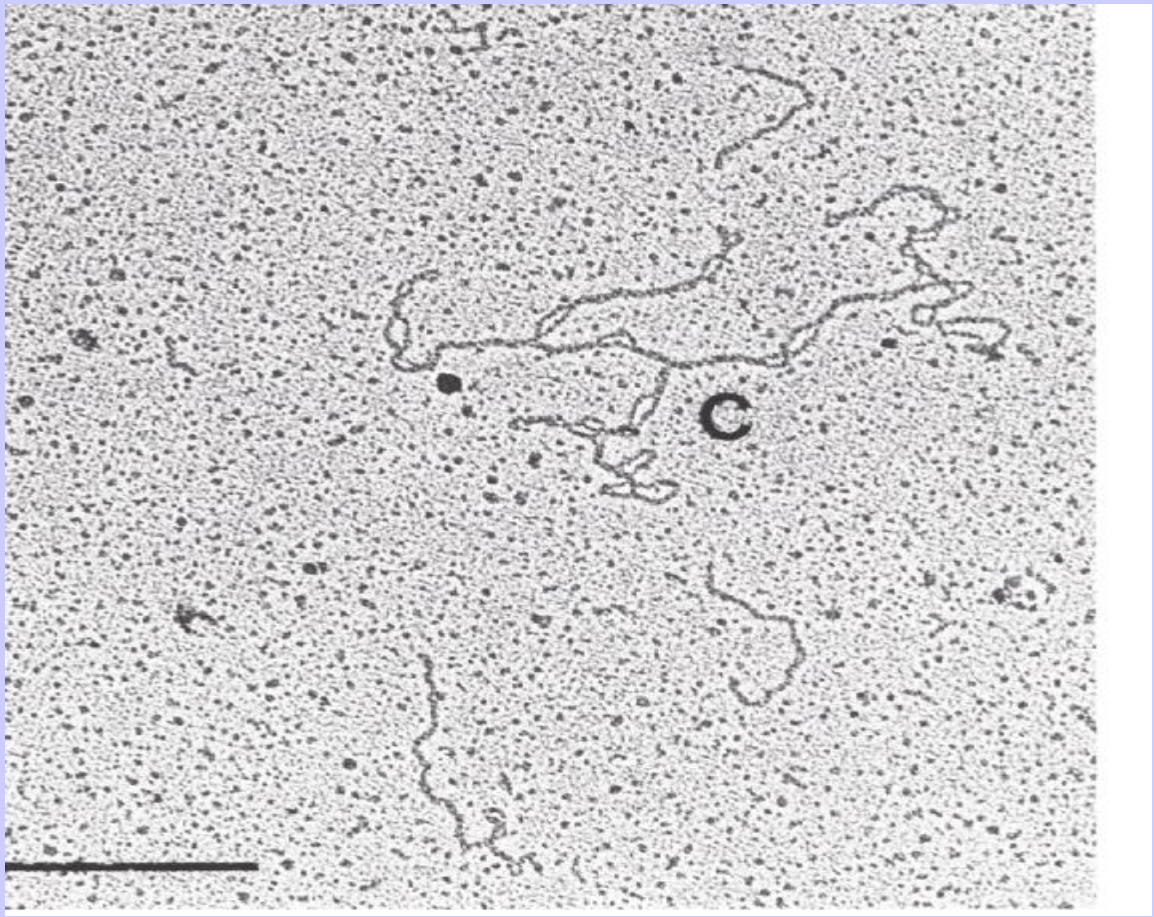


Genome in Flux

Bacteriophages are Parasites of Spirochetal Parasites

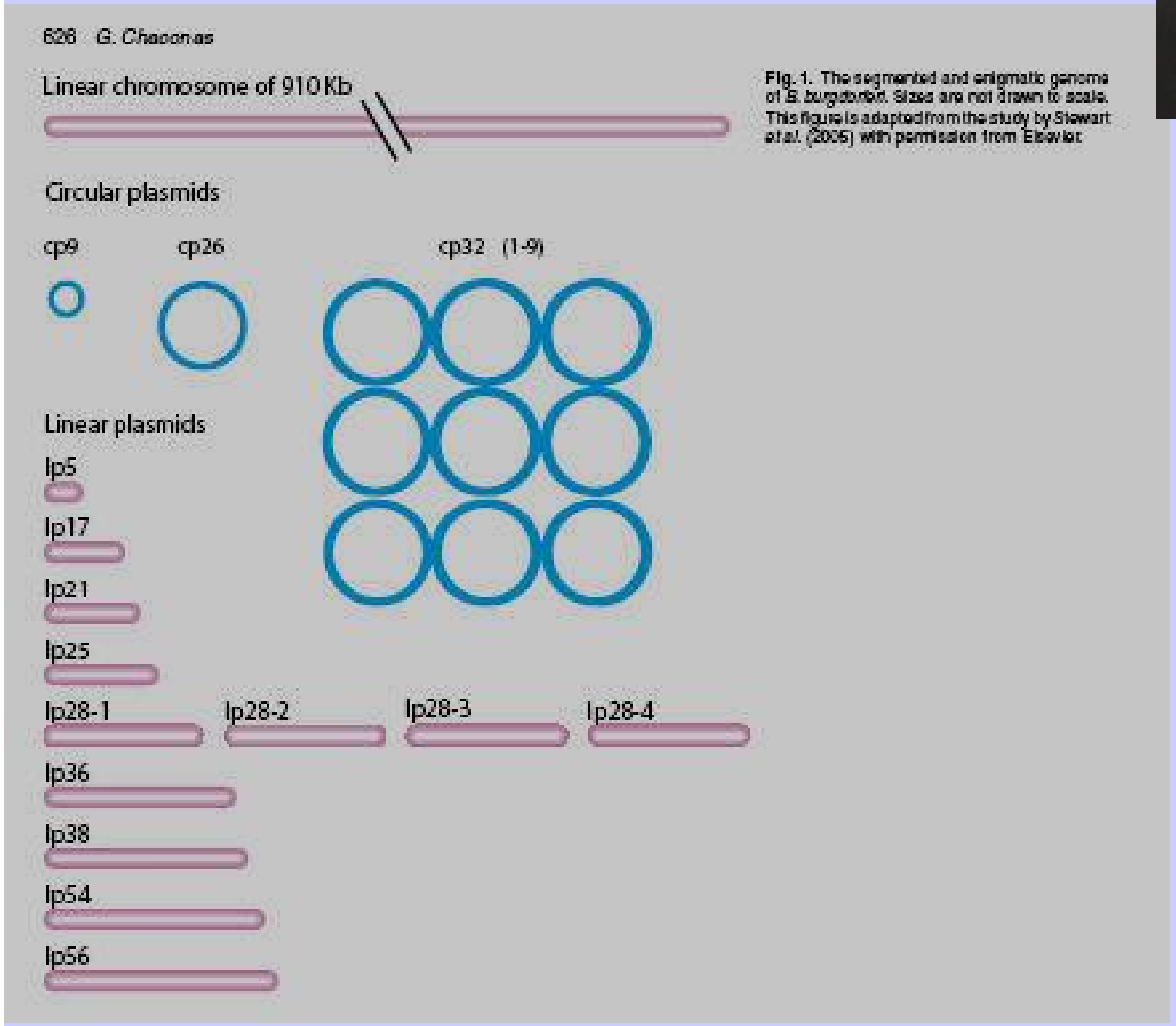


Genome in Flux



DNA in Blebs of Borrrelia burgdorferi

Genome in Flux



Chromosome and Plasmids

Genome in Flux

DNA “Inside” the Cell Wall Region

- Chromosome (S)
- Plasmids (S)
- Copy Number Issues

<u>Chromosome to Plasmid</u> “Shuffle” Like a Deck of cards

Plasmids as “Mini- Chromosomes”

DNA “ Outside” of the Cell wall region ***(“Blebs with DNA”)***

Genome in Flux

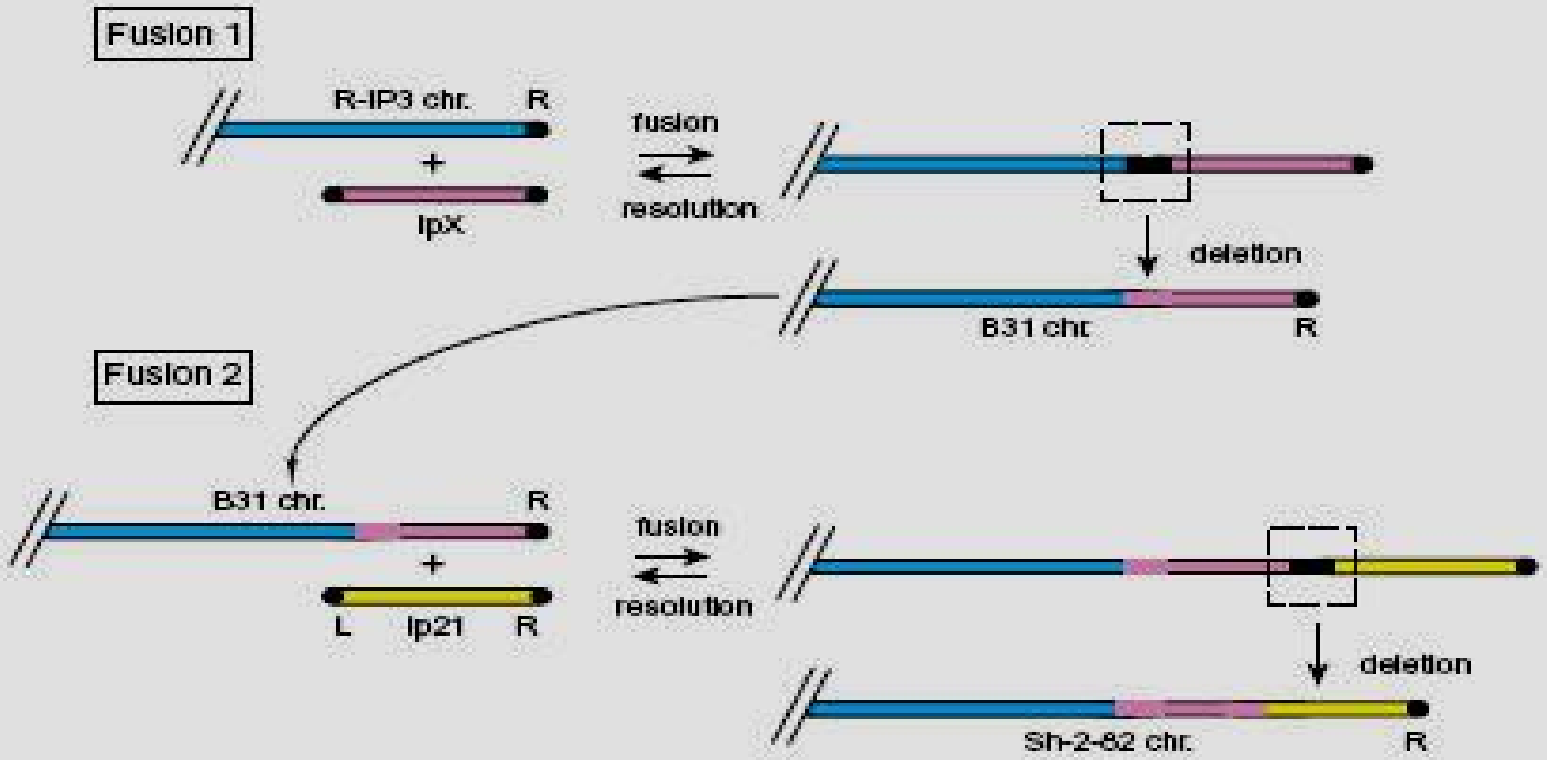
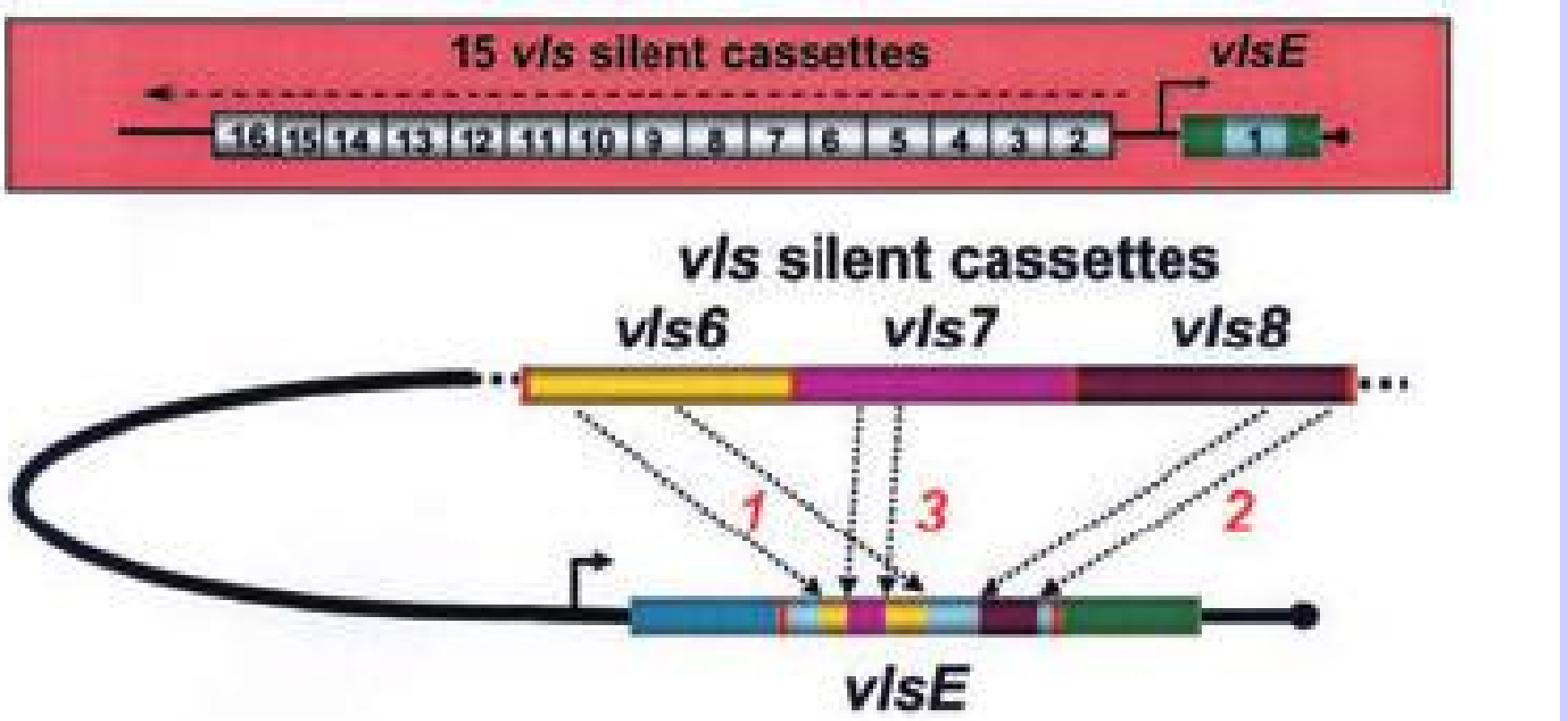


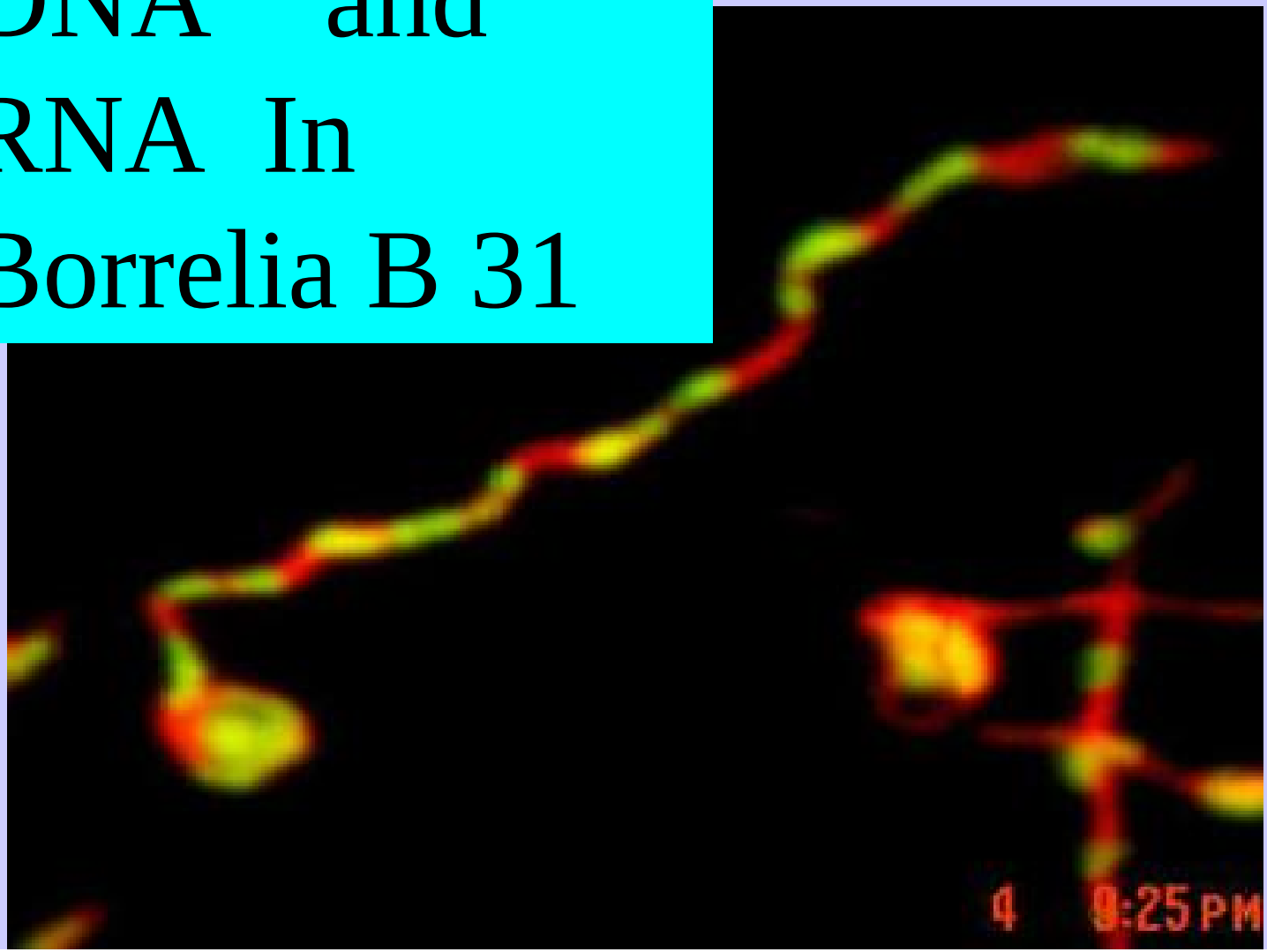
Fig.5. Telomere exchange by RecT-mediated telomere fusion. Fusion 1 links an unknown linear plasmid (lpX) to the right end of the *B. burgdorferi* R-IP3 chromosome to generate the structure of the right-end telomere found in the B31 chromosome. The identity of lpX is not clearly discernible and the right end of the B31 chromosome shares homology with several linear plasmids (Casjens *et al.*, 1997; 2000). Fusion 2 shows a telomere exchange that converts the right end of B31 to the right end observed for the Sh-2-82 chromosome through fusion with lp21 (see Casjens *et al.*, 1997; Huang *et al.*, 2004b). Successive rounds of telomere fusion with deletion formation can also explain the many examples of telomere exchange observed in the *B. burgdorferi* linear plasmids (Casjens *et al.*, 2000). This figure has been adapted from the study by Kobryn and Chaconas (2005).

Genome in Flux



Genome in Flux

DNA and
RNA In
Borrelia B 31



DNA in Flux model

Module #11
Blebs of
Borrelia
as Weapons

Blebs as Weapons

Blebs

Are

Released

From Borrelia

Blebs as Weapons

"Shedding" of "Blebs" from Outer Membrane

DNA in Blebs – Origins??? Participation in "fluxes"

"Blebbing" as a Verb and as an Adjective

"Blebs as Projectiles"

Big Cysts from Little Blebs – Electron Microscopy

Blebs as Weapons

Blebs as Weapons

Module #12
Flagellae of Borrelia
as
Hypodermic Needles

Flagellae Hypodermic Needle

Flagellin Structure

- Hollow Core
- “Cap”
- Rotor Base
- Inner Fla B

Type III Secretion

Type IV Secretion

Flagellae Hypodermic Needle

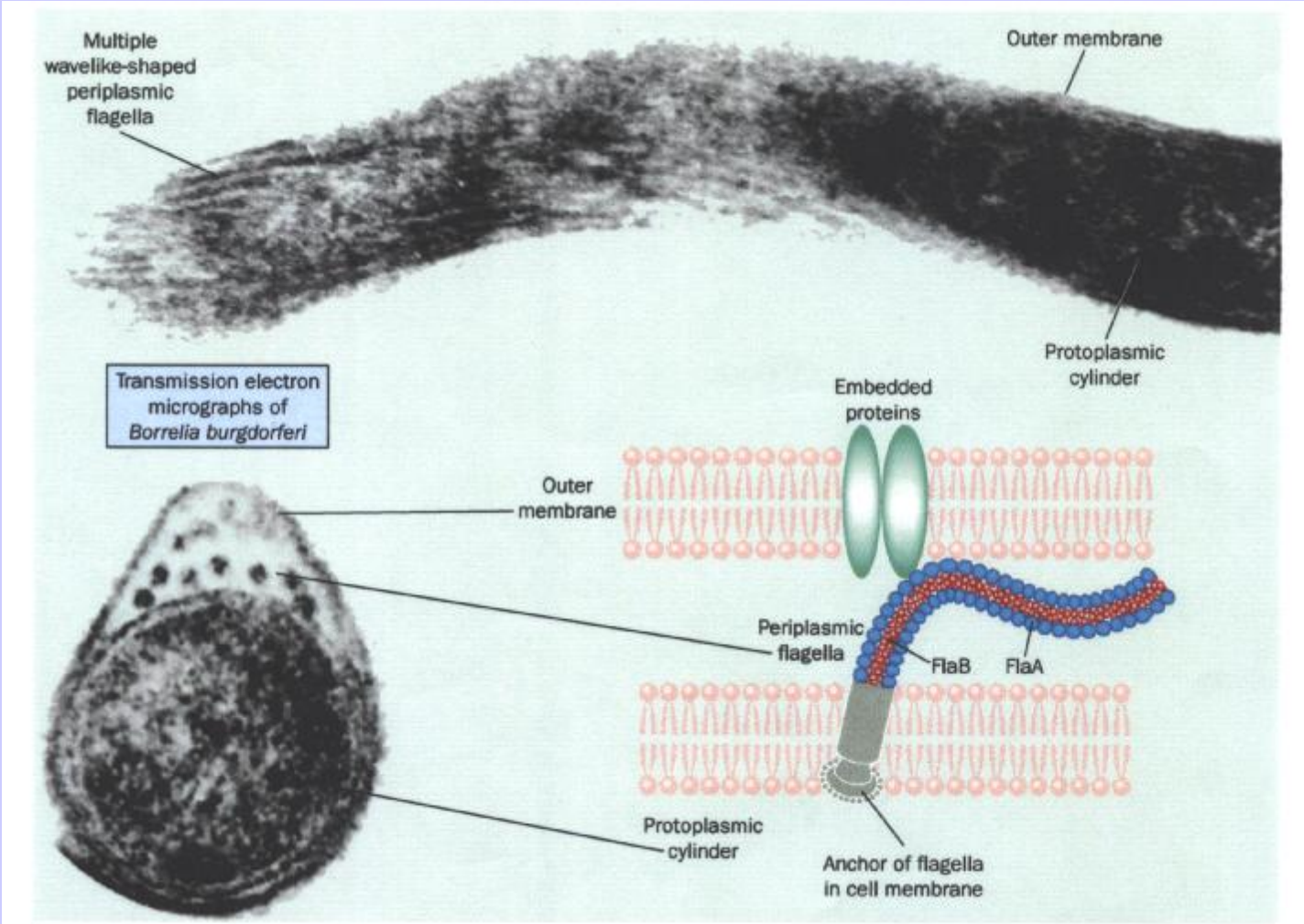


Figure 1. Ultrastructural morphology of B burgdorferi *electron micrographs and schematic line drawings illustrate the structural morphology of*

Module #13
Borrelia infections
Inside of Neurons
Which
Transit Across
Synapses

Infections Inside of Neurons , Schwann cells or Glia

(*** documents Transsynaptic transmission of Infection)

***Rabies Virus (Ref 3,4)

***Pseudorabies Virus (Ref 5,6,7)

***Herpes Zoster (Varicella Zoster) Virus (Ref 8,9)

***Herpes Simplex Virus (Ref 10)

Measles Virus

***Variant Measles (SSPE Agent) Virus (Ref 11,12)

***West Nile Virus (Ref 13)

Poliomyelitis Virus

***Adenovirus (Ref 14)

***Simian B Virus (Ref 15,16)

Canine Distemper Virus

***Influenza Virus H5N1 (Ref 17)

Tick Borne Encephalitis virus

***Prion Agent of Creutzfeldt Jacob (Ref 18)

***Prion Agent of Transmissible Mink Encephalopathy (Ref 19)

Prion Agent of Variant CJD (Mad Cow Disease)

Kuru Agent

***Scrapie Agent (Ref 20)

Leprosy (Mycobacterium Leprae)

***Listeria infection of Trigeminal Nerve in Mouse (Ref 21)

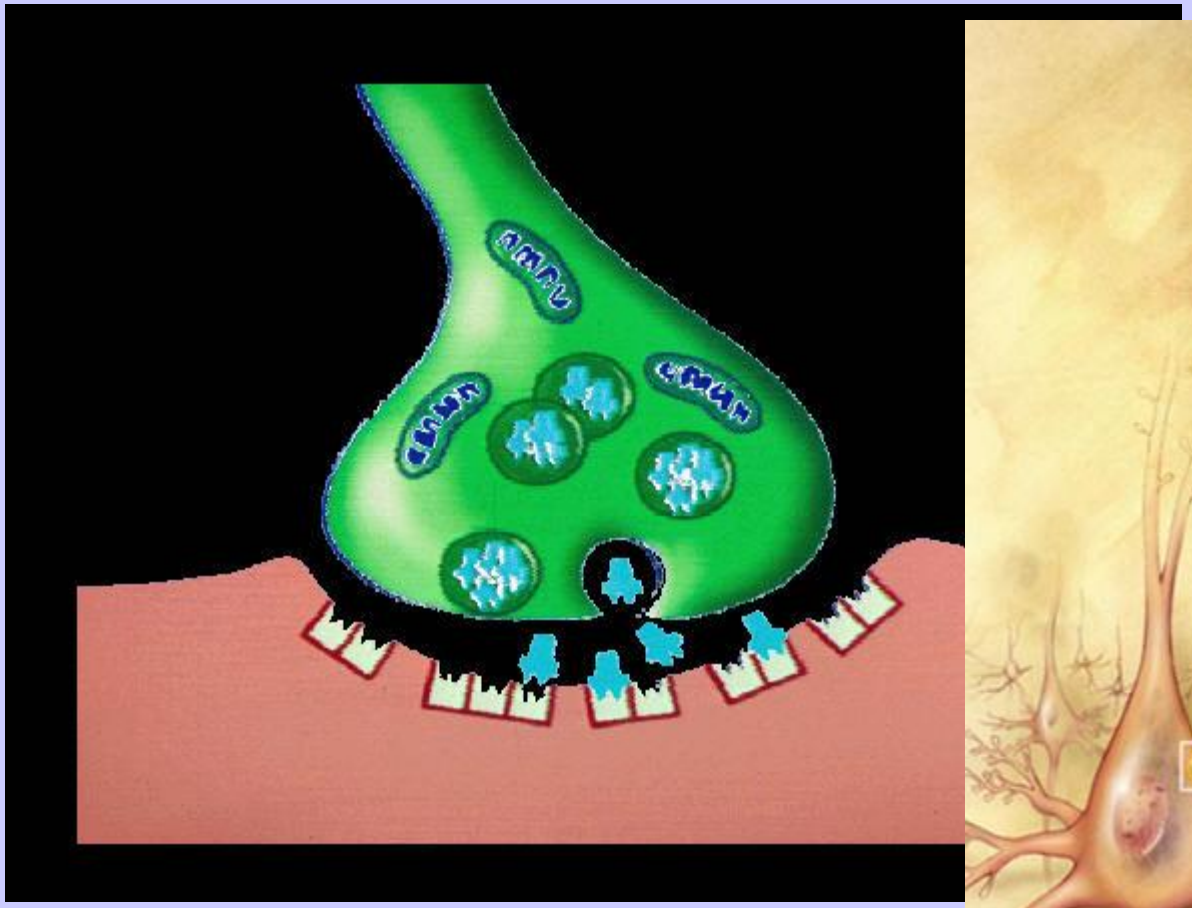
***Treponemal Spirochetes (Oral) via Trigeminal Nerve to Brain (Ref 22)

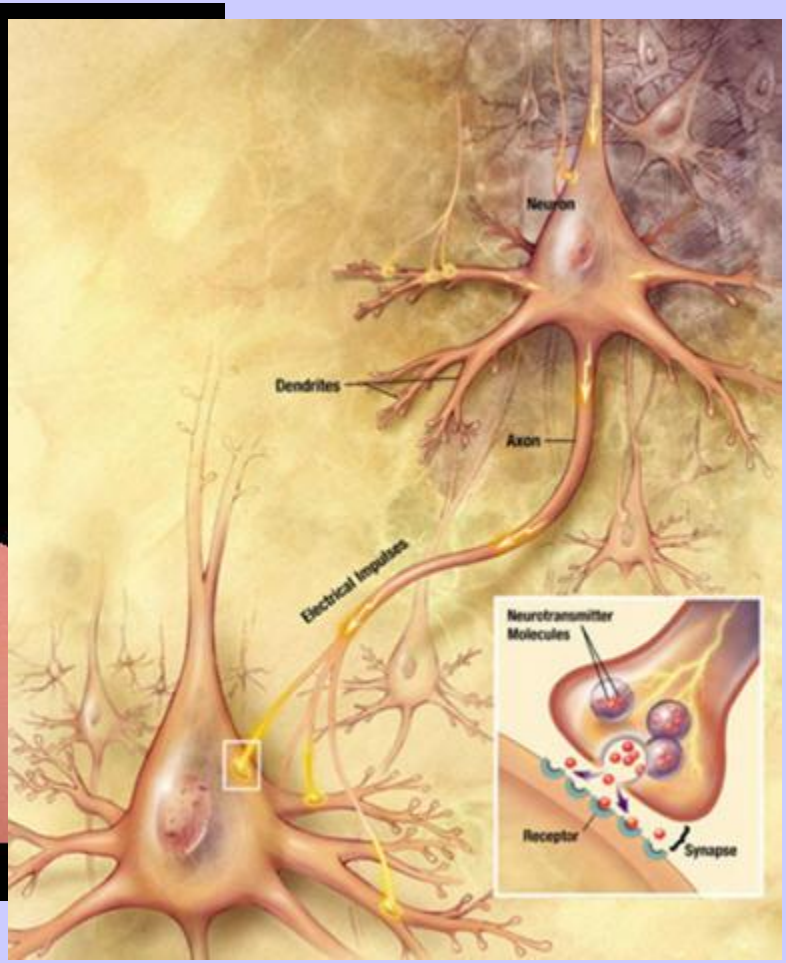
***Vesicular Stomatitis Virus (Ref 23,24)

Various Mosquito borne encephalitis viruses

Infections Inside Neurons Schwann cells or Glial cells

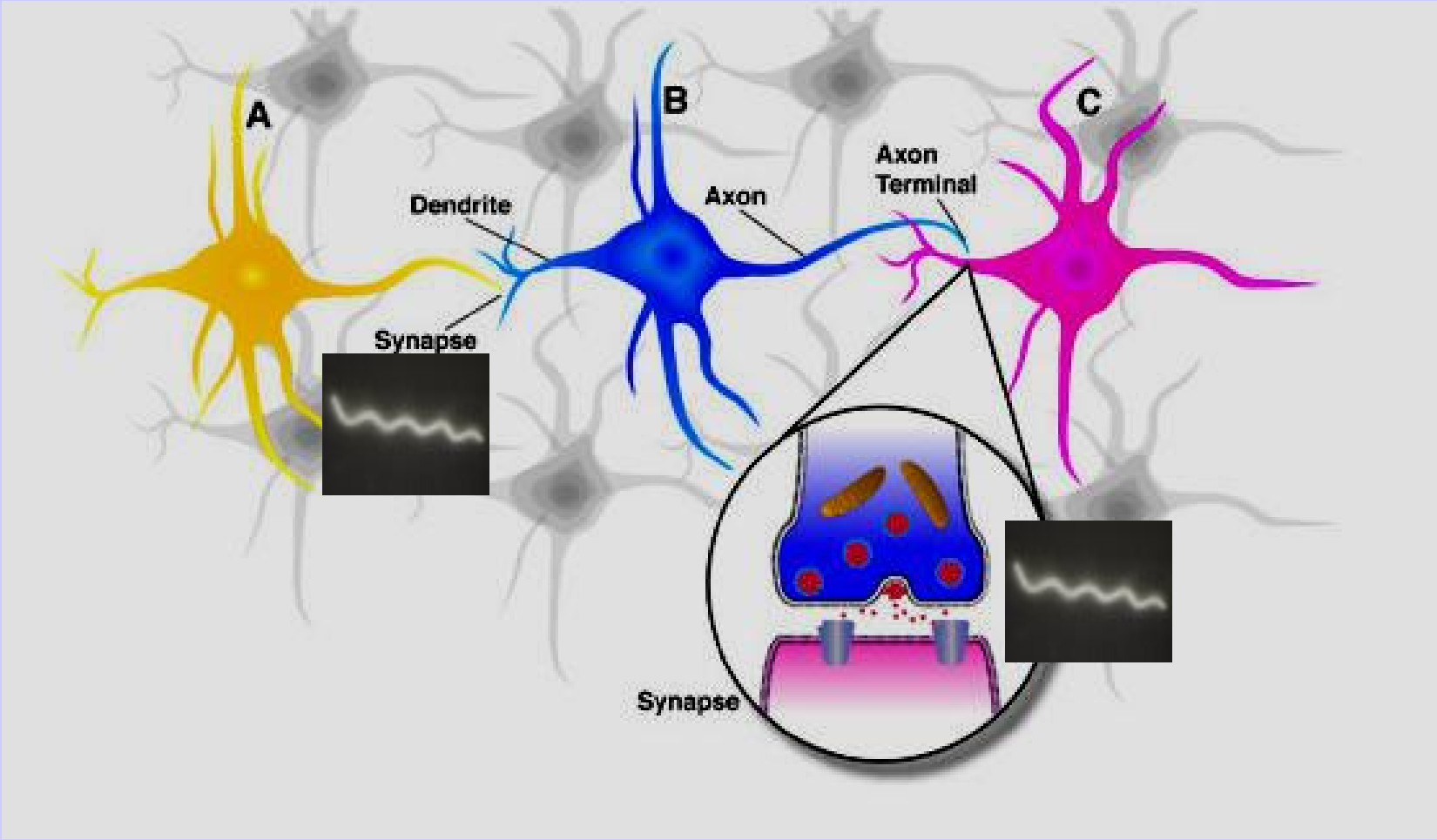
Trans-Synaptic Borreliosis



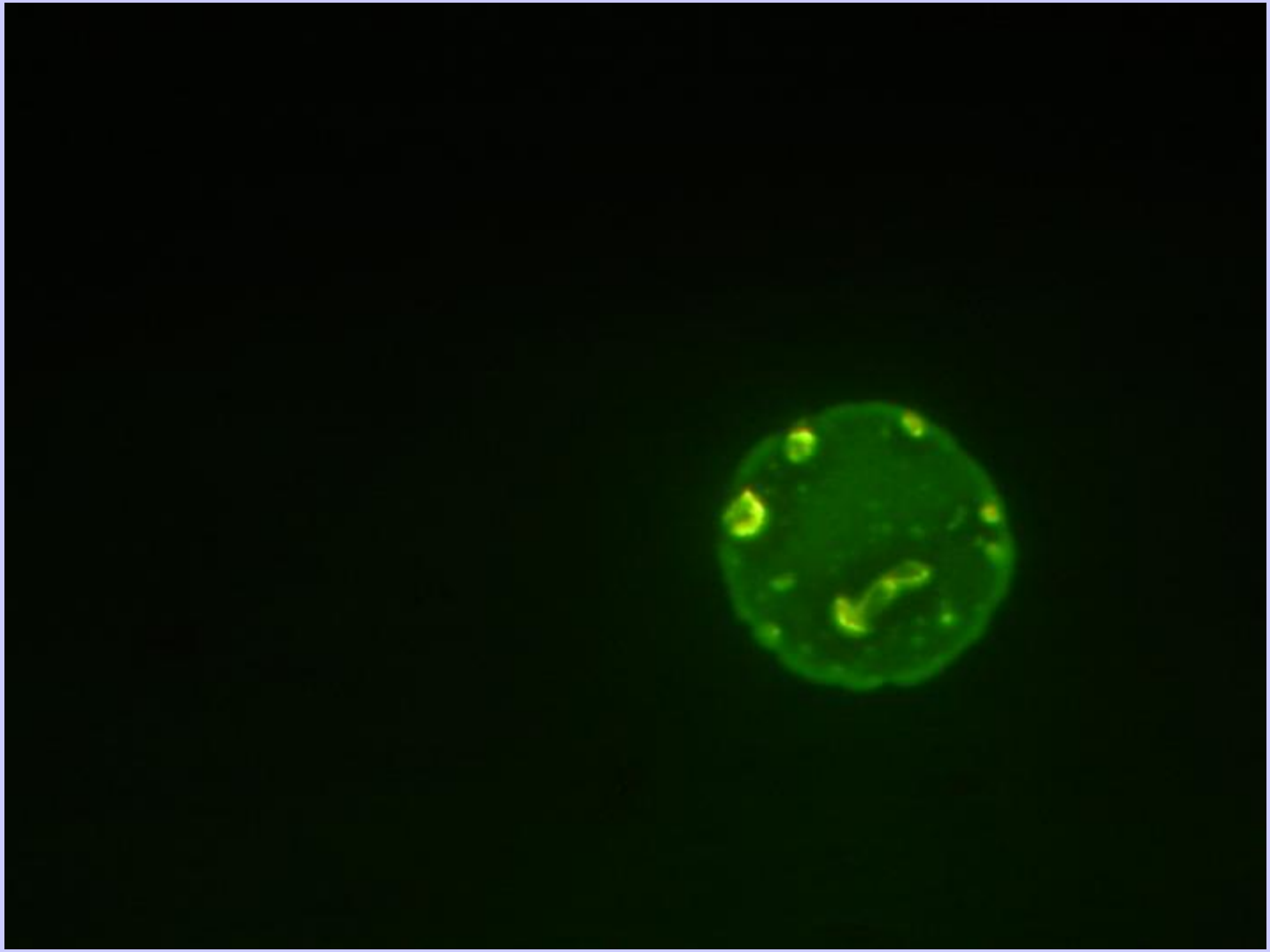


Module #14
Neural Network
Borrelia Infection

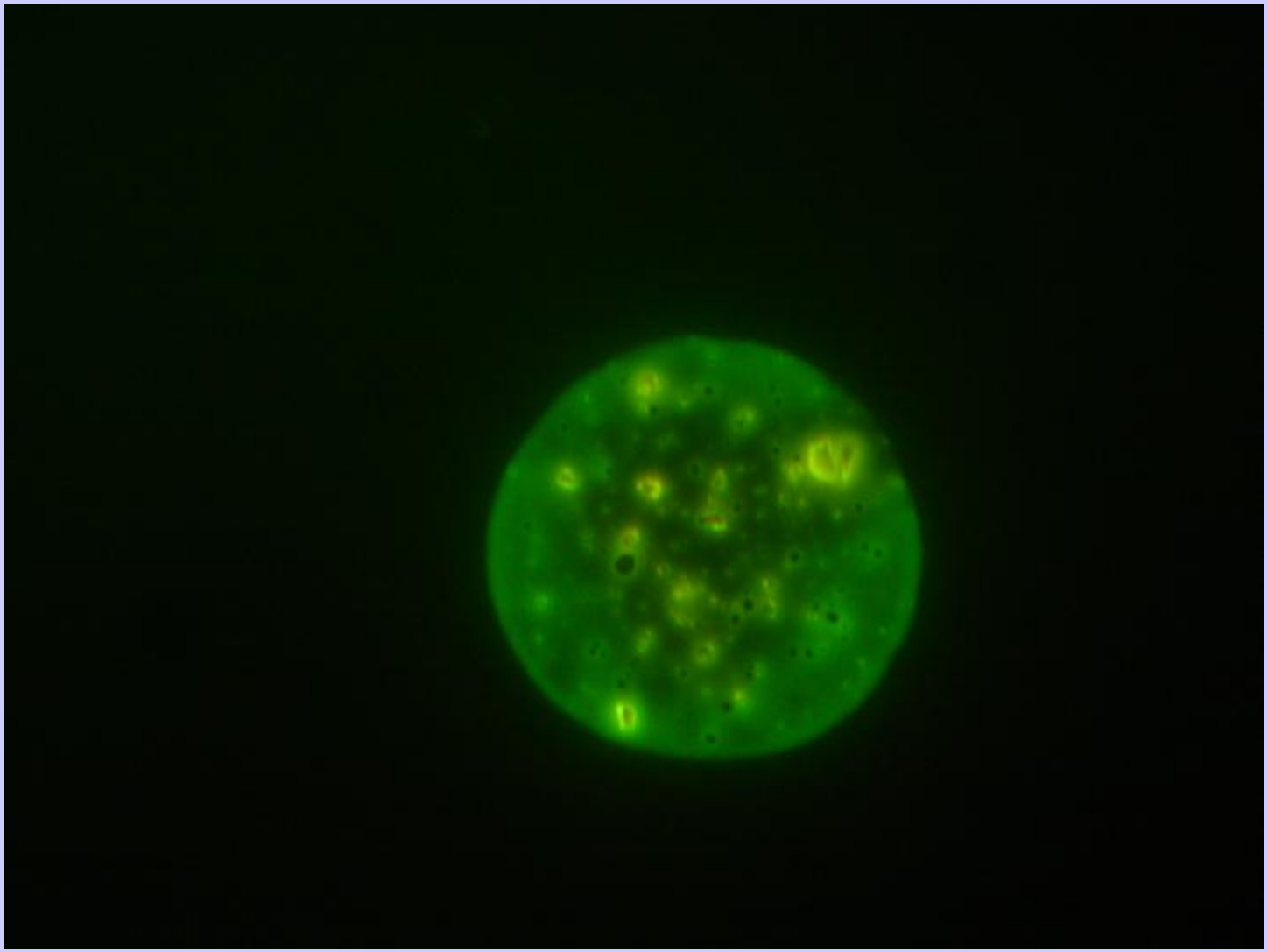
Borrelia Infection Neural Networks

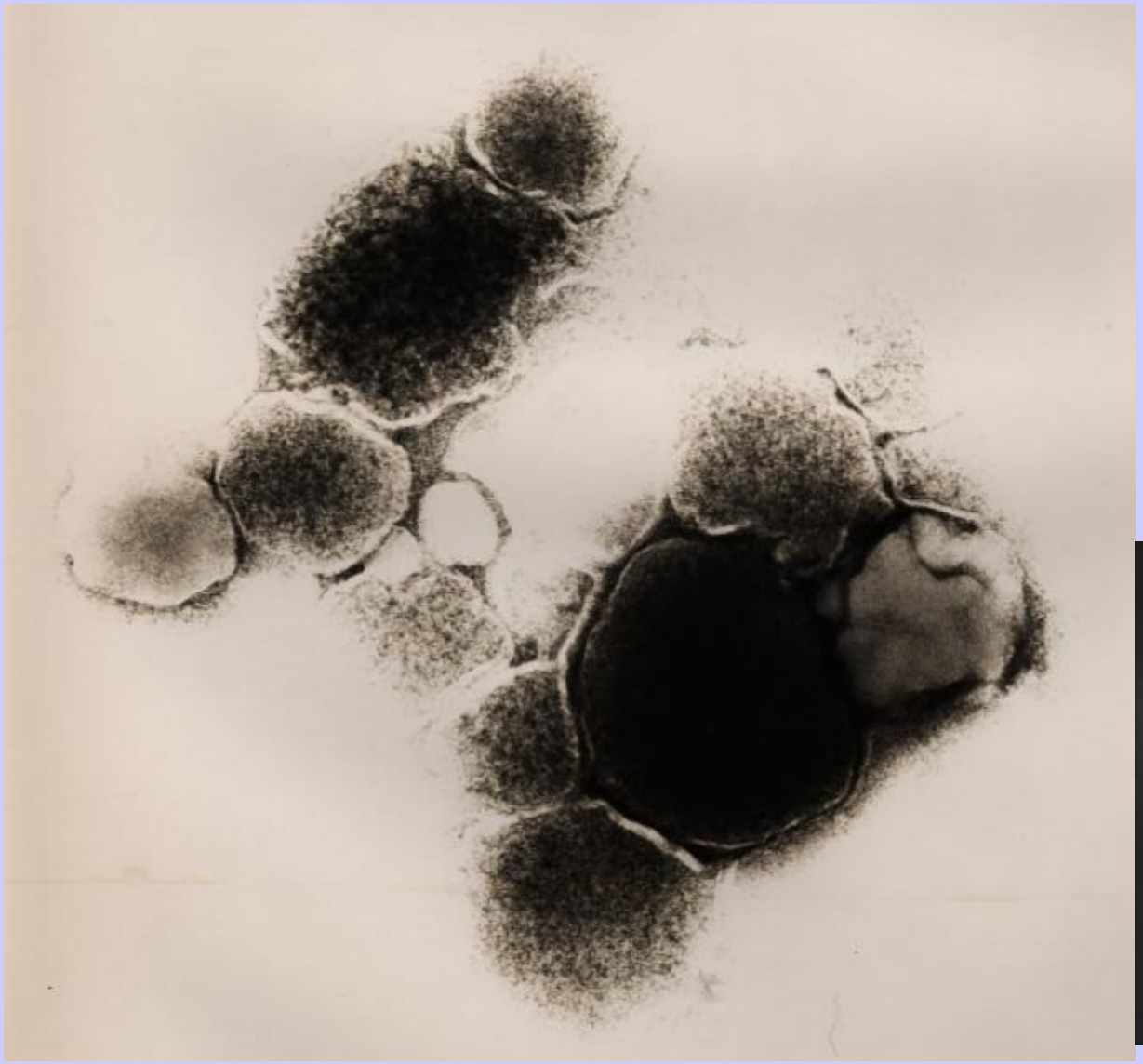


Borrelia Image Review



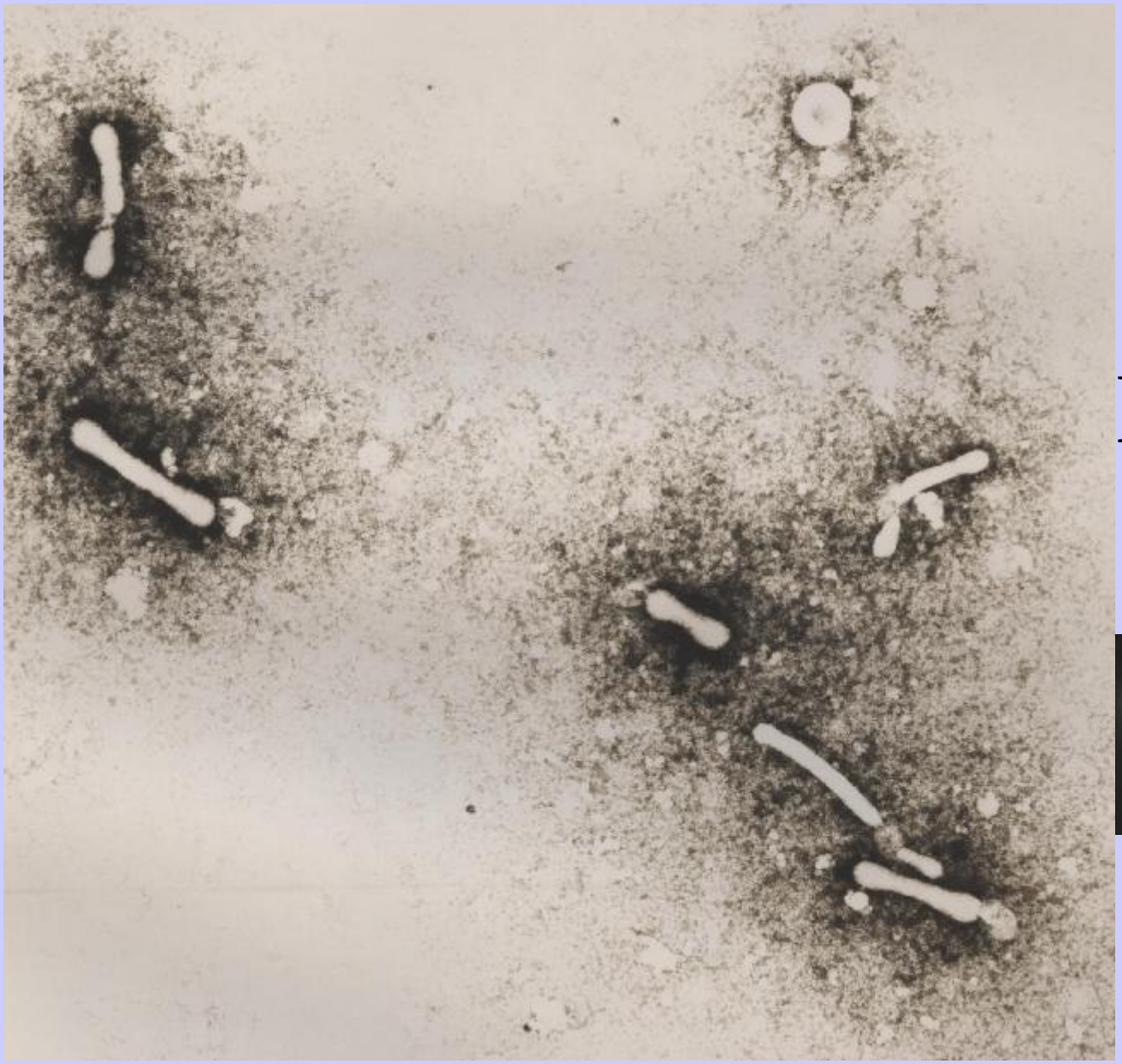
Borrelia Image Review





Blebs and L forms

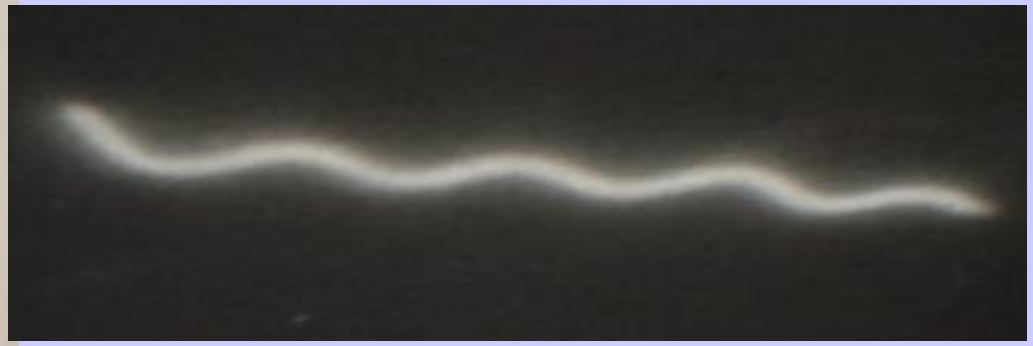
L Forms of Borrelia



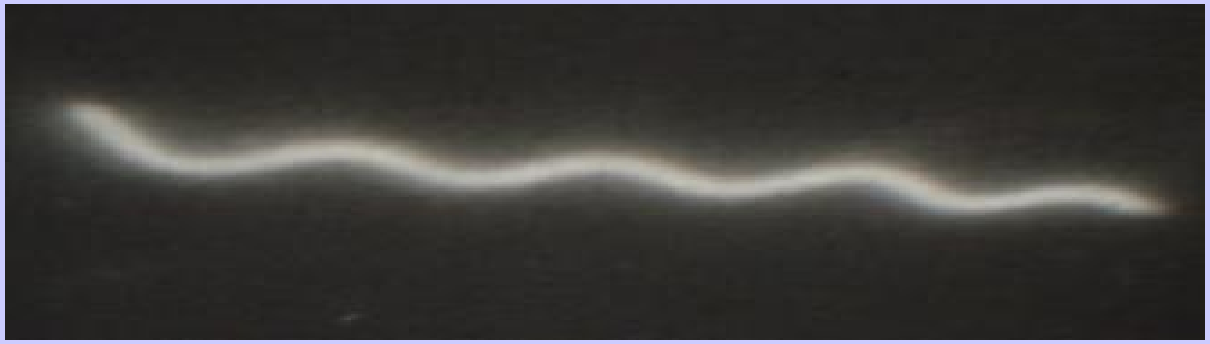
L forms of Borrelia

L form Borrelia

Borrelia
L Form
UltraStructure

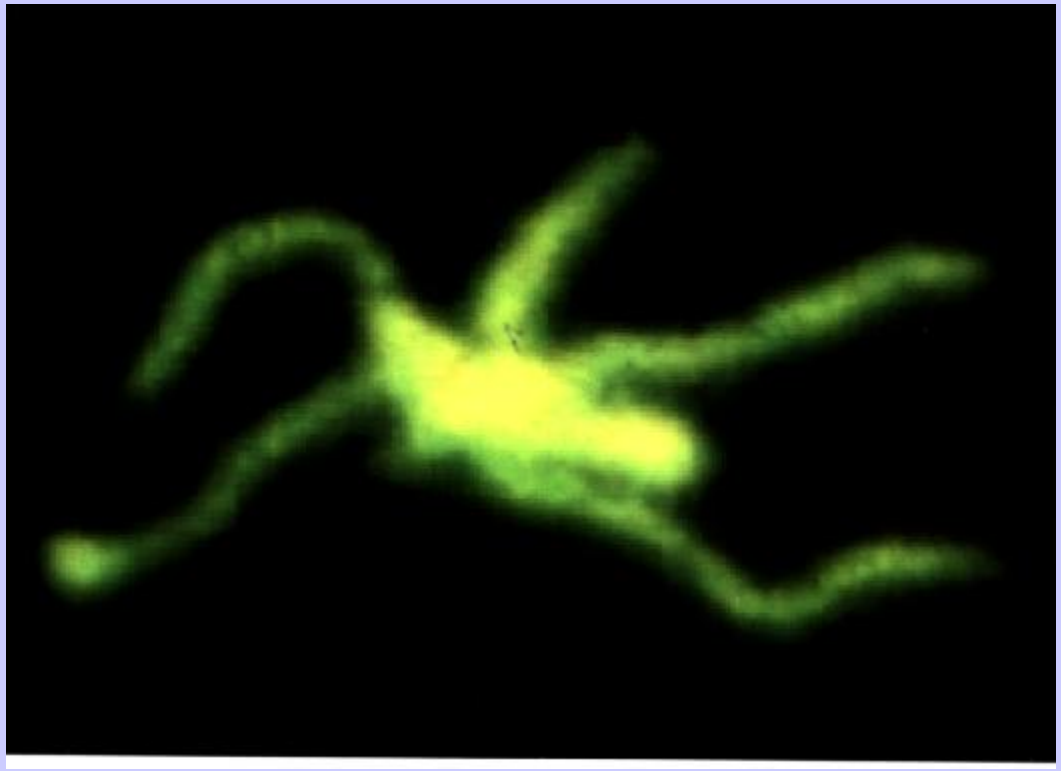


Borrelia Bleb Model

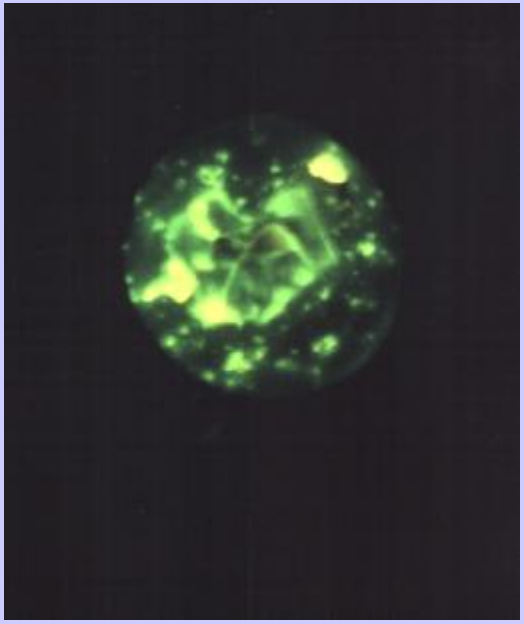


Ring Form of Borrelia

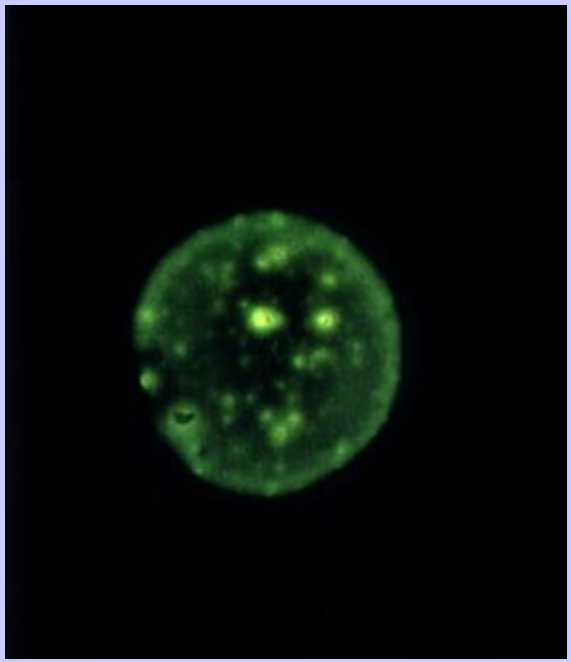
Aberer and Duray Borrelia form



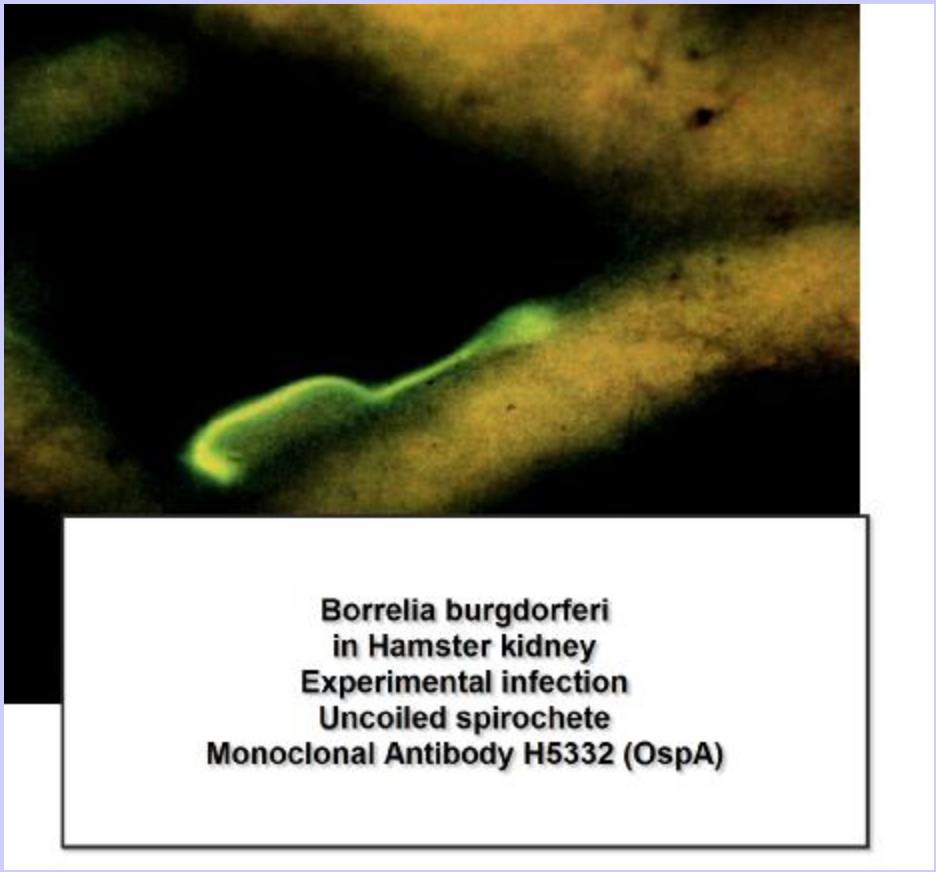
Cystic Borrelia with Granules



Cystic Borrelia with Granules

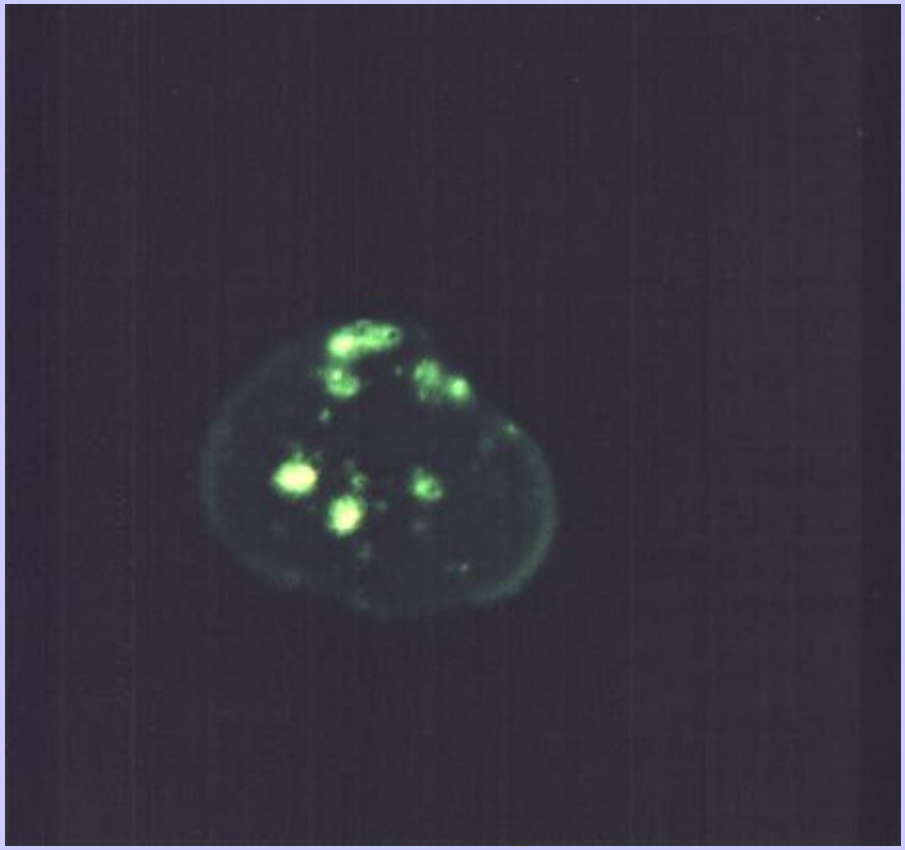


Experimental Borrelia Infection

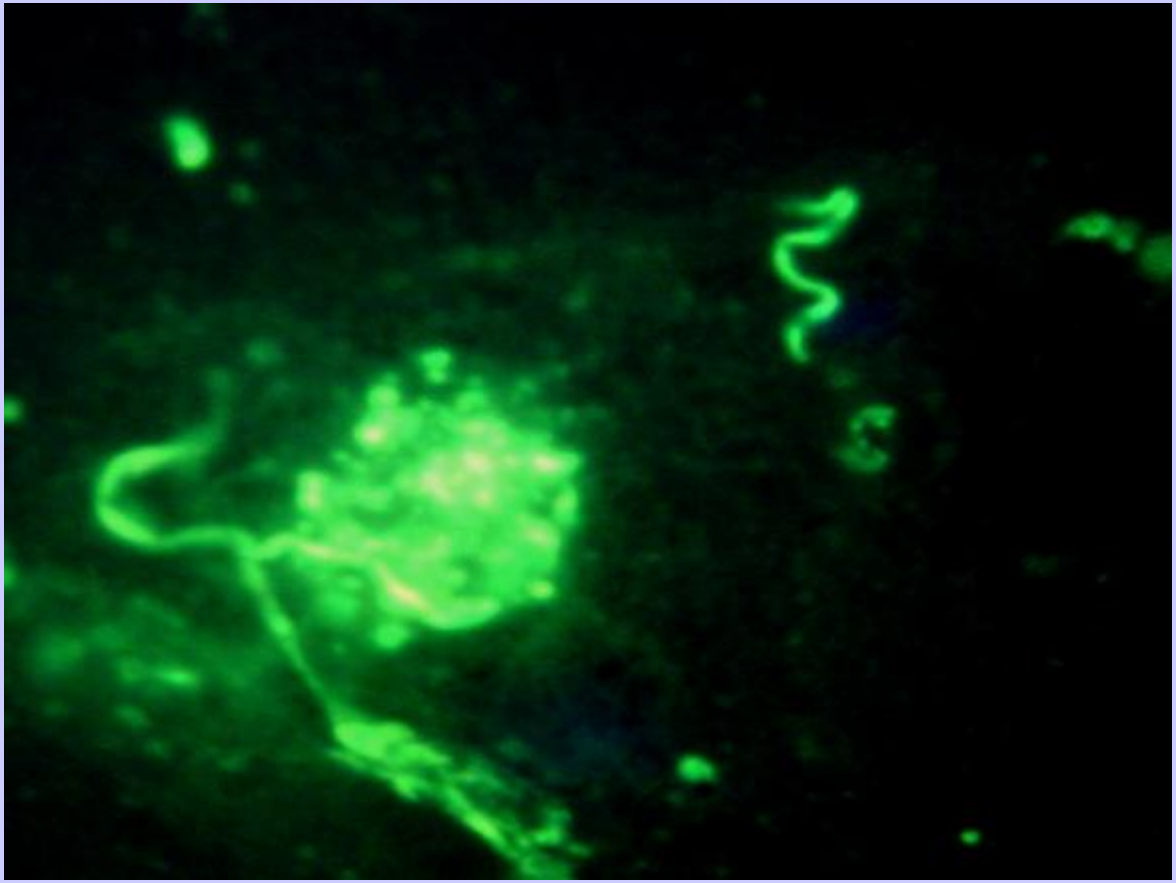


Cystic Form of Borrelia

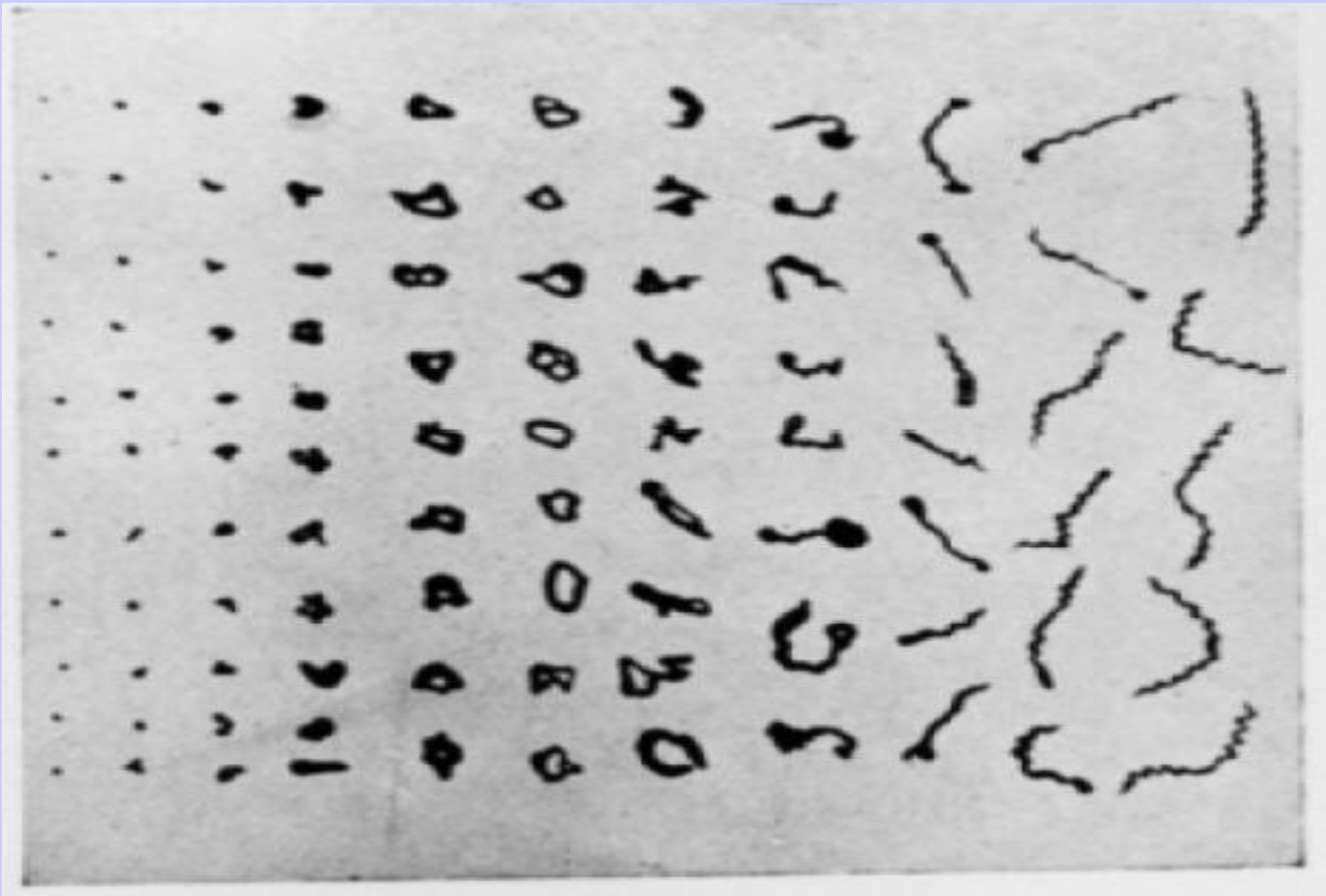
Cystic Form of Borrelia



Cystic Form of B31 Borrelia



Perfect Spirochetes – All of These



The Research Support of the Turn the Corner Foundation

and the Lyme Disease Association

and the Time For Lyme

All Gratefully Acknowledged