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08 October 2013

Special bacteriology

Introduction (taxonomy)

Gram positive rods

Corynebacterium spp.

Listeria monocytogenes

Erysipelothrix rhusiopathiae

Lactobacillus spp.

Normal flora of the vagina

Probiotics

Classification of Bacteria

(Murray 5th Chapter 2., page 7.) R. H. Whittaker

Formal rank

Regnum = Kingdom

Divisio = Division
(Phylum)

Classis = Class

Ordo = Order

Familia = Family

Genus = Genus

Species = Species

Example (Taxon)

Prokaryotae

Firmicutes

Thallobacteria

Actinomycetales

Mycobacteriaceae

Mycobacterium

Mycobacterium tuberculosis

➤ **Some repetition...**

❖ *Infection*

PROCESS! .

- **local – general**
- **toxic disease, intoxication**
- **allergic disease**
- **Immune regulation disorder; autoimmune?**

poisoning

- food
- pyrogenic substances
(Microbe multiply outside macroorganism!)

Special bacteriology

how to speak about a bacterium?

1. morphology
2. cultivation
3. resistance
4. biochemical features
5. Ag - structure
6. groups/types (e.g. phage typing)
7. pathology, virulence factors, pathogenesis
8. clinical findings, diseases (immunity)
9. diagnosis (microbiological)
10. treatment, prophylaxis, prevention

descriptive

Role in disease

Gram positive rods

Non spore-forming

AEROB

Corynebacteria

C. diphtheriae

diphtheroids

C. jeikeium

Listeria monocytogenes

Erysipelothrix rhusiopathiae

ANAEROB

Lactobacillus spp.

Spore-forming

AEROB

Genus: Bacillus

B. anthracis

B. cereus

B. subtilis

ANAEROB

Genus: Clostridium

C. tetani

C. botulinum

C. difficile

gasgangrene Clostridia

Historical overview I.

CORYNEBACTERIUM DIPHTHERIAE

Bretonneau 1821

Clinical characterisation of diphtheria

Klebs 1883

Detecting the bacterium

Loeffler 1884

Isolating the bacterium

Roux and Yersin 1888

Discovering the diphtheria toxin

Behring and Kitasato

1890-1892

- Discovering the diphtheria antitoxin
- Antitoxic immunity (therapy and prevention)

Roux 1894

Treatment with antitoxin

Historical overview II.

CORYNEBACTERIUM DIPHTHERIAE

Emil von Behring 1901

Nobel prize

Behring 1913

Active immunisation I.
with toxin-antitoxin mix

Schick 1913

Skin test

Ramon 1923

Active immunisation II.
Anatoxin = toxoid

Freeman 1951

PHAGE (lysogenia,
toxin production)

Historical overview 1.

CORYNEBACTERIUM DIPHTHERIAE

Bretonneau 1821

Clinical characterisation of diphtheria



Pierre Bretonneau
1778 - 1862

1825 tracheotomy

Klebs 1883

Detecting the bacterium



Theodor Albrecht Edwin Klebs
1834 - 1913

Historical overview 2.

CORYNEBACTERIUM DIPHTHERIAE

Loeffler 1884

Isolating the bacterium

Others:

Methylene blue staining

Media

Loeffler's coagulated serum-agar



Friedrich Loeffler 1852 - 1915

Historical overview 3.

CORYNEBACTERIUM DIPHTHERIAE

Roux and Yersin 1888

Discovering the diphtheria toxin



Pierre Paul Émile Roux
1853 – 1933
Director Pasteur Institute
1904 - 1933



Alexandre Yersin
1863 - 1943

Historical overview 4.

CORYNEBACTERIUM DIPHTHERIAE

Behring and Kitasato

1890-1892

- Discovering the diphtheria antitoxin
- Antitoxic immunity (therapy and prevention)



Shibasaburo Kitasato
1852 – 1931



Emil Adolph von Behring
1854 - 1917

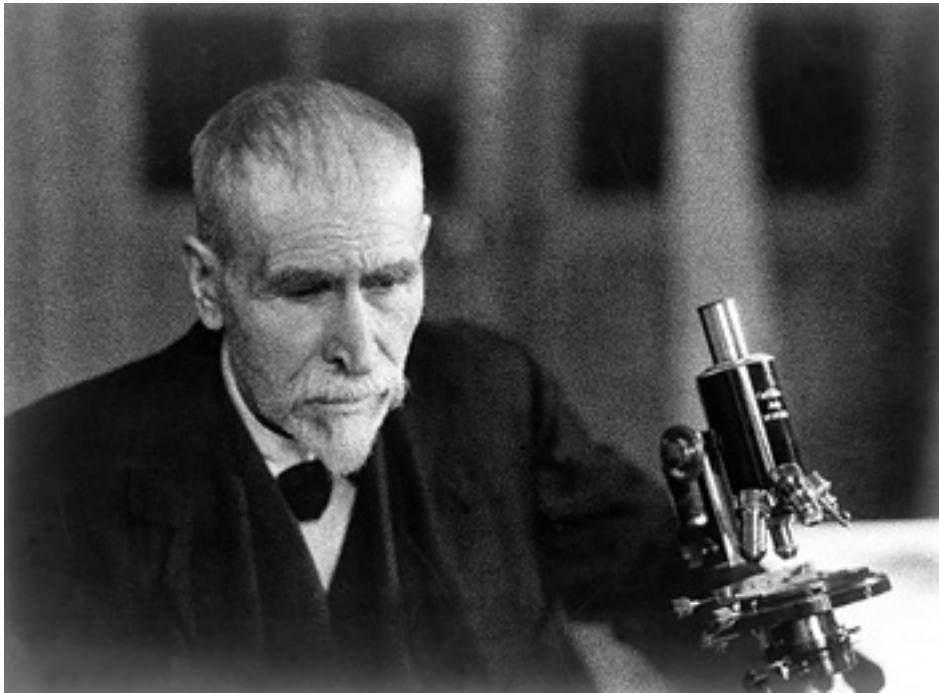


Historical overview 5.

CORYNEBACTERIUM DIPHTHERIAE

Roux 1894

Treatment with antitoxin



Historical overview 6.

CORYNEBACTERIUM DIPHTHERIAE

Emil von Behring 1901

Nobel prize



1940



Historical overview 7.

CORYNEBACTERIUM DIPHTHERIAE

Behring 1913

Active immunisation I.
with toxin-antitoxin mix



1914

Historical overview 8.

CORYNEBACTERIUM DIPHTHERIAE

Schick 1913

Skin test

Béla Schick

1877 Balatonboglár – 1967 USA

www.liveauctioneers.com



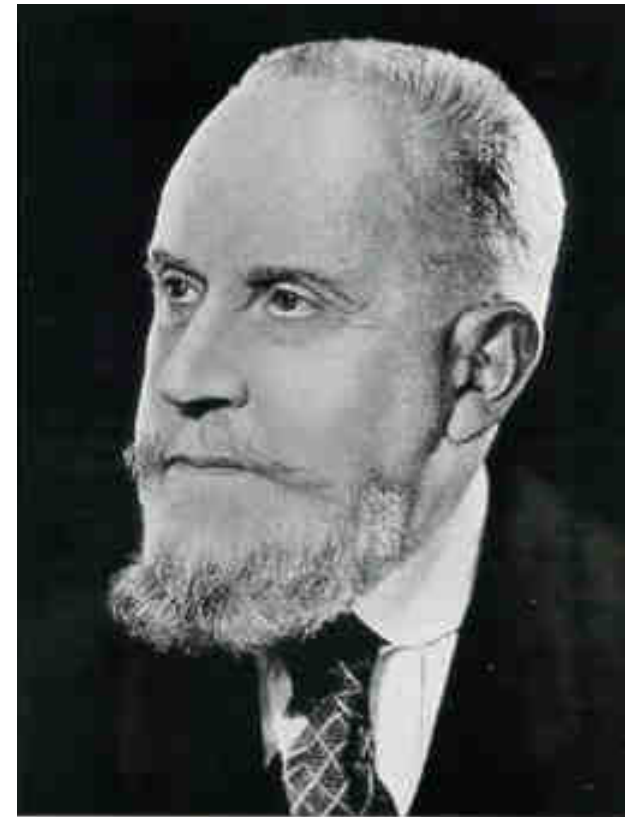
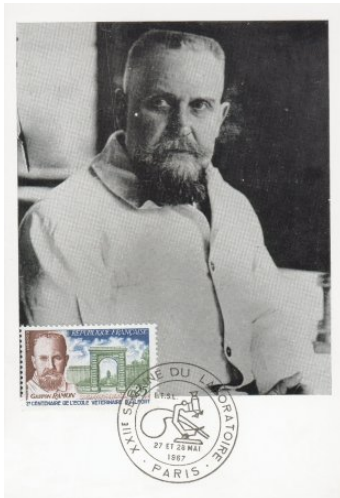
Historical overview 9.

CORYNEBACTERIUM DIPHTHERIAE

Ramon 1923

Active immunisation II.

Anatoxin = toxoid



GASTON-LÉON RAMON
- 1886 - 1963

Gaston Léon Ramon
1886 - 1963

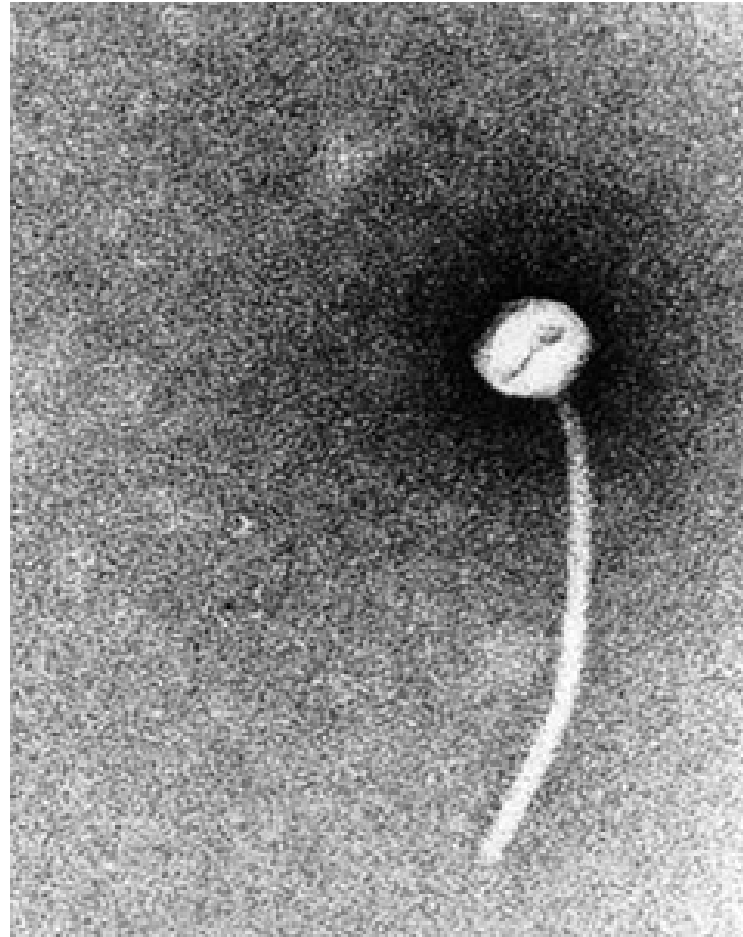
Historical overview 10.

CORYNEBACTERIUM DIPHTHERIAE

Freeman 1951

PHAGE

(lysogenia,
toxin production)



B phage

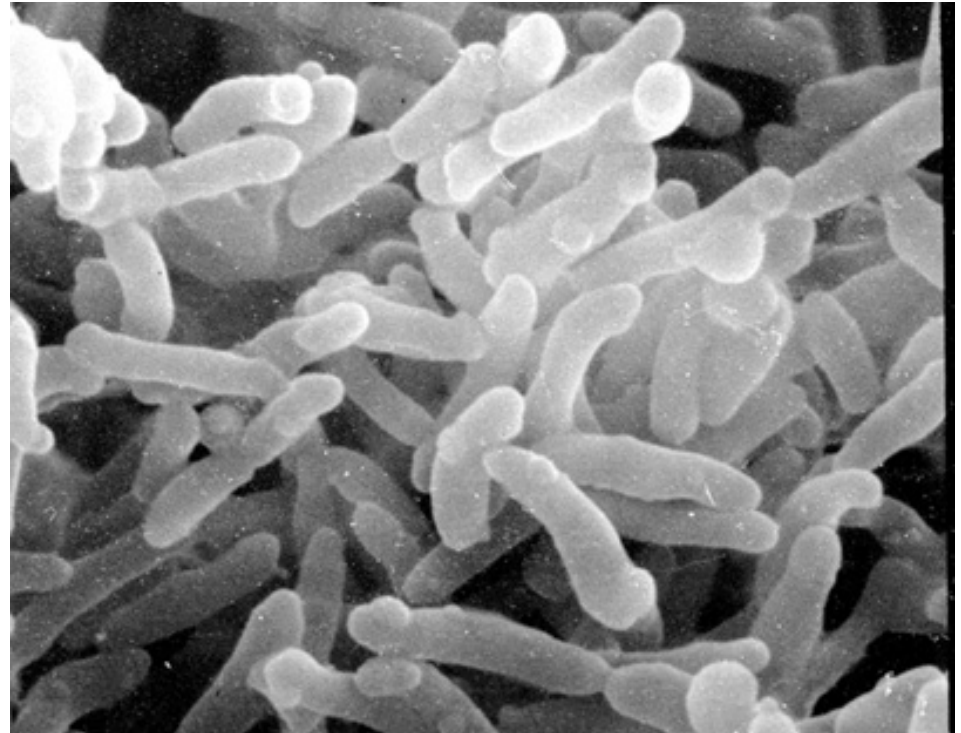
Corynebacterium diphtheriae-1

Morphology:

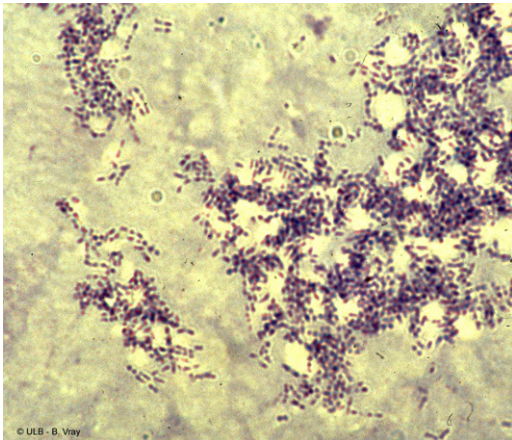
Gram + rod

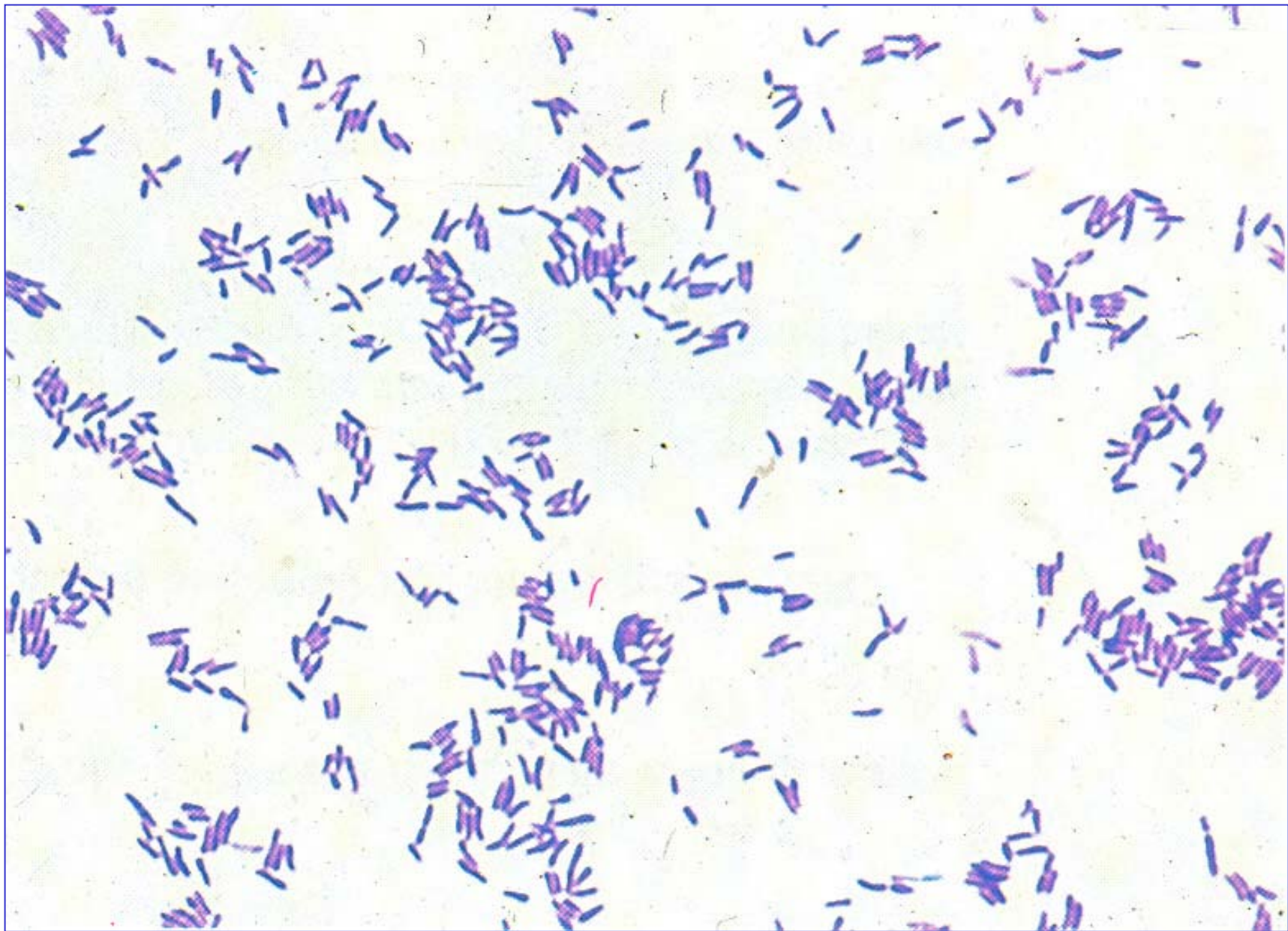
Neisser- staining

(Babes-Ernst
granules,
metachromatic)



EL-MI





Gram-staining/festés

Neisser staining

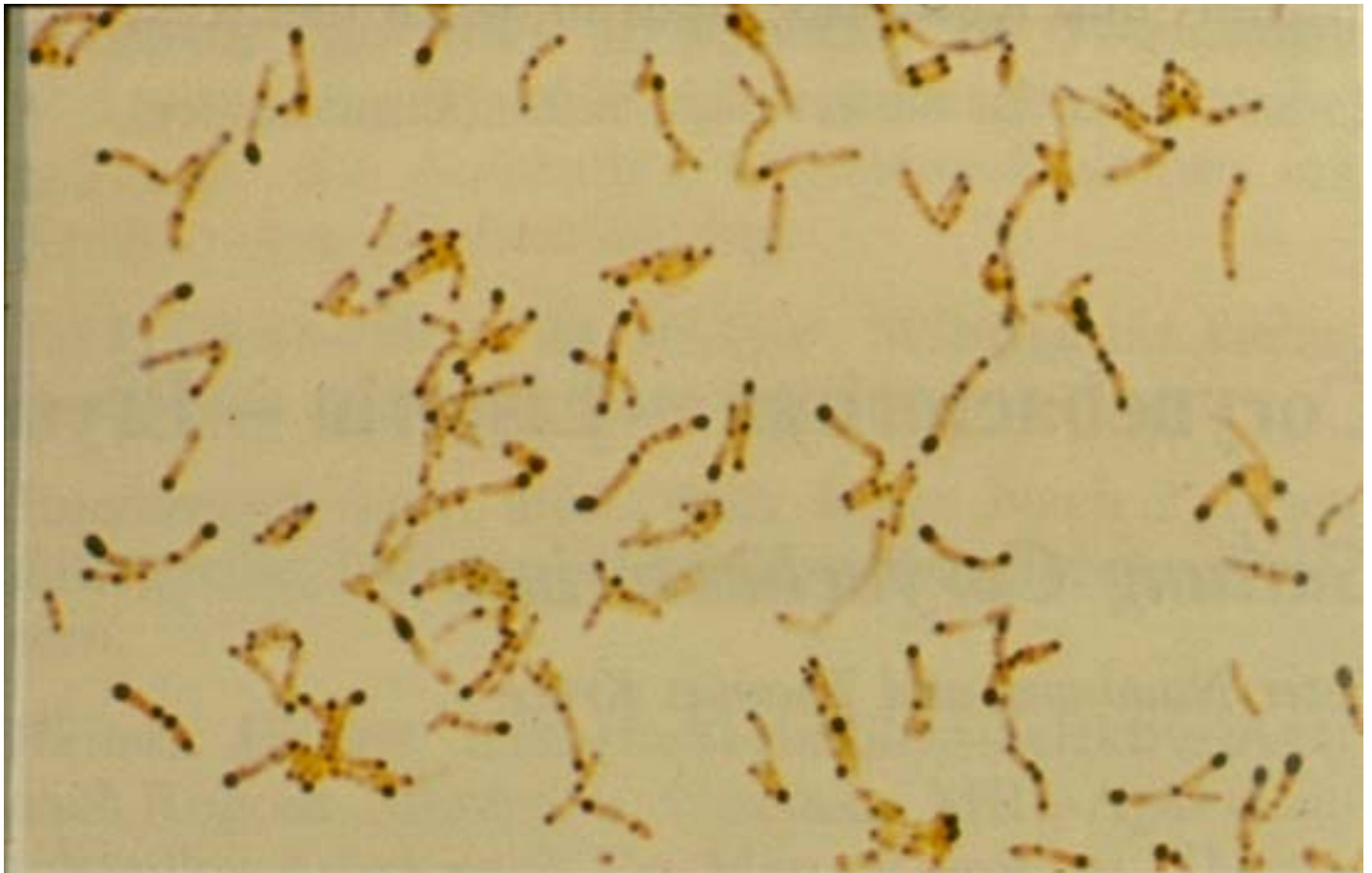


Abb. 2.52 **a** u. **b** *Corynebacterium diphtheriae*. **a** Gram-Präparat. Unregelmäßig, zum Teil in X- oder V-Form gelagerte, teilweise keulenförmige Stäbchen, Vergr. etwa 1600fach. **b** Neisser-Färbung. Gelbbraune Stäbchen mit zahlreichen, vielfach endständigen, schwarzblauen Polkörperchen. Vergr. etwa 1600fach

Corynebacterium diphtheriae-2

Cultivation:

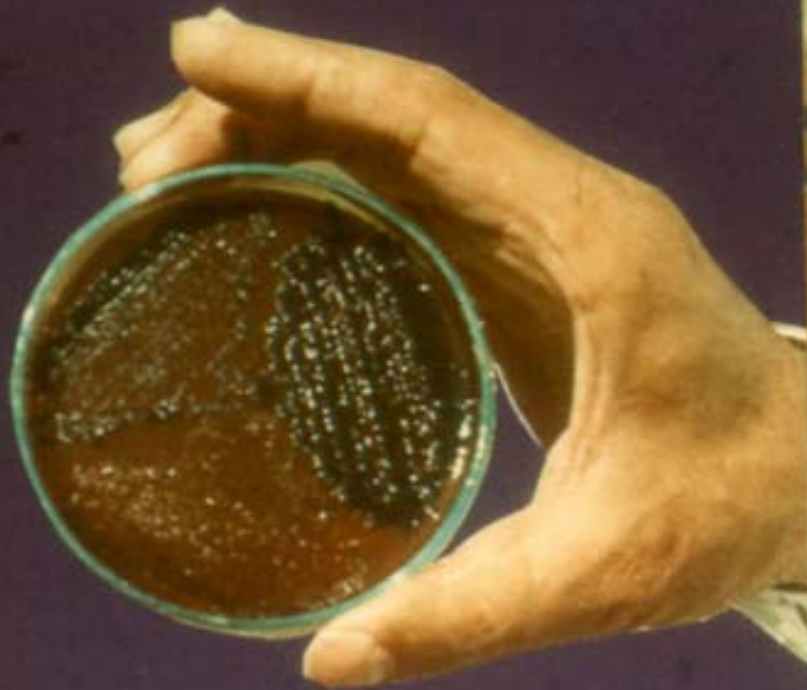
SPECIAL

Löffler-medium, serum agar

Clauberg-medium

- gravis
- mitis
- intermedius

112. *C. DIPHTHERIAE GRAVIS, MITIS*
ÉS INTERMEDIUS TELEPTÍPUSAI
CLAUBERG-FÉLE TÁPTALAJON

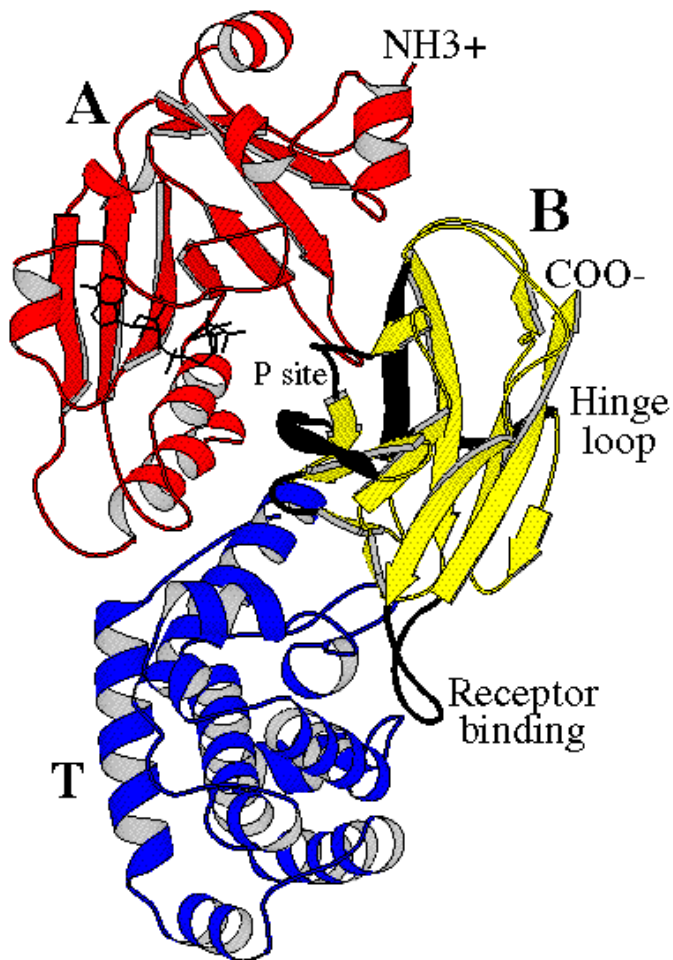


Clauberg (K-tellurite) medium - colony morphology

Corynebacterium diphtheriae-3

Pathogenicity, virulence:

EXOTOXIN!!!
AB toxin



Corynebacterium diphtheriae-4

Pathogenesis-1

source: **carrier**

(ill or reconvalescent or healthy)

penetration:

mucosal surfaces (nose, throat etc.)

skin (wounds!)

transmission: **drop infection, contact**

(direct, indirect)

Corynebacterium diphtheriae-4

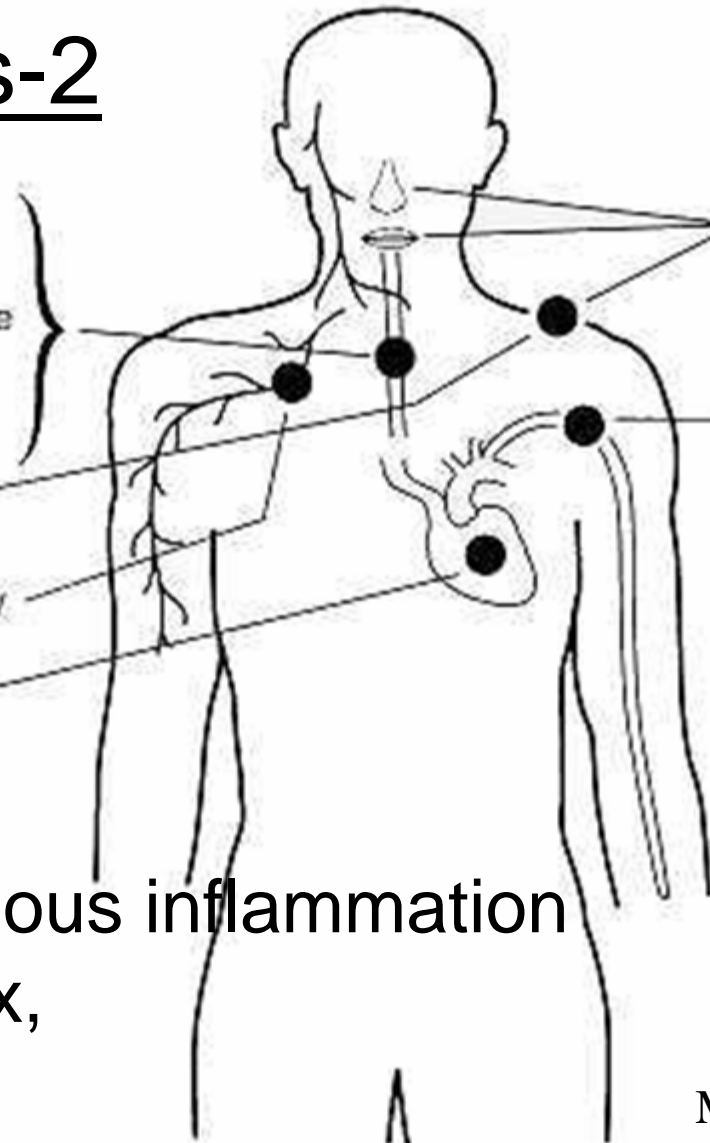
Pathogenesis-2

3. Disease

- Pharyngeal diphtheria
- Pharyngitis
- Hypoxia - pseudomembrane obstruction
- Fever
- Lymphadenitis
- Cutaneous diphtheria
- Systemic complications
 - Toxic peripheral neuropathy
 - Toxic myocarditis and congestive heart failure

- 1. Entry
- 4. Exit

- 2. Spread - bacteria rarely become disseminated but toxin becomes blood borne



Bacterium remains local!!

locally:

pseudomembraneous inflammation on tonsils, pharynx, larynx, trachea

Corynebacterium diphtheriae-4

Pathogenesis-3

3. Disease

Pharyngeal diphtheria

Pharyngitis

Hypoxia - pseudomembrane
obstruction

Fever

Lymphadenitis

Cutaneous diphtheria

Systemic complications

Toxic peripheral neuropathy

Toxic myocarditis and
congestive heart failure

1. Entry

4. Exit

2. Spread - bacteria
rarely become
disseminated but
toxin becomes
blood borne

Toxaemia!

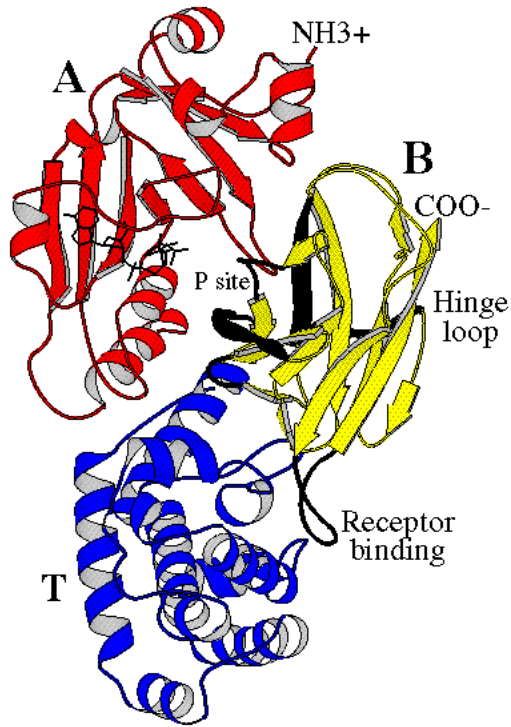
generalisation:

exotoxin ⇒ intoxication ⇒ degeneration

in the parenchymal organs,

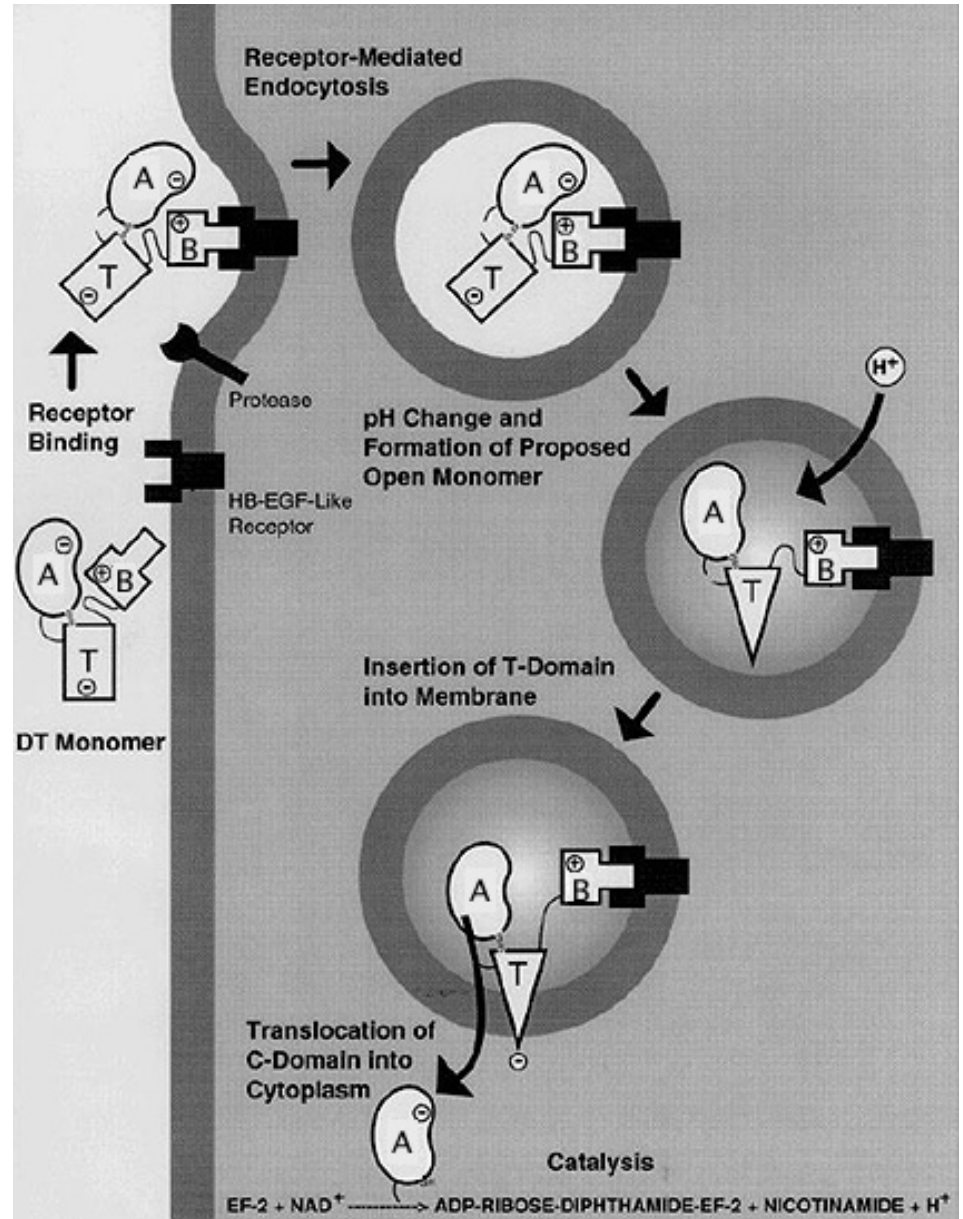
necrosis (acute cause of death if in the heart)

C. diphtheriae - exotoxin

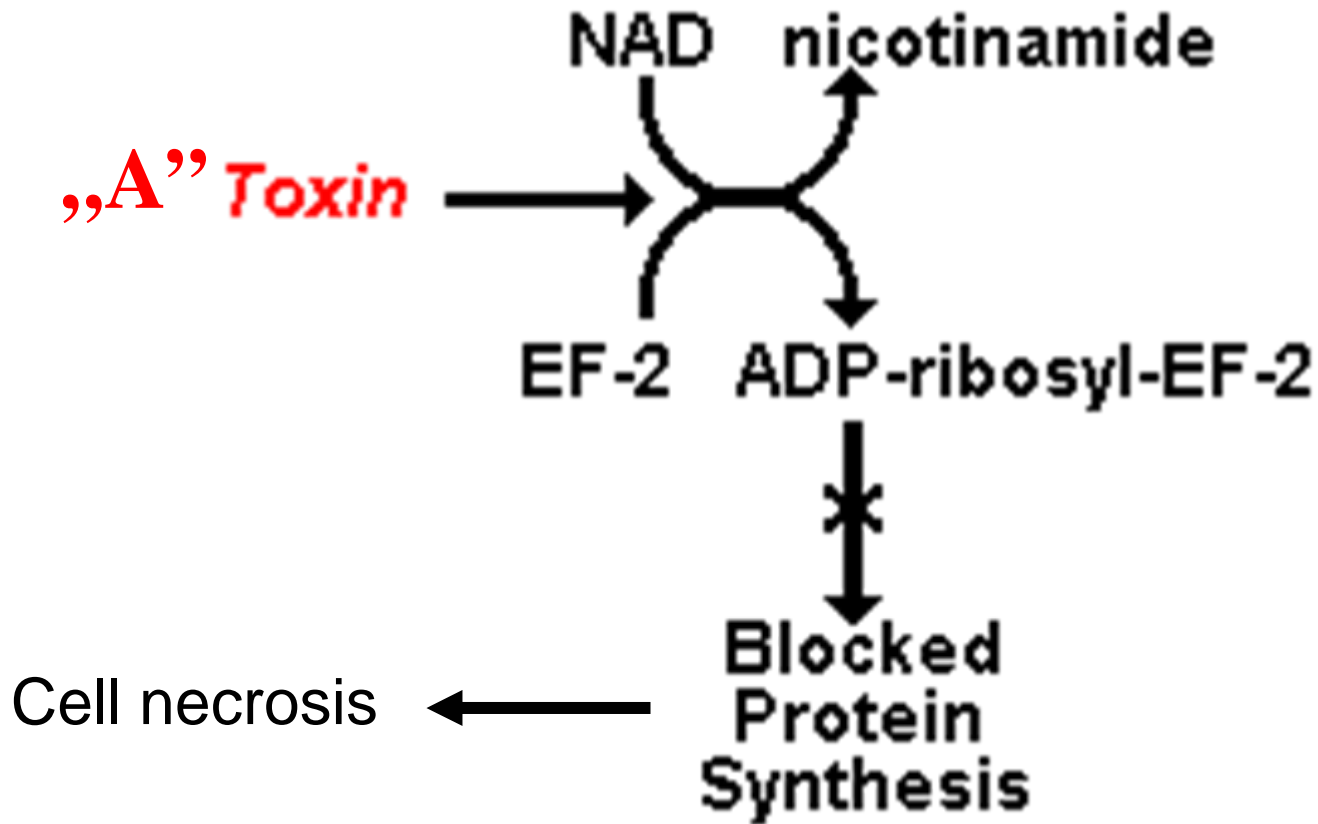


A domain: toxic (catalytic)
B domain: receptor binding
T domain: transmembrane

<http://textbookofbacteriology.net>



C. diphtheriae - exotoxin



Corynebacterium diphtheriae-5

Clinical finding: Diphtheria ("torokgyík")





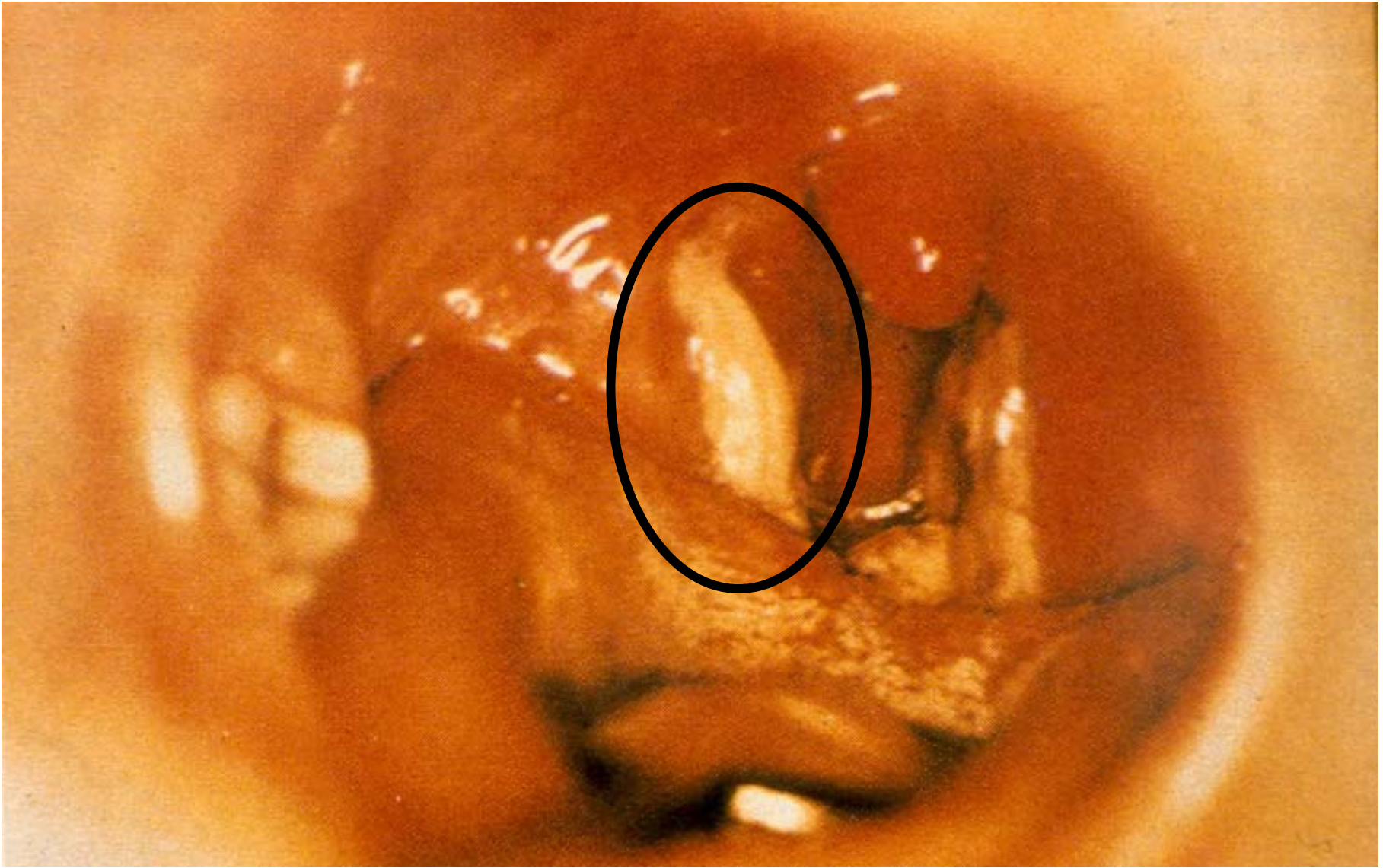
**193. DIPHTHERIÁS ALHÁRTYA TONSILLÁKON (a)
(b) GÉGEFŐBEN ÉS LÉGCSŐBEN**

Diphtheria - pseudomembrane



On tonsils

Tonsillardiphtherie.



Diphtheria - tonsils/tonsillák

torok
throat



Rachendiphtherie.

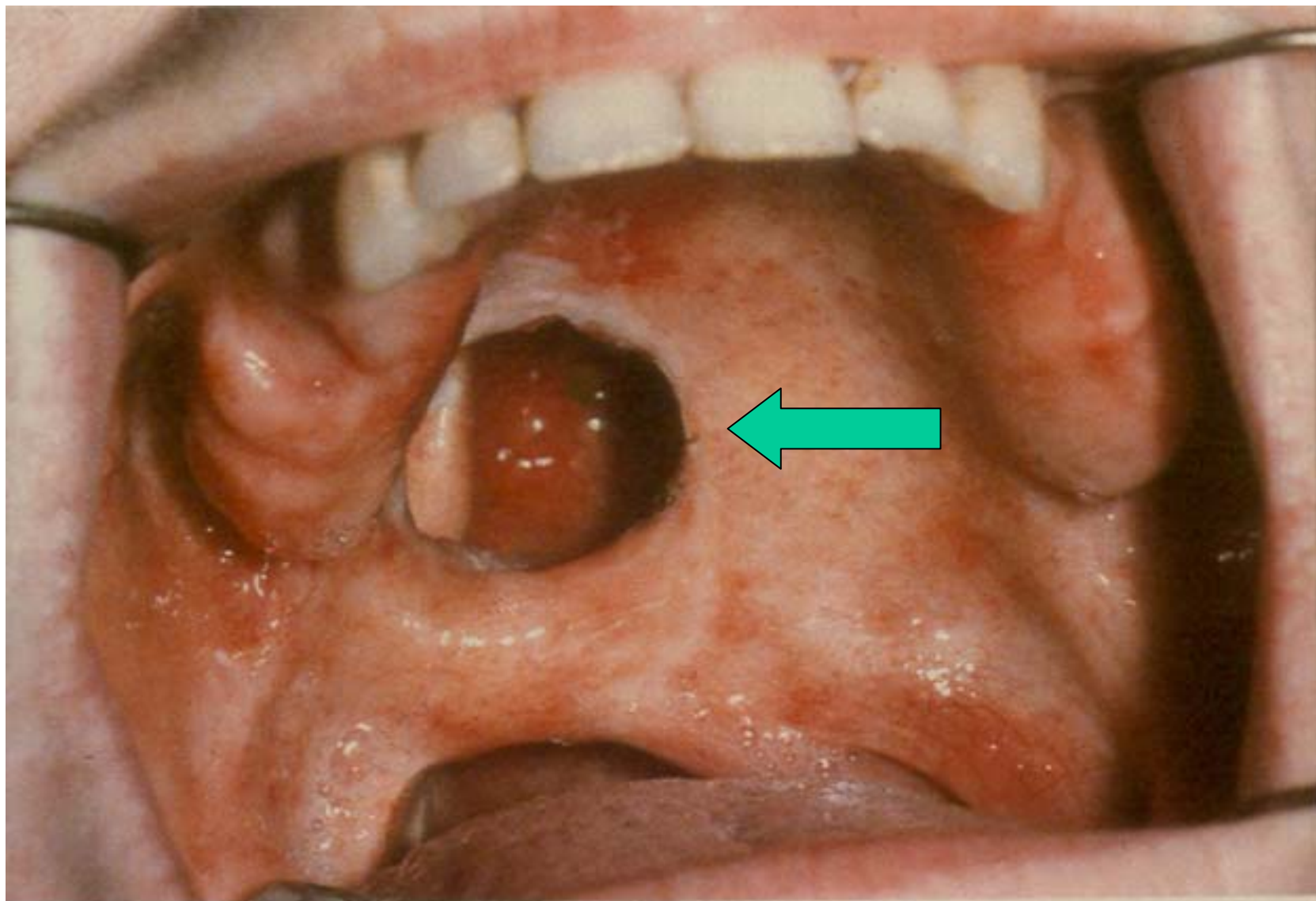


Fig. 1.38 Perforation of the soft palate, a late effect of pharyngeal diphtheria. By courtesy of Dr C. J. Meryon.

Lágyszájpad - perforáció



orr

Nasendiphtherie.

nose

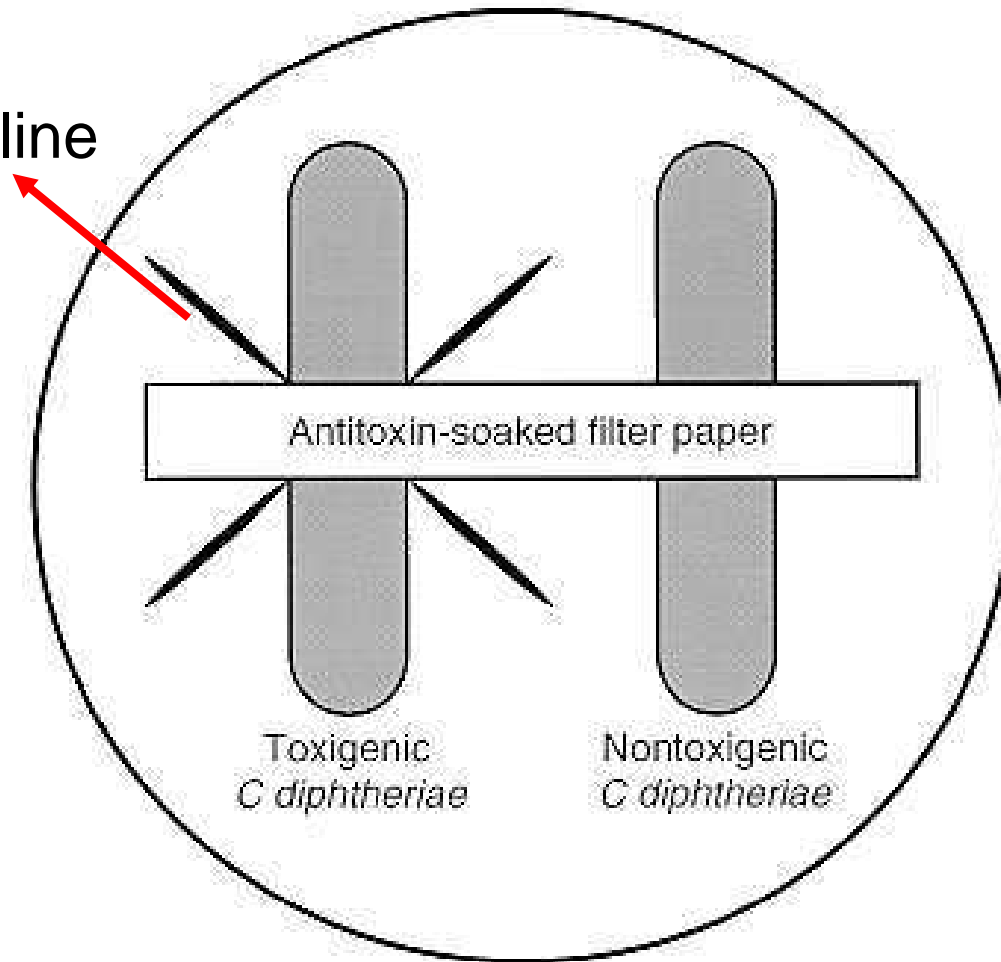
Corynebacterium diphtheriae-6

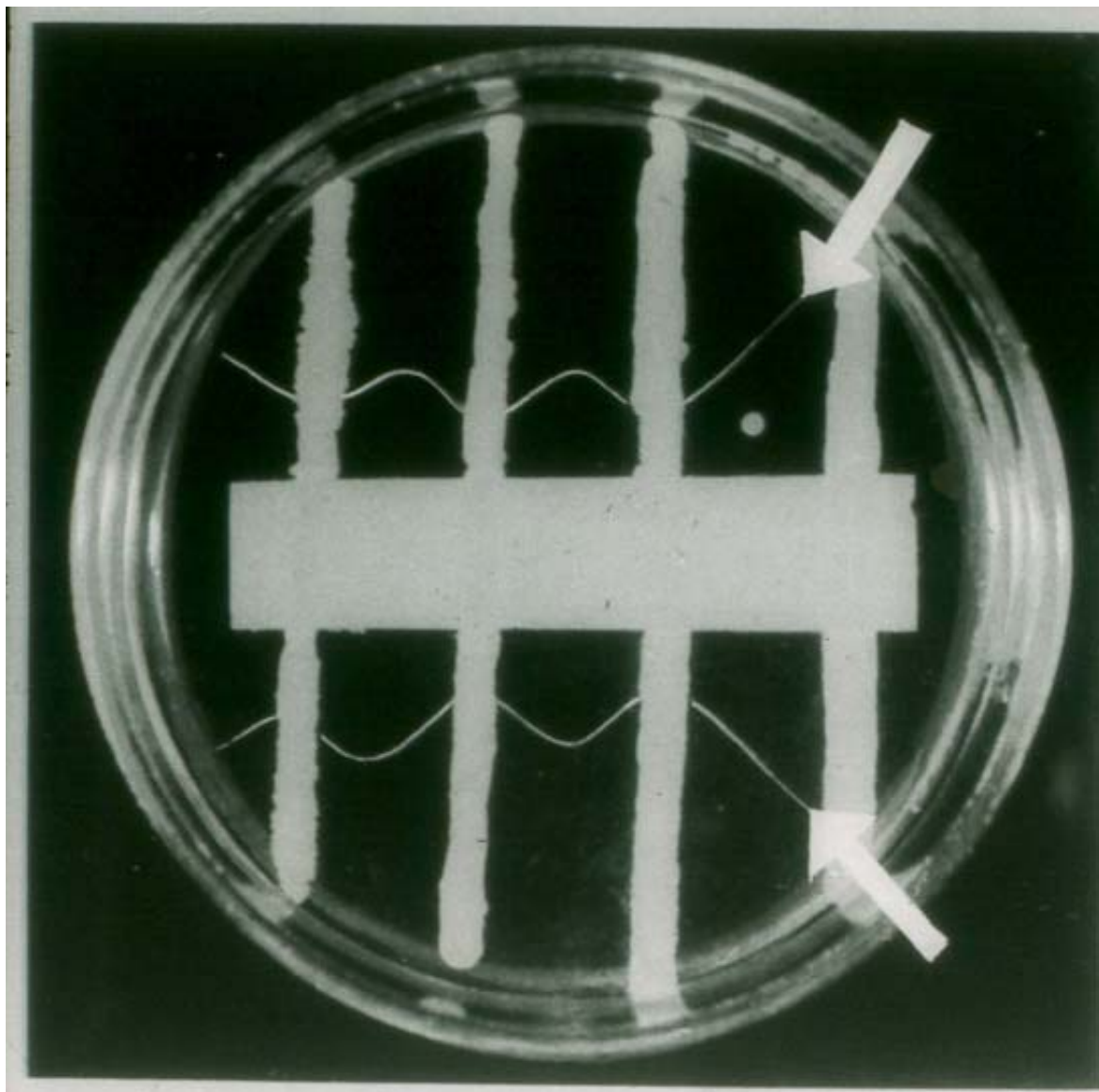
diagnosis:

- Clinical specimen direct microscopically
(Neisser-staining)
- cultivation → distinction between a pathogen (+)
and a pathogen (-) species! (sugar fermentation)
- virulence tests
(toxin production?)
 - ★ Elek - test (in vitro - paper strip)
 - ★ Römer - test (in vivo - guinea pig)
 - ★ PCR – toxin gene

Elek – test (agargel precipitation)

Precipitation line





114. ELEK-PRÓBA
(Di. toxintermelés
kimutatása)

Elek -test

**113. A RÖMER-FÉLE TOXINPRÓBA POZITÍV EREDMÉNYE
TENGERI MALAC BŐRÉN**



Römer test (guinea-pig)

Corynebacterium diphtheriae-7

Clinical findings - diff. dg.:

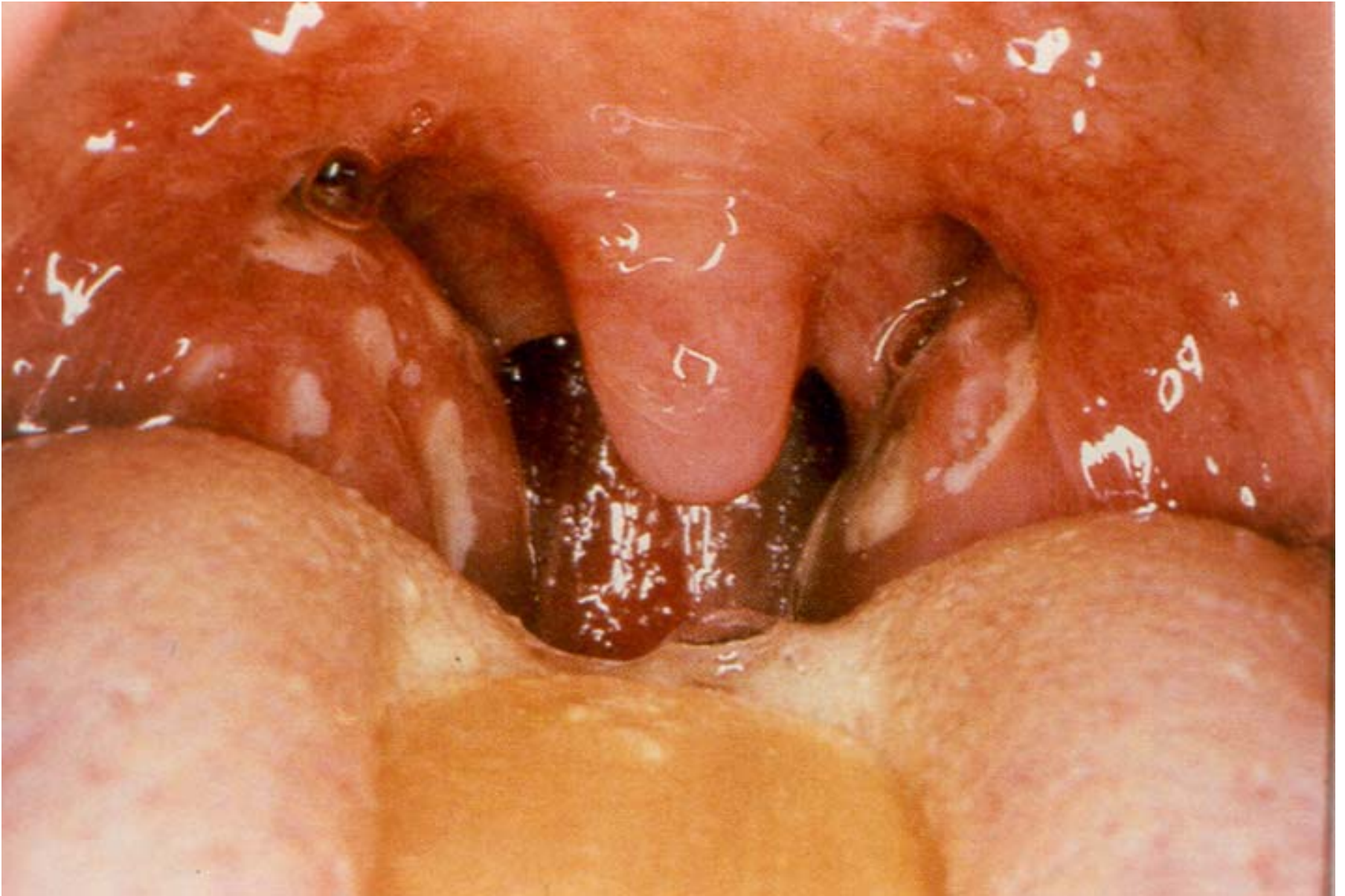
1. Tonsillitis follicularis

(S. pyogenes)

2. Plaut - Vincent angina

3. Soor (yeast)

4. Mononucleosis infectiosa



Tonsillitis follicularis - *S. pyogenes*

Corynebacterium diphtheriae-7

Clinical findings - diff. dg.:

1. Tonsillitis follicularis

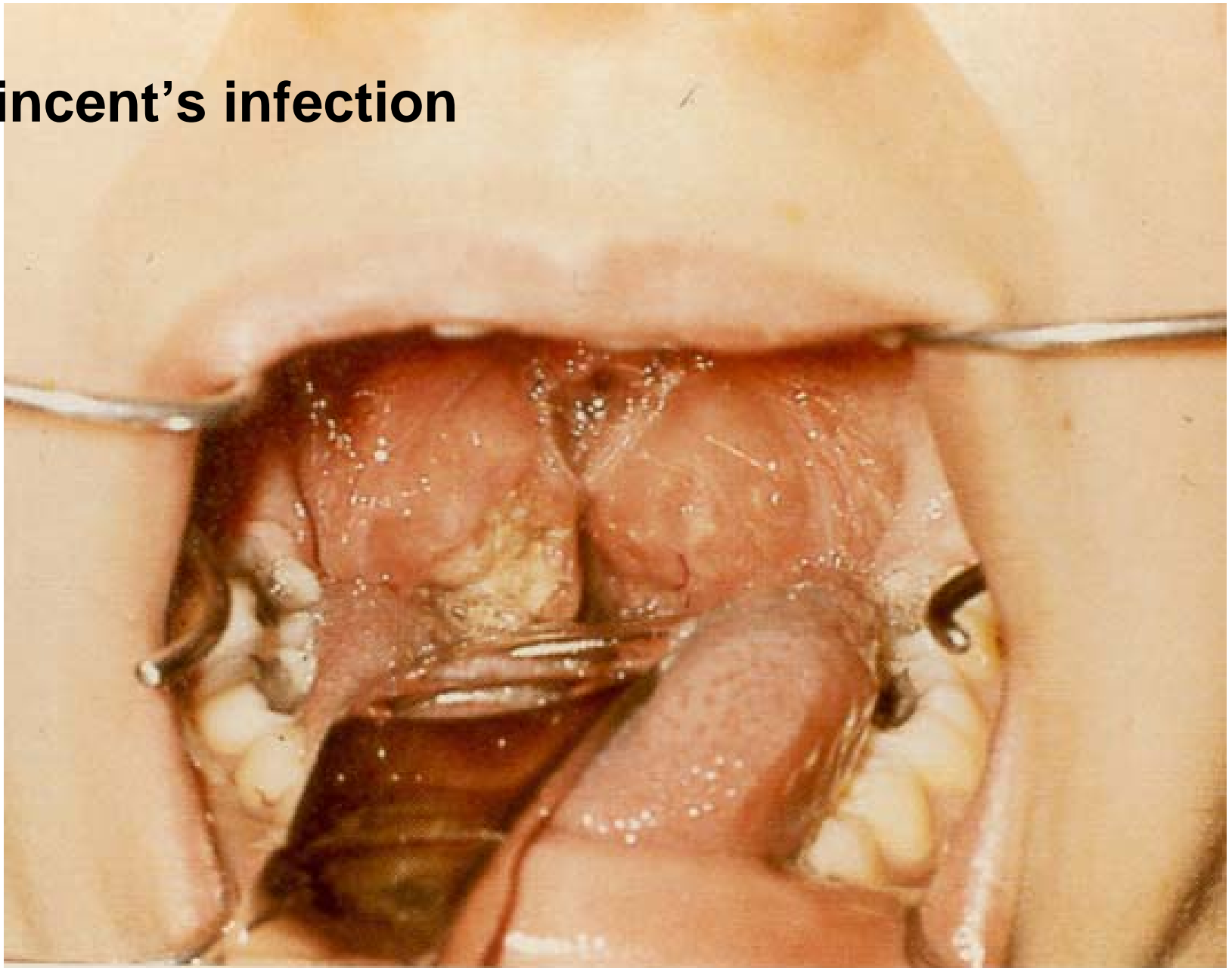
(*S. pyogenes*)

2. Plaut - Vincent angina

3. Soor (yeast)

4. Mononucleosis infectiosa

Vincent's infection



Angina Plaut-Vincent.

Corynebacterium diphtheriae-7

Clinical picture - diff. dg.:

1. Tonsillitis follicularis
(*S. pyogenes*)
2. Plaut - Vincent angina
- 3. Soor (yeast)**
4. Mononucleosis infectiosa

Soor (yeast)



Corynebacterium diphtheriae-7

Clinical findings - diff. dg.:

1. Tonsillitis follicularis

(*S. pyogenes*)

2. Plaut - Vincent angina

3. Soor (yeast)

4. **Mononucleosis infectiosa (EBV)**



Mononucleosis infectiosa

Corynebacterium diphtheriae-8,9

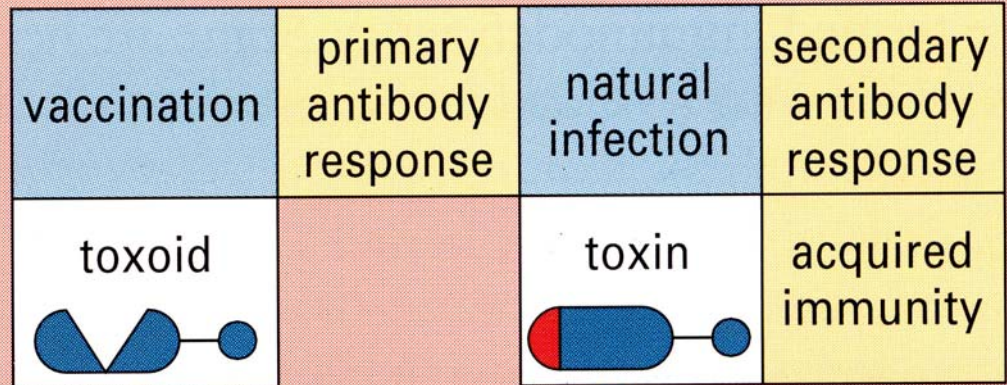
immunity: antitoxic

prophylaxis: DPT

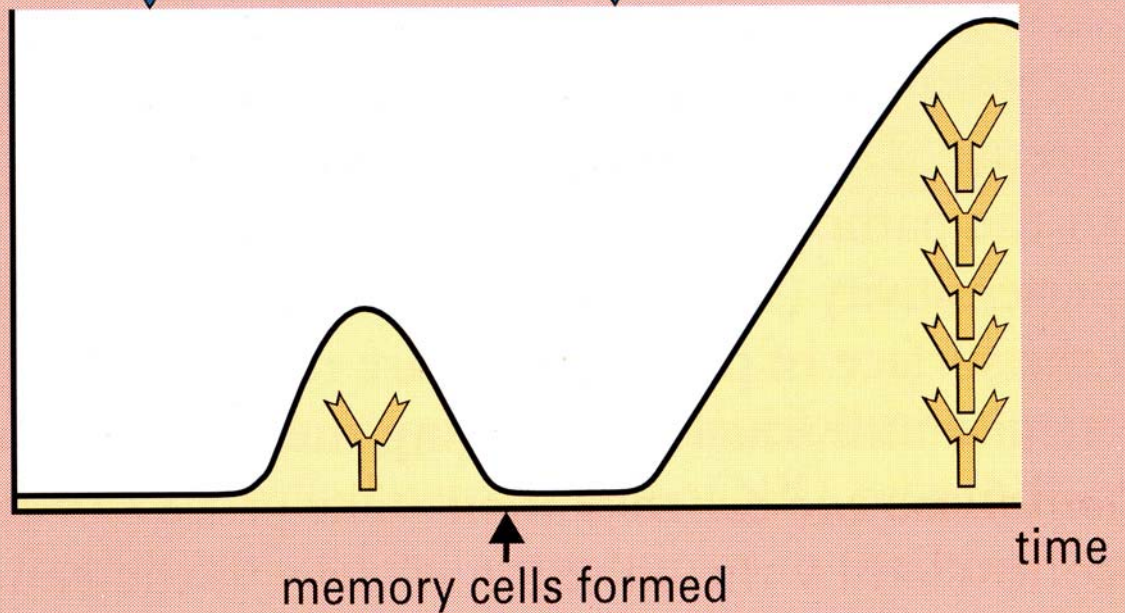
ACTIVE IMMUNIZATION

(TOXOID!!)

Principle of vaccination



antibody response



Active immunization

Corynebacterium diphtheriae-8,9

immunity: antitoxic

prophylaxis: DPT

ACTIVE IMMUNIZATION

(TOXOID!!)

treatment: Penicillin, Erythromycin +

**immediate passive immunization =
serumtherapy (Behring)**

other diphtheroids

See: Murray 5th box 27-4
Normal flora members

Corynebacterium jeikeium

Nosokomial – intensive care units
especially after gram negative bacteria caused sepsis
immunosuppressed patients
multiresistant

Listeria monocytogenes

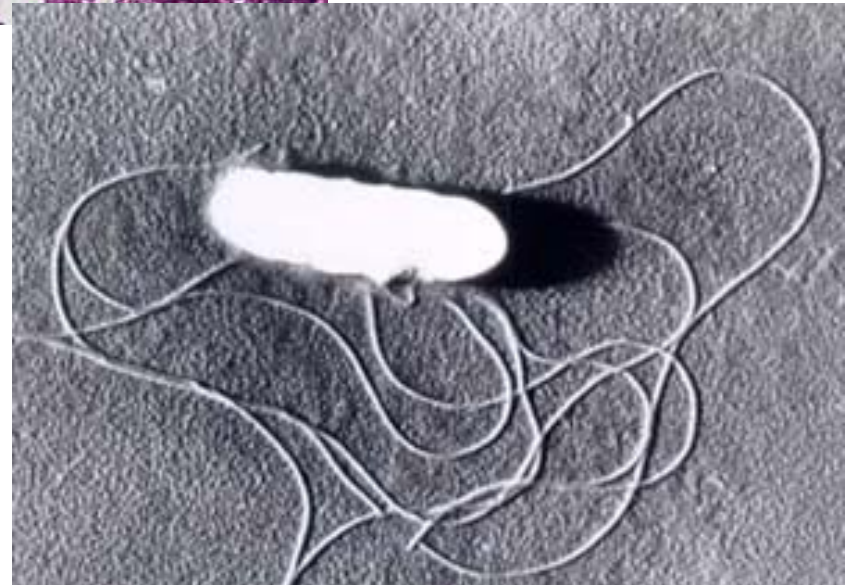
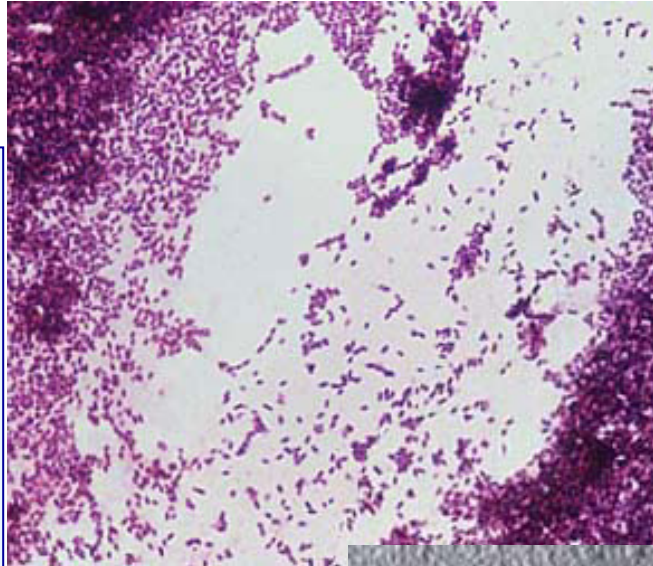
morphology:

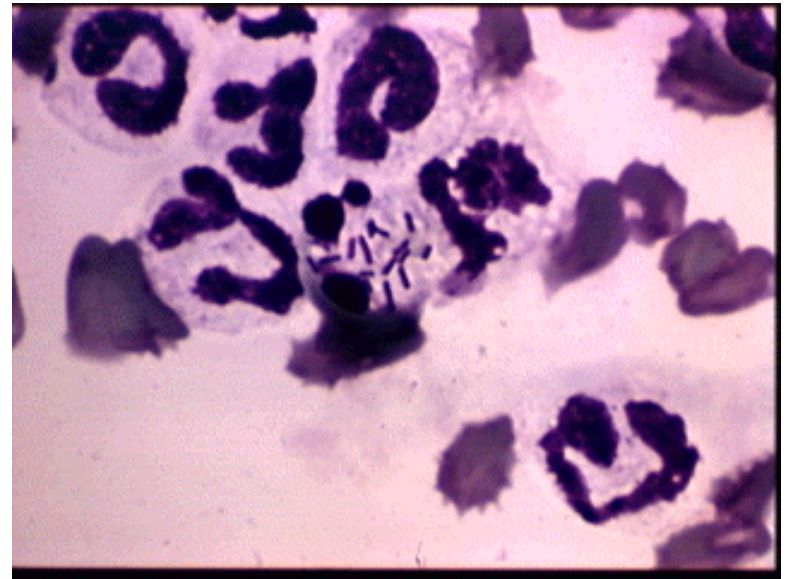
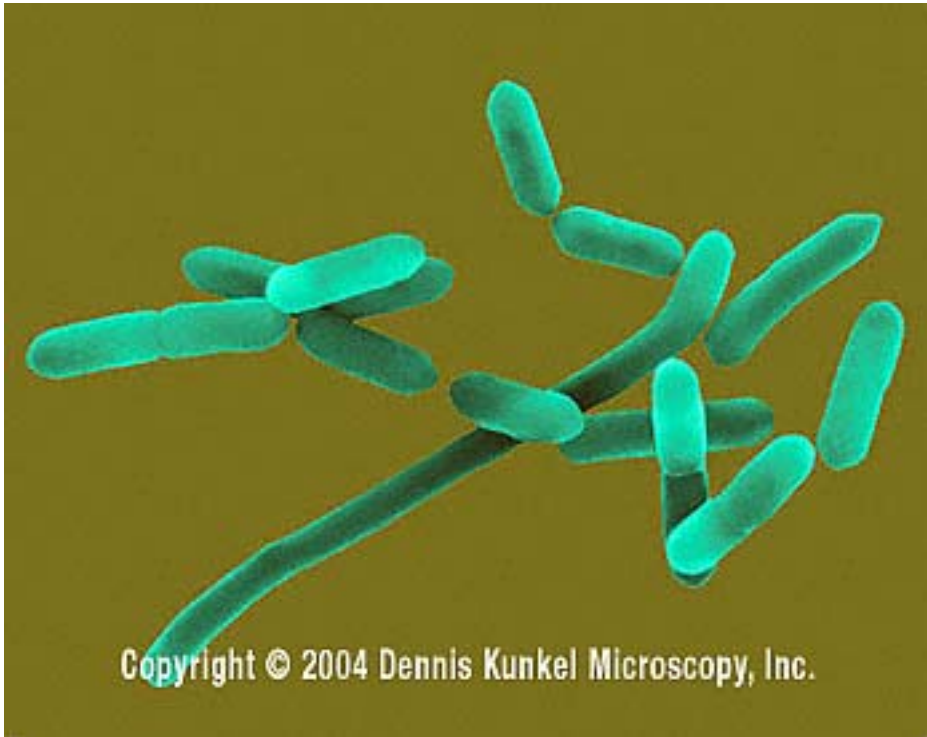
Gram + rod

cultivation:

blood agar

β -haemolysis



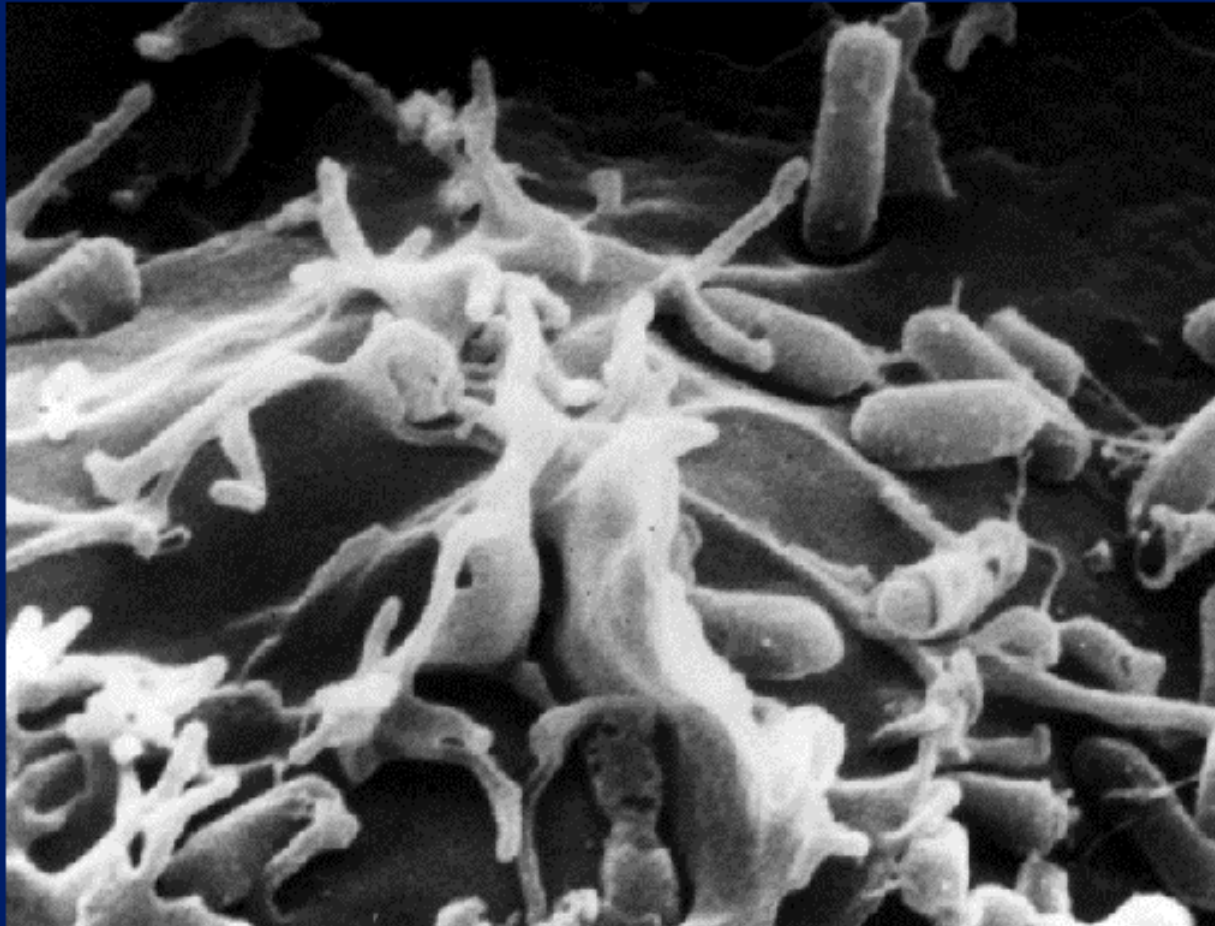


<http://textbookofbacteriology.net>,
<http://pathmicro.med.cs.edu>

www.med.sc.edu:85



Listeria on a Macrophage [SEM]



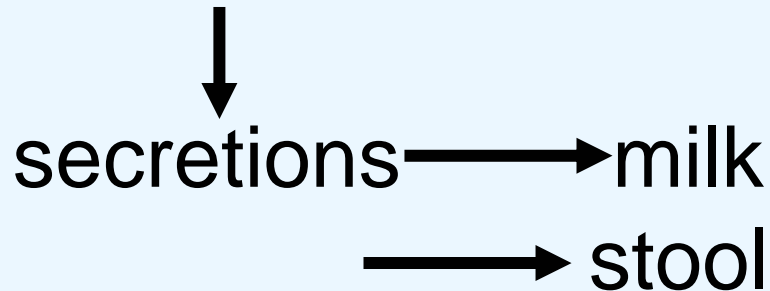
Listeria monocytogenes

resistant: to (1) heat, (2) cold (+4°C, fridge!),
(3) salt concentration

pathogenicity: animals and humans

pathogenesis, infection:

source: - carrier **animal**, healthy or ill



penetration: contaminated food, soil

transmission: oral, aerogen, **transplacental!!**
(during delivery as well)

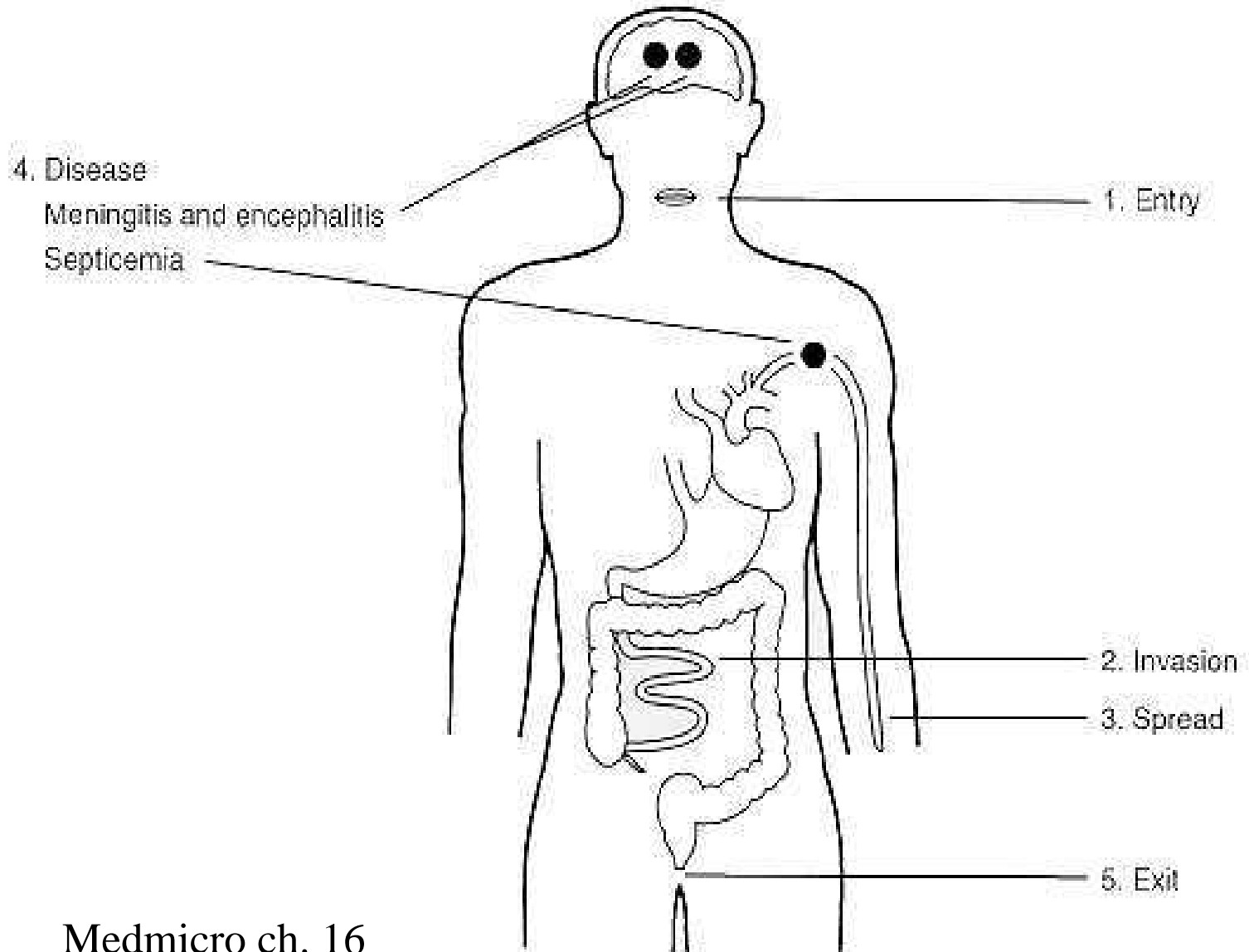
Listeria



www.fehd.gov.hk <http://pathmicro.med.cs.edu>,
www.med.sc.edu:85

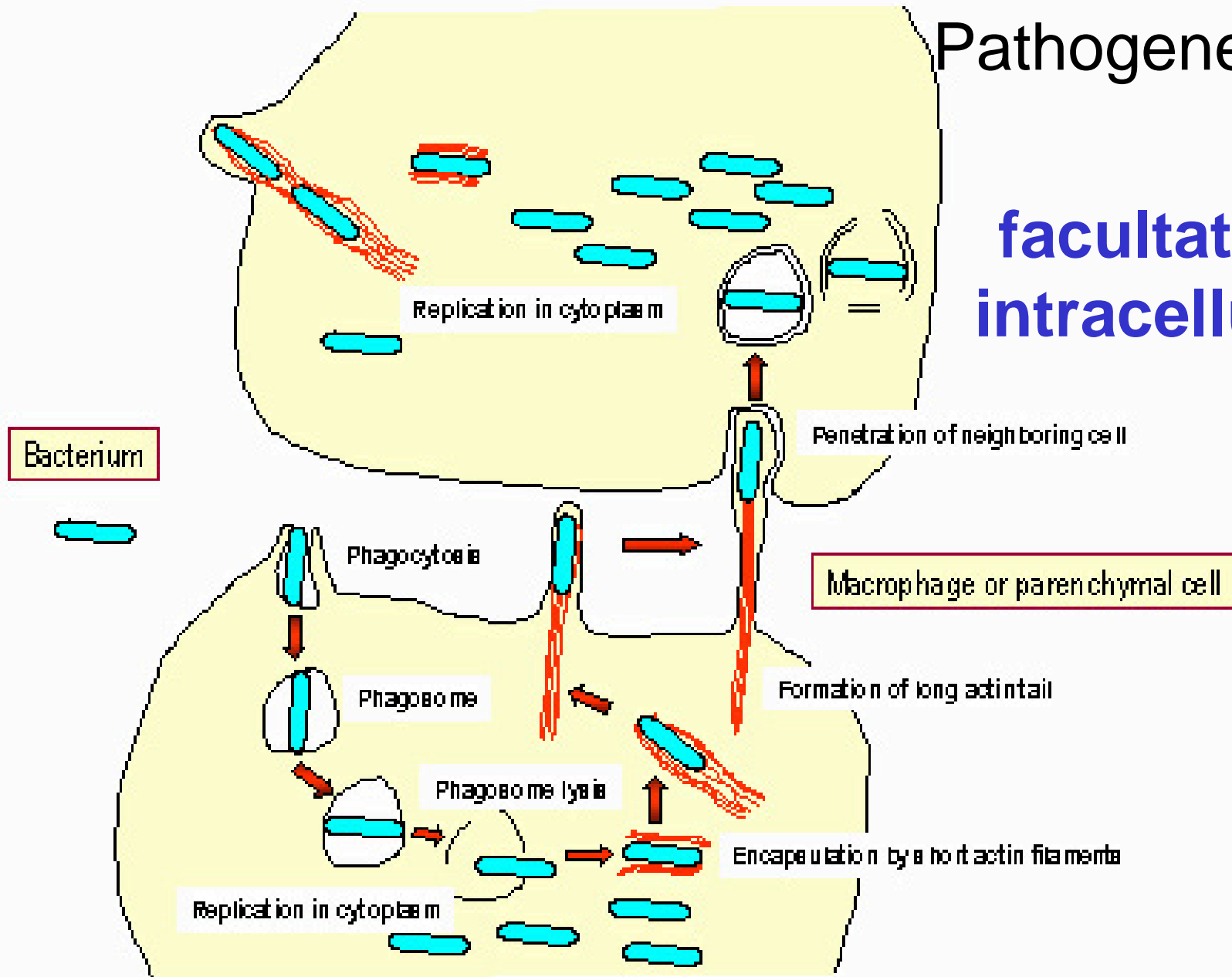


Pathogenesis-1



Pathogenesis-2

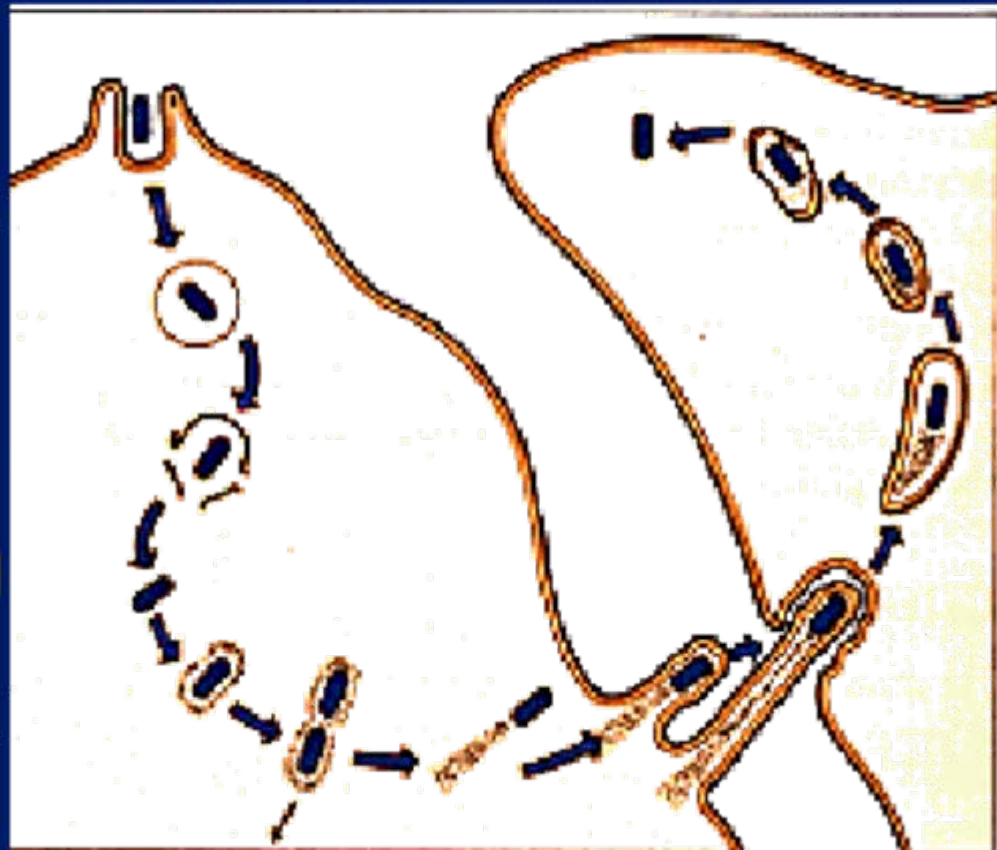
**facultative
intracellular**



Microbial Gene Expression, Virulence Factors and the Life-cycle of *Listeria monocytogenes*

Internalin
[entry]

Listeriolysin-O
LLO
[membrane lysis]



LLO
&
P-lipase
[lysis]

Act-A
[Actin-polymerization]

Listeria monocytogenes

clinical findings: **Listeriosis**

* local: granuloma

* general: meningitis, encephalitis,
endocarditis

* **Listeriosis in pregnant:**

AB, premature, dead

* **Listeriosis in new-born:**

granulomatosis infantis septica



*Granulomatosis
infanti septica*

Neonatal listeriosis

**nosokomial
meningitis
(story)**

Listeria monocytogenes

diagnosis: serology, occupation!
(anamnesis, patients history)

prophylaxis: exposition

★ keep all the rules!

★ treatment of sick animals

treatment:

Penicillin et al., Aminoglycosides !

(PIC: Ampicillin + Netromycin)

Erysipelothrix rhusiopathiae

morphology:

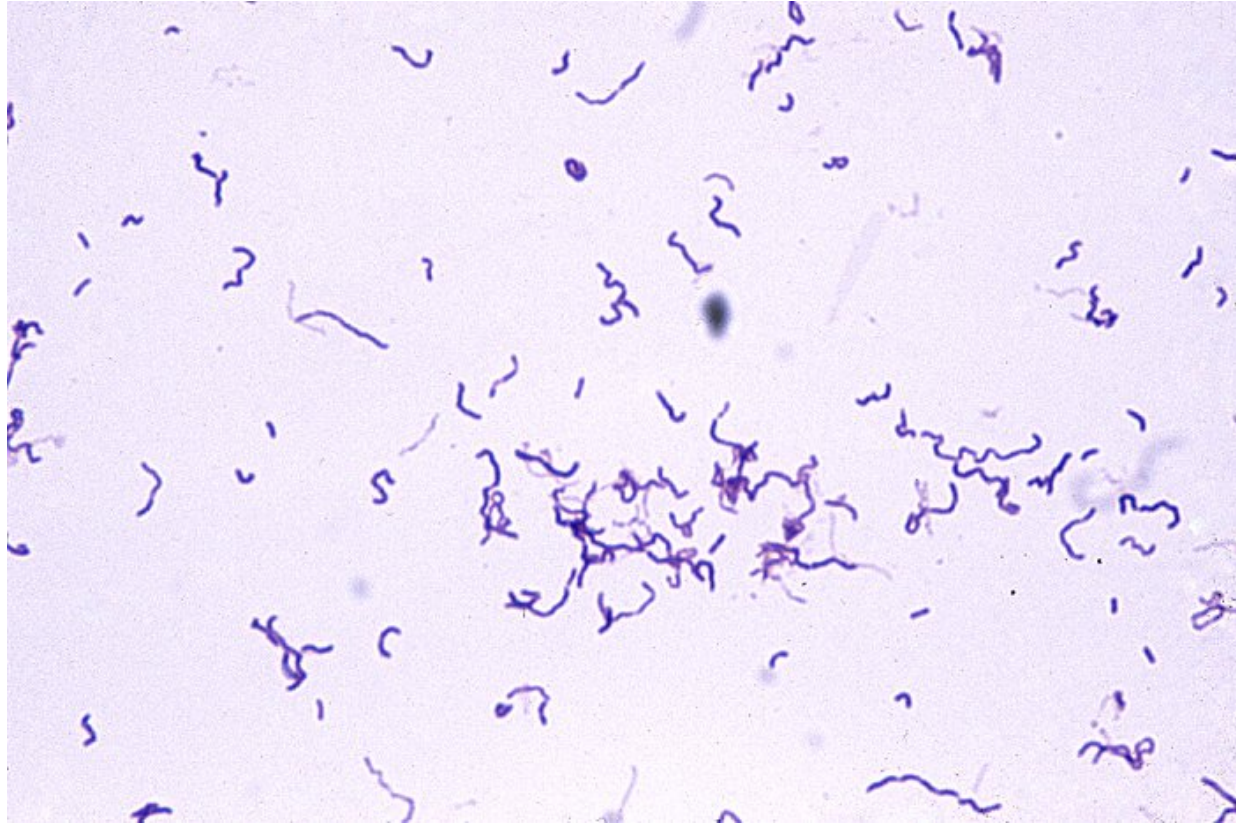
Gram + rod

cultivation:

blood agar

5-10% CO₂

tiny colonies



Erysipelothrix rhusiopathiae

pathogenicity: animals and humans

pathogenesis, infection:

source: - carrier **animal**, adult pigs – stool, sheep, cattle, poultry, fish etc. - healthy or ill

penetration: **WOUNDS** (scratch or puncture)

transmission: only from animals
no human to human!
(over 50 species can be carriers!)



Erysipelothrix rhusiopathiae

clinical finding

ANIMALS

Swine erysipelas

Joint ill (sheeps)

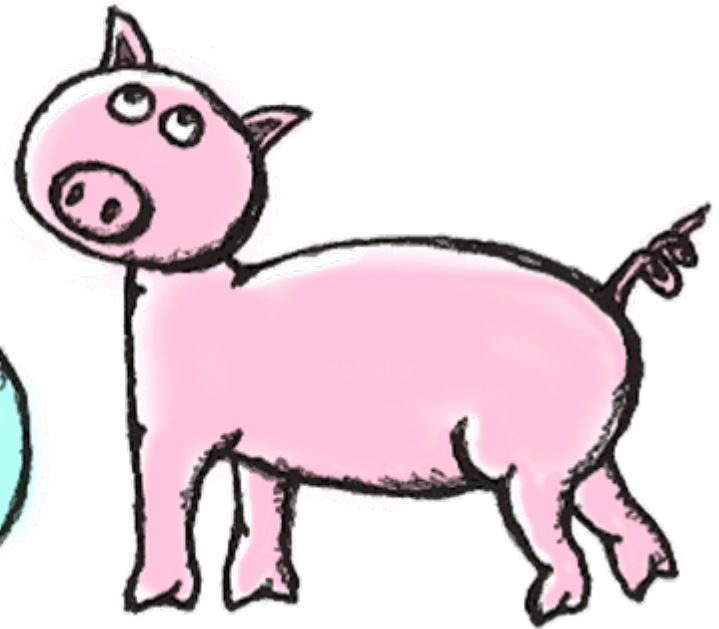
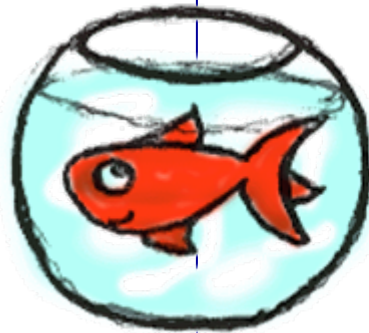
Sudden death

(in birds)

HUMANS

Erysipeloid

Sepsis, endocarditis



„Le rouget du porc”



ANIMALS

Swine erysipelas







Erysipelothrix rhusiopathiae

diagnosis: isolation

prophylaxis:

HUMANS: exposition

★ keep all the rules!

★ Animal vaccination

treatment:

Penicillin et al.



Lactobacillus spp.

Normal flora of the vagina
Probiotics

morphology:

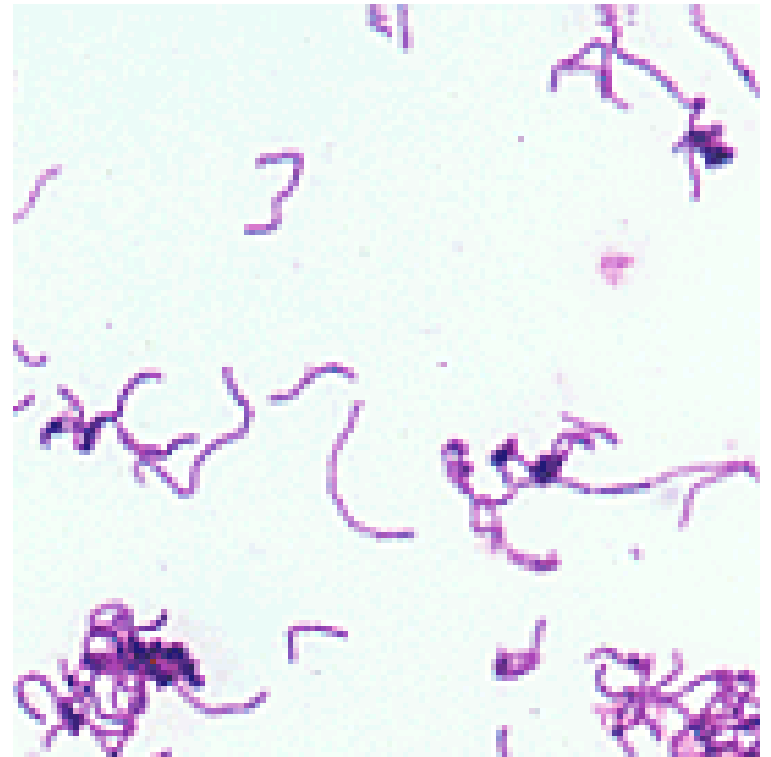
Gram + rod (anaerobic)

cultivation:

Rogosa medium

Pathology:

role in dental decay
(cariogenesis)



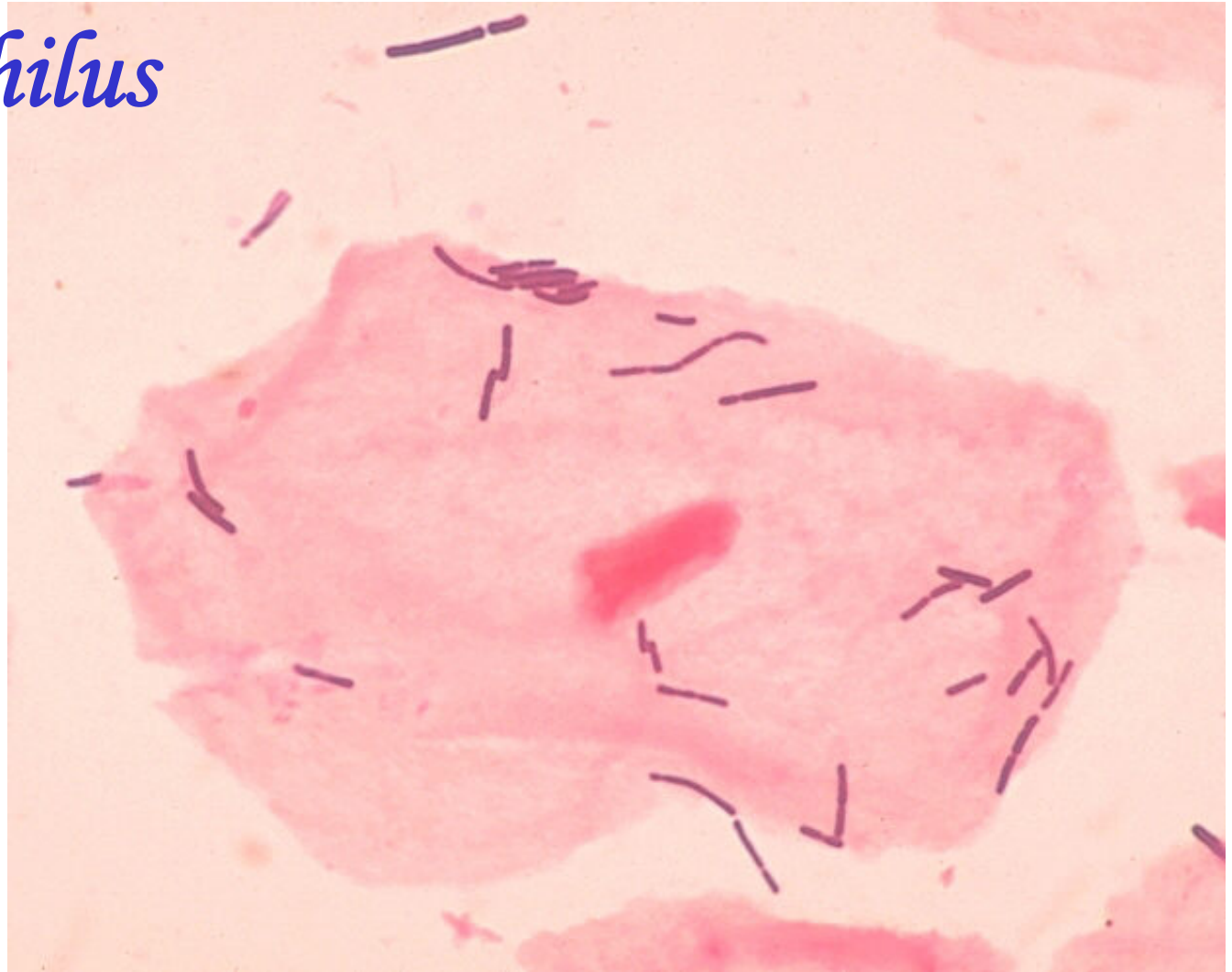
Lactobacillus spp. Normal flora of the vagina

L. acidophilus

lactic acid

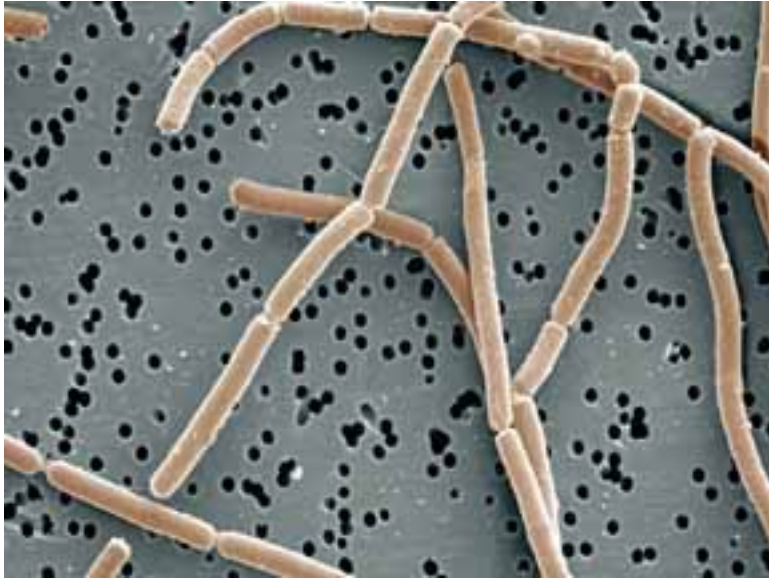


low pH

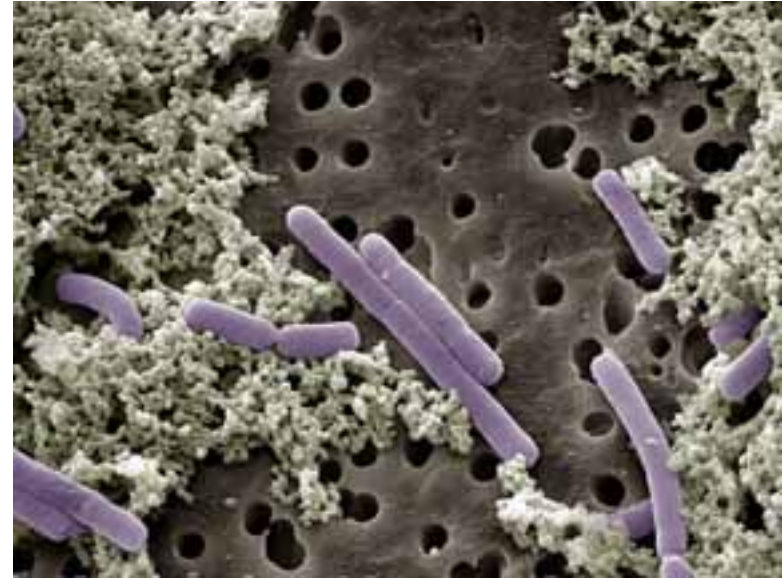


Vagina,
epithelial cell

Lactobacillus spp.



L. bulgaricus



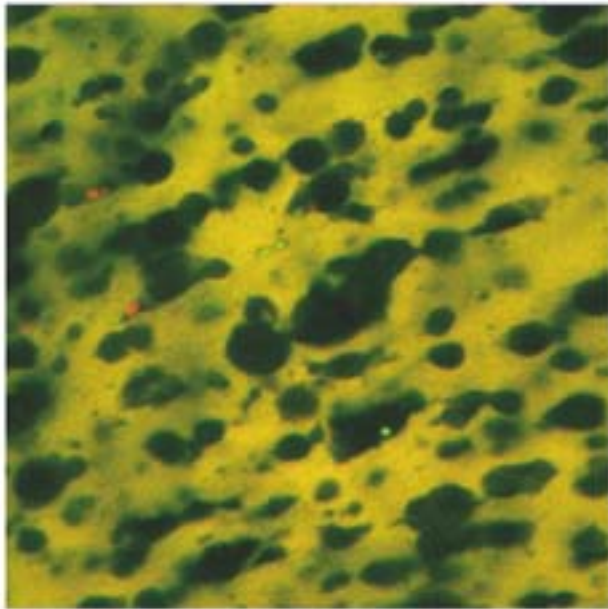
L. casei

**Food industry
Probiotics**

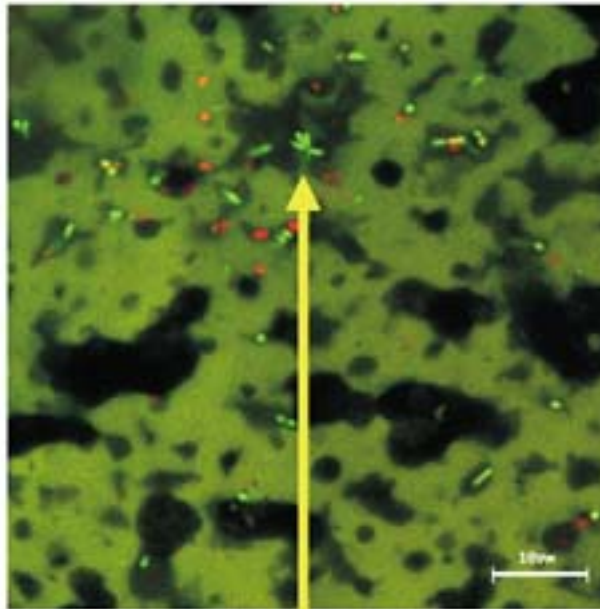


L. brevis

Control Cheese



Probiotic Cheese



Live probiotic culture (green)

www.teagasc.ie



soutecz.abo.cz, www.velvethammer.hu,
www.eltean.com

Probiotics

- are live microorganisms administered in adequate amounts which confer a beneficial health effect on the host
- are our „friendly bacteria”
- are products with live active bacteria cultures

Prebiotics

- are nondigestible food ingredients that beneficially affect (stimulate) the normal flora bacteria in the colon

Probiotics

Synbiotics

Prebiotics

Living bacteria
in the diet

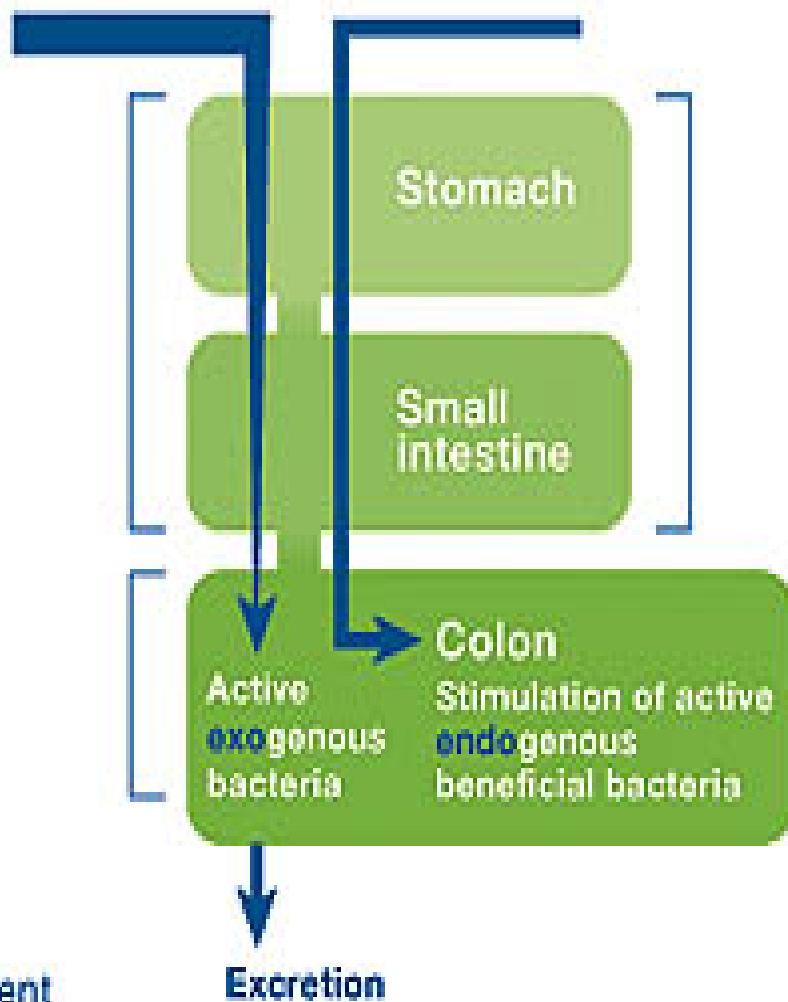


Survival*



Fixation*
Activity*

* Strain-dependent



Inulin or
oligofructose
in the diet



Total transfer
to the colon

Each gram
of oligofructose
or inulin



Stimulates
the
bifidobacteria

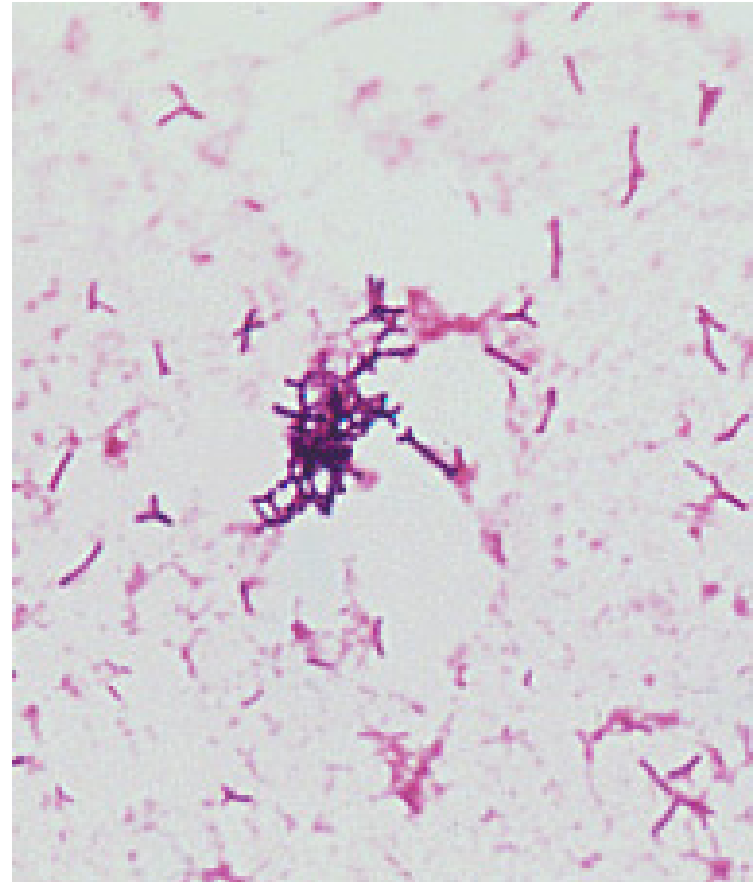
Bifidobacteria

B. bifidum

**Predominant:
breast-fed babies**

B. longum

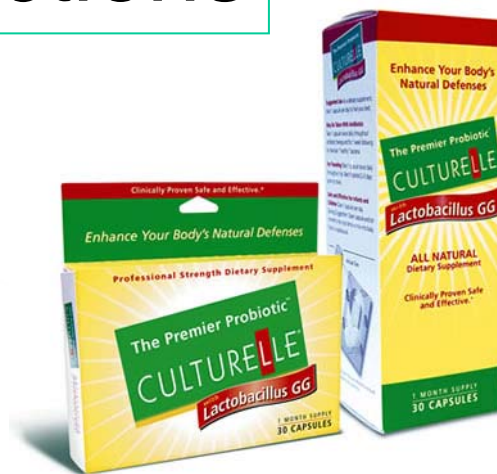
Both: probiotic



**Gram + , lactic acid rods
of *B. bifidum***

Probiotic bacteria: *Lactobacilli* in therapy

Diarrhoeal diseases
Vaginal candidiasis
Bacterial vaginosis
Urinary tract infections



The end

