

## CHAPTER 05: STUDY of the MAJOR BACTERIAL GROUPS

### 5.6. RICKETTSIAS and CHLAMYDIAS

#### 1. Introduction

In the first edition of the Bergey's Manual, the genera *Chlamydia* and *Rickettsia* were grouped together, as both are **Gram-negative intracellular parasites**. In the second edition, *Rickettsia* are placed within the *α-proteobacteria*, while the genus *Chlamydia* is included in the phylum *Chlamydiae*, classified with genetically similar bacteria whose cell walls do not contain peptidoglycan.

#### 2. *Chlamydia*

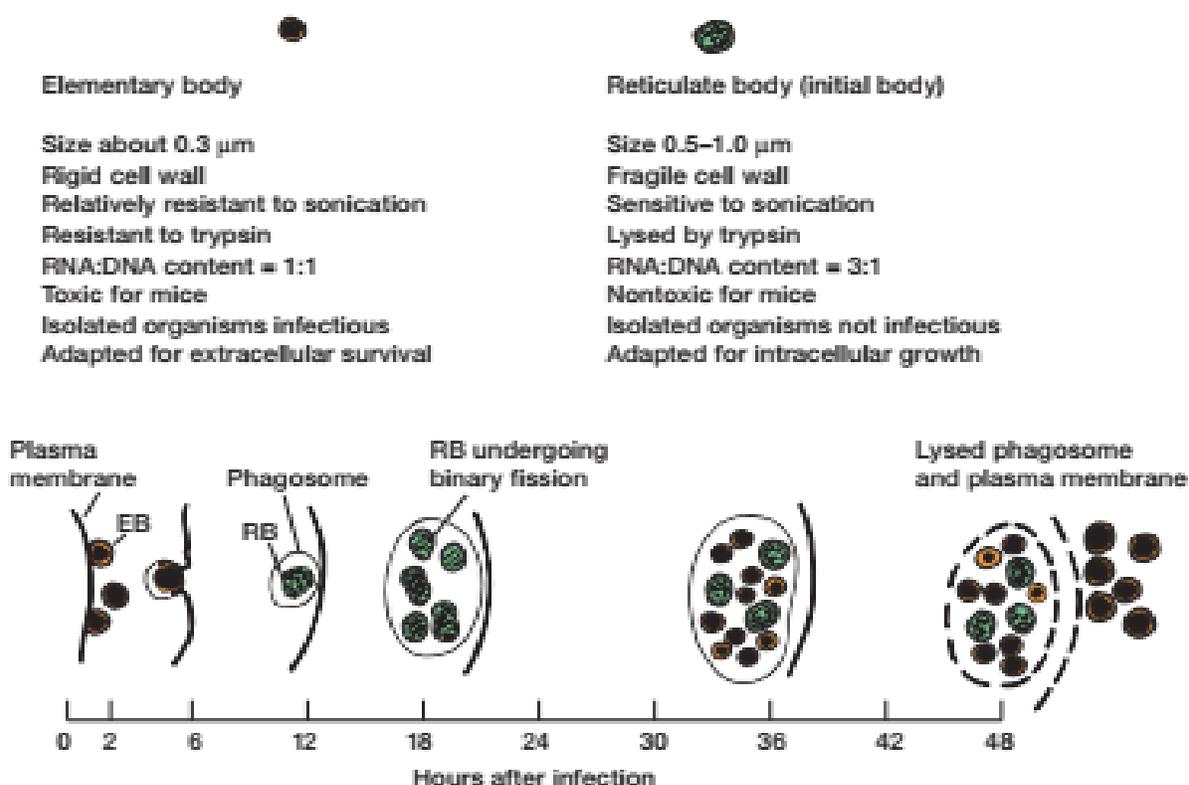
The Gram-negative *Chlamydiae* are **obligate intracellular parasites**. That is, they must grow and reproduce within host cells. Although their ability to cause disease is widely recognized, many species grow within protists and animal cells without adverse effects (natural reservoir for the *Chlamydiae*).

The phylum *Chlamydiae* has a single class *Chlamydiai* and one order *Chlamydiales*. The order *Chlamydiales* consists currently of eight families, type family *Chlamydiaceae*. The genus *Chlamydia* is by far the most important and best studied.

*Chlamydiae* are nonmotile, coccoid bacteria, ranging in size from 0.2 to 1.5 μm. They reproduce only within cytoplasmic vesicles of host cells by a unique developmental cycle involving the formation of two cell types: **elementary bodies** and **reticulate bodies**. Although their envelope resembles that of other Gram-negative bacteria, the cell wall differs in lacking muramic acid and a peptidoglycan layer. Elementary bodies achieve osmotic stability by cross-linking their outer membrane proteins, and possibly periplasmic proteins, with disulfide bonds. *Chlamydiae* are extremely limited metabolically, relying on their host cells for key metabolites. This is reflected in the size of their genome, the G + C content is 41 to 44%.

*Chlamydia* reproduction begins with the attachment of an **elementary body (EB)** to the cell surface (**Fig. 01**). Elementary bodies are 0.2 to 0.6 μm in diameter, contain electron dense nuclear material and a rigid cell wall, and are infectious. The host cell phagocytoses the EB, which are held

in inclusion bodies where the EB reorganizes itself to form a **reticulate body (RB)** or **initial body (IB)**. The RB is specialized for reproduction rather than infection. Reticulate bodies are 0.6 to 1.5  $\mu\text{m}$  in diameter and have less dense nuclear material and more ribosomes than EBs; their walls are also more flexible. About 8 to 10 hours after infection, the reticulate body undergoes binary fission and RB reproduction continues until the host cell dies. A chlamydia-filled inclusion can become large enough to be seen in a light microscope and even fill the host cytoplasm. After 20 to 25 hours, RBs begin to differentiate into infectious EBs and continue this process until the host cell lyses and releases the *Chlamydiae* 48 to 72 hours after infection.



**Figure 01:** Chlamydial life cycle.

Chlamydial metabolism is very different from that of other Gram-negative bacteria. It had been thought that chlamydiae cannot catabolize carbohydrates or other substances or synthesize ATP. *Chlamydia psittaci*, one of the best-studied species, lacks both flavoprotein and cytochrome electron transport chain carriers, but has a membrane translocase that acquires host ATP in exchange for ADP. Thus, chlamydiae seem to be energy parasites that are completely dependent on their hosts for ATP. The *C. trachomatis* genome sequence indicates that the bacterium may be able to synthesize at least some ATP. Although there are two genes for ATP/ADP translocases,

there also are genes for substrate-level phosphorylation, electron transport, and oxidative phosphorylation. When supplied with precursors from the host, RBs can synthesize DNA, RNA, glycogen, lipids, and proteins. Presumably the RBs have porins and active membrane transport proteins (little is known about these). They also can synthesize at least some amino acids and coenzymes. The EBs have minimal metabolic activity and cannot take in ATP or synthesize proteins. They are designed exclusively for transmission and infection.

There are three described species of *Chlamydiae* that commonly infect humans:

**1. *Chlamydia trachomatis*:** causes ocular infection, which may lead to blindness (trachoma), a disease still found in tropical and subtropical endemic areas, and it causes genitourinary tract infections, which may cause tubal factor infertility (fallopian tube obstruction), which is a serious complication of chronic salpingitis. Chlamydia infection is a common sexually transmitted infection (STI) in humans caused by the bacterium *Chlamydia trachomatis*.

*C. trachomatis* infection can be effectively cured with antibiotics once it is detected. Current guidelines recommend: azithromycin, doxycycline, erythromycin, or ofloxacin. Agents recommended for pregnant women include erythromycin or amoxicillin.

**2. *Chlamydia pneumoniae*:** causes a form of pneumonia. This human biovar of *Chlamydia pneumoniae* is a common respiratory pathogen with manifestations of pharyngitis, bronchitis, and pneumonia and is associated with atherosclerosis and coronary heart disease.

**3. *Chlamydia psittaci*:** causes psittacosis (infection acquired from a psittacine birds). The systemic infection in birds is often asymptomatic. Humans may contract *Chlamydia psittaci* infection by the airborne route, either by direct contact with infected birds or indirectly by inhalation of dust contaminated with excreta of infected birds.

### **3. *Rickettsia***

The genus *Rickettsia* is placed in the order *Rickettsiales* (rod-shaped, coccoid or irregularly shaped bacteria with typical Gram-negative cell walls and no flagella. Multiply only inside host cells) and family *Rickettsiaceae* of the  $\alpha$ -proteobacteria class. The members of this family are mainly diplococcus-shaped, but can also be rod shaped or coccoid. Gram negative. Obligately intracellular. Intimately associated with arthropod hosts. No flagella or endospores occur. No member of the family has yet been cultivated in cell-free media. Some species can be parasitic in

man and other vertebrates, causing diseases (e.g., typhus) that are transmitted by arthropods (lice, fleas, ticks and mites). Some are confined to the invertebrate host as pathogens or symbionts. DNA G+C% = 29–33. Type genus: *Rickettsia*.

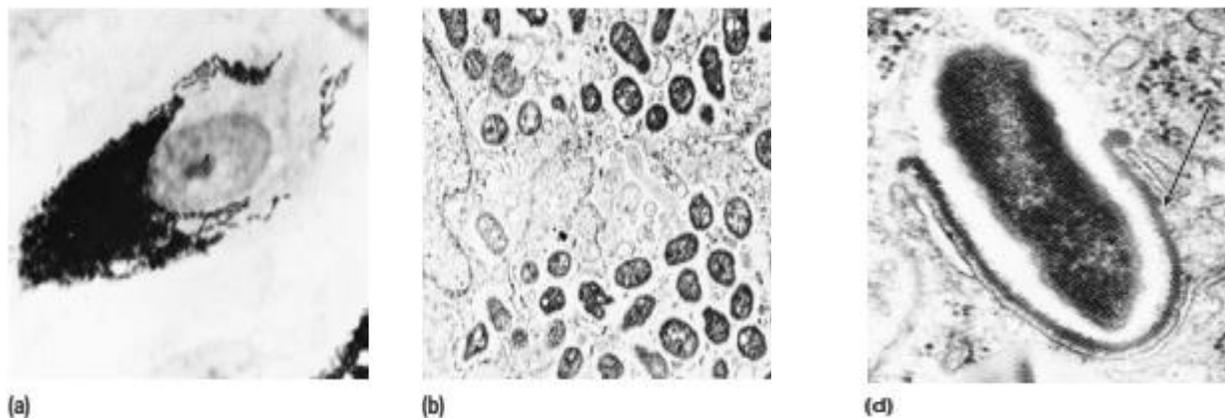
Short, often paired rods, 0.3–0.5 x 0.8–2.0  $\mu\text{m}$ . The rickettsial envelope has a typical Gram-negative structure. The cells are often surrounded by a protein microcapsular layer and slime layer. The organisms are **obligately intracellular and reside free in the cytoplasm of the eucaryotic host cell**, where they divide by **binary fission**. Rickettsiae of the Spotted Fever Group (SFG) may also reside in the nucleus of the eucaryotic host cells. Rickettsiae are **closely associated with arthropods** (ticks, mites, fleas, lice, and other insects) **for their maintenance in nature**. Their natural cycle usually involves both a vertebrate and an invertebrate host. For some, the arthropod host is both a reservoir (primary hosts) and a vector. **Transovarian transmission** of the agent from the infected female to the next generation is the essential mechanism for the maintenance of many species. Rickettsiae derive energy from the metabolism of glutamate via the citric acid cycle, but do not utilize glucose. They transport and metabolize phosphorylated compounds but do not synthesize or degrade nucleoside monophosphates.

The rickettsial plasma membrane has carrier-mediated transport systems, and host cell nutrients and coenzymes are absorbed and directly used. For example, rickettsias take up both  $\text{NAD}^+$  and uridine diphosphate glucose. Their membrane also has an adenylate exchange carrier that exchanges ADP for external ATP. Thus, host ATP may provide much of the energy needed for growth. This metabolic dependence explains why many of these organisms must be cultivated in the yolk sacs of chick embryos or in tissue culture cells.

Rickettsiae are inoculated into the skin in the saliva during feeding by an infected tick, mite, or flea or by scratching of rickettsia-laden feces deposited by an infected louse or flea. Rickettsiae are distributed throughout the body via the bloodstream where they enter their principal target, endothelial cells.

In the interaction of rickettsiae with host cells, the entry process involves three steps: (1) attachment, (2) internalization and (3) escape from the phagosome. Rickettsiae adhere to the host cell by means of a **rickettsial adhesin** and a host cell receptor. Once phagocytosed by the host cell, rickettsiae rapidly escape from the phagosome prior to its fusion with a lysosome (**Fig. 02**). Within the cytosol, rickettsiae acquire nutrients, ATP, amino acids, and nucleic acid precursors from the

host by active transport mechanisms and replicate slowly. **Rickettsiae are etiological agents of typhus and spotted fevers in humans.** There are 21 recognized species. Type species: *Rickettsia prowazekii*.



**Figure 02: *Rickettsia*.**

[Rickettsial morphology and reproduction. [(a) A human fibroblast filled with *Rickettsia prowazekii* (31,200). (b) A chicken embryo fibroblast late in infection with free cytoplasmic *R. prowazekii* (313,600). (d) *R. prowazekii* leaving a disrupted phagosome (arrow) and entering the cytoplasmic matrix (3 46,000)]

A number of *Rickettsia* species cause severe disease in humans. Before the advent of the broad-spectrum antibiotics, epidemic typhus and Rocky Mountain spotted fever; caused by *R. prowazekii* and *R. rickettsii*, respectively; had a very high case fatality rate.

Other named rickettsiae and their diseases include: *R. typhi* (murine typhus), *R. conorii* (boutonneuse fever, Mediterranean spotted fever, Astrakhan fever, Israeli spotted fever), *R. sibirica* (North Asian tick typhus), *R. australis* (Queensland tick typhus), *R. akari* (rickettsialpox), *R. japonica* (Japanese spotted fever), *R. africae* (African tick bite fever), *R. honei* (Flinders Island spotted fever), and *R. felis* (flea-borne spotted fever).

Rickettsiae are naturally resistant to most classes of antimicrobial agents. Contemporary methods determine rickettsial susceptibility to antimicrobial drugs in cell culture. Even the most effective antimicrobial activities against *Rickettsia* are bacteriostatic rather than bactericidal.

- The antimicrobial drugs that are most effective, i.e., that have the lowest minimal inhibitory concentrations, belong to the following classes: **tetracyclines, chloramphenicol, rifampin, fluoroquinolones** (*pefloxacin, ofloxacin, ciprofloxacin*) and some, but not all, **macrolides** (*josamycin, azithromycin, clarithromycin*);

- **Doxycycline** is the drug of choice in most clinical settings, except for patients who are pregnant or hypersensitive to tetracyclines;

- **Josamycin** has been shown to be an effective treatment of boutonuse fever during pregnancy;

- **Ciprofloxacin, ofloxacin, pefloxacin, clarithromycin, and azithromycin** have been reported to ameliorate the course of boutonuse fever;

- The results of treatment with **rifampin** have been less conclusive;

*R. massiliae*, *R. aeschlimannii*, *R. montanensis*, and *R. rhipicephali* are relatively resistant to **rifampin**;

- *Rickettsia prowazekii* is susceptible to **erythromycin** *in vitro*;

- *Rickettsia* species are resistant to **aminoglycosides** and to **penicillin** and other **β-lactam** antimicrobial agents.