34.1 INTRODUCTION

*Mycoplasma* species are the smallest free-living organisms. These organisms are unique among prokaryotes in that they lack a cell wall, hence lack fixed shape or size and also lack Gram stain reaction and their lack of susceptibility to beta-lactams. Because of their plasticity, they can pass through bacterial filters of 450nm pore size and have often been mistaken for viruses. *Mycoplasma* organisms are usually associated with mucosal surfaces of respiratory and urogenital tracts. They rarely penetrate the submucosa, except in the case of immunosuppression or instrumentation, when they may invade the bloodstream and disseminate.

Species most commonly associated with infections are *Mycoplasma pneumoniae*, *Mycoplasma hominis*, *Mycoplasma genitalium*, and *Ureaplasma* species.

Culture: They can be cultivated on fluid (PPLO broth) or solid media (PPLO agar) enriched with 20% horse or human serum and yeast extract and addition of antibiotics as selective agents. Colonies appear after incubation for 2-6 days and are about 10-600 µm in size with a typical “fried egg” appearance. Colonies
Mycoplasma and L-Forms

may be seen with a hand lens but are best studied after staining by Dienes method. For this, a block of agar containing the colony is cut and placed on a slide. It is covered with a cover slip on which an alcoholic solution of methylene blue and azure has been dried. Colonies cannot be picked with loops; instead subculture is carried out by cutting a agar block with colonies and rubbing it on fresh plates In the liquid medium no turbidity is appreciated.

Fig. 34.2: Dienes stain

OBJECTIVES

After reading this lesson, you will be able to:

- describe the structure of Mycoplasm & L.form
- explain the pathogenesis of Mycoplasm & L. Form

34.2 PATHOPHYSIOLOGY

*M pneumoniae* causes community-acquired atypical pneumonia, tracheobronchitis or bronchiolitis. Pneumonia develops in only 5-10% of persons who are infected. Acute pharyngitis may also occur.

After inhalation of respiratory aerosols, the organism attaches to host epithelial cells in the respiratory tract. It produces adhesions and other accessory proteins which mediate attachment, followed by induction of ciliostasis, local inflammation and tissue destruction that may be mediated by liberation of hydrogen peroxide. Recently, *M pneumoniae* has been shown to produce an exotoxin: community-acquired respiratory disease toxin (CARDS) that is also believed to play a major role in the damage to the respiratory epithelium. The organism replicates intracellularly, which contribute to chronicity of illness and difficult eradication.

Spread of infection throughout households is common. the incubation period is 2-3 weeks.
Antimicrobials

Oral erythromycin or one of the newer macrolides such as azithromycin or clarithromycin have long been the Drug of Choice for mycoplasmal respiratory tract infections. Tetracycline and its analogues are also active. Fluoroquinolones such as levofloxacin or moxifloxacin exhibit bactericidal antimycoplasmal activity but are generally less potent in vitro than macrolides. As would be predicted by the lack of a cell wall, none of the beta-lactams is effective against \textit{M} \textit{pneumonia}.

In addition to the administration of antimicrobials for the management of \textit{M} \textit{pneumoniae} infections, other measures (eg, cough suppressants, antipyretics, analgesics) should be administered as needed to relieve other systemic symptoms.

INTEXT QUESTIONS 34.1

1. Mycoplasma lack ............
2. Mycolasma commonly causes ............
3. Mycoplasma produces ............... \& ............... which mediate attachment
4. \textit{M}.\textit{pneumoniae} produce ............... toxin

34.3 L-FORM BACTERIA

L-form bacteria, also known as L-phase bacteria, L-phase variants, and cell wall-deficient (CWD) bacteria, are strains of bacteria that lack cell walls.[1] They were first isolated in 1935 by Emmy Klieneberger-Nobel, who named them “L-forms” after the Lister Institute in London where she was working.

Two types of L-forms are distinguished: unstable L-forms, spheroplasts that are capable of dividing, but can revert to the original morphology, and stable L-forms, L-forms that are unable to revert to the original bacteria.

L-forms can be generated in the laboratory from many bacterial species that usually have cell walls, such as Bacillus subtilis or Escherichia coli. This is done by inhibiting peptidoglycan synthesis with antibiotics or treating the cells with lysozyme, an enzyme that digests cell walls.

Some of the species of L-form bacteria that have been implicated in chronic disease include Bacillus anthracis, Treponema pallidum, Mycobacterium tuberculosis, Helicobacter pylori, Rickettsia prowazekii, and Borrelia burgdorferi.
Mycoplasma and L-Forms

Although L-forms can develop from Gram-positive as well as from Gram-negative bacteria, in a Gram stain test, the L-forms always colour Gram-negative, due to the lack of a cell wall.

INTEXT QUESTIONS 34.2

1. L-form are ............... deficient bacteria
2. L-forms that are unable to revert to the original bacteria are ............... 
3. L-forms can be generated in laboratory from bacterial species that have ............... 
4. Due to the lack of cell wall l-forms ............... on staining

WHAT YOU HAVE LEARNT

- *Mycoplasma* species are the smallest free-living organisms
- These organisms lack a cell wall and hence lack Gram stain reaction
- Mycoplasmal organisms are habitant of mucosal surfaces of respiratory and urogenital tracts & cause infections in immunosuppressed patients.
- *M. pneumoniae* causes community-acquired atypical pneumonia, tracheobronchitis or bronchiolitis
- L-form bacteria, also known as L-phase bacteria, L-phase variants, and cell wall-deficient bacteria.
- L-form bacteria are two types namely unstable L-forms, which can revert to the original morphology.
- Stable L-forms, L-forms that are unable to revert to the original bacteria.

ANSWERS TO INTEXT QUESTIONS

34.1

1. Cell wall
2. Atypical pneumonia
3. Adhesions & proteins
4. Community acquired respiratory disease

34.2
1. Cell wall
2. Stable l-forms
3. Cell wall
4. Gram negative