# **Legionella Species (Legionnaires’ Disease)**

**About:**

* *Legionella pneumophila* (and related species) are **Gram-negative rods** that are **thin, pleomorphic**
* **do not Gram stain well** in tissues (they often require silver stain).
* **aerobic**
* **Motile (single polar)**
* **fastidious**, requiring L-cysteine for growth
* Growth only on BCYE and not on blood agar > Legionella + Francisella tularensis.
* **Weak oxidase positive,** c**atalase** positive
* They are found in water systems and cause atypical pneumonia (Legionnaires’ disease) and a milder form (Pontiac fever).
* *L. pneumophila* liquefies gelatin
  + Many labs rely on **urine antigen testing** (for L. pneumophila serogroup 1) or PCR on respiratory samples, rather than culture, for rapid diagnosis.

**Laboratory Isolation:**

* Colonies
  + 3–5 days,
  + small (1-3mm),
  + white or blue-green,
  + ground-glass appearance + internal speckling, described as a “cut-glass” appearance.

**Key Identification Tests:**

* **direct fluorescent antibody (DFA)** test on sputum (specific but not very sensitive).
* **serological latex agglutination**
* **Species differentiation:** requires biochem or serology; *L. pneumophila* can be further serotyped.
* Other species (like *L. micdadei, L. bozemanii*, etc.) exist but are less common.

**If you suspect Legionella in culture:**

* MALDI-TOF also identifies Legionella from culture.

**Clinical significance:**

* Legionella causes severe pneumonia often with multilobar involvement, hyponatremia, and other systemic symptoms.
* acquired from inhalation of aerosolized water (AC cooling towers, showers).
* Because it’s fastidious, diagnosis might rely on urine antigen or PCR. But culture is useful for epidemiology (to compare isolates from environment).
* Treatment is with macrolides or fluoroquinolones (it doesn’t respond to β-lactams due to intracellular location and βlactamases).

# **Campylobacter Species (Curved Microaerophilic Gram-Negative Rods)**

**About:**

* *Campylobacter* are **curved, S-shaped or “gull-wing” Gram-negative rods**.
* **Motility:** Darting motility on wet prep, polar flagella
* **microaerophilic** (require reduced oxygen, ~5% O₂, and often increased CO₂).
* The most common pathogens: **C. jejuni** (major cause of bacterial diarrhea), **C. coli**, and less commonly *C. fetus* (extraintestinal systemic infections).

**Culture Requirements:**

* selective media such as **Campy-BAP or Skirrow’s medium** containing antibiotics to inhibit normal flora. -
* Incubation at **42°C** for intestinal Campylobacters (like *C. jejuni, C. coli*), in **microaerophilic conditions**, whilst *C. fetus* grows better at 37°C and not well at 42°C.

**Key Tests:**

* **Oxidase: Positive**. - **Catalase: Positive** (most Campy).
* **Hippurate Hydrolysis: Positive** for *Campylobacter jejuni subsp. jejuni*; negative for *C. coli* and most others. The **hippurate test** is the classic way to identify *C. jejuni* (blue = positive).
* **Nitrate reduction:** Positive for most.
* **Urease:** Negative (this differentiates from *Helicobacter pylori*, which is similar shape but urease positive)
* **Nalidixic Acid & Cephalothin Susceptibility:** Traditional testing to differentiate species:
  + *C. jejuni* and *C. coli* are **susceptible to nalidixic acid** (30 μg disk) and **resistant to cephalothin**.
  + *Campylobacter fetus* is **resistant to nalidixic acid** and **susceptible to cephalothin**.
* **Hydrogen sulfide:** Campylobacters do not produce H₂S in TSI (though they typically aren’t inoculated to TSI in practice).
* **Skirrow’s tests:** Some labs might do indoxyl acetate hydrolysis (Campy are positive for indoxyl acetate hydrolysis, except C. fetus).

**Identification Algorithm (stool culture scenario):**

* After 48h in microaerophilic 42°C, suspect colonies are tested: **oxidase +, Gram stain shows curved rods**.
* Do **hippurate test**:
  + If positive identify as *C. jejuni*.
  + If negative likely *C. coli* (especially if from stool, since C. coli second most common in diarrhea).
  + Confirm *C. coli* vs others by other biochem or MALDI if needed, but often lab may just report

**Identification Algorithm (blood culture scenario):**

* If the isolate was from blood culture (incubated at 37°C), suspect *C. fetus*.
* Use cephalothin susceptibility (C. fetus is usually cephalothin susceptible) or MALDI.

**Clinical:**

* *C. jejuni* is a leading cause of diarrhea, often from undercooked poultry.
* It causes a bloody, inflammatory diarrhea and cramps, sometimes pseudoappendicitis.
* A notable complication is **GuillainBarré syndrome** (especially with certain serotypes like O:19).
* *C. fetus* causes bacteremia particularly in immunocompromised; it has a protein capsule (S-layer) that helps it evade complement.
* Therapy for severe Campy is with macrolides (azithromycin) as first-line.
* Fluoroquinolone resistance is common in Campylobacter in some areas.

# **Helicobacter Species (Curved Urease-Positive Rods)**

* *Helicobacter pylori* is the main human pathogen, causing peptic ulcers and gastric carcinoma risk.
* **curved or spiral Gram-negative rods**, similar in shape to Campylobacter
* multiple sheathed polar flagella.
* **microaerophilic**
* Catalase and urease positive > CLO test
* enriched media: Skirrow’s.
* Corkscrew motility
* Warthin-Starry silver stain on biopsy

**Other Helicobacters:**

* *H. cinaedi, H. fennelliae* > cause bacteremia and GI issues especially in immunocomp, or cellulitis
* They are urease negative and more related to Campy physiologically.

**Differentiation from Campylobacter:**

* *H. pylori* is **urease positive**, while Campylobacter are urease negative.
* *H. pylori* grows at 37°C, not 42°C (Campy jejuni prefers 42).
* *H. pylori* is typically found in stomach biopsy, not stool isolation.
* If a curved rod is urease positive from gastric biopsy, it’s Helicobacter. If from stool and urease neg, likely Campy. -
* *H. pylori* is also **nalidixic acid resistant and cephalothin sensitive** (like C. fetus).

**Clinical:**

* *H. pylori* infection is diagnosed by non-culture methods mostly (urease test on biopsy, breath test, serology, stool antigen test, or molecular).
* Treatment is with combination of antibiotics (clarithromycin, amoxicillin or metronidazole) and proton pump inhibitor (triple therapy).
* Culture is usually only done if needed for susceptibility testing (e.g., suspected resistance).