# Identifying Gram Negative Bacteria

## Facultative anaerobe, oxidase negative, catalase positive

**Non-lactose fermenter:**

Shigella, Salmonella (except arizonae), Proteus, Providentia, Morganella, Hafnia, Serratia, Yersinia, klebsiella rhinoscleromatis.

**Lactose fermenter:**

Klebsiella, Enterobacter (most), Citrobacter (most), E coli.

Fine GNR:

Haemophilus

## Facultative anaerobe, Oxidase positive, Catalase positive

Vibrio, Aeromonas, Plesiomonas, Aggregatibacter, pasteurella, haemophilus, Capnocytophaga, Campylobacter..

## Facultative anaerobe, oxidase positive, catalase negative

Kingella, Eikenella, Cardiobacterium, Haemophilus.

## Facultative anaerobe, Oxidase negative, Catalase negative

Aggregatibacter aphrophilus, Haemophilus pittmaniae, Capnocytophaga, Gardnerella.

## Aerobic, Oxidase negative, Catalase positive

Acinetobacter, Stenotrophomonas (slow +), Chryseomonas, Bordetella.

## Aerobic, Oxidase positive, Catalase positive

Neisseria, Moraxella, Bordetella, Burkholderia, Brucella, Pseudomonas aeruginosa, Stenotrophomonas (slow +)

# Details

## Facultative anaerobe, oxidase negative, catalase positive – Non lactose fermenter

### Shigella

History of diarrhoea
Cat 3 room (S dysenteriae)
API 20E
Serology (do in Cat 3)
Infection control
Inform public health – S dysenteriae, outbreak etc.

### Salmonella

Diarrhoea.
History of travel – enteric fever, may not have diarrhoea, may have constipation.
Cat 3 room.
XLD – Pink colony with black centre (S typhi may not produce H2S/black centre).
DCA – Transparent colony with black centre (S typhi – no blackcentre).
Selenite broth – do not forget to plate the next day.
Use either XLD or DCA.
May have chromogenic plate – eg SM2.
API 20E.
Agglutination.
Infection control.
Public health – outbreak, imported case, enteric fever…

Agglutination – some common Salmonella
S typhi – O9, D, Vi,
S paratyphi A – O2,
S paratyphi B – O4, b,
S paratyphi C – O6, O7, Vi,
S enteritidis – O9, g,
S typhimurium – O4, i,
S montevideo – O6, O7, g,
S virchow – O6, O7,
S dublin – O9, g,
S newport – O6,

S java – O4, b,
S hadar – O6,
S muenchen – O6, d,
S stanley – O4, d.

**Proteus**

UTI, renal stone, bacteraemia.
Swarming growth, do not swarm on CLED/MacConkey.
API 20E
Spot indole – P mirabilis is indole neg; P vulgaris is not.
Tigecycline R.

**Morganella, Providentia,**

UTI
API 20E

**Hafnia**

Can produce colourless colony on Sorbitol MacConkey agar.
Latex for E coli O157 – negative
API 20E.

**Serratia**

May produce red colony (S marcescens)
API 20E

**Yersinia**

Diarrhoea, lymphadenopathy, appendicitis like picture.
Y pestis is the causative organism of Plague, bioterrorism.
Small GNCB, may have bipolar staining.
CIN agar – bulls eye colony (transparent colony with red centre)
API 20E.

## Facultative anaerobe, oxidase negative, catalase positive – Non lactose fermenter

**Klebsiella**

API 20E

Amox – R

Pip-taz R, coamoxiclav sensitive – K1 hyperproducer.

**E coli**

API 20E

National enhanced mandatory surveillance for Bacteraemia.

**Enterobacter**

API 20E

Co-amox R/don't use

(Some strains may be lactose non-fermenter – but API 20E should be OK)

**Citrobacter**

API 20E

Coamox R/don't use

Some strains may not ferment lactose.

Foul smelling colony.

## Facultative anaerobe, oxidase negative, catalase positive – fine gram negative rod

**Haemophilus**

Grayish flat colony – poor/no growth on blood agar, grows on chocolate agar.

X & V plate.

API NH.

Nitrocefin test for beta-lactamase.

X&V:

X only – ducreyi.

V only – parainfluenzae, parahaemolyticus, paraaphrophilus (allpara).

X&V – influenzae, heamolyticus.

## Facultative anaerobe, oxidase positive, catalase positive

**Vibrio:**

Diarrhoea, travel, seafood, skin lesion, fish tank.
Curved GNR, motile, history
(Don't do oxidase for TCBS).
(Check TCBS ASAP. and document the colour of the colony – it may change on standing).
Don't forget to plate alkaline peptone water.
API 20NE.

Ask for Pteridine 0129 discs (10mcg, 150mcg) – optional.

Vibrio cholerae – agglutination O1 and O139 (ref lab).
Infection control.
Public health.

**TCBS**
**Yellow colony –**
V cholerae, alginolyticus, cincinnatiensis, fluvialis, furnissi. metschnikovii, carchariae.
**Green colony** – V damsela, hollisae (may not grow on
TCBS), vulnificus, parahaemolyticus, mimicus, carchariae.
**Blue-green** – Pseudomonas,
**yellow-green** – Proteus.
**Yellow** – Enterococcus.
**Pteridine**
O10 S/O150 S – V cholerae,
O10 R/O150 R – Aeromonas or V cholerae,
O10 R/O150 S – parahaemolyticus, alginolyticus, fluvialis, furnisii.

**Aeromonas**
Coliform,
Lactose fermentation – varies, could be NLF.
Occasionally beta haemolytic.
API 20NE.

**Plesiomonas**
Travel, seafood, pink colony on XLD, yellow on TCBS.
API 20 NE.

**Aggregatibacter actinomycetemcommitans**

Endocarditis.
GNCB, oxidase could be variable.
Prolonged incubation needed (Ask how long incubated for). Pinpoint colony after 24hr.
Firm adherent star shaped colony, non-motile, indole negative,urease negative.
Don't need X/V.
Do not grow on MacConkey.
***Aggregatibacter segnis album***

**Pasteurella**

Animal bite, pet (may not have a bite history)
Gram negative coccobacilli.
No growth on MacConkey/CLED.
Penicillin sensitive.
API 20NE.

**Haemophilus (all except ducreyi)**

Fine GNR.
Do not grow/poor growth on blood agar.
X&V.
API NH.
Nitrocefin.
Urease (Pittmaniae – negative).

**Capnocytophaga**

Human/animal bite.
Fusiform shaped organism.
Yellowish colony, spreading or gliding edge.
Aminoglycoside R.

**Campylobacter**

Diarrhoea, barbeque, summer months.
Microaerophilic, 42 deg C, seagull shaped bacteria in gram stain –
use carbol fuschine as counterstain or if strongly suspecting, just put G. violet, wash, iodine, wash and look for the shape.
Typical shape and oxidase + is enough to identify it.

## Facultative anaerobic, oxidase positive, catalase negative

**Kingella kingae**

Straight rod, pair/short chain.
May produce spreading colony or pits agar.
Beta haemolytic.
Spot Indole negative.
Penicillin sensitive.
Do not grow on Mac.
Twitching motility.

**Eikenella corrodens**

50% pits agar.
Pale yellow colony, clear centre and spreading around, slight greencolour around the colony,
Indole negative.
Chlorine like smell.

**Cardiobacterium**

Pleomorphic GNR (some part of the rod may stain positive),
May pit agar.
Indole positive.
Penicillin sensitive.
Tear drop cells in rosette/picket fence.

**Haemophilus (ducreyi, parainfluenzae, parahaem, pittmanie)**

Fine GNR,
Do not grow/poor growth on blood agar.
X&V.
API NH.
Nitrocefin.

## Facultative anaerobic, oxidase negative, catalase negative

**A. aphrophilus**
Short rod, Lactose fermenter.
Granular, yellowish colony.
GN coccobacillus.
Urease neg, oxidase sometimes positive.
**H pittmanie**
Non-lactose fermenter.
X&V, API NH.

**Capnocytophaga**
Human/animal bite.
Fusiform shaped organism.
Yellowish colony, spreading or gliding edge.
Aminoglycoside R.
**Gardnerella**
Unlikely from BC as SPS (Sod plyanetholsulphonate in BC bottle inhibits it.
Urease and indole neg.
Beta haemolytic.
Hip+.

## Aerobic, oxidase negative, catalase positive

**Acinetobacter**
Non-motile,
May retain stain and look gram positive.
Haemolytic.
API 20NE.

**Stenotrophomonas**
Motile.
Meropenem R.
Ammonia like smell.
DNase +, Aesculin +.
API 20NE.

**Chryseomonas**
API 20NE.

**Bordetella**
Typical history.
Coccobacilli.
NPS/ NPA for PCR
Antiserum agglutination,
Ref lab.

## Aerobic, oxidase positive, catalase positive

**Pseudomonas aeruginosa**
Typical smell.
Pigmentation – green.
Metallic sheen on the growth.
API 20NE.

**Neisseria**
Cocci, diplococci, Pen S.
API NH.
NYCA medium.
Tributyrin negative.

**Gonococcus –**
Presumptive id= growth on selective media + appropriate colonial morphology + typical GN diplococci (long axis parallel) + Oxidase positive.
Medicolegal – presumptive + 3 tests (biochemical, immunological and molecular – ref lab).
High risk population (GUM) – Presumptive + 1 or 2 tests, Low risk population (GP) – presumptive + 2 tests.

**Moraxella catarrhalis**
Plump GNC.
Smooth, flat buff colony,
Hockey puck sign (colony can be pushed intact on agar surface).
Tributyrin test +.
API NH.

Nitrocefin (90% +)=pen R.
DNAse +

**M lacunata, osloensis, atlantae**

GNCB.
May pit agar, may be haemolytic.
Penicillin sensitive.
API NH.

**Burkholderia cepacia**

If BC – h/o Chr Granulomatous disease.
CF
Cepacia plate – pink colony.
May be slow oxidase +.
API 20NE.

**Brucella**

Very small coccobacilli.
Ref lab

**Bordetella**

Typical history.
Coccobacilli.
NPS/ NPA for PCR
Antiserum agglutination,
Ref lab.