Phenotypic Identification of Yeasts

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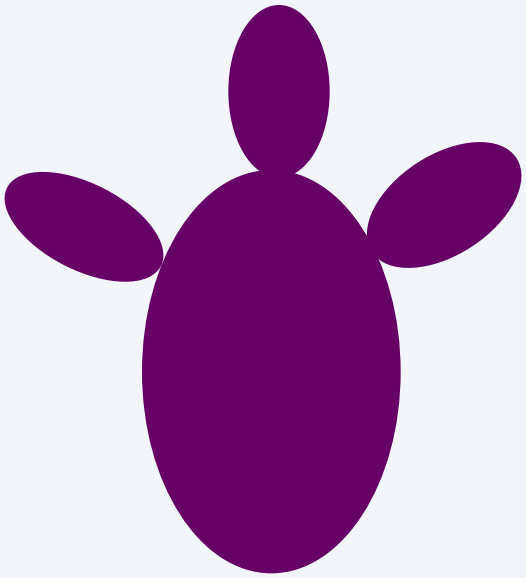
Terminology

- **Yeast:** morphological state of a fungus characterized by unicellular growth.
- (most labs would see ~20 out of 650spp.).
- **Blastoconidium:** an asexual conidium that forms by a blowing out or budding process.
- **Pseudohyphae:** a string of elongated blastoconidia formed by some yeasts that resemble a hypha-like filament.

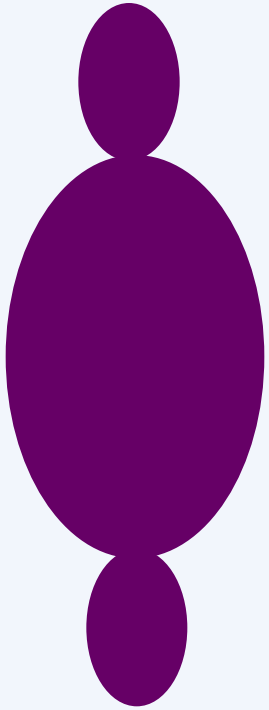
Yeasts - Budding

Narrow based

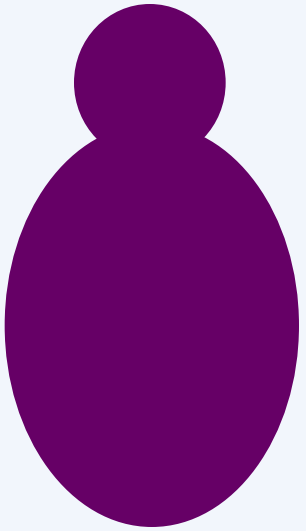
Broad based



multilateral

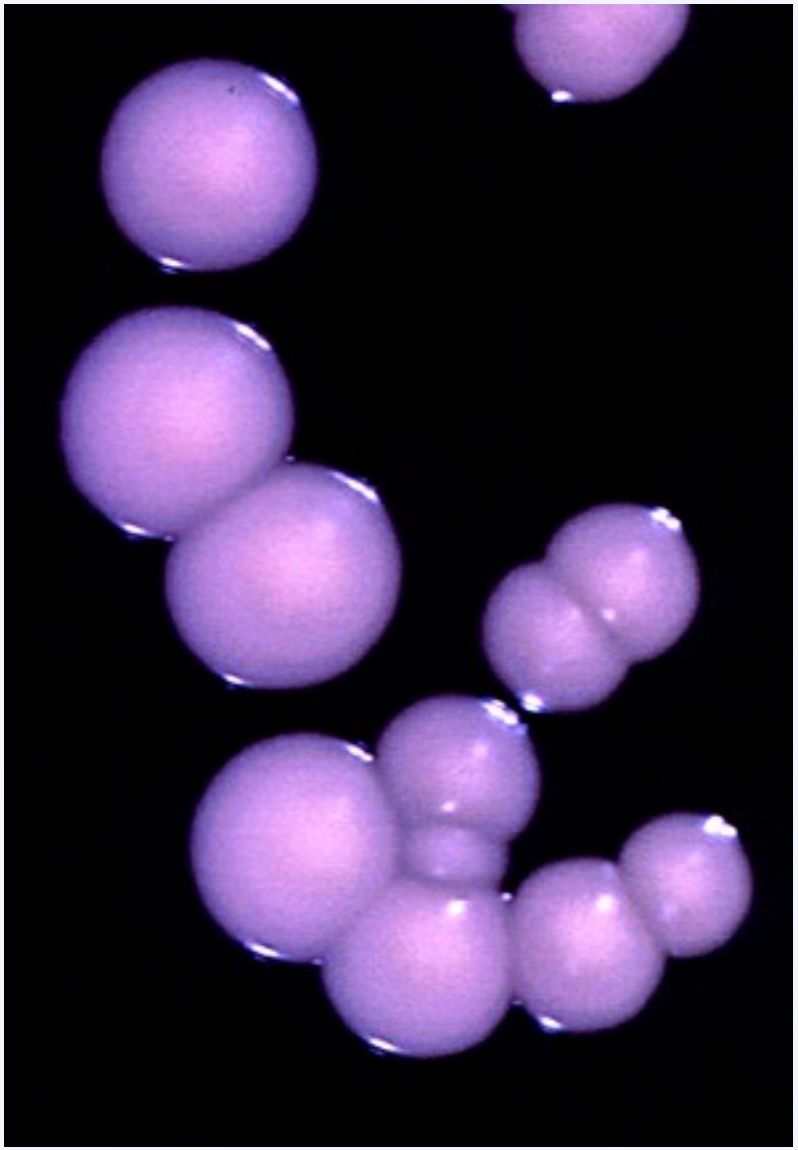


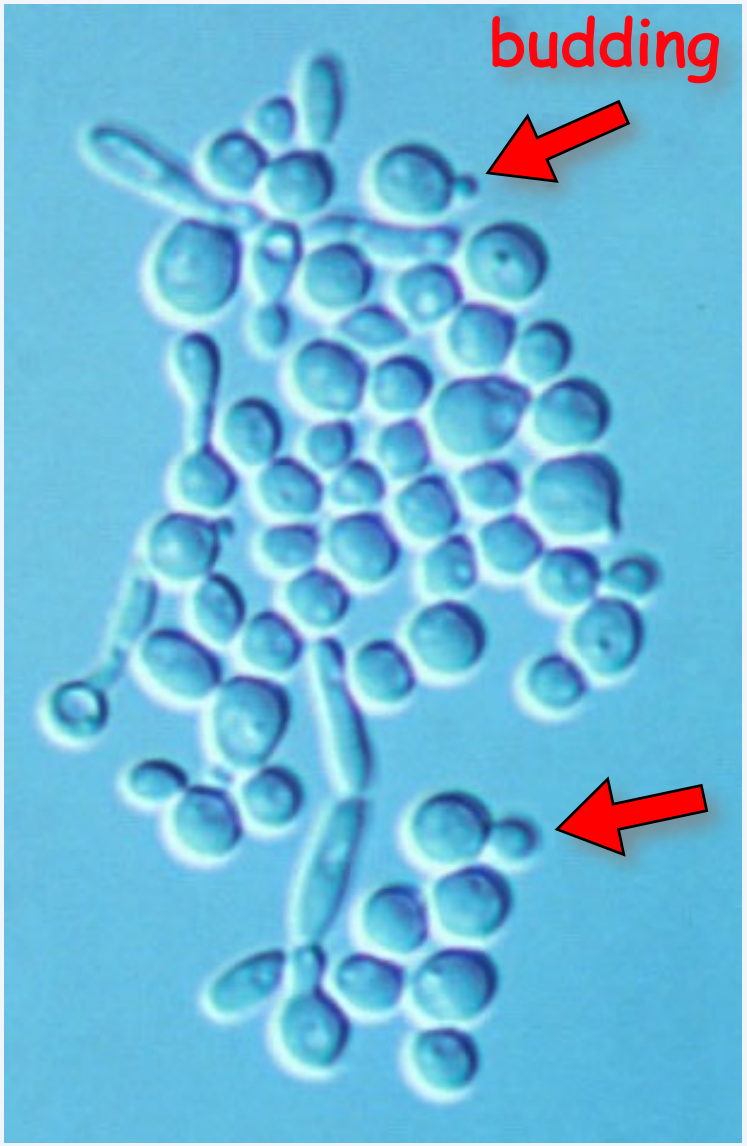
bipolar



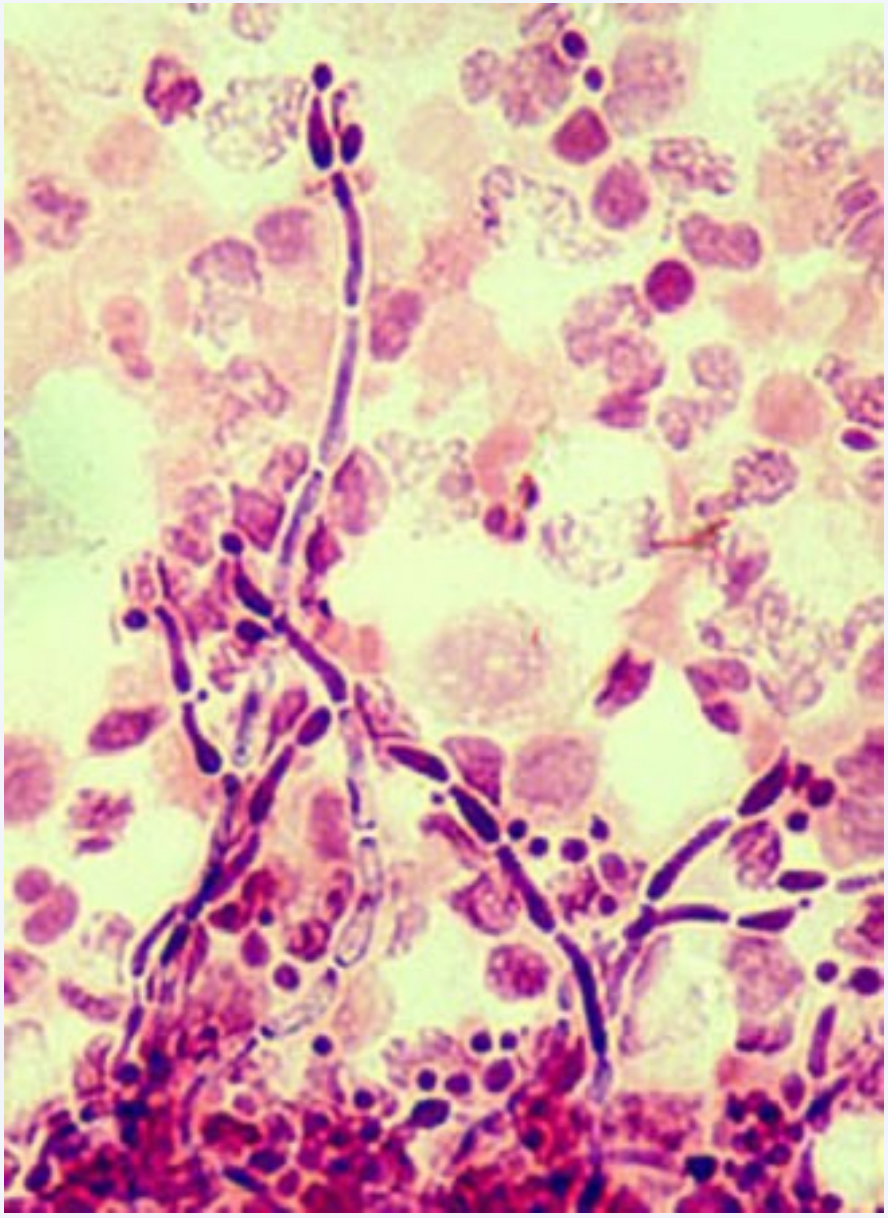
unipolar

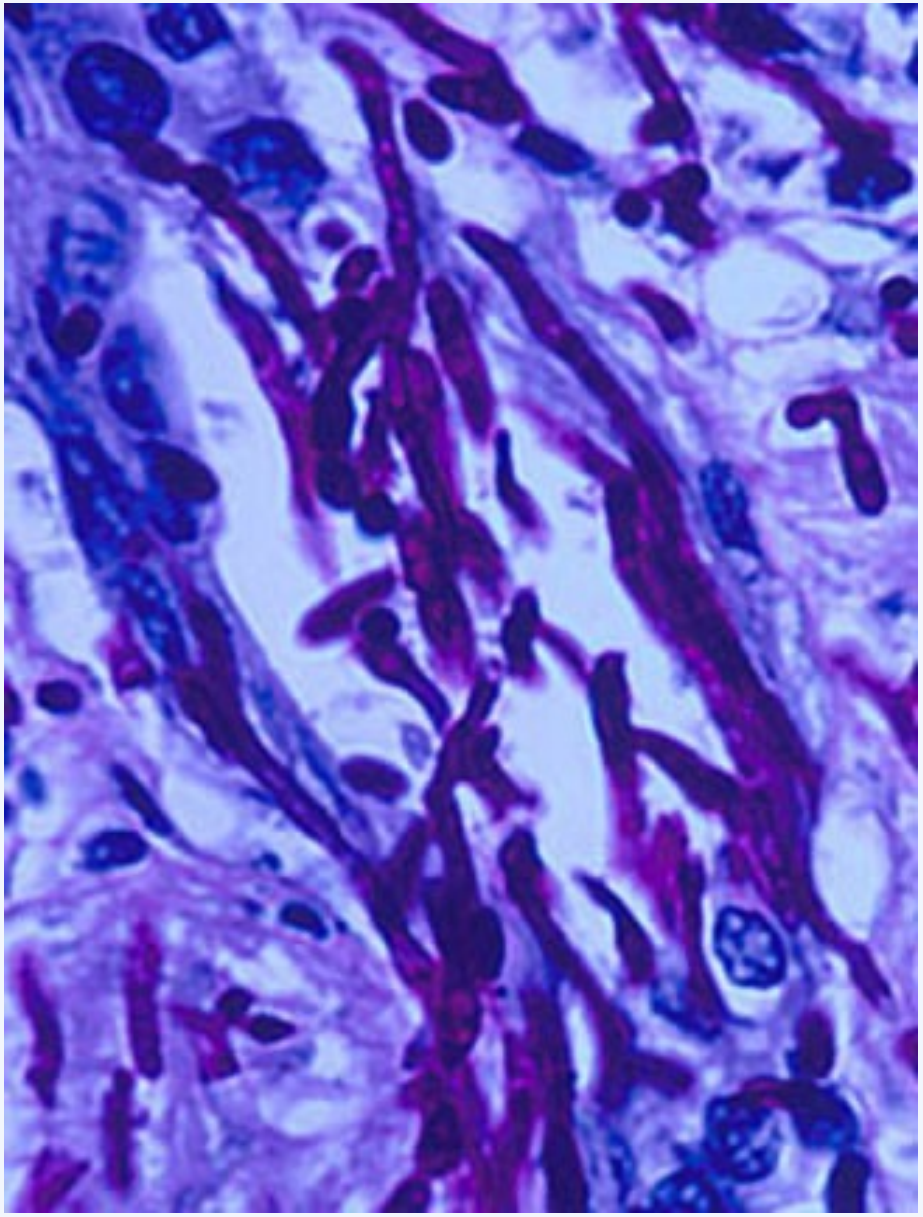
Yeasts - *Candida albicans*



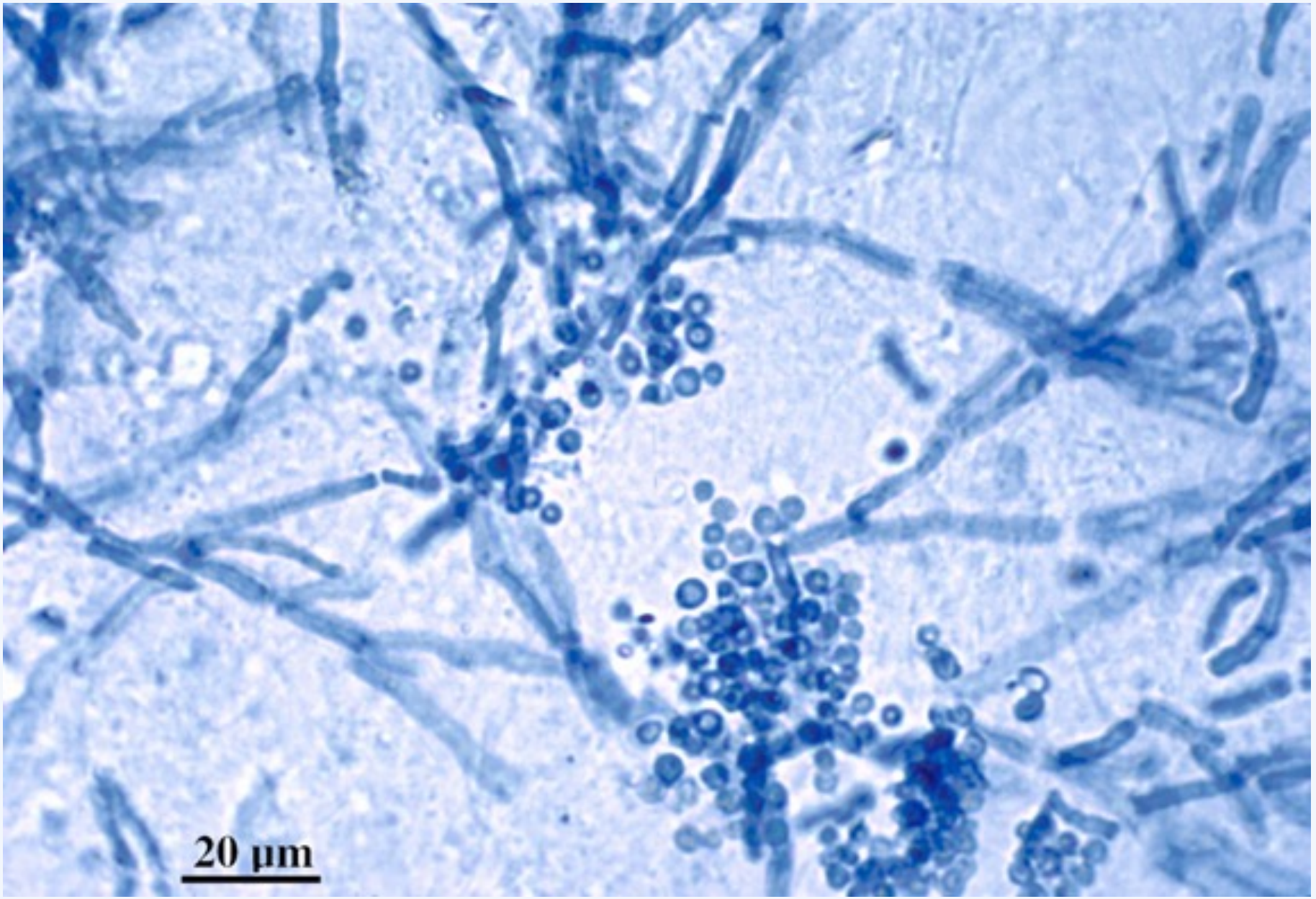


Pseudohyphae of *Candida albicans*



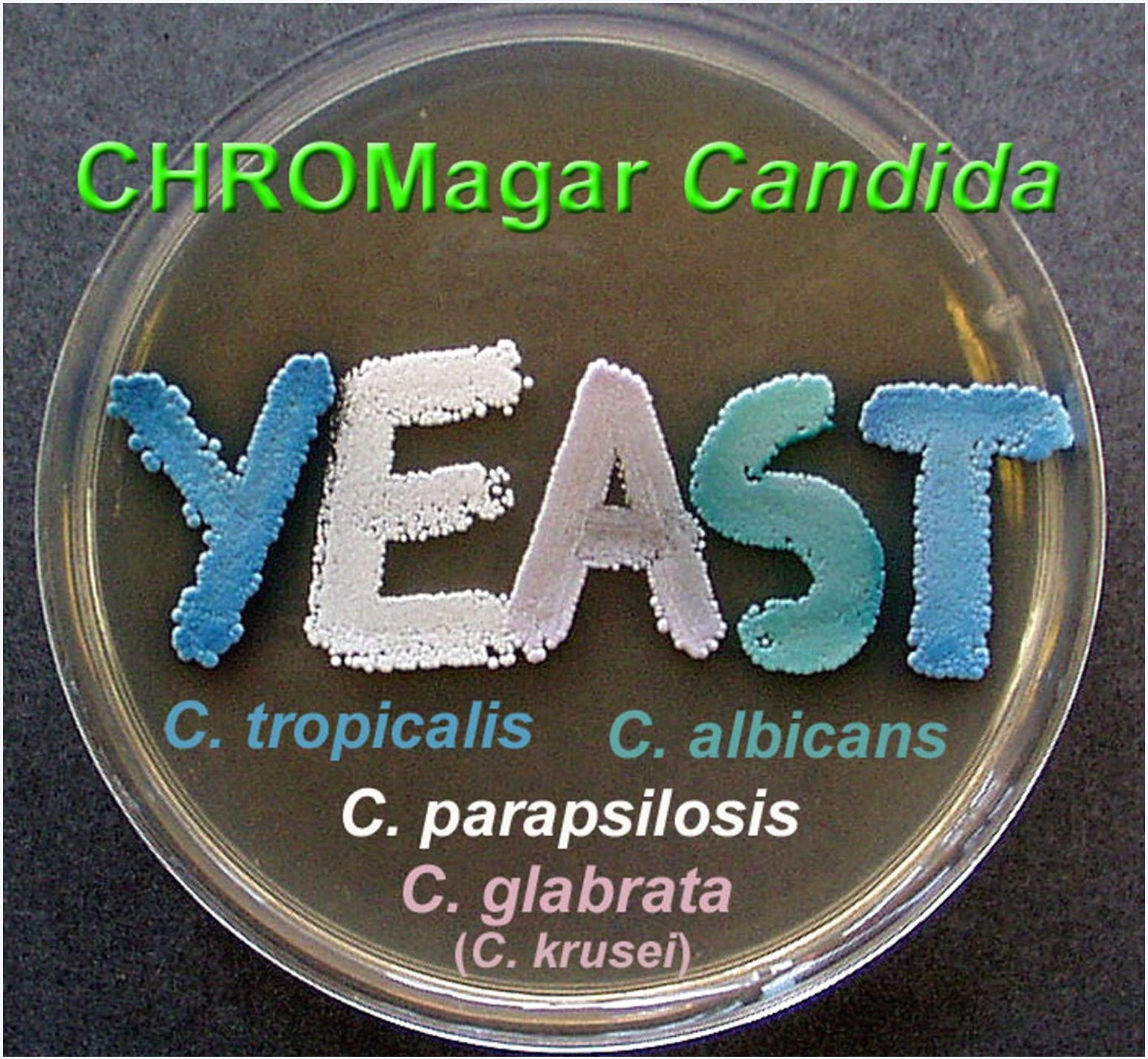


Malassezia furfur



Paracoccidioides brasiliensis


Blastomyces dermatitidis


CHROMagar Candida
YEAST
C. tropicalis
C. albicans
C. parapsilosis
C. glabrata
(C. krusei)

Mixed *Candida* blood isolates

2.8% patients presented with mixed *Candida* infections. 19 patients

73.1% of mixed cases had a *C. glabrata* and/or *C. krusei* isolate.

Species combinations	No	%
albicans/glabrata	8	30.8
parapsilosis/glabrata	1	3.8
parapsilosis/tropicals/glabrata	1	3.8
dubliniensis/glabrata	1	3.8
pelliculosa/glabrata	1	3.8
parapsilosis/krusei	4	15.4
albicans/krusei	1	3.8
tropicalis/krusei	1	3.8
tropicalis/glabrata/krusei	1	3.8
albicans/parapsilosis	5	20
albicans/tropicalis	1	3.8
albicans/dubliniensis	1	3.8
Total	26	100

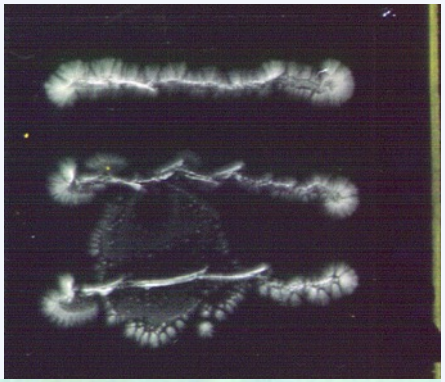
Identification of Yeasts: - the germ tube test.

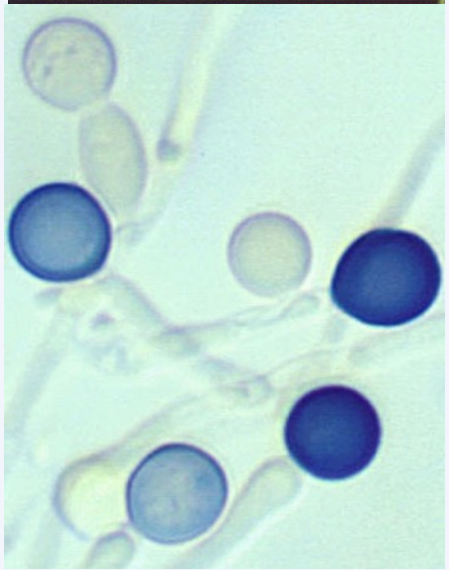
- lightly inoculate 0.5 ml of serum containing 0.5% glucose, incubate at 35^{o}C for 2-3 hrs.
- Positive = *Candida albicans*
  = *C. dubliniensis.*

Cheap and rapid test, 90% accuracy for *C. albicans*, misses *C. dubliniensis* (3%), ? rare *C. tropicalis* strains.

Does not detect *C. glabrata*?

Identification of Yeasts: - Dalmau morphology plate

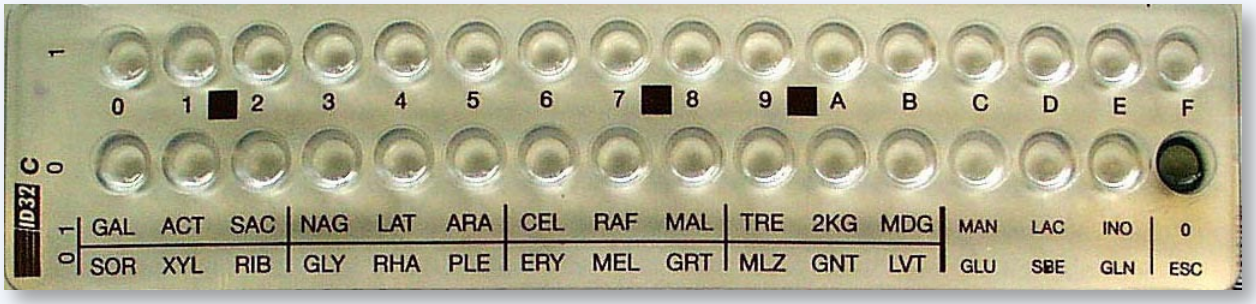




- Stimulate production of pseudohyphae to distinguish between *Candida* and *Torulopsis.* No longer taxonomically valid.
- Production of chlamydoconidia to support identification of *Candida albicans.*
- Regrettably morphology rarely used today!

Identification of Yeasts: - sugar assimilation tests.

- Assimilation, fermentation, hydrolysis, enzyme detection etc. Similar to many bacterial identification systems.
- Allows identification to 90-95% accuracy by either manual or semi-automated systems.
- 48-72 hours (may take 4-5 days for late sugars).
- Many commercial systems available e.g. API 20C, ID 32C, Auxacolor, Vitek, Biolog, BCCM/Allev, Microscan etc.
- Good databases now available.



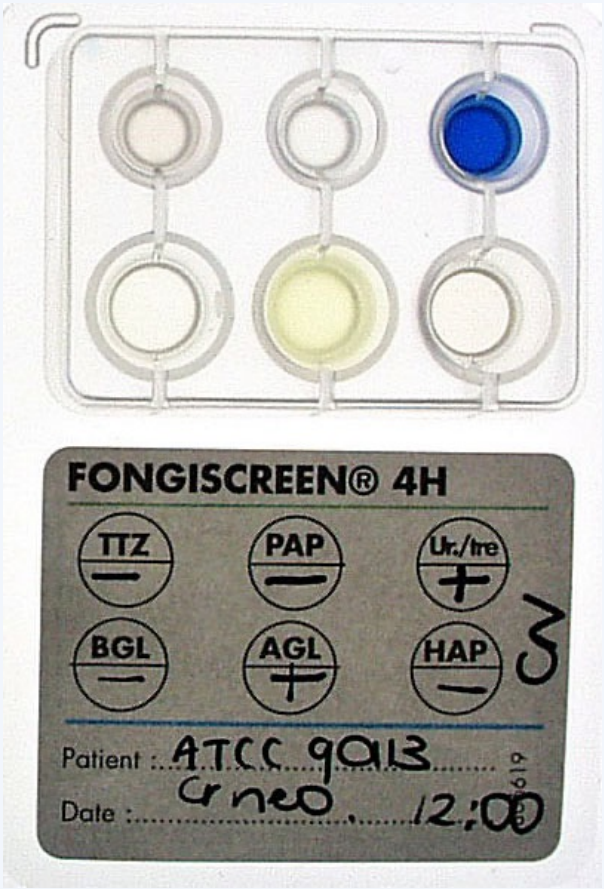
Must have a negative and positive glucose control.

Auxacolor yeast ID strip

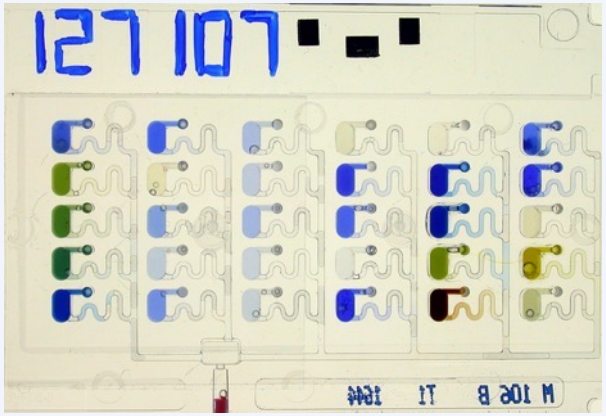
Colorimetric sugar assimilation test system including actidione resistance and phenoloxidase test. 48 hr test using 24-48 hr culture.

Fongiscren ® 4H

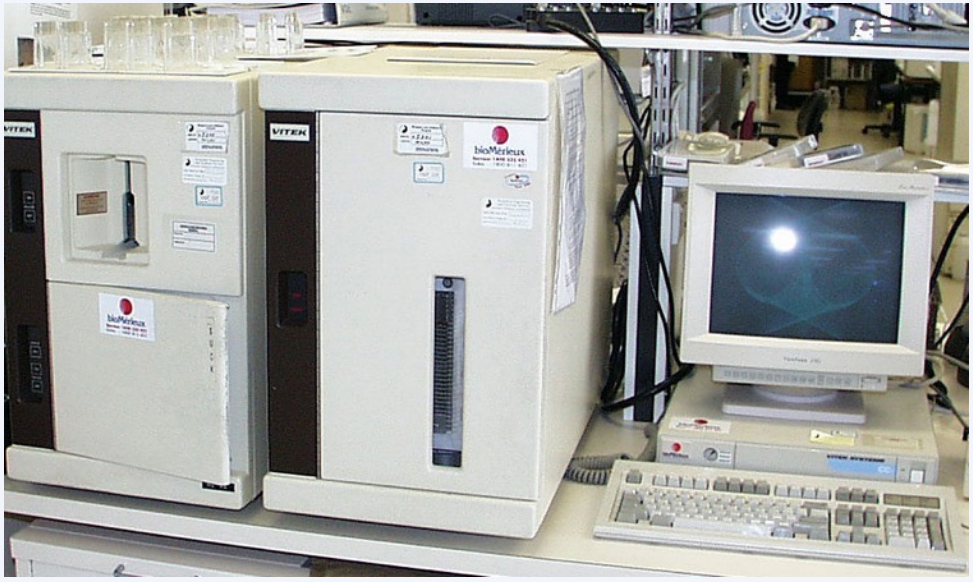
Rapid 4 hr enzyme test for the identification of *C. albicans, C. glabrata C. tropicalis, C. neoformans.* Need 48-72 hr culture.

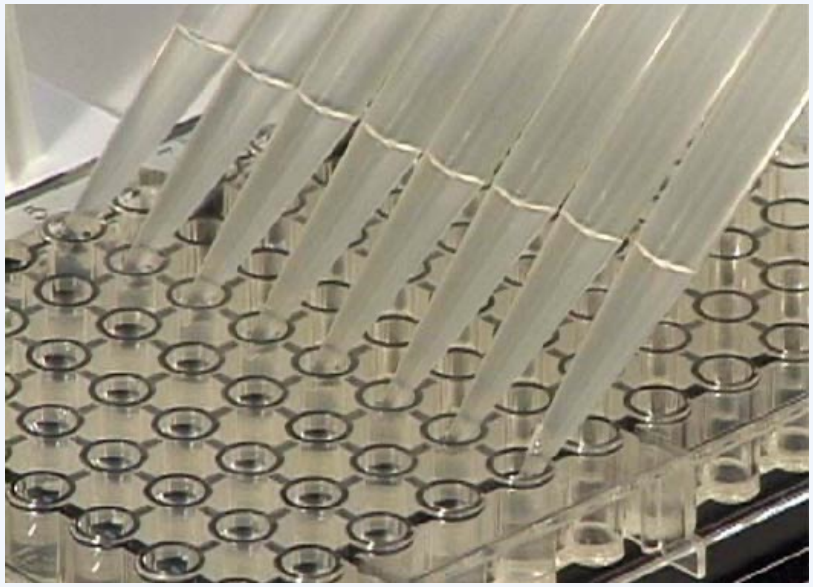


Automated yeast ID systems



Vitek II system
BCMM/ALLEV system
BiOLOG system



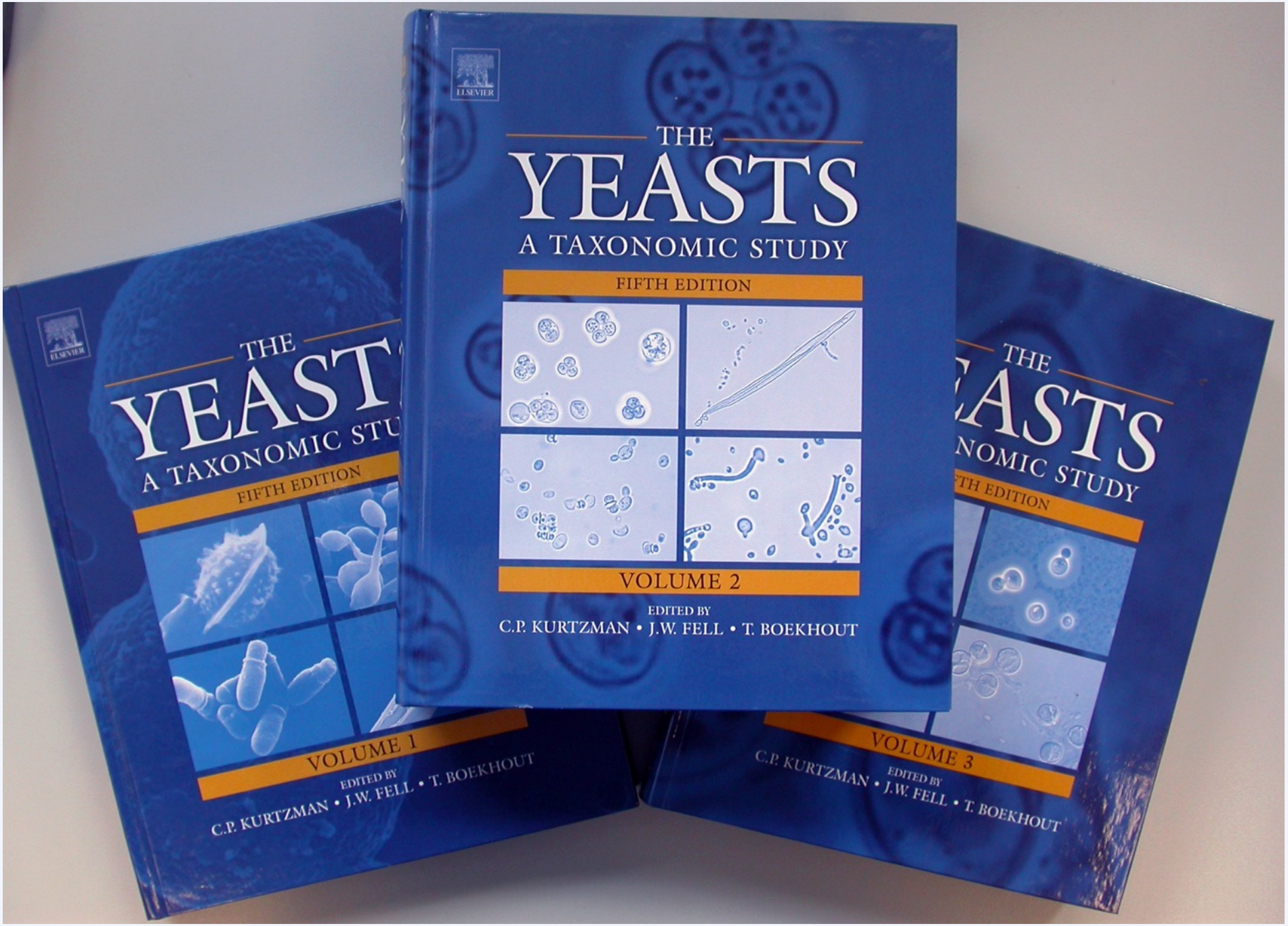


Newer Methods!

- **MALDI-TOF MS:** The Bruker MALDI-TOF database is useful for identification of most clinical yeasts. The MALDI-TOF Vitek MS has been reported to misidentify some yeasts, notably *Candida metapsilosis* as *Candida parapsilosis* (Nobrega *et al*. 2014).
- **Molecular Identification:** ITS sequencing is useful for the identification of most clinical yeasts.

Limitations

- Most rapid and/or commercial yeast identification systems have ~90% accuracy?
- You will always get one isolate that will not give a clear identification?
- Delayed sugar assimilations.
- Streak for purity and repeat test. ?Mixed culture.
- Use full CBS yeast scheme? But best now to go for ITS sequencing (especially if from a sterile site).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How far to go with yeast identification?

- A positive germ tube test is presumptive of *Canada albicans*, but need to be aware of *C. dubliniensis.*
- CHROMagar presumptive for *C. albicans, C. parapsilosis, C. tropicalis* and *C. glabrata/C. krusei*? OK for screening and rapid ID from non-sterile sites.
- All encapsulated yeasts from any site should be identified to species level [*C. neoformans*].
- Full identification of all yeast isolates from a sterile body fluid or tissue, especially immunosuppressed patients or those with chronic or recurrent infection should be considered.