TAXONOMY AND IDENTIFICATION OF *CANDIDA* AND *CRYPTOCOCCUS*: CONVENTIONAL METHOD AND AUTOMATION SYSTEMS

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OUTLINE

- General characteristics
- Yeast identification
- *Candida*
- *Cryptococcus*
- Other yeast
  - *Trichosporon, Rhodotorula*
  - *Malassezia, Prototheca*

Definition of Yeasts

- Eukaryote –cells have defined nucleus and nuclear membrane
- Single cell (vs mould mostly multicellular and forms mycelium) that reproduces by budding
- Reproduce by sexual and asexual process
- Yeast- appearance is generally moist looking with smooth round colonies

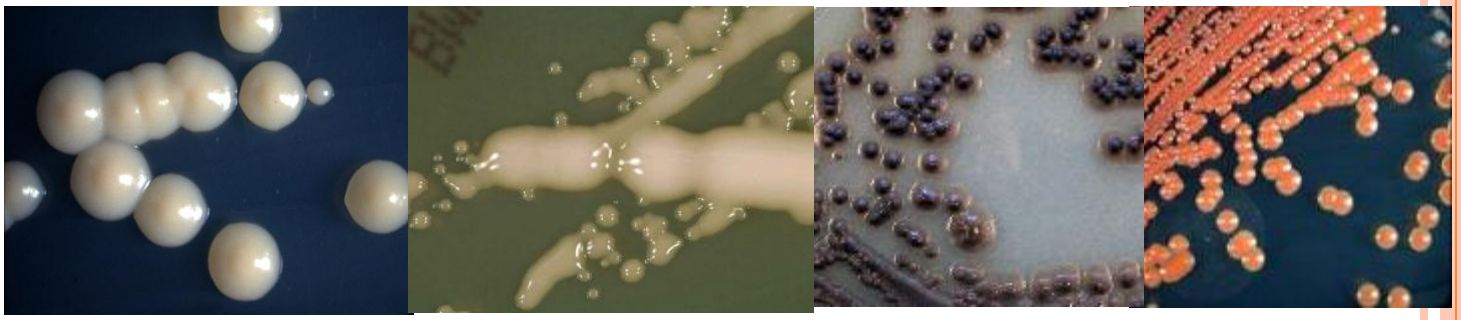
IDETIFICATION OF YEAST

- Conventional method
  - Gross morphology
  - Microscopic morphology (wet mount, Gram stain, India ink)
  - Germ tube
  - Morphology, cornmeal tween 80 agar (Dalmau plate)
  - Chromogenic agar
  - Biochemical tests (Assimilation, API/ATB, Vitek etc, Urease, Phenol oxidase)
- DNA sequencing
- MALDI-TOF mass spectrometry

GROSS MORPHOLOGY

Table 8.5–1 General list of yeast species or genera based on colony color[a]

White, cream, or tan	Brown or black	Salmon, pink, or red
Blastoschizomyces	*Aureobasidium*	*Rhodotorula*
Candida	*Phaeoannellomyces elegans*	*Sporobolomyces*
Cryptococcus	*Hortaea werneckii*[b]	
Geotrichum[d]	*Phaeococcomyces exophialae*[c]	
Hansenula	*Ustilago*	
Kloeckera		
Malassezia		
Prototheca[d]		
Saccharomyces		
Trichosporon		
Ustilago		
Teleomorphs[e]		
Debaryomyces		
Kluyveromyces		
Pichia		



[a] Based on growth on Sabouraud glucose agar (Emmon's modification), potato dextrose agar, malt extract agar, or BHI agar. Note that all *Malassezia* spp. except *Malessezia pachydermatis* require exogenously supplied lipid (e.g., olive oil) for growth.

[b] Previously *Exophiala werneckii.*

[c] A second anamorphic (asexual) form (i.e., synanamorph) of *Exophiala (Wangiella) dermatitidis.*

[d] Not a yeast but produces yeastlike colonies on mycological media.

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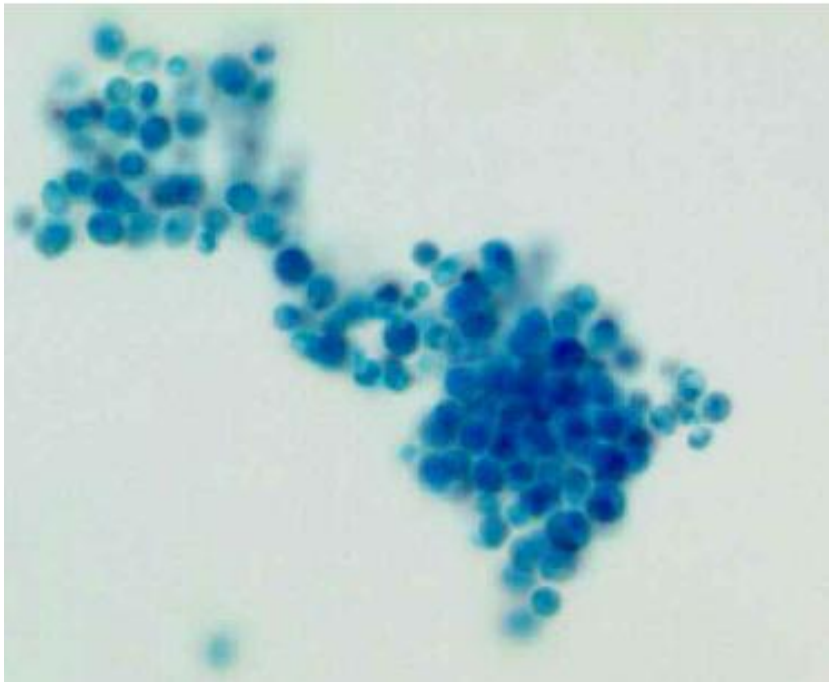
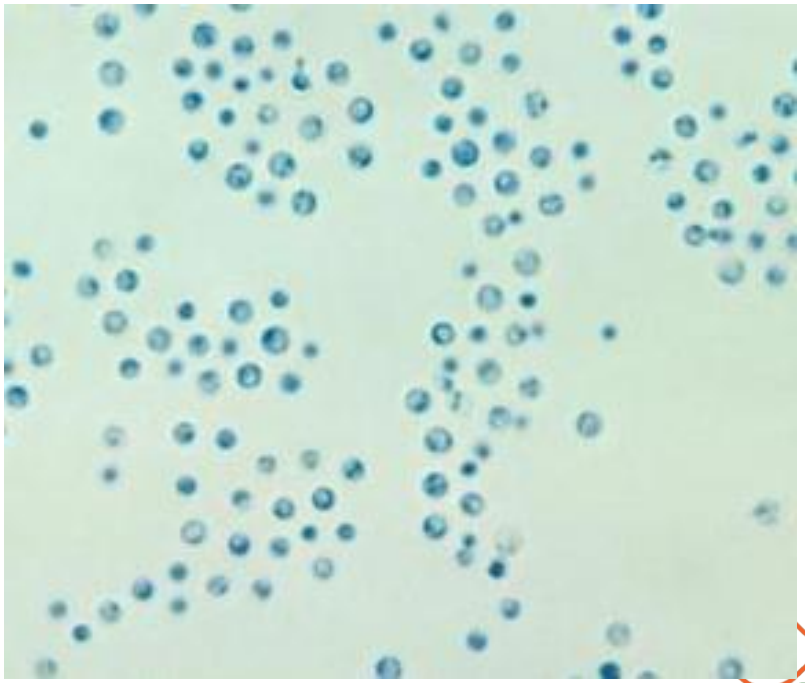
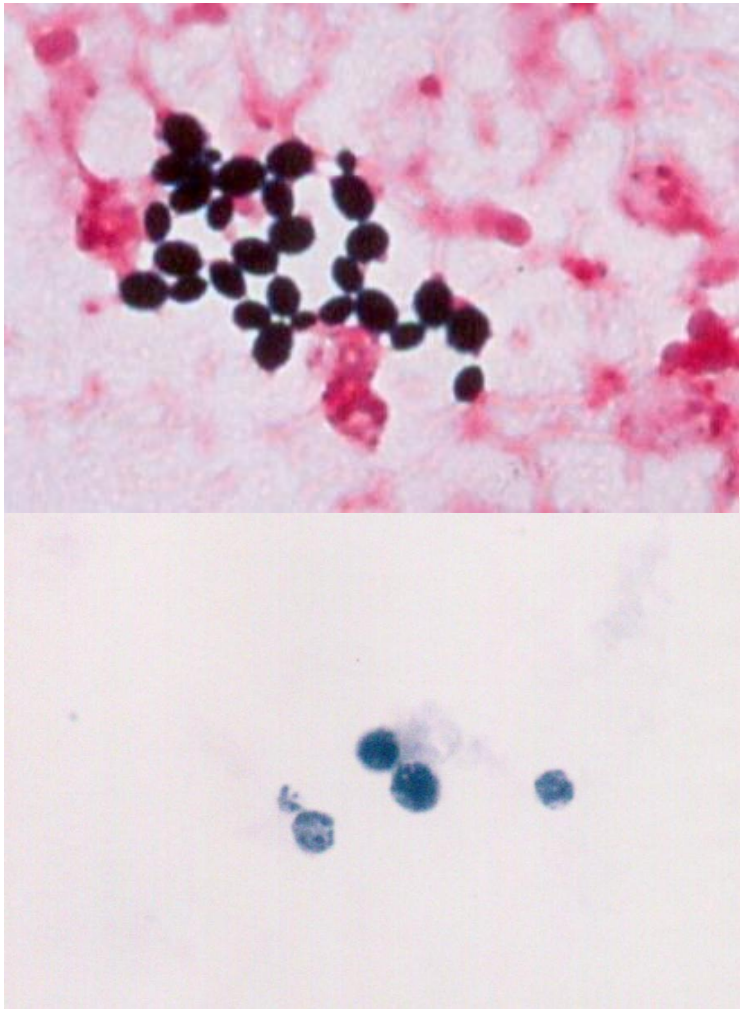
MICROSCOPIC EXAMINATION

- Do microscopic examination -perform a wet mount or Gram stain
  - Methylene blue, lactophenocotton blue

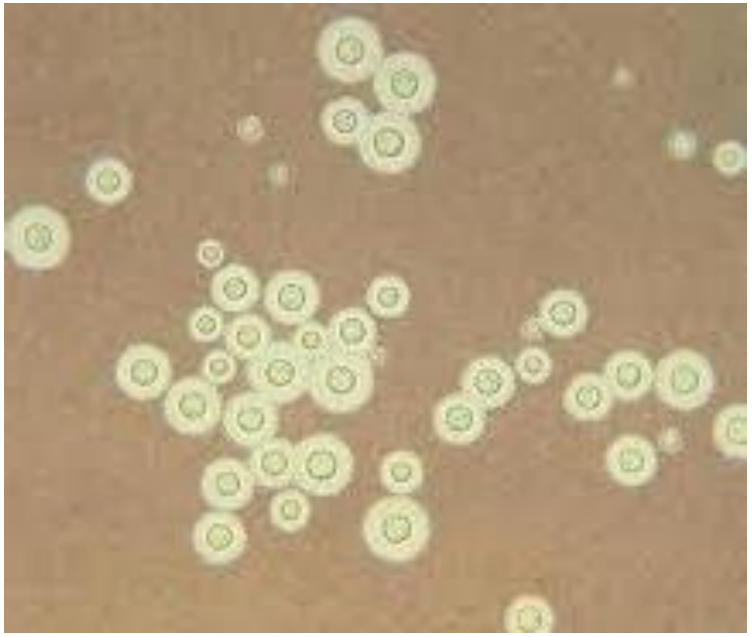




GRAM STAIN

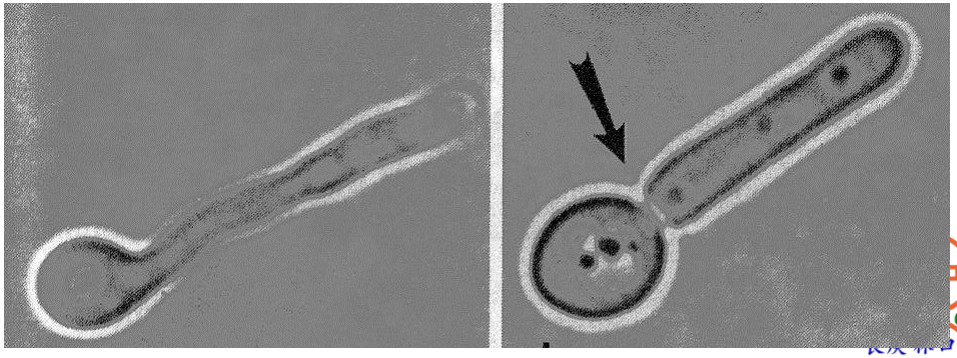


INDIAN INK



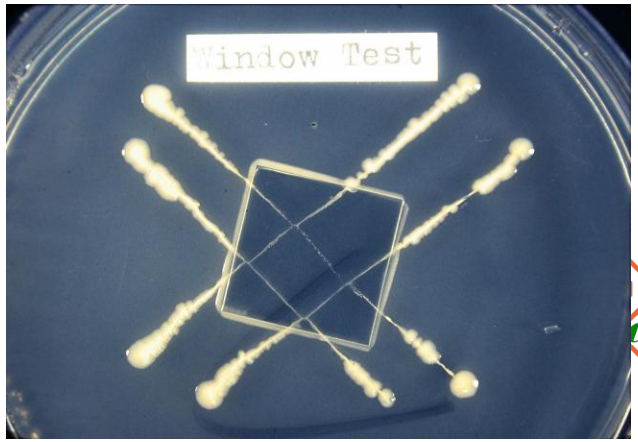
GERM TUBE TEST

- Procedure –inoculate a light inoculum of yeast from a young culture into 0.5 mL serum, incubate at 35°C for 2-3 hours. Look for tube like structures appearing from blastoconidia (no constriction at the point of formation).
- *Candida albicans* and *C. dubliniensis* form germ tubes.



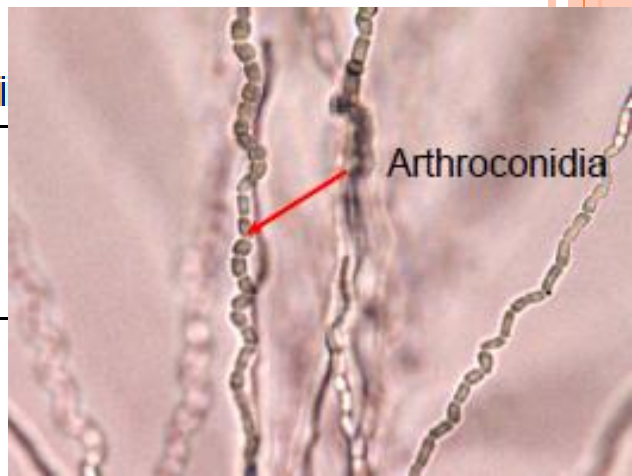
YEAST MORPHOLOGY

- Perform yeast morphology. Use Cornmeal agar with 1% Tween 80 (Dalmau plate).
  - chlamydospore (chlamydoconidia) –tentatively identifies as Candida albicans.
  - Other features seen include blastoconidia, pseudohyphae, hyphae, arthroconida.
  - Incubate at 22-26℃ for 24 hours, or 48 hours if necessary.
- Can narrow down to genus level.



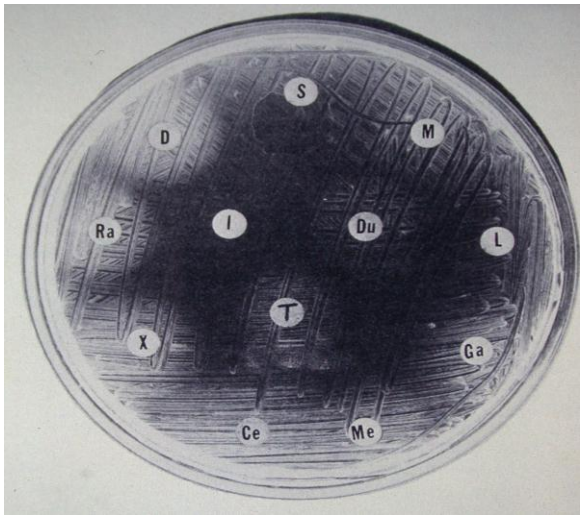
YEAST MORPHOLOGY

Structures observed	Probable Identific
Pseudohyphae (occasional hyphae) Blastoconidia Chlamydospores	Candida albicans Candida dubliniensis
Pseudohyphae (occasional hyphae) Blastoconidia	Candida species
Blastoconidia	Candida species (C. glabrata) Cryptococcus species Rhodotorula species Saccharomyces speci
Hyphae & pseudohyphae Blastoconidia Arthroconidia	Trichosporon species
Hyhpae Arthroconidia	Geotrichum species



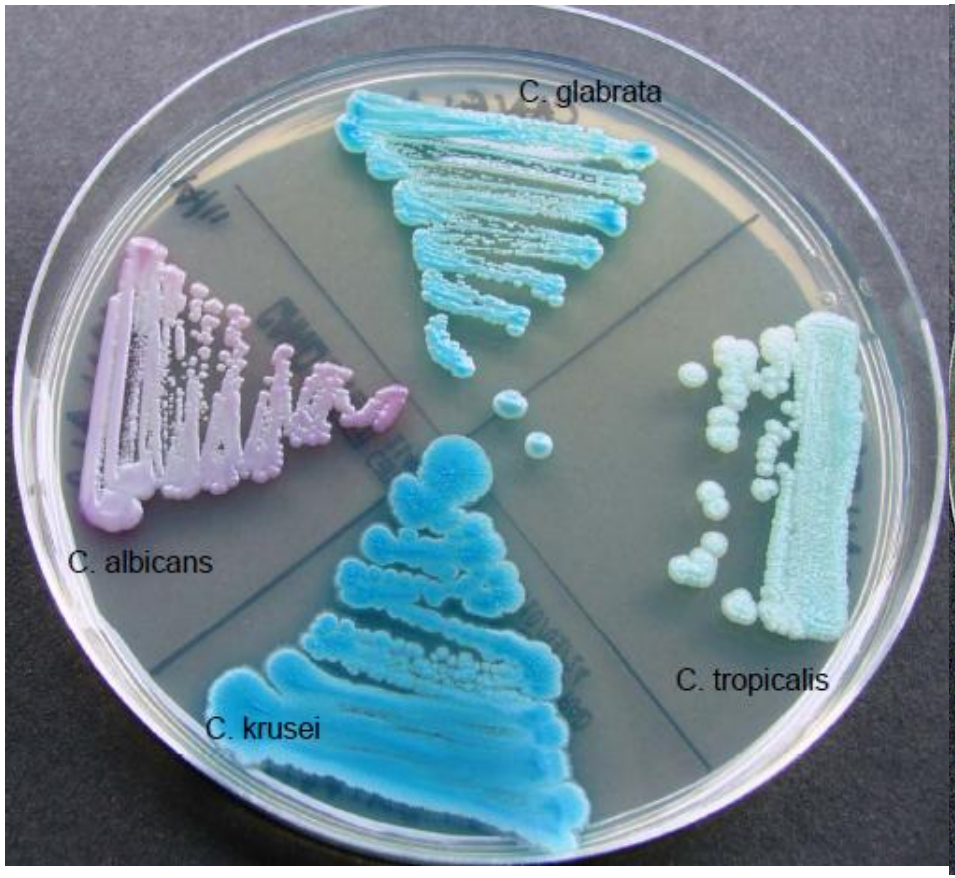
BIOCHEMICAL TEST

- Biochemical tests
  - Urease,
  - Phenol oxidase
    - Caffeic acid agar/Bird seed agar/niger seed agar
  - Rapid trehalose
  - Carbon assimilation
  - API/ATB
  - Vitek 2



CHROMOGENIC AGAR

- If resources or expertise are limited.
- mixed culture with more than 1 type of Candida spp.



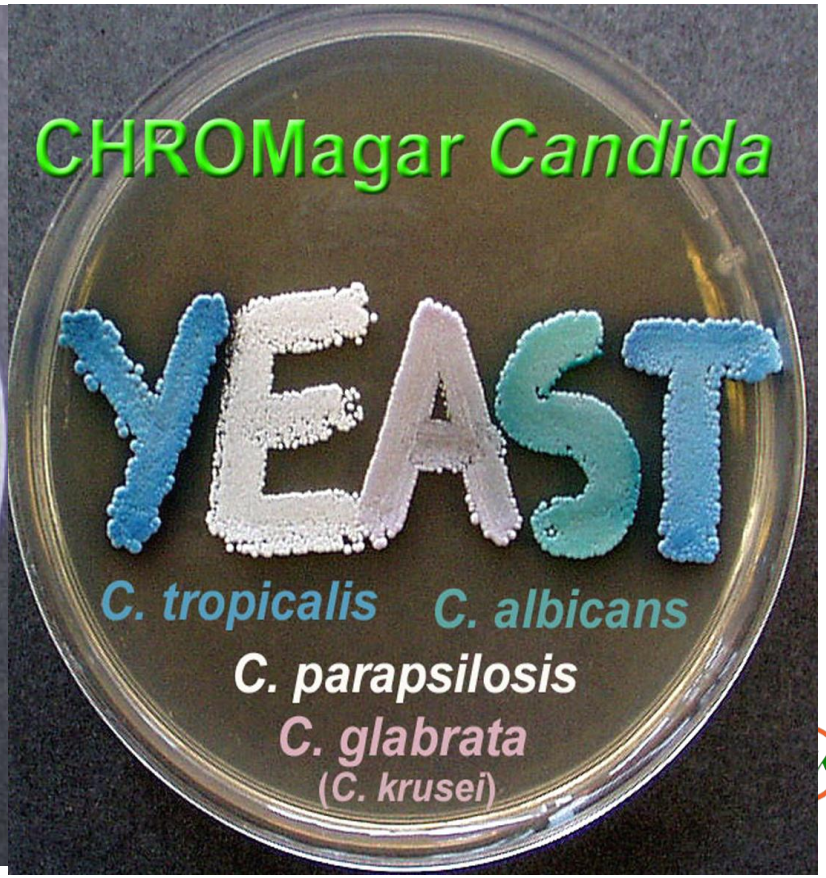


Table 8.6–1 Presumptive identification tests for yeasts on primary culture

Test	Item no.[a]	Organism
CHROMagar	8.5.VI.A	*C. krusei, C. albicans, C. tropicalis, Trichosporon* spp.
Germ tube	8.6.IV.A	*C. albicans* complex[b]
C. albicans screen	8.6.IV.B	*C. albicans, C. dubliniensis*
Rapid urease	8.6.IV.C	*Cryptococcus* spp., *Rhodotorula* spp., *Trichosporon* spp. (variable), *C. krusei* (variable), *Malassezia pachydermatis, Candida lipolytica* (i.e., basidiomycetous yeasts)
Rapid nitrate reductase	8.6.IV.D	*C. albidus* (+), *C. neoformans* (−), *C. terreus* (+)
Caffeic acid disk	8.6.IV.E	*C. neoformans*
Rapid trehalose assimilation	8.6.IV.F	*C. glabrata*
India ink	8.3.V.D	*Cryptococcus* spp.

[a] In this handbook.

[b] This complex consists of *C. albicans, C. dubliniensis, C. stellatoidea* (obsolete), and *Candida africanum* sp. nom. inval.

CARBON ASSIMILATION

Table 8.8–4 Culture and biochemical characteristics of yeasts frequently isolated from clinical specimens[a]

Species	Growth at 37°C	Pellicle in broth	Pseudo- or true hyphae	Chlamydospores	Germ tubes	Capsule, India ink	Assimilation of[b]:												Fermentation of:						Urease	KNO_3	Phenol oxidation	Ascospores
							Glucose	Maltose	Sucrose	Lactose	Galactose	Melibiose	Cellobiose	Inositol	Xylose	Raffinose	Trehalose	Dulcitol	Glucose	Maltose	Sucrose	Lactose	Galactose	Trehalose				
C. albicans	+	−	+	+[c]	+	−	+	+	+•	−	+	−	−	−	+	−	+	−	F	F	−	−	F	F	−	−	−	−
Candida catenulata	+•	−	+	−	−	−	+	+	−	−	+	−	−	−	+	−	−	−	F•	−	−	−	−	−	−	−	−	−
C. dubliniensis	+	−	+	+[c]	+	−	+	+	+	−	+	−	−	−	+•	−	+	−	F	F	−	−	F	F	−	−	−	−
Candida famata	+	−	−	−	−	−	+	+	+	+•	+	+	+	−	+	+	+	+•	W	−	W	−	−	W	−	−	−	−•
C. glabrata	+	−	−	−	−	−	+	+	−	−	−	−	−	−	−	−	+	−	F	−	−	−	−	F	−	−	−	−
Candida guilliermondii	+	−	+	−	−	−	+	+	+	−	+	+	+	−	+	+	+	+	F	−	F	−	F•	F	−	−	−	−•
Candida inconspicua	+	−	−	−	−	−	+	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
C. kefyr	+	−	+	−	−	−	+	−	+	+	+	−	+•	−	+•	+	−•	−	F	−	F	F•	F	−	−	−	−	−
Candida krusei[d]	+	+	+	−	−	−	+	−	−	−	−	−	−	−	−	−	−	−	F	−	−	−	−	−	+•	−	−	−•
Candida lambica	+•	+	+	−	−	−	+	−	−	−	−	−	−	−	+	−	−	−	F	−	−	−	−	−	−	−	−	−•

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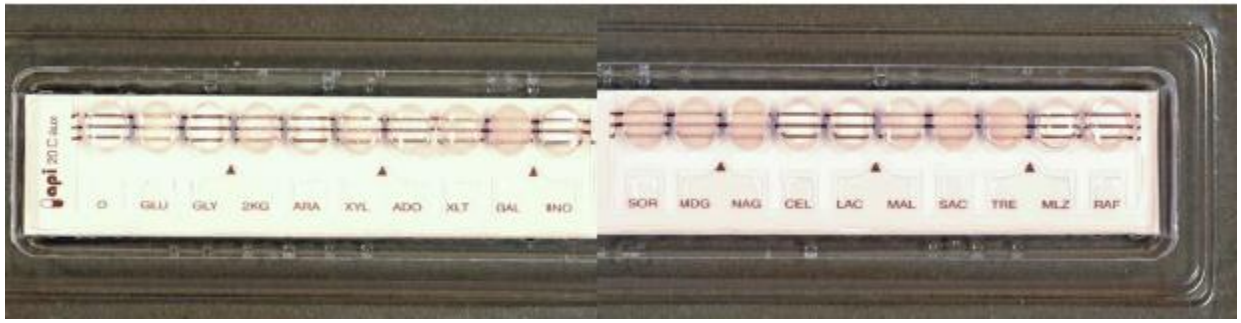
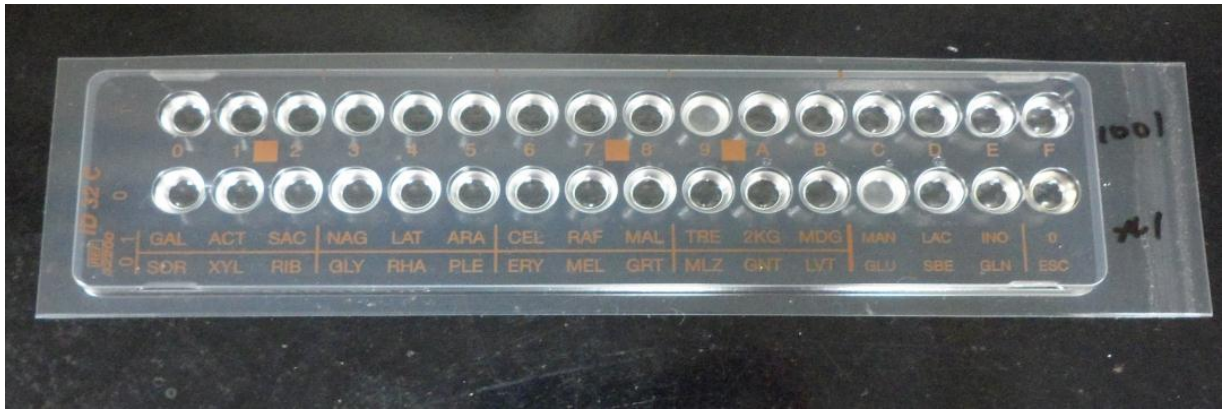
API/ATB-API 20C AUX /ATB 32C





ID 32 C V3.0
Candida albicans 1
Candida albicans 2
Candida boidinii
Candida catenulata
Candida colliculosa
Candida dattila
Candida dubliniensis
Candida famata
Candida glabrata
Candida globosa
Candida guilliermondii
Candida hellenica
Candida holmii
Candida inconspicua/norvegensis
Candida intermedia
Candida kefyr
Candida krusei
Candida lambica
Candida lipolytica
Candida lusitaniae
Candida magnoliae
Candida melibiosica
Candida membranifaciens
Candida norvegica
Candida parapsilosis
Candida pelliculosa
Candida pulcherrima
Candida rugosa
Candida sake
Candida silvicola
Candida sphaerica
Candida tropicalis
Candida utilis
Candida valida
Candida zeylanoides
Cryptococcus albidus
Cryptococcus curvatus
Cryptococcus humicola
Cryptococcus laurentii
Cryptococcus neoformans
Cryptococcus terreus
Cryptococcus uniguttulatus
Debaryomyces etchellsii/carsonii
Debaryomyces polymorphus
Geotrichum capitatum
Geotrichum spp
Kloeckera apis/apiculata
Kloeckera japonica
Kodamaea ohmeri
Pichia farinosa
Rhodotorula glutinis
Rhodotorula minuta
Rhodotorula mucilaginosa
Saccharomyces cerevisiae
Saccharomyces kluyverii
Sporobolomyces salmonicolor
Stephanoascus ciferrii
Trichosporon inkin
Trichosporon asahii
Trichosporon mucoides
Williopsis saturnus
Zygosaccharomyces spp

API 20 C AUX V4.0
Candida albicans 1
Candida albicans 2
Candida boidinii
Candida colliculosa
Candida dubliniensis
Candida famata
Candida glabrata
Candida guilliermondii
Candida kefyr
Candida krusei/inconspicua
Candida lusitaniae
Candida magnoliae
Candida norvegensis
Candida parapsilosis
Candida pelliculosa
Candida rugosa
Candida sphaerica 1
Candida sphaerica 2
Candida tropicalis
Candida utilis
Candida zeylanoides
Cryptococcus albidus
Cryptococcus humicola
Cryptococcus laurentii
Cryptococcus neoformans
Cryptococcus terreus
Cryptococcus uniguttulatus
Geotrichum capitatum
Geotrichum klebahnii
Kloeckera spp
Kodamaea ohmeri
Pichia angusta
Prototheca wickerhamii
Rhodotorula glutinis
Rhodotorula minuta
Rhodotorula mucilaginosa 1
Rhodotorula mucilaginosa 2
Saccharomyces cerevisiae 1
Saccharomyces cerevisiae 2
Sporobolomyces salmonicolor
Stephanoascus ciferrii
Trichosporon asahii
Trichosporon inkin
Trichosporon mucoides

Malassezia ??

CGLM 長庚林口

VITEK

Disadvantage -not good for some yeasts

eg *Trichosporon* misidentified at *Cryptococcus laurentii*

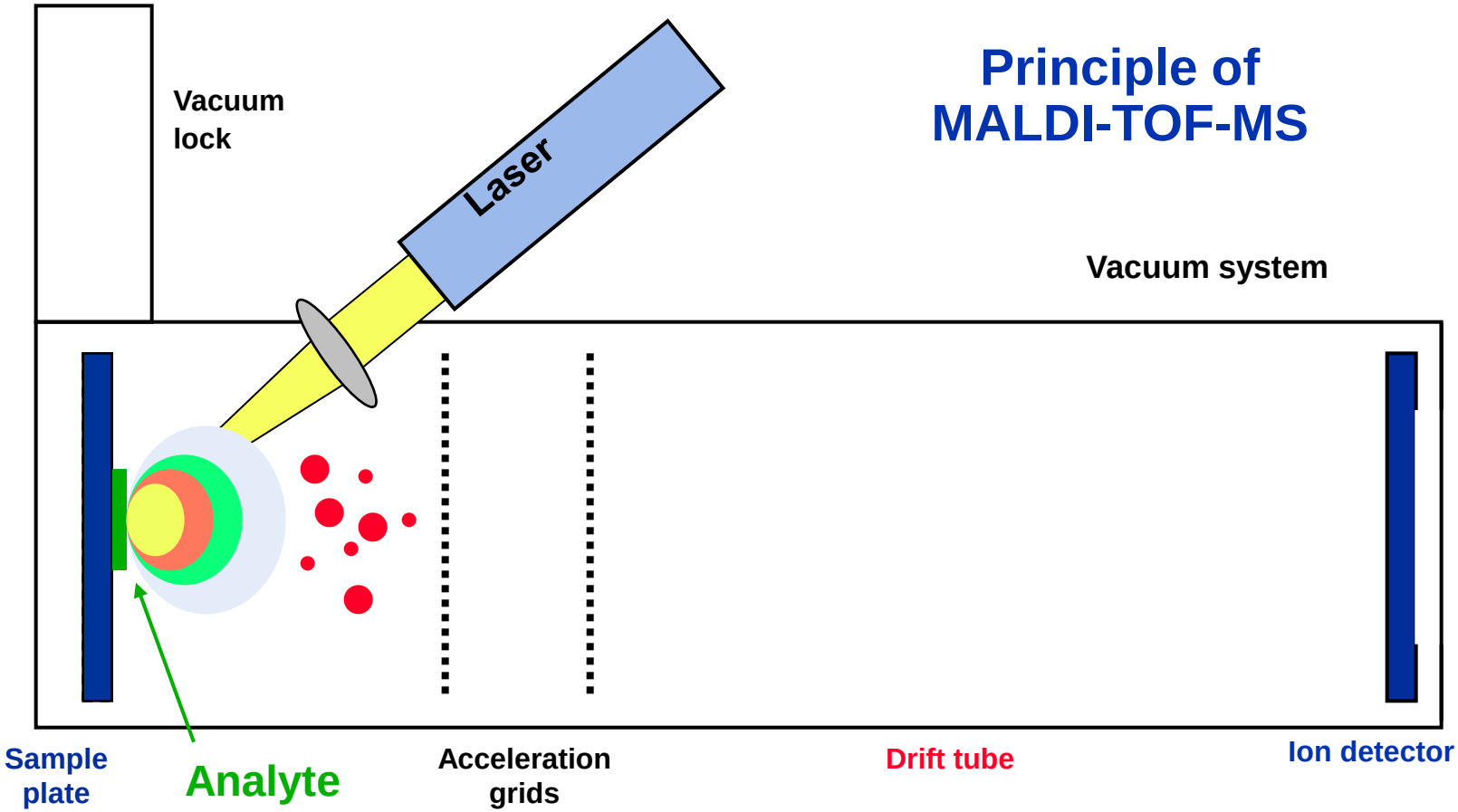
Vitek® 2 YST
ID Card

YEAST	
Candida albicans	*Candida utilis*
Candida boidinii	*Candida zeylanoides*
Candida catenulata	*Cryptococcus albidus*
Candida colliculosa	*Cryptococcus laurentii*
Candida dubliniensis	*Cryptococcus neoformans*
Candida famata	*Cryptococcus terreus*
Candida freyschussii	*Cryptococcus uniguttulatus*
Candida glabrata	*Geotrichum capitatum*
Candida guilliermondii	*Geotrichum klebahnii*
Candida haemulonii	*Kloeckera spp*
Candida inconspicua	*Malassezia furfur*
Candida intermedia	*Malassezia pachydermatis*
Candida kefyr	*Pichia farinosa*
Candida krusei	*Kodamaea ohmeri*
Candida lambica	*Prototheca wickerhamii*
Candida lipolytica	*Prototheca zopfii*
Candida lusitaniae	*Rhodotorula glutinis*
Candida magnoliae	*Rhodotorula minuta*
Candida norvegensis	*Rhodotorula mucilaginosa*
Candida parapsilosis	*Saccharomyces cerevisiae*
Candida pelliculosa	*Sporobolomyces salmonicolor*
Candida pulcherrima	*Stephanoascus ciferrii*
Candida rugosa	*Trichosporon asahii*
Candida sake	*Trichosporon inkin*
Candida sphaerica	*Trichosporon mucoides*
Candida tropicalis	*Zygosaccharomyces bailii*

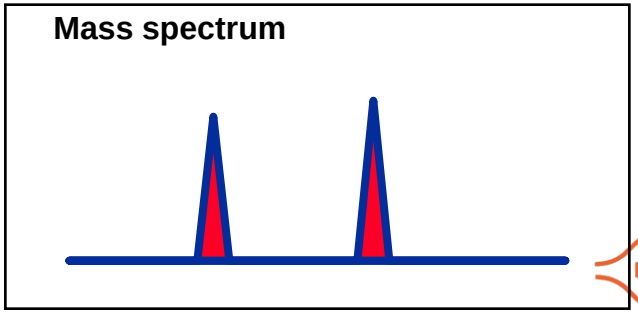
MALDI-TOF MS

- Matrix-assisted laser desorption/ ionization time of flight mass spectrometry
- Used to analyze samples of many types, including solutions of organic molecules, nucleic acids, proteins, and whole microorganisms

Principle of MALDI-TOF-MS



$$\frac{m}{z} = \frac{2t^2K}{L^2}$$

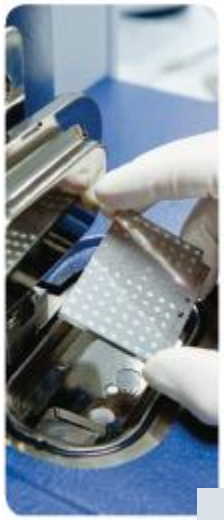


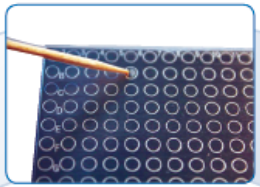
MALDI-TOF MS

- MALDI-Biotyper system- Bruker Daltonics (Leipzig, Germany) (who have recently partnered with Becton-Dickinson, Franklin Lakes, NJ)
- Vitex-MS - Shimadzu Corp. (Kyoto, Japan) and bioMerieux SA (Marcyl'Etoile, France)

MALDI-BIOTYPER SYSTEM

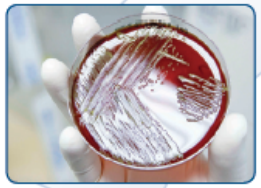
microorganisms.





Smear & Add Matrix

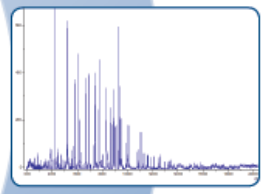
Create Project at The Bench



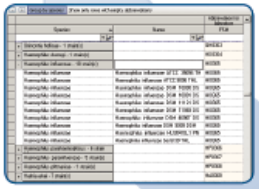
Select a Colony

- Microdilution Technique for Antimicrobial Susceptibility Testing
- Kirby-Bauer Antibiotic Testing
- Automation Systems
- Laboratory Information System

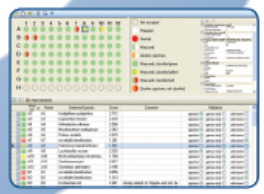
Open Microbiology Concept



Generate MALDI-TOF Spectrum



Smart Translation Table for LIMS



Match Profile with Library

Final Review by Microbiologist

CE-IVD Workflow

MALDI-TOF database

- *Candida* spp. - >60
- *Cryptococcus* spp. – 20
- *Geotricum* spp. -3
- *Saccharomyces*
- *Malassezia spp*
- *Pichia spp. – 10*
- *Prototheca*
- *Rhodotorula spp – 6*
- *Trichosporon spp -20*
- *Alternaria alternata*
- Aspergillus spp. – 8
- Epidermophyton
- Exophiala
- Fusarium -2
- Microsporum spp. –2
- Mucor
- Paecilomyces - 2
- Penicillium spp. – 3
- Phoma - 2
- Rhizopus
- Sporothrix
- Trichophyton spp - 4

CGLM
長庚林口

YEAST + formic acid

Meaning of Score Values

Range	Description	Yeast
		81
2.300 ... 3.000	highly probable species identification	77.8%
2.000 ... 2.299	secure genus identification, probable species identification	
1.700 ... 1.999	probable genus identification	16.0%
0.000 ... 1.699	not reliable identification	6.2%

Table 2 Routine identification performance of matrix-assisted laser desorption/ionization time-of-flight mass spectrometry for yeasts and nontuberculous mycobacteria (NTM).

Microorganism	Total no.	No. (%) of isolates identified to		No. (%) of unidentified isolates		
		Species level	Genus level	Total	No peaks found	No reliable identification
Yeast	81	63 (77.8)	76 (93.8)	5 (6.2)	4 (4.9)	1 (1.2)
Candida glabrata	19	19 (100.0)	19 (100.0)	0 (0.0)	0 (0.0)	0 (0.0)
Candida tropicalis	16	12 (75.0)	15 (93.7)	1 (6.3)	1 (6.3)	0 (0.0)
Candida albicans	15	11 (73.3)	15 (100.0)	0 (0.0)	0 (0.0)	0 (0.0)
Candida parapsilosis	14	9 (64.3)	12 (85.7)	2 (14.3)	2 (14.3)	0 (0.0)
Others	17	12 (70.6)	15 (88.2)	2 (11.8)	1 (5.9)	1 (5.9)

J Microb Immun Infec (2016) in press

APPLICATION OF MALDI-TOF MS TO THE IDENTIFICATION OF YEASTS IN THE CLINICAL LABORATORY

TABLE 1 Methods for analysis of isolated yeast colonies

Method	Study enrollment	Positive ID (%)[a]	Correct ID (%)[a]	Reference
Tube-based extraction (Cultured for 48 h at 30°C on SAB. One to 5 colonies were extracted using 75% EtOH, followed by suspension in a 50/50 mixture of 70% formic acid and ACN.)[b]	267 yeast isolates (250 *Candida* spp.)	247/267 (92.5)	247/247 (100)	Marklein et al. (32)
	1,192 yeast isolates (1,007 *Candida* spp.)	1,171/1,192 (98.2)	1,163/1,171 (99.3)	Bader et al. (31)
	241 yeast isolates (193 *Candida* spp.)	225/241 (93.4)	225/225 (100)	Dhiman et al. (29)
On-plate extraction (Cultured for 48 h at 30°C on SAB. One colony was transferred directly to MALDI-TOF MS analysis plate and overlaid with 70% or 25% formic acid.)	167 yeast isolates	135/167 (80.8)	135/135 (100)	Van Herendael et al. (34)
	90 yeast isolates (71 *Candida* spp.)	86/90 (95.6)	89/90 (98.9)	Theel et al. (33)
	192 yeast isolates (182 *Candida* spp.)	185/192 (96.3)	184/185 (99.5)	Iriart et al. (35)

[a] ID, identification. Number of isolates identified to the genus or species level (numerator) according to defined score interpretation guidelines out of the total number of isolates tested (denominator).
[b] EtOH, ethanol; ACN, acetonitrile.

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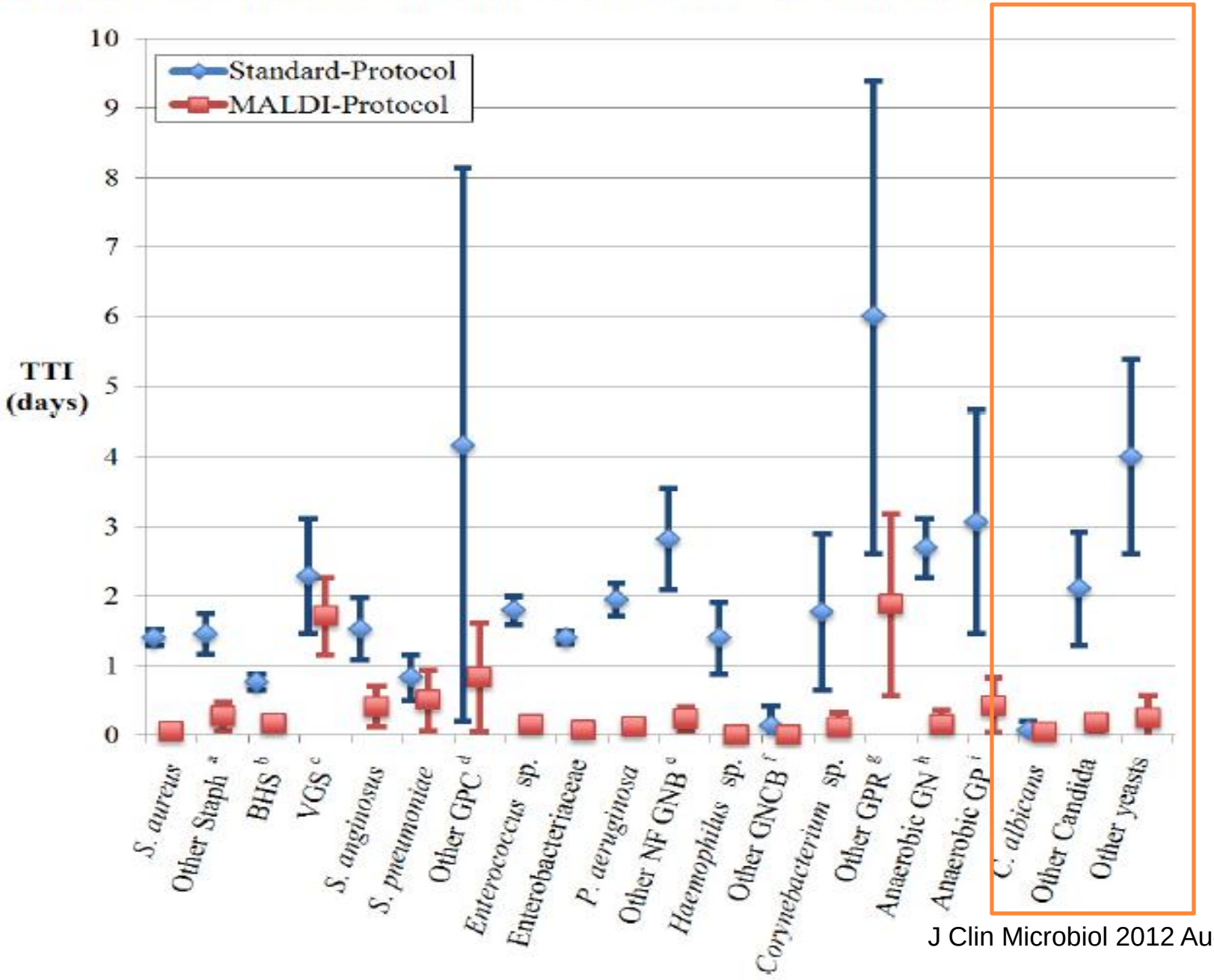
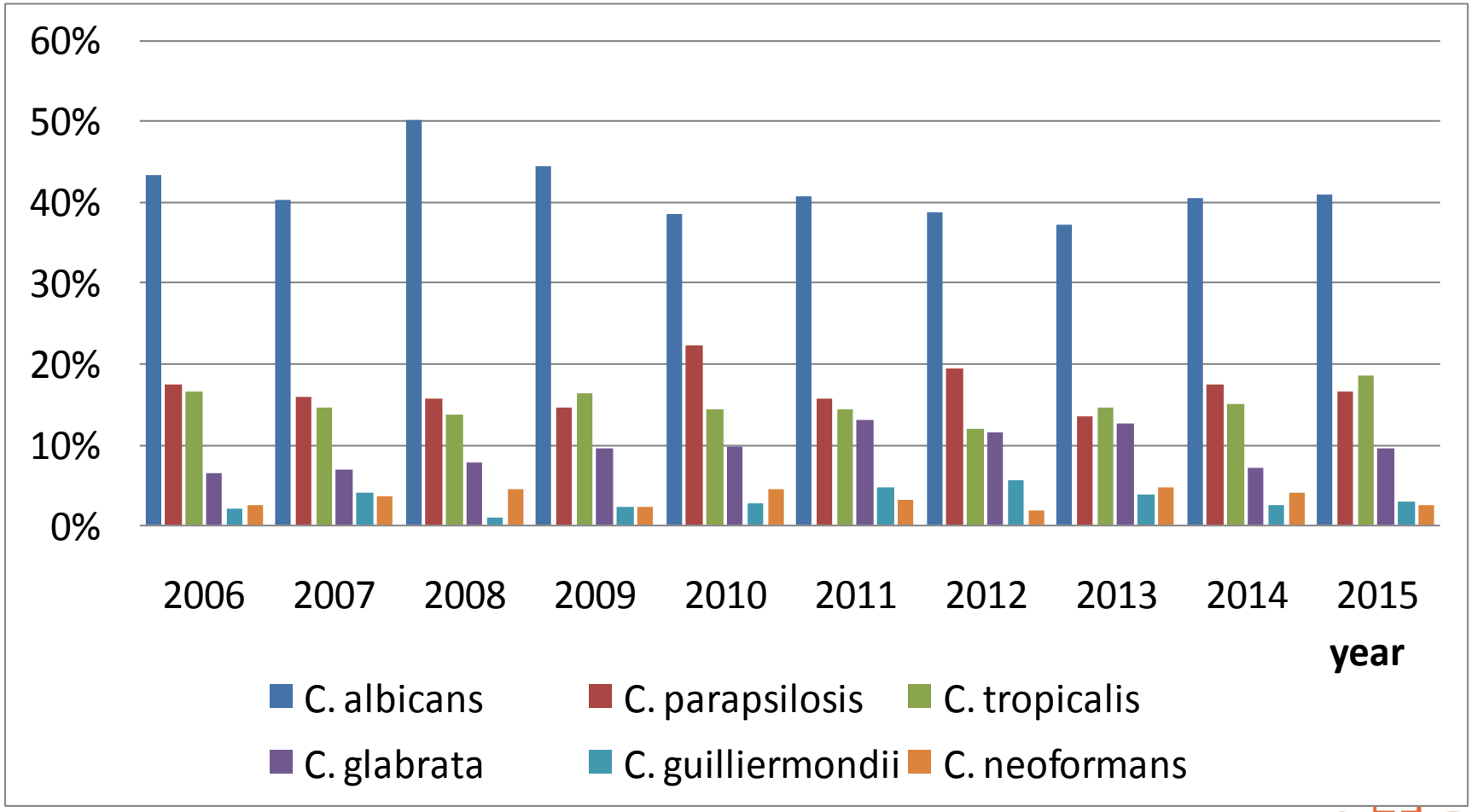
Figure 1: Time to identification (TTI) and 95% CI by MALDI- and standard-protocol

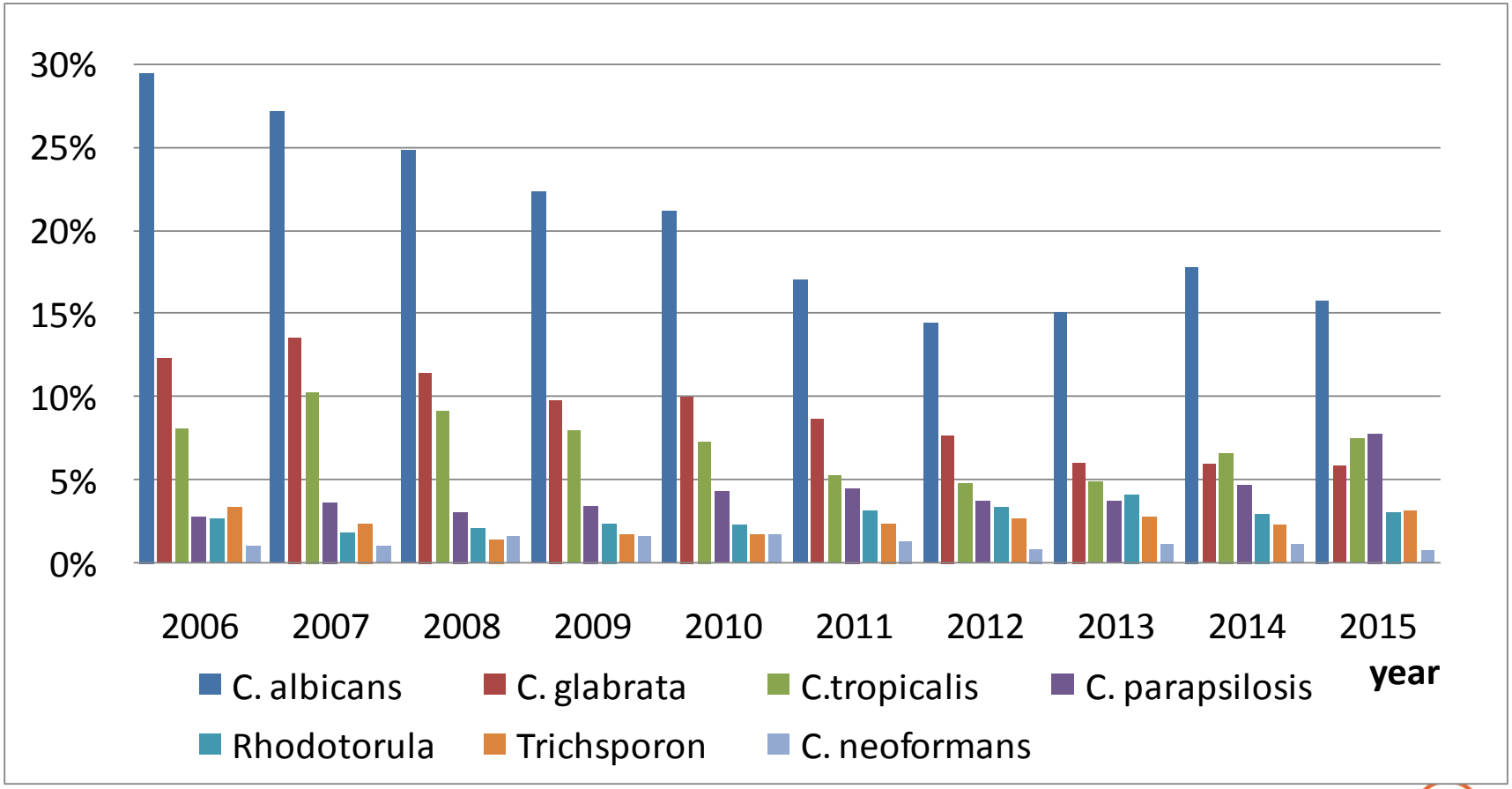


J Clin Microbiol 2012 Aug

CULTURE DATA (BLOOD)



CULTURE DATA (FUNGUS)



TAXONOMY AND IDENTIFICATION

- one fungus two names
  - sexual (teleomorph) and asexual (anamorph) forms of a fungus

Curr Fungal Infect Rep (2012) 6:170–177

Anamorph name	Teleomorph name (if present)	Older name
Blastoschizomyces capitatus	*Dipodascus capitatus*	*Geotrichum capitatum*
Candida ciferrii	*Trichomonoascus ciferrii*	*Stephanoascus ciferrii*
Candida famata	*Debaryomyces hansenii*	
Candida fermentati	*Meyerozyma caribbica*	*Pichia caribbica*
Candida guilliermondii	*Meyerozyma guilliermondii*	*Pichia guilliermondii*
Candida guilliermondii var. *membranifaciens*	*Kodamaea ohmeri*	*Pichia ohmeri*
Candida kefyr	*Kluyveromyces marxianus*	
Candida krusei	*Pichia kudriavzevii*	*Issatchenkia orientalis*
Candida lipolytica	*Yarrowia lipolytica*	
Candida lusitaniae	*Clavispora lusitaniae*	
Candida palmioleophila	None	
Candida pelliculosa	*Wickerhamomyces anomalus*	*Pichia anomala* *Hansenula anomala*
Candida utilis	*Lindnera jadinii*	
Cryptococcus gattii	*Filobasidiella bacillispora*	*Cryptococcus neoformans* var. *gattii*
Cryptococcus neoformans	*Filobasidiella neoformans*	*Cryptococcus neoformans* var. *grubii* (serotype A) and *C. neoformans* var. *neoformans* (serotype D)

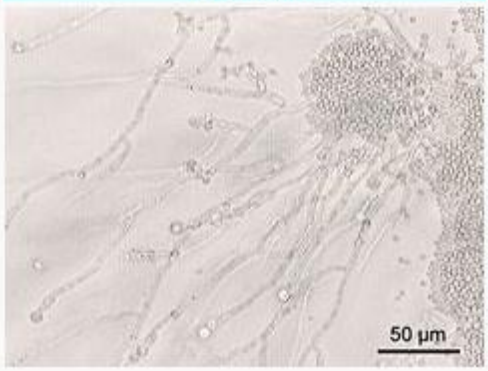
Curr Fungal Infect Rep (2012) 6:170–177

MALDI-TOF database

- *Candida albicans*
- *Candida albicans (africana)*
- *Candida dubliniensis*
- *Candida auris*
- *Candida glabrata*
- *Candida parapsilosis*
- *Candida tropicalis*
- *Candida krusei (Issatchenkia orientalis)*
- *C. guilliermondii (Meyerozyma guilliermondii)*
- *Cryptococcus gattii*
- *Cryptococcus neoformans*
- *Cryptococcus neoformans var_grubii*
- *Cryptococcus neoformans var_neoformans*

Candida and Other Ascomycetous Yeast

Candida



Candida albicans at 200X magnification

Scientific classification

Kingdom:	Fungi
Division:	Ascomycota 子囊菌門
Class:	Saccharomycetes
Order:	Saccharomycetales
Family:	Saccharomycetaceae
Genus:	***Candida*** Berkh. (1923)

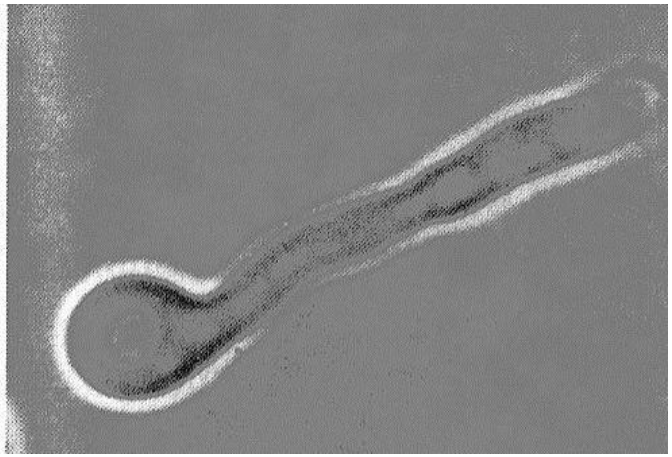
Type species

Candida vulgaris Berkh. (1923)

- a genus of yeasts
- the most common cause of fungal infections worldwide
- ~ 200 species
- *Candida* appears as large, round, white or cream colonies

C. albicans

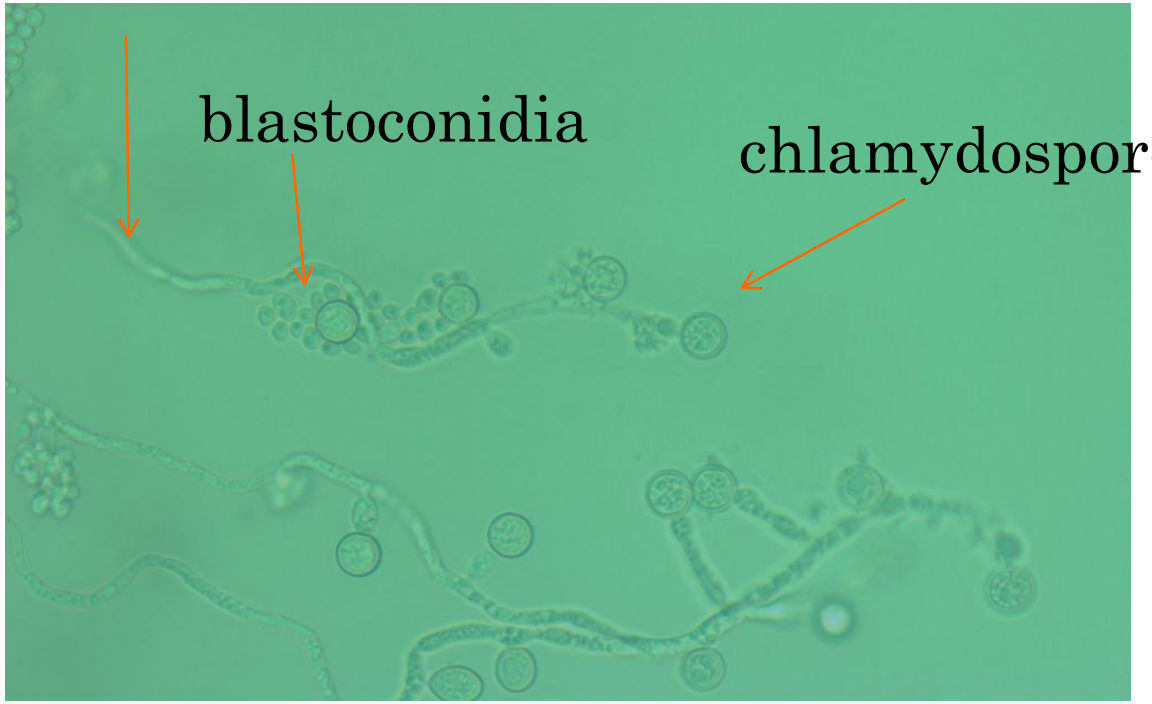
- Growth at 25, 37 and 42-45 ℃
- Germ tube
- susceptible to antifungal agents



C. albicans

- On cornmeal agar
  - Blastoconidia
  - Pseudohyphae
  - Chlamydospore

pseudohyphae



C. albicans ver. africana

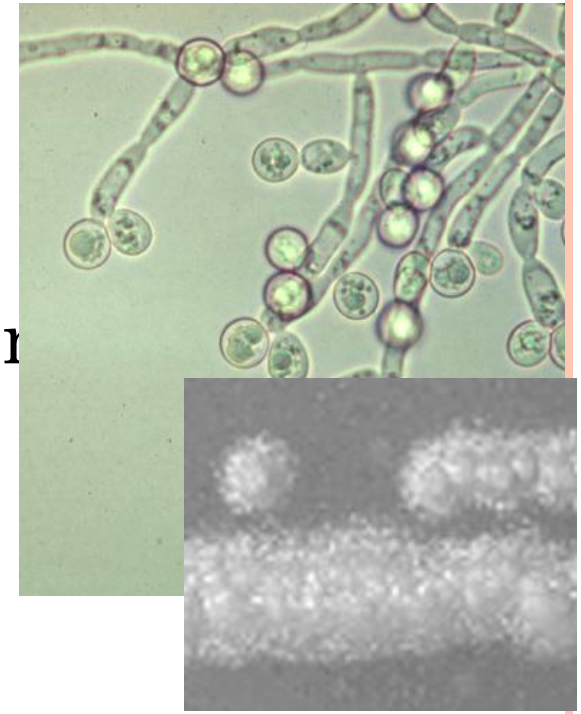
- susceptible to antifungal agents
- biovar of *C. albicans*
  - germ tubes (+)
  - Chlamydospores on cornmeal (-)
- Jan~Jun.2016 in CGMH

name	No.	%
C. albicans /*C. africana*	1011	99.5%
C. dubliniensis	5	0.5%

C. dubliniensis

- AIDS patient
- resistance to fluconazole (frequently or <5%)
- phenotypic similarities to *C. albicans*
  - ChromAgar
  - Germ tubes (+)
  - Chlamydospores on cornmeal (+)
- A European study - 52/2,589 isolates (2.0%).
- In USA – 15 *C. dubliniensis* / 1,251 *C. albicans* (1.2%)

C. dubliniensis



- CAP 2010-A F01(1046 participar laboratories)
  - 411 (39.4 %) → *C. dubliniensis* ✓
  - 445 (42.6 %) → *C. albicans* X

Characteristics	*C. albicans*	*C. dubliniensis*
Germ tube	+	+
Terminal Chlamydospores	Single or two	In pair, triple, cluster
Growth at 45℃	+	-
Pal's agars	Smooth colonies	hyphal fringe colonies

C. parapsilosis

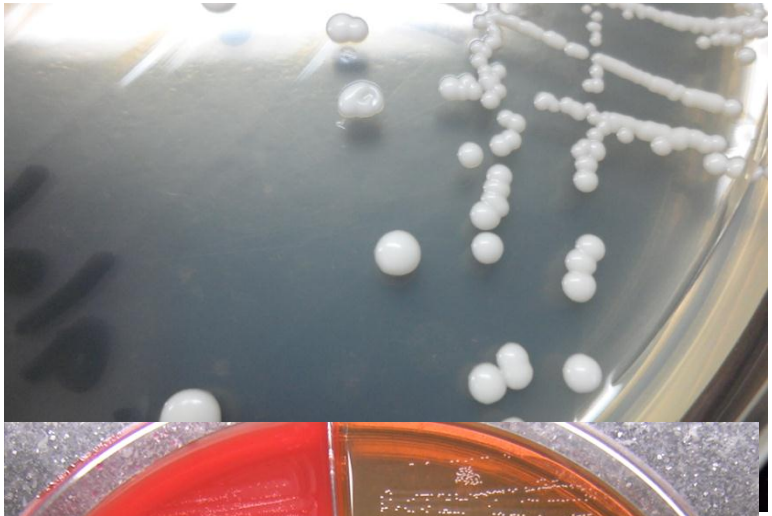
- similar morphology (DNA-based methods)
  - *C. orthopsilosis* and *C. metapsilosis*
  - low prevalence (~1 %) in most bloodstream isolate collections surveyed

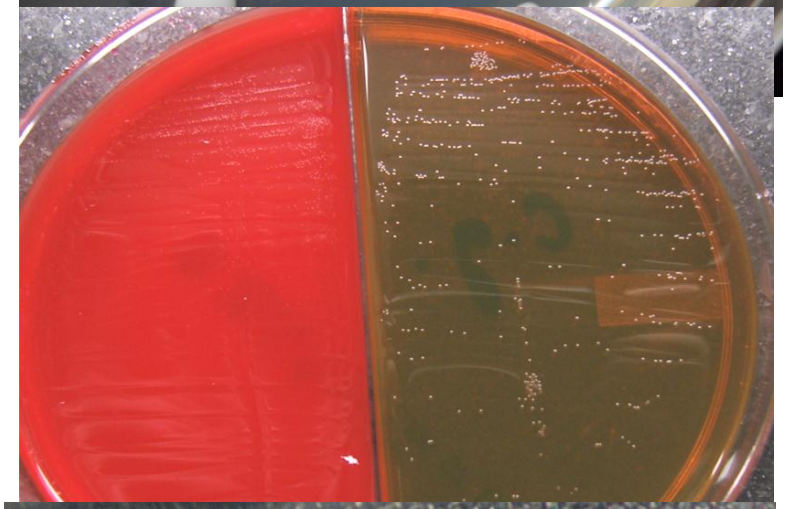
- Jan~Jun.2016 in CGMH

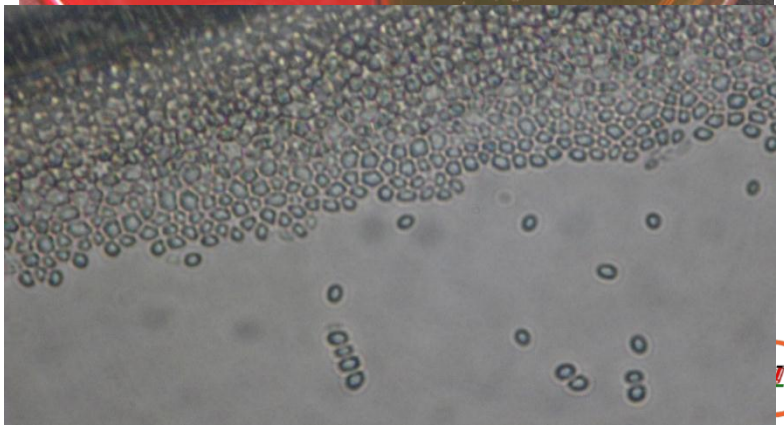
name	No.	%
C. parapsilosis	305	78.2%
C. metapsilosis	49	12.6%
C. orthopsilosis	36	9.2%

C. glabrata

- decreased susceptibility to azoles
- grows a bit more slowly than the other species of *Candida*.
- Growth better on EMB than blood
- blastospore, no pseudohyphae
- Rapid trehalose (+)







C. grabrata

- *Candida bracarensis* and *C. nivariensis* are genetically related to *C. glabrata*.
  - 1 % to 2 % of isolates

- Jan~Jun.2016 in CGMH

name	No.	%
C.glabrata	416	99.5%
C. nivariensis	2	0.5%

C. krusei (Issatchenkia orientalis)

- innate resistance to fluconazole
- reduced susceptibility to amphotericin B
- pseudophyphae with elongate blastoconidia forming a cross matchsticks or treelike.

C. auris

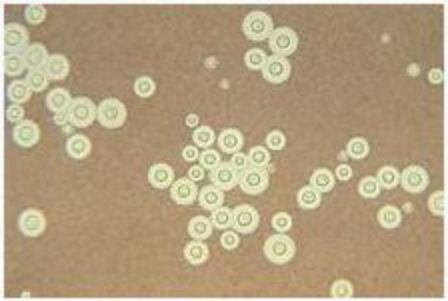
- first isolated from the external ear canal of a patient in Japan in 2009
  - bloodstream infections, wound infections, and otitis
- *Candida auris* is an emerging multidrug-resistant (MDR) yeast
  - resistant to the first-line antifungal drug fluconazole
  - rapidly evolve to develop resistance, so specific antifungal susceptibility testing is indicated
- its propensity for transmission between hospital patients.
- It has caused prolonged hospital outbreaks in India, Pakistan, Venezuela, and Colombia.

C. auris

- Germ tube (-)
- On cornmeal agar - pheudohyphae
- API AUX 20C and VITEK-2 YST - misidentify *C. auris as C. haemulonii, Saccharomyces cerevisiae or Rhodotorula glutinis.*
- molecular sequencing or MALDI-TOF
- Jan~Jun.2016 in CGMH
  - No=0

Cryptococcus Species

Cryptococcus



Cryptococcus neoformans

Scientific classification

Kingdom:	Fungi
Phylum:	Basidiomycota 擔子菌門
Class:	Tremellomycetes
Order:	Tremellales
Family:	Tremellaceae
Genus:	***Cryptococcus*** Vuill.

Type species

Cryptococcus neoformans

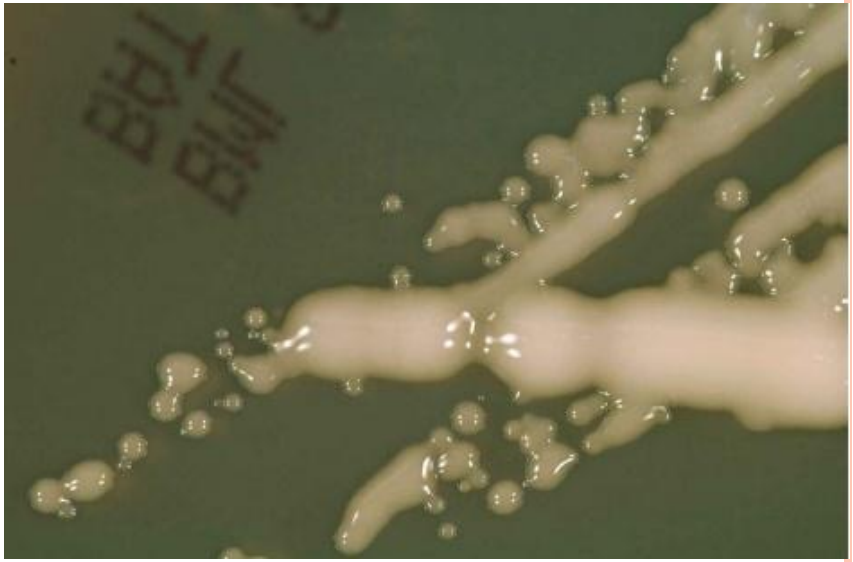
Synonyms

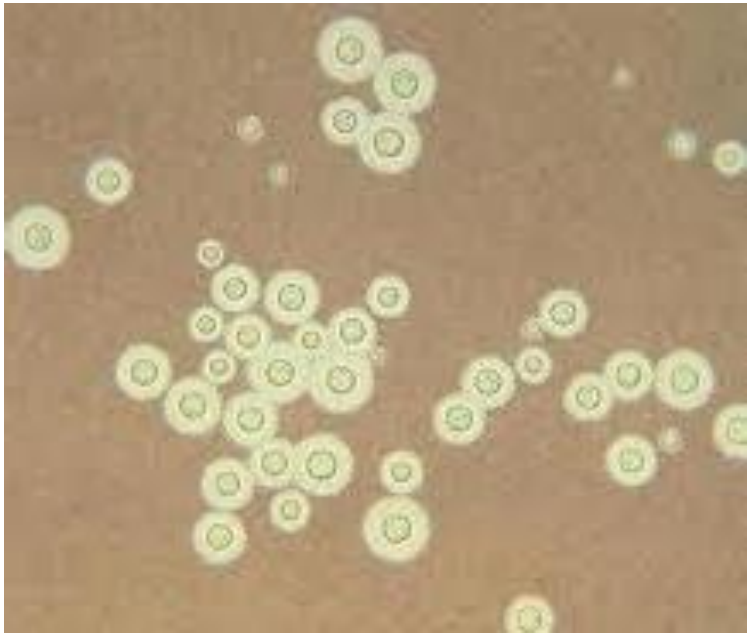
Filobasidiella

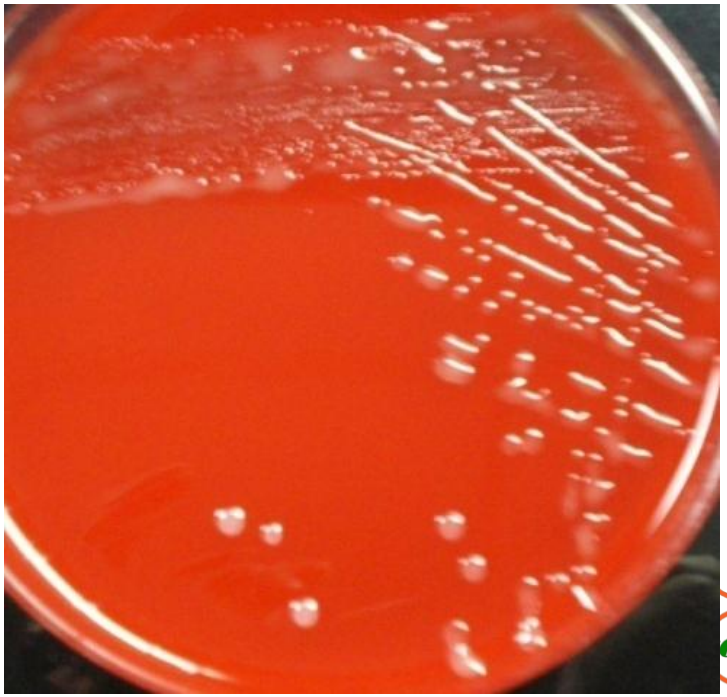
- ~37 recognized species of *Cryptococcus*
- The sexual forms or teleomorphs of *Cryptococcus* species are filamentous fungi in the genus *Filobasidiella.*
- cryptococcosis
  - *Cryptococcus neoformans*
  - *Cryptococcus gattii*

C. neoformans

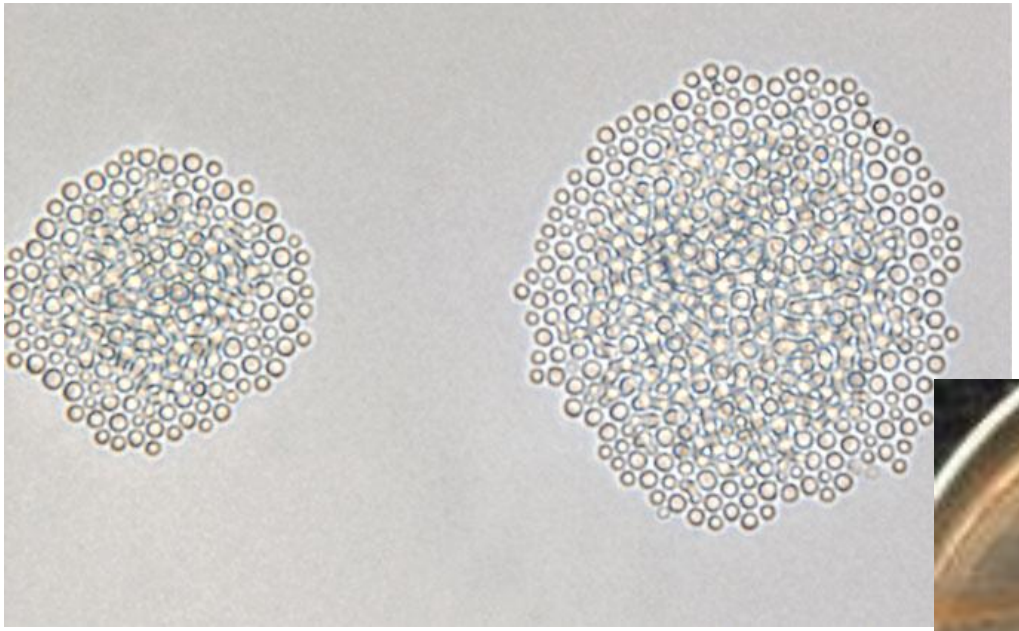
- Mucoid colony
- India ink (+)







C. neoformans



No psedohyphae

Urease(+)

phenol oxidase (+)

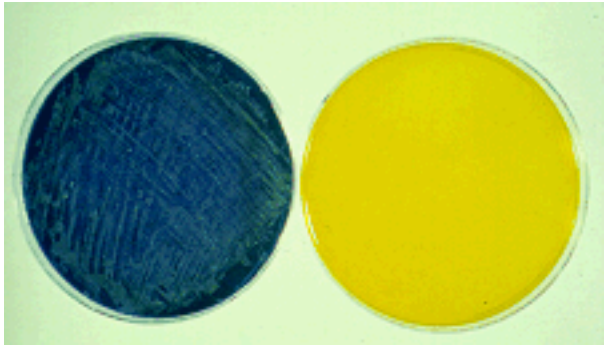
C. neoformans

- *C. neoformans var. neoformans* (serotype D)
  - more geographically restricted, found mostly in Mediterranean Europe
- *C. neoformans var. grubii* (serotype A or hybrid serotype AD.)
  - worldwide distribution
  - in soil which has been contaminated by bird excrement
  - AIDS

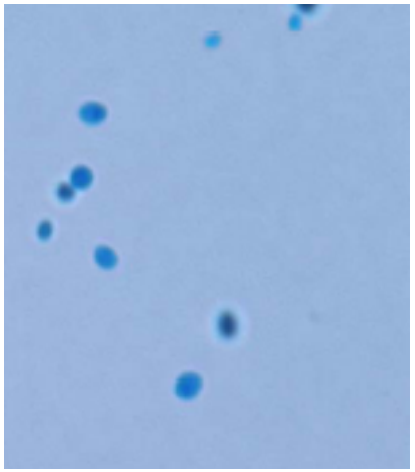
name	No.	%
Cryptococcus neoformans _var_grubii	38	100%

C. gattii

- *C. gattii* (serotype B,C)
  - (formerly *Cryptococcus neoformans* var *gattii*) is endemic to tropical parts of the continent of Africa and Australia (eucalyptus)
  - causing disease (cryptococcosis) in non-immunocompromised people
- Canavanine-glycine-bromthymol blue (CGB) agar
  - Blue - *Cryptococcus gattii*



Rhodotorula



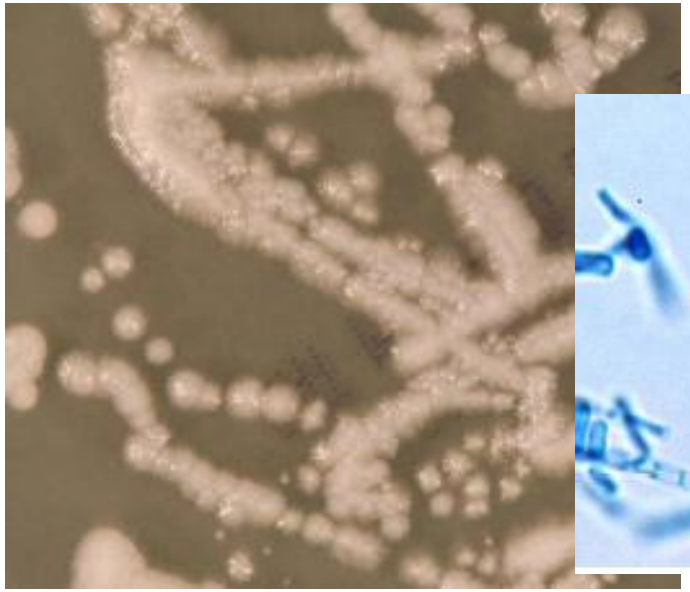
Scientific classification	
Kingdom:	Fungi
Division:	Basidiomycota
Class:	Urediniomycetes
Order:	Sporidiales
Family:	*incertæ sedis*
Genus:	***Rhodotorula*** F.C.Harrison (1927)
Type species	
Rhodotorula glutinis (Fresen.) F.C.Harrison (1928)	

- Smooth, pinkish colonies.
- Yeast morphology –forms only blastoconidia
- Urease test –will be positive.

Trichosporon

Trichosporon	
Scientific classification	
Kingdom:	Fungi
Phylum:	Basidiomycota
Subphylum:	Agaricomycotina
Class:	Tremellomycetes
Order:	Tremellales
Family:	Trichosporonaceae
Genus:	***Trichosporon*** Behrend, 1890
Type species	
Trichosporon beigelii (Küchenm. & Rabenh.) Vuill.	

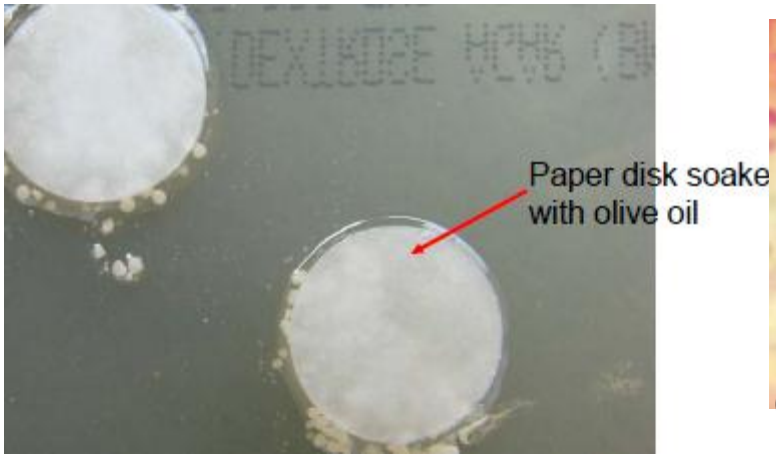
- Dry, rough and opaque white.
- Forms arthroconidia, hyphae and blastoconidia on yeast morphology
- Urease test –will be positive.

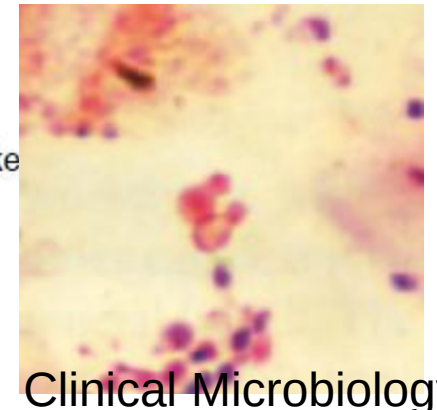


Malassezia furfur

Scientific classification	
Kingdom:	Fungi
Division:	Basidiomycota
Subdivision:	Ustilaginomycotina
Class:	Exobasidiomycetes
Order:	Malasseziales
Family:	Malasseziaceae
Genus:	***Malassezia***

- Pityriasis vericolor
- needs olive oil for growth
- Slow growing
- Small white colonies
- Wet mount –shows blastoconidia with budding
- API 20C AUX - misidentified- *C. lipolytica*



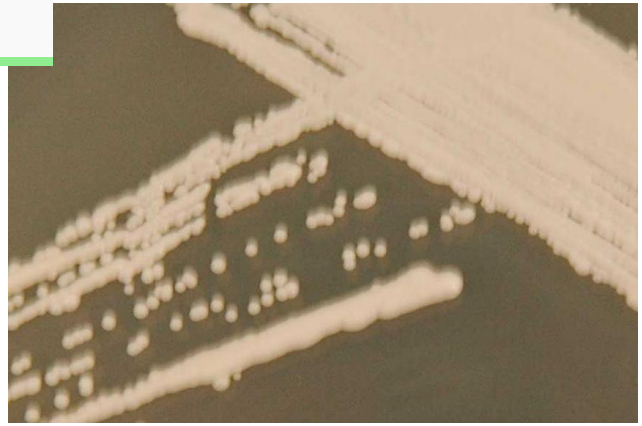


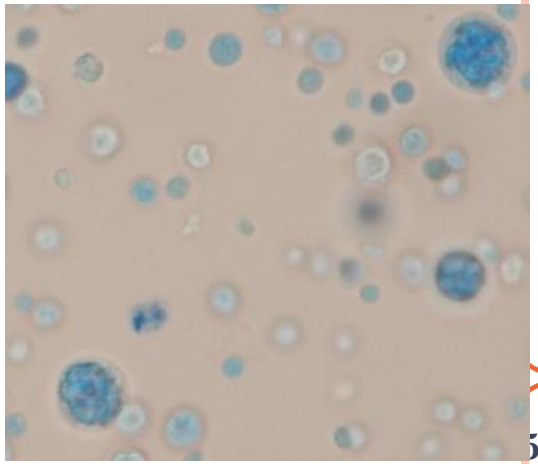
Clinical Microbiology Newsletter 2004;26:14

Prototheca 綠藻

Prototheca	
Scientific classification	
Domain:	Eukaryota
Kingdom:	Viridiplantae
Phylum:	Chlorophyta
Class:	Trebouxiophyceae
Order:	Chlorellales
Family:	Chlorellaceae
Genus:	***Prototheca*** Krüger, 1894[1]

- a genus of algae
- found worldwide in sewage and soil
- Protothecosis
  - a disease found in dogs, cats, cattle, and humans
  - a wound or abrasion





JAN.2016~JUN.2016 YEAST ISOLATED FROM CGMH

name	No.	%
C.albicans/C.africana	1011	40.2%
C. dubliniensis	5	0.2%
C.glabrata	416	16.5%
C. nivariensis	2	0.1%
C .tropicalis	399	15.9%
C. parapsilosis	305	12.1%
C. orthopsilosis	36	1.4%
C. metapsilosis	49	1.9%
C. guilliermondii	84	3.3%
C. guilliermondii _var_membranaefaciens	3	0.1%
Cryptococcus neoformans _var_grubii	38	1.5%
Rhodotorula mucilaginosa	28	1.1%
Trichosporon asahii	25	1.0%
Trichosporon mucoides	21	0.8%

name	No.	%
R. minuta	13	0.5%
Trichosporon faecale	10	0.4%
C. haemulonii	9	0.4%
C. lusitaniae	8	0.3%
C. krusei	7	0.3%
C. duobushaemulonii	5	0.2%
Trichosporon japonicum	5	0.2%
Trichosporon montevideense	5	0.2%
C. kefyr	4	0.2%
C. intermedia	3	0.1%
C. fabianii	3	0.1%
C. lipolytica	3	0.1%
Cryptococcus diffluens	3	0.1%
Kluyvera ascorbata	3	0.1%
other	15	0.6%
總計	**2516**	

長庚 林口

THANKS FOR YOUR ATTENTION