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Support of the laboratory for the diagnosis of skin infections: The lab point of vue

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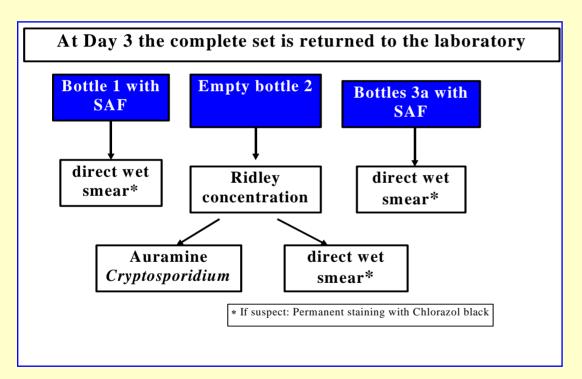
A few words about stool collection procedures for parasites

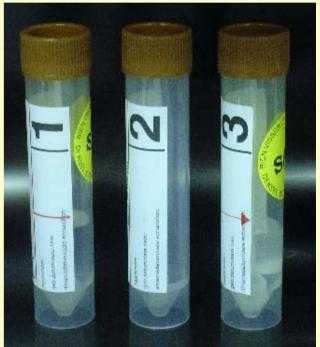


Triple Faeces Test

- •SAF preservation fluid for ova and trophozoites.
- •Samples collected during 3 consecutive days.
- We request to provide the stool with clinical information.

TRIPLE FAECES TEST





Recovery of parasites

comparison of one fresh stool specimen vs TFT :

from 10.33% to 28.07% positive cases, for both pathogenic and non-pathogenic protozoans.

Vandenberg et al. submitted to CMI. 2006

Diagnosis of skin infection: is the bacteriology lab useful?



NORMAL OR RESIDENT SKIN FLORA

- 1.5m² skin normally colonized within the first few days after birth predominantly with
 - Coagulase negative Staphylococci
 - Corynebacterium spp.
 - Micrococcus spp.
 - Propionibacerium spp.

And less frequently

- Acinetobacter spp. (25% population)
- Staphylococcus aureus (external nares of 30% pop.)
- Saprophytic Mycobacteria
- Malassesia
- Candida spp.
- Enterobacteriaceae
- Bacillus spp.
- **–**

P. Murray et al. Clinical Microbiology Handbook. 8th ed. 2003.

PREANALYTICAL STEPS

- The purpose of a sampling must be defined prior to its collection.
- Don't swab to know IF there is a microbial skin infection.
- Don't forget the microbiologist can be associated to the prescription.

HELPFUL CLINICAL INFORMATION FOR THE MICROBIOLOGIST.

- Origin of the lesion: trauma, animal or human bite, gardening wound, decubitus ulcer, genital ulcer, post surgical infection, ...
- Travel.
- Duration of the illness: acute vs chronic.
- Location of the lesion.
- Underlying disease : diabetes, HIV, immunocopromised patient...

WHY?

To inoculate appropriate selective culture media and choose the specific growth conditions.

• To search the most probable involved pathogens including viruses, fungi, mycobacterias and parasites.

HENCE

Don't hesitate to call the microbiologist to discuss the best approach to diagnosis prior to sending specimens to the laboratory.



WHAT IS THE ROLE OF THE MICROBIOLOGIST?

- Trying to find the aetiological agent of an infection.
- To discuss together with the clinician over the most useful tests to reach this goal: bacterial and viral cultures but also serology, specific stainings, direct immunofluorescence, molecular biology, ...
- To perform ID and AST on relevant isolates only:
 - many wound infection are polymicrobic, there is little need to identify them all.
 - AST on non relevant specimens may lead to an inappropriate (over)use of antbiotics.

WOUND SAMPLES

COLLECTION OF CLINICAL SPECIMENS (1)

- Superficial wounds are always colonized by commensal flora.
- Before swabbing, clean wound surface with 70% alcohol or non bacteriostatic sterile saline.
- Prefer pus or fluid aspirate in a syringe, deep swabbing or punch biopsy of the leading edge of the lesion.

JM. Miller. A guide to specimen management in clinical microbiology. 2nd ed. 1999. H. Isenberg. Clinical Microbiology Procedures Handbook, 2nd ed. 2004.

COLLECTION OF CLINICAL SPECIMENS (2)

- Syringes must be capped with a sterile closure.
 Syringes with needles in place are inacceptables!
- Virus cultures require specific transport media.
- Don't forget to specifically request yeasts and molds and mycobacterial cultures.



Tinea corporis

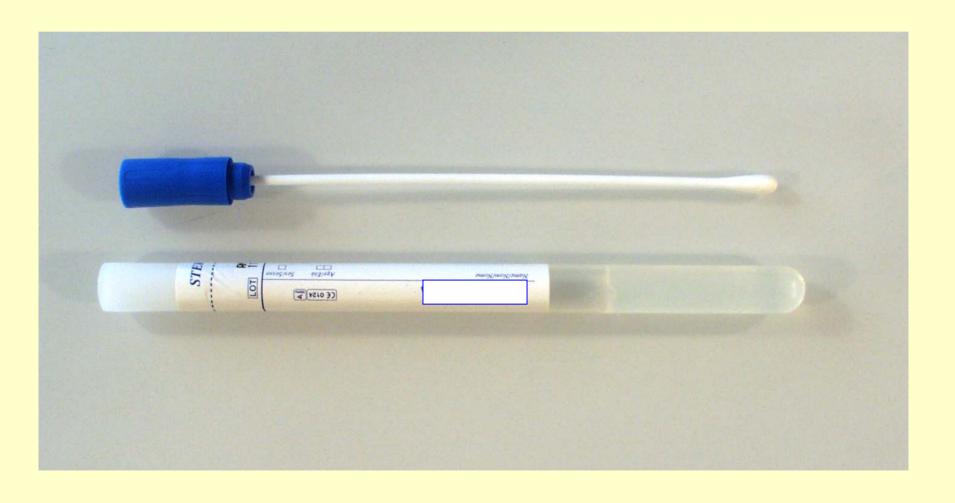


M. marinum

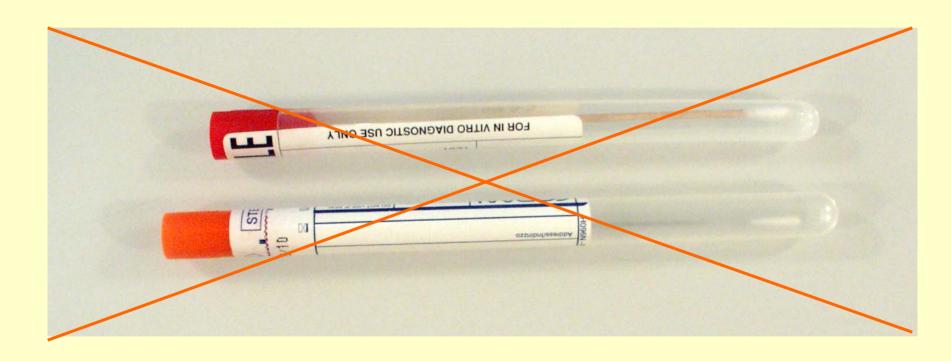
COLLECTION OF CLINICAL SPECIMENS (3)

 For bacteriological cultures, always use swabs with transport medium (gel or wet sponge) to prevent drying of the specimen and to preserve the bacteria.

Swab with transport medium



DRY SWABS



SHOULD NEVER BE USED FOR BACTERIOLOGICAL CULTURE

COLLECTION OF CLINICAL SPECIMENS (4)

- If anaerobic bacteria are suspected to be involved in an infectious process, prefer fine needle aspiration or tissue biopsy since most ordinary swabs are not valid transport media for these pathogens.
- Samples for diagnostic and epidemiologic surveillance (eg MRSA) are to be differentiated.

NEVER USE A SWAB IF PUS OR LIQUID SECRETIONS ARE AVAILABLE

PLEASE REMEMBER

Homepathy is based on dilutions principle:

The more diluted, the more efficient!

Microbiology is quite different:

The greater amount of microorganisms,

the better recovery of them in culture

• Insufficient material may yield false-negative results.

• The more different analysis you want, the greater amount of infected material you have to provide!

REMEMBER ALSO

• Some microorganisms are not cultivable.



2nd Syphilis





Lyme disease







Leprosy





Myiasis





REMEMBER ALSO

- Some microorganisms are not cultivable.
- Rare diseases are rare but yet do really exist!



Anthrax : Bacillus anthracis





Tularemie

WORST SPECIMENS FOR BACTERIOLOGICAL CULTURE (1)

- Superficial swabbing of a wound :
 - Yield many squamous epithelial cells and little PMNs on microscopic examination.
 - Is of little diagnostic value because of presence of many skin commensals.
 - Isolated organism(s) may only be colonizing ones not involved in the infective process.

The presence of epithelial cells indicates contamination of the specimen with skin microbiota and compromises the significance of the culture results.

Superficial swabbing of a wound

→ Usually no identification and no antimicrobial susceptibility testing on such samples.

WORST SPECIMENS FOR BACTERIOLOGICAL CULTURE (2)

 Don't submit decubitus ulcers swabs in order to establish an aetiological diagnosis.

These wounds are always colonized with resident, transient or fecal flora.

But this culture is useful to look for presence of MRSA.

REJECTION CRITERIA FOR MICROBIOLOGICAL SPECIMENS: SOME EXAMPLES

- Specimens recieved in fixative. Exception: stool for parasites.
- Dry swab.
- Only one swab for multiple requests.
- Anaerobic cultures on decubitus ulcer material.

BEST SPECIMENS FOR BACTERIAL CULTURE

- From clinically infected or deteriorating wounds.
- Whose result will influence therapy.
- Specimens collected prior to initiation of therapy.

BEST SPECIMENS FOR BACTERIAL CULTURE

- Tissues collected during surgery.
- Aspirates through intact skin by needle and syringe or fine-needle biopsy eventually after irrigation with non bacteriostatic saline.
- Samples from viable infected tissues avoiding necrotic aeras.

SOME EXAMPLES

CLINICAL IMPRESSIONS AND MAIN POTENTIAL PATHOGENS

Impetigo, echhyma	Strep. pyogenes, Staph. aureus
Folliculitis, furuncles	Staph. aureus
Erysipelas	Strep. pyogenes
Cutaneous abcess	Staph. aureus, Staph. epidermidis, Streptococcus spp., Propionibacterium spp., Peptostreptococcus spp., Leishmania tropica or brasiliensis
Nodules, papules, subcutaneous tissue involvement	Various bacteria, mycobacteria, yeasts and molds, viruses, parasites
Tinea	Various dermatophytes



Impetigo: Staphylococcus aureus



Impetigo: Streptococcus βA

DIABETIC FOOT INFECTION







DIABETIC FOOT INFECTION

Culture for infecting organisms remains problematic:

- Deep tissue cultures (surgically obtained) seem to provide the most accurate results.
- Poor correlation was demonstrated between deep tissue culture and other modalities (ulcer swabs, draining material, ...).
- Most patient have several isolates, all of them are not involved in the infective process.



ERISYPELAS



Acute skin infection caused by *Streptococcus pyogenes*.

Culture of the lesions

- lacks of sensitivity
 - Needle aspiration of the lesion → 5% culture +
 - Punch biopsy of the skin → 20% culture +
- is frequently contaminated with normal skin flora and Gram stain is not contributive.
- its interest is mainly epidemiological.

Blood cultures

- seldom positive (5% in febrile patients).



TAKE HOME MESSAGE

Overuse of microbiology lab can lead to its misuse

CONCLUSION

