Centrifuging Nail Specimen Debris Increases Sensitivity of Fungus Detection

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Objectives—Nail Unit Processing

• Nail bed/matrix
• Nail plate
• New method for fungal detection
Need to be involved in lab prep

Clinical

| Surgery | Grossing | Embedding | Slide |

Laboratory

Need to be involved on this side
Goal in Submitting Nail Bed and Matrix Biopsy

- Orientation of tissue
- Clear information to laboratory
- Reproducible among different laboratories
Idea—Orientation of Tissue

Cartoon of Eyes
Idea—Orientation of Tissue

Cartoon of Eyes
Inking to maintain orientation
Orientation of soft tissue

Matrix  Bed

Orientation is helpful whether the lesion is inflammatory or neoplastic
Place biopsy in cassette
Cover with sponge to hold in place
Submit in an excess of 10% fresh formaldehyde.
Summary of Matrix/Bed Biopsy

• Use cartoon diagram
• Ink one end for orientation
Submit nail plate separately from soft tissue
Pigment in the nail plate

- Blood
- Melanin
- Fungus
- Exogenous pigment
- Medications
Blood

• Evident on standard H&E.
  – Benzidine stain works but not necessary
    • Carcinogenic
  – Perl’s iron stain does not work in the nail.
    • Iron not released from heme.

• May still be a subjacent neoplasm or dermatitis (fungus).
Nail fungus diagnostics

- Calcofluor White Stain
- PAS
- KOH
- Culture

Nail sampling is an issue

- Subungual debris is better than nail plate for sampling.
  - Issue—Not all patients have subungual debris.

New Method: Centrifuging Debris

- Formalin sample is reduced in volume in a large volume centrifuge.
- Pellet moved to Cytospin (Fisher HealthCare) centrifuge.
- Debris is centrifuged directly onto a glass slide.
CytoSpin Centrifuge
Centrifuge with slide
Centrifuge with slide
Study—204 Cases

• 132 cases positive on initial nail PAS
• 72 cases negative
  – 52 cases centrifuged after negative nail PAS
  – 20 cases (10%) not centrifuged, usually because nail was submitted dry.
Study—204 Cases

• 18 (of 52) cases positive on centrifuge
• Total 150 positive cases—nail and centrifuge
Study—204 Cases

- Sensitivity increase of 12%.
- 74% nails positive for fungus
Study—52 Centrifuged Cases

• 18 cases positive on centrifuge
• 34 negative on centrifuge
  – Bacteria—20 cases
  – No bacteria—14 cases
Study—Bacteria

• 59% of negative cases were associated with bacteria, usually cocal forms.

• Suggests that bacteria, probably Staphylococcus species, are pathogenic in nail disease.
Study—Conclusion

• Positive cases—sensitivity increase of 12%
• Negative centrifuge cases often associated with bacteria (59%)
Conclusion

• Cartoon orientation of bed/matrix biopsies
• Separate nail plate—blood issues
• New fungal diagnostics via centrifuging
Gratitude:

Phoebe Rich, Oregon
Monica Lawry, California
Hendrik Uyttendaele, New York
Brandon Stokes—Centrifuge Technique
Rodd Takiguchi—Centrifuge Study