Sexually Transmitted Diseases

Genitourinary Core
Computer Assisted Instruction
2008

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Session Composition

- 10 Stations
  - Each contains a single disease
  - Questions for review
Stations

- **Viral Diseases**
  - #1 Genital herpes
  - #2 Genital warts
  - #3 HIV infection

- **Bacterial Diseases**
  - #4 Chlamydia
  - #5 Gonorrhea
  - #6 Syphilis
  - #7 Bacterial vaginosis

- **Fungal Disease**
  - #8 Vaginal candidiasis

- **Parasitic Diseases**
  - #9 Trichomoniasis
  - #10 Crab lice infestation
Viral STDs

- Genital herpes
- Genital warts
- HIV infection
Station #1
Genital Herpes

- **Etiological agents**
  - *Herpes simplex* viruses (HSV)
    - Type 1
    - Type 2
  - Viral characteristics
    - DNA virus
    - Similar to other herpes viruses
      - Virus establishes latency in dorsal root ganglia after 1° infection
      - Reactivation of infection may occur
      - Cannot be cured
        - Control for reactivation
Genital Herpes

Transmission

- Direct viral contact
  - Genital secretions
    - During active infection
- About 25% of general population is sero-positive
Genital Herpes

**Clinical features**

- **1° infection**
  - Genital tract vesicular lesion
  - Fever and lymphadenopathy

- **Recurrent infection**
  - **Frequency**
    - Varies
    - Range from 0 to 12 or more infections yearly
  - Milder symptoms
  - Shorter duration of lesion healing
Genital Herpes

**Laboratory diagnosis**

- Specimen source
  - Lesion scrapping
  - Vesicular fluid
- Test methods
  - Direct exam or
  - Culture
  - Both are confirmatory tests

- **Direct exam**
  - Fluorescent antibody stain
    - Commercial stain
      - Fluorescein-labeled HSV-1 or HSV-2 antibody
    - Confirmatory

Apple-green fluorescent HSV infected cells,
Original magnification (OM) 400x
**Genital Herpes**

- **Culture**
  - Conventional tissue cell culture
  - Characteristic CPE produced
    - Cells round and subsequently detach from tube
    - Fluorescent stain of cells
      - To identify viral species
  - Confirmatory

CPE = cytopathic effects

**Normal tissue culture cells**, **OM 400x**

**Viral CPE suggesting herpes**, **OM 400x**
Genital Herpes

**Unique facts**

- *Herpes simplex* viruses
  - Also associated with oral cold sores
  - Both Type 1 and Type 2 HSV can be associated with oral and genital herpes
- Herpes infection is highly contagious
  - When active lesions are present
- Herpes infection is controlled but not cured
Which immunological test is commonly used on direct smears to identify *Herpes simplex* virus and to distinguish Type 1 from Type 2-caused disease?

- Fluorescent antibody test
  - Use commercially-available fluorescein-labeled HSV-1 and HSV-2 antibody
  - Considered confirmatory
Station #2
Genital Warts

- **Etiological agent**
  - Human papillomavirus (HPV)

- **Two main types of HPV**
  - **Cutaneous type**
    - Most associated with warts on….
      - Feet
      - Hands
      - Arms
      - Head
  - **Mucosa type**
    - Affect anogenital epithelium
    - > 40 mucosa types known
    - Some associated with epithelial cancers
      - Types 16, 18, 45, 56, 58, 59, & 68
      - Called high risk types
Genital Warts

**Transmission**
- Direct contact
  - Through sexual exposure

- Genital warts......
  - The most common viral sexually transmitted disease in the US
    - Specific numbers unknown
      - Not a nationally reportable disease
Genital Warts

Clinical features

– Called condyloma acuminatum
– Affects anogenital tract epithelium usually
  • May also affect upper respiratory tract
– Hyperkeratotic lesions of 2 types
  • Flat
  • Attached
    – By a broad stalk-like peduncle
– Gender specific symptoms
  • Males
    – Located on penile shaft or perianal area
  • Females
    – Perianal area, vagina, and cervix
    – Cervical infection frequently flat and not easily detected
Genital Warts

- **Laboratory diagnosis**
  - HPV cannot be grown in cell culture

  - Colposcopic exam
    - Macroscopic appearance of warts are diagnostic
    - However, a majority of genital HPV infections are the flat type
      - Not visible to the unaided eye
      - Usually found in the cervix

  - Specimens
    - Exfoliated cell samples
      - From PAP smear in females
    - Tissue biopsy
Genital Warts

- **Laboratory diagnosis**
  - Direct exam
    - Cell scrapping or biopsy
  - Biopsy
    - **Koilocytes** in tissue
      - Characterized by...
        - Peri-nuclear clearing
        - Increase in density of surrounding rim of cytoplasm

Cervical bx, H & E, OM 400x
Genital Warts

- **Cell scrapping**
  - Papanicolaou (PAP) smear
  - Images of cervical specimen using the PAP smear (OM 100x)

- **HPV Typing:** Can be done using nucleic acid probes directed against specific viral types. Not currently routine

- **Normal**
  - Low grade squamous epithelial lesions with no dysplasia
  - Characteristic of HPV infection

- **Moderate dysplasia**
  - Cervical carcinoma
Genital Warts

Unique facts

- Given the role specific HPV types play in pathogenesis of cervical cancer ..........
  - It is likely that typing will some day become standard of care to predict risk of cancer
- A HPV vaccine is now available
  - Recommend to begin immunization of females at age 11-12 years
Station #2
Questions

- Which characteristic cell type is observed in tissue infected with the HPV?
  - Koilocytes

- End Station #2
Station #3
HIV Infection

- Etiological agent
  - Human immunodeficiency virus
    - Types 1 and 2
      - HIV-1
      - HIV-2
  - Classified as a Retrovirus
HIV Infection

**Transmission**
- Sexual intercourse
- Connately from mother to child
- Postnatally by breast feeding to child
- Parenteral inoculation
  - IV drug abuser

- Most frequent route of transmission
  - Vaginal infection of women by unprotected sexual intercourse
HIV Infection

Clinical features
- CD4 T lymphocytes
  - Represent the ultimate target
  - Infection culminates in…..
    - Severe immunosuppression
    - Opportunistic infections
    - Cancer
    - Ultimately leads to premature death
- Disease types
  - Symptomatic
    - Called acquired immunodeficiency syndrome (AIDS)
  - Asymptomatic
    - Called HIV infection
- Illness progresses over the course of many years
HIV Infection

- Laboratory diagnosis
  - Specimen
    - Serum
      - Diagnosis is performed by serological testing
HIV Infection

- **Laboratory diagnosis**
  - **Initial screen**
    - Enzyme linked immunosorbent assay (ELISA)
      - To detect HIV antibody in serum
      - Inexpensive test
      - Automated test for large volume screening
    - False-positive results possible
      - Requires confirmation testing
HIV Infection

- **Laboratory diagnosis**
  - **Confirmation test**
    - Western blot assay
      - Utilizes commercially prepared paper strips containing HIV protein antigens
        - Proteins have been electrophoresed and separated by size
      - **Test procedure**
        - Place patient’s serum over the strip
        - **Specific** antibodies if present will react with **specific** proteins on strip
        - Reactions are visualized as “bands”
      - **Interpretation**
        - Presence of two or more bands of major proteins are diagnostic for HIV infection
        - Major proteins include: gp120, gp160, p41, and p24
HIV Infection

Western blot assay examples

Each strip represents one patient....protein size is indicated on the left.

- Patient’s 1, 4, 9 & 11 have HIV infection
- Patient’s 3 & 8 are negative
- Patient’s 2 and 5 are indeterminate (require repeat testing after waiting for 3-4 weeks)
HIV Infection

Interesting facts

- Disease diagnosis most frequently done by observing an IgG immune response
- Several companies offer kits that can detect both specific antibodies and antigens
  - Many “over the counter” test kits are available to the general public
- AIDS is the 4th most common reportable disease in the U.S.
  - 2005 totals (top 5)
    - Chlamydia 976,445 cases
    - Gonorrhea 339,593
    - Salmonellosis 45,322
    - AIDS 41,120
    - Syphilis 33,278

(CDC, MMWR, Vol 54, March 30, 2007)
Station #3
Questions

• **How is an HIV infection confirmed?**
  – Screen for IgG antibodies to HIV
  – Confirm screen-positive results

• **Name the common tests used for screening and confirmation.**
  – Screening = ELISA test
  – Confirmation = Western blot assay

• **End Station #3**
Bacterial STDs

- Chlamydia
- Gonorrhea
- Syphilis
- Bacterial vaginosis
Station #4
Chlamydia

- **Etiological agent**
  - *Chlamydia trachomatis*
    - Small intracellular bacterium
    - 0.3 µ in diameter
      - Recognized as one of smallest bacterium known

- Not part of the normal flora
  - True pathogen
Chlamydia

**Transmission**
- Sexual exposure
- Neonate during exposure to infected birth canal

- Reportable disease
  - Most common reportable disease in U.S.
  - About 1 million new cases each year reported
    - Suspect this only represents 25% of actual cases
Clinical features

- Bacteria attach to mucosal cells
  - Urethra
  - Cervix
  - Rectum
  - Eyes
  - Oral-pharyngeal area
- Produces a nongonococcal urethritis or cervicitis
  - Males
    - Usually symptomatic
  - Females
    - Usually asymptomatic
- Clinical features identical to gonorrhea
Chlamydia

- **Laboratory diagnosis**
  - Specimen sources
    - Urethral swab
      - Males
    - Cervical swab
    - Urine
      - Both females and males
      - Becoming more common because of the noninvasive collection procedure
    - Other sources
      - Throat
      - Rectum
      - Eye
  - **Direct exam**
    - Not commonly done
    - Bacteria not observed on Gram stain
Chlamydia

**Laboratory diagnosis**
- **Culture**
  - Difficult
    - Requires cell culture
    - *C. trachomatis* acts like a virus
      - Will only grow *in vitro* in living cells
      - Not on standard bacteriological media
  - Required for unusual specimens
    - Throat, rectum, and eye
    - Non-culture methods only FDA approved for ...
      - Cervix
      - Urethra
      - Urine
  - Confirmed in cell culture by FA staining using specific antibody
    - Detect chlamydial inclusions

FA stained cell culture

OM 100x

↑

Chlamydial inclusion

OM 400x
Chlamydia

- **Laboratory diagnosis**
  - Non-culture tests
    - Mainstay of diagnosis
    - PCR-based automated tests
      - Useful for high volume testing
    - Approved for....
      - Urethral
      - Cervical
      - Urine specimens
    - Highly sensitive (>95%)
      - <5% false negatives
    - Highly specific (<99%)
      - <1% false positives
  - Dual testing common
    - Detect for both chlamydia and gonorrhea at same time
Chlamydia

- **Facts**
  - *Chlamydia trachomatis* is a bacterium with viral characteristics
    - Similarities to virus
      - Obligate intracellular parasite
      - Requires living cells to grow *in vitro*
      - Small size
    - Differences from virus
      - Contain both DNA and RNA
      - Can treat infections with antibacterial agents
Station #4
Questions

• When is culture to diagnose chlamydia most appropriate?
  – To evaluate throat, eye, and rectal specimens

• How common is chlamydia in the U.S.?
  – Most common reportable disease

• End Station #4
Station #5
Gonorrhea

- **Etiological agent**
  - *Neisseria gonorrhoeae*
    - Gram negative coccobacillus
  - Not part of normal flora
    - True pathogen

Gram stain from culture,
OM 1000x
Gonorrhea

Transmission

- Sexual exposure
- Neonate in eyes during birth

- Reported by CDC as 2\textsuperscript{nd} most common reportable disease in U.S.
  
  - About 400,000 cases reported per year
    - May represent less than half of the actual cases
Gonorrhea

**Clinical features**
- Identical to chlamydia
- Males
  - Symptomatic with urethritis
- Females
  - Asymptomatic with cervicitis
Gonorrhea

**Laboratory diagnosis**

- Specimen
  - Urethral swab
    - Male
  - Cervical swab
    - Female
  - Urine
    - Female and male
  - Other specimens
    - Throat
    - Rectum
    - Eye

(Similar to chlamydia)
Gonorrhea

**Laboratory diagnosis**

- Direct exam
  - Unlike chlamydia, direct Gram stain is useful
  - Gram-negative intracellular diplococci
    - Pyogenic infection (PUS)
    - Diagnostic in males
    - Presumptive in females

Urethral exudate, Gram stain, OM 1000x
Gonorrhea

- Laboratory diagnosis
  - Culture
    - Fastidious organism
      - Requires direct plating of specimen to medium after collection
      - Sensitive to cooling
        - Keep plate and specimen at room temperature
      - Sensitive to atmospheric changes
        - CO₂-enriched environment during transport
        - Candle jar is a useful method
    - Requires enriched medium to grow in vitro
      - Chocolate agar
      - Contains enrichments needed for growth
  - Selective medium
    - Useful to detect *N. gonorrhoeae* from specimens contaminated with normal flora
      - Medium contains antimicrobial agents
    - Example is Modified Thayer Martin agar
      - Chocolate agar base
      - Contains vancomycin (inhibit gram-positive bacteria), colistin (inhibit gram-negative bacteria), and nystatin (inhibit yeast)
Gonorrhea

- Laboratory diagnosis
  - Identification from culture
    - Requirement for fastidious growth
      - Growth only on chocolate agar
      - Growth only in the presence of a CO₂ enhanced atmosphere
    - Carbohydrate acidification
      - Biochemical tests
        - Observe for the ability to ferment a pattern of sugars
        - Classic method for culture ID
Gonorrhea

- Laboratory diagnosis
  - Non-culture tests
    - Test in combination with chlamydia detection
    - PCR-based assays
      - The mainstay of diagnosis
    - Confirmatory
Gonorrhea

- **Interesting facts**
  - Nucleic acid amplification tests (NAATs)
    - Are the mainstay of diagnosis
  - NAATs can be performed on urine
    - Eliminates “invasive” procedures necessary for specimen collection
    - More people will consider getting tested
Station #5
Questions

- *Neisseria gonorrhoea* is a fastidious bacterium that is sensitive to which environmental conditions?
  - Cooling and atmospheric air

- Describe the classic Gram stain picture of gonorrhea.
  - Gram-negative intracellular diplococci
  - Many white blood cells (PMNs)

- End Station #5
Station #6
Syphilis

- Etiological agent
  - *Treponema pallidum*
    - Spirochete
    - Up to 20 µ in length
  - Obligate parasites of humans
    - No known animal or environmental reservoirs
  - True pathogen

Darkfield, Wet prep
OM 400x
Syphilis

Transmission
- Sexual intercourse
  - Direct contact with active lesions
- Transplacental
  - Infected mother to fetus
Syphilis

**Clinical features**

- Wide variety of clinical manifestations

- 1° syphilis
  - At site of infection
    - Lesion characterized by…
      - Ulceration
      - Generally painless
    - Lesions called chancres
Syphilis

Clinical features

- 2° syphilis
  - Disseminated infection
    - Six weeks to 6 months after 1° disease
    - Multiple papular lesions
      - Palms of hands
      - Soles of feet
      - Other locations
    - Patchy hair loss common
Syphilis

Clinical features

- Latent syphilis
  - Interval between or following episodes of 1° and 2° syphilis
  - About 75% of persons untreated will remain in this stage for life
- Tertiary syphilis
  - Occurs in about 25% of untreated cases
  - Characterized by chronic inflammatory granulomas
    - Called gumma
  - Affects...
    - Central nervous system
    - Aortic valve
    - Thoracic aorta
    - Skin
    - Bone
Syphilis

**Laboratory diagnosis**

- Specimen
  - 1° syphilis
    - Aspirate from chancre
  - 2° and tertiary syphilis
    - Serum to detect a serological response
Syphilis

- **Laboratory diagnosis**
  - **Direct detection**
    - 1° syphilis
      - Dark-field microscopic exam
        - Spirochete in lesion aspirate

Dark-field exam, chancre, OM 1000x
Syphilis

- **Laboratory diagnosis**
  - **Culture**
    - Organism cannot be cultured *in vitro*
  - **Sero logical**
    - To diagnose
      - Test for immunological response to infection
      - Usually involved 2nd or latent syphilis
    - Two methods used
      - Screen
        - Using a non-treponemal test
        - Some false positive results occur
      - Confirm
        - Using a more specific treponemal test
Syphilis

- Laboratory diagnosis
  - Non-treponemal screen
    - Measure antibody directed against a non-specific antigen
      - Antigens called reagin
    - Highly sensitive
      - But low specificity
        - False positive results common
  - Require positive results to be confirmed
  - Test methods used
    - Venereal Disease Research Laboratory (VDRL) test
    - Rapid Plasma Reagin (RPR) test
    - ELISA test
Syphilis

- **Laboratory diagnosis**
  - Treponemal tests
    - To confirm screen positive results
    - Detect anti-treponemal antibody
      - Highly specific
      - Most common test
        - Fluorescent-treponemal antibody-absorption (FTA-ABS) test
      - Performed by overlaying slide containing commercially obtained *T. pallidum* with serum from patient
      - Subsequently stain with fluorescent-labeled antihuman reagent

FA, Shows fluorescing spirochetes to indicate specific antibody is present OM 400x
Interesting facts

- Syphilis is one of most common STDs in the U.S.
  - Over 30,000 new cases each year
- Diagnosis is usually by a serological test
  - To detect an antibody response.
- *T. pallidum* cannot be cultured *in vitro*
Screen for syphilis is done using which test method?
- VDRL, EIA, or RPR test

Why is confirmation testing required?
- Screening tests detect a non-specific antibody associated with syphilis and thus may be a false-positive result

Which test is used to confirm a screen positive result?
- FTA-ABS test

End Station #6
Station #7
Bacterial Vaginosis

- **Etiological agents**
  - *Gardnerella vaginalis*
    - Coryneform gram-positive rod
  - Various anaerobes

- Unknown how these bacteria interact to produce infection
  - Decrease in normal flora vaginal *Lactobacillus* one possibility
- Organisms considered normal vaginal flora
  - But for not completely understood reasons overgrow in individuals with BV
Bacterial Vaginosis

Transmission

- BV is an endogenous infection in females

- *G. vaginosis* may be recovered from urethra of males
  - Disease association is however questionable
Bacterial Vaginosis

Clinical features

- Inflammatory response
  - Production of a discharge with pungent fishy odor

Cervix covered by frothy discharge
Bacterial Vaginosis

- **Laboratory diagnosis**
  - Specimen
    - Vaginal secretions
  - Direct detection
    - Visualization of “clue cells”
      - Squamous epithelial cells peppered with *G. vaginalis*

Vaginal secretion wet mount, OM 400x; Clue cells present
Bacterial Vaginosis

**Laboratory diagnosis**

- **Culture**
  - Not useful for diagnosis

**Clinical diagnosis**

- Vaginal secretions
  - pH > 4.5
  - Fishy amine odor following application of 10% KOH
  - Presence of “clue cells”
Bacterial Vaginosis

**Interesting facts**
- The gold standard for the diagnosis of BV
  - Direct examination of vaginal secretions
  - Culture not useful
    - *G. vaginalis* can be also be recovered from healthy females
Station #7

Questions

- Describe the major cell type observed in the vaginal secretions of women with BV?
  - Clue cells
    - Squamous epithelial cells with characteristic stripping caused by adherent *G. vaginalis*

- End Station #7
Fungal STD

- Vaginal candidiasis
Station #8
Vaginal Candidiasis

- **Etiological agent**
  - *Candida albicans*
  - Other *Candida* species may also be involved
  - Characterized as a yeast fungus
  - Present as normal flora
    - Overgrow during disruption of normal bacterial flora
      - Antibiotic usage
      - Pregnancy
      - Immune deficiency
      - Endocrine disturbance
Vaginal Candidiasis

**Transmission**

- **Endogenous**
  - From normal flora

- Transmission to male
  - Not common
  - May lead to urethritis or balanitis (inflammation of the penis)
Vaginal Candidiasis

- Clinical features
  - Thick milky vaginal discharge
  - Inflammation
  - Disease also called *vaginal thrush*
Vaginal Candidiasis

- **Laboratory diagnosis**
  - Specimen source
    - Vaginal secretions
  - Direct exam
    - Wet preparation or Gram stain
      - Budding yeast
      - Pseudohyphae (characteristic of *Candida* species)

Wet preparation, budding yeast mixed with squamous epithelial cells, OM 400x
Vaginal Candidiasis

- **Laboratory diagnosis**
  - Direct exam
    - Gram stain
    - Budding yeast with pseudohyphae

Gram stain, OM 400x
Vaginal Candidiasis

- **Laboratory diagnosis**
  - Culture
    - Sabouraud dextrose agar
      - Classic fungal media
    - Not generally necessary for diagnosis

SAB agar culture, OM 10x
Vaginal Candidiasis

- **Interesting facts**
  - *Candida albicans* is the most common cause of candidiasis
    - >85% of cases
  - Yeast infections are most frequently diagnosed by direct exam
    - Culture is time consuming and not necessary for patient management
Which unique characteristic identifies *Candida* species in clinical specimens?
- Presence of pseudohyphae within the budding cells

End Station #8
Parasitic STDs

- Trichomoniasis
- Crab lice infestation
Station #9
Trichomoniasis

- **Etiological agent**
  - *Trichomonas vaginalis*
    - Classified as a protozoal parasite
      - In contrast, the helminths are the true worms
    - Not normal flora
Trichomoniasis

Transmission
  - Direct contact to infected individual
  - Males
    - Most frequently asymptomatic
    - May however develop prostatitis
Trichomoniasis

- **Clinical features**
  - Not a reportable disease
    - Estimated that over 5 million females may be infected
  - Characteristics
    - Vaginal discharge
      - Described as greenish, frothy, and foul smelling
    - Intense vaginal and vulvar pruritus
Trichomoniasis

- **Laboratory diagnosis**
  - Differentiate from candidiasis and bacterial vaginosis
    - Patient management very different
  - **Direct exam**
    - Wet preparation
      - Vaginal and urethral discharge
Trichomoniasis

- **Direct exam**
  - Examine immediately after collection
    - Actively motile organism with movement of an undulating membrane

T. vaginalis → OM 100x
Trichomoniasis

- Laboratory diagnosis
  - Direct detection of the protozoan

Giemsa stain, trophozoite (name of the adult)
Trichomoniasis

**Interesting facts**

- *T. vaginalis* is one of the few parasites that can be cultured *in vitro*
  - Culture however, is not cost effective for patient care
  - Direct detection is reliable and simple

- “Trich” is frequently diagnosed on-site after collection
  - Rarely are samples submitted to the laboratory for testing

- *T. vaginalis* does not produce a cyst form
  - Only a trophozoite
Which structure of *Trichomonas vaginalis* is detected in genital secretions from an infected individual?

- Trophozoite form

End Station #9
Station #10
Crab Lice Infestation

- Etiological agent
  - *Phthirus pubis*
    - Referred to as the crab louse

- Classified as an arthropod “of the annoying” type
  - In contrast to those associated with disease transmission (vectors)
Crab Lice Infestation

Transmission
- Direct sexual contact with infected individual
- Males and females
  - Equally affected
Crab Lice Infestation

Clinical features

- Lice require blood for survival
  - When feeding an unexplained dermatitis occurs in the genital tract
    - Due to repeated feeding and chronic exposure to louse excreta
Crab Lice Infestation

- Laboratory diagnosis
  - Specimen
    - Pubic hair
  - Direct exam
    - Nits on the hair shaft
      - Known as larval eggs
Crab Lice Infestation

- **Laboratory diagnosis**
  - Adults
    - May be observed
      - on the skin
      - on cloths or
      - in the bed
  - Nits
    - On the hair
Crab Lice Infestation

- **Comparison**
  - Head lice
  - Body lice
  - Crab lice
- **All three lice are body site restricted**

*Pediculus humanus var. capitis*, head lice
Crab Lice Infestation

Interesting fact

- Lice infestation can occur by …..
  - Sharing infested clothing,
  - Exposure to infested bedding, or
  - By direct contact
Name the three species of lice associated with lice infestation.

- Body louse, *Pediculus humanus var corporis*
- Head louse, *Pediculus humanus var capitis*
- Crab louse, *Phthirius pubis*

End Station #10
Conclusion

- This completes the computer-assisted instruction for the sexually transmitted diseases.

- For questions or comments about this educational experience please e-mail
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