Infectious Encephalitis
with focus on findings from
California Encephalitis Project

Carol Glaser, DVM, MPVM, MD
Encephalitis and Special Investigations Section
Division of Communicable Disease Control
California Department of Public Health

Outline

- Background
  - Encephalitis
  - California Encephalitis Project (CEP)
- Case vignettes
  - Present as unknown
  - Highlight agent-specific findings
  - Lessons learned
- Approach to unknowns
  - Overall findings
  - Clinical Profiles
  - Powerpoint modified, different than handout

Encephalitis Background

- Severe CNS illness: significant morbidity & mortality
- Hospital and long term rehab costs
- Causative agent often not identified
- Frustrating for patient and family

Encephalitis - Viral

- Togavirus: EEE, VEE, WEE
- Flavivirus: SLE, WN, JV, Dengue
- Bunyaviruses: LaCrosse
- Paramyxoviridae: mumps, measles
- Arenaviruses: LCM, Machupo, etc.
- Enteroviruses: Polio, coxsackie, etc.
- Reoviruses: CTF
- Rhabdovirus: rabies
- Filoviridae: Ebola, Marburg
- Retroviridae: HIV
- Herpes: HSV1/2, VZV, EBV, CMV, HHV6
- Adenoviruses
Non-Viral Causes

- Rickettsia
- Bacteria
- Fungi
- Parasites
- Prions
- Non-infectious “mimickers”

With so many pathogens…. Where do you begin?

California Encephalitis Project (CEP)

- 1998 – present
- CDPH Viral and Rickettsial Disease Laboratory
- Funding from CDC Emerging Infections Program
- Cases referred from MDs throughout CA
  - not population-based (e.g., large sampling throughout CA)
  - biased toward more severe and diagnostically difficult cases
- Similar projects in TN, NY

Encephalitis Case Definition

- Hospitalized w/ encephalopathy (depressed or ALOC ≥24 hrs)
  - AND
  - 1 or more of the following:
    - fever (38°C)
    - seizure(s)
    - focal neurological findings
    - CSF pleocytosis
    - EEG findings c/w encephalitis
    - abnormal neuroimaging
- Exclusions: <6 months old or immunocompromised
Specimens

- CSF
- Acute serum
- Respiratory sample (NP/throat swab)
- Convalescent serum (10-14 days > acute serum)

CEP Testing Algorithm

- Molecular, serologic, isolation
- Multiple specimen types (CSF, sera, respiratory, brain if available)
- Core testing:
  - Arboviruses (WNV, SLE, WEE)
  - Herpesviruses (HSV1, HSV2, VZ, EBV, HHV6)
  - Enteroviruses
  - Respiratory viruses (Flu A/B, Paraflu 1-3, adenovirus, HMPV)
  - Mycoplasma pneumoniae
- Expanded testing - exposures, clinical symptomatology, laboratory

Expanded Testing

- Based on exposures, travel, symptomatology, lab values
- Examples:
  - Exposures
    - Foreign travel - other arbos, rabies
  - Laboratory values
    - Low WBC/platelets -- rickettsia
    - Eosinophilia -- parasites
Case Vignettes

Case 1

Adult male with fever, headache, nausea

43 y/o white male presented with headache and nausea for several days and fever of 102°F
- Diagnosed with viral meningitis

Several days later, presented with worsening symptoms, febrile to 103°F, AMS
- Admitted

Social and exposure history
- Firefighter
- Married with 3 healthy children
- 4 dogs
- Southern CA resident, no recent travel

Admit labs, neuroimaging
- LP #1: 300 WBC cells/mm³ (90% mono)
- Protein = 674 mg/dL
- Glucose = 42 mg/dL
- MRI #1: Normal
**Case 1**

- Hospital Day #3 – waxing and waning pattern and then became unresponsive
  - CT showed evidence of brainstem herniation
  - s/p R-sided ventriculostomy → improved

- A few days later - patient unarousable again
  - s/p L-frontal ventriculostomy

**Case 1 – Labs**

<table>
<thead>
<tr>
<th>Test</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>CSF bacterial Cx</td>
<td>Neg</td>
</tr>
<tr>
<td>CSF AFB smears</td>
<td>Neg</td>
</tr>
<tr>
<td>CSF fungal smear/Cx</td>
<td>Neg</td>
</tr>
<tr>
<td>CSF Enterovirus PCR</td>
<td>Neg</td>
</tr>
<tr>
<td>CSF HSV PCR</td>
<td>Neg</td>
</tr>
<tr>
<td>CSF N. mening Ag</td>
<td>Neg</td>
</tr>
<tr>
<td>CSF GBS Ag</td>
<td>Neg</td>
</tr>
<tr>
<td>CSF S. pneumonia Ag</td>
<td>Neg</td>
</tr>
<tr>
<td>CSF cytology</td>
<td>Neg</td>
</tr>
<tr>
<td>CSF Hib Ag</td>
<td>Neg</td>
</tr>
<tr>
<td>CSF viral Cx</td>
<td>Neg</td>
</tr>
<tr>
<td>Lyme serologies</td>
<td>Neg</td>
</tr>
<tr>
<td>CSF Lyme PCR</td>
<td>Neg</td>
</tr>
<tr>
<td>West Nile IgM ELISA</td>
<td>Neg</td>
</tr>
<tr>
<td>Cocci CF/ID</td>
<td>Neg</td>
</tr>
<tr>
<td>HIV RNA</td>
<td>Neg</td>
</tr>
<tr>
<td>PPD</td>
<td>Neg</td>
</tr>
</tbody>
</table>

**Case 1 – Readmission**

- Patient presents to hospital 3 days after discharge with similar symptoms as first admission
  - Diffuse headache
  - Nausea (significant weight loss)
  - Dizziness, diploplia
  - Afebrile

- LP:
  - WBC=280 (93%L, 7%PMNs)
  - Glucose= 56mg/dL
  - Protein=440 mg/dL

- Remained in hospital ....

- MRI brain HD#40 suspicious for fungal etiology

- Initiated anti-fungal Rx

- Some improvement and he was discharged ~ 6 weeks after admit (on fluconazole)
T2 MRI sequence demonstrates multifocal hyperintensities with mass effect in:

- Left frontal cortex
- Right caudate
- Left temporo-occipital lobe
- Splenium of corpus callosum

**Case 1 — CEP Contacted**

- Given the clinical course (several weeks), neuroimaging, and LP results....
  - Viral etiology--unlikely
  - Fungal etiology--possible
  - *Mycobacterium tuberculosis*--possible
  - *Balamuthia mandrillaris*--possible

**Case 1 — CEP Test Results**

- *Balamuthia mandrillaris* testing positive

**Balamuthia mandrillaris - Background**

- *Balamuthia* is a free-living amoeba
- Found in soil and water
- Worldwide distribution
- Inhalation or direct contamination of skin lesion
- Granulomatous encephalitis - affects immunocompetent and immunocompromised
**Balamuthia Encephalitis**

- Initially believed to be “very rare”; likely not-so-rare but under-diagnosed

**CEP’s Balamuthia Cases**

- 12 cases reported in CA since 1990, 9 deaths
- Differential-
  - Tumor/lymphoma
  - Mycobacterium TB
  - Neuro-cysticercosis, fungal
  - Atypical ADEM
- Age range: 1-72 yrs (median 12 years)
- 82% Hispanic
- 91% Male
- LP (Median): 171 WBC cells/mm³, Protein 131mg/dL, Glucose 39 mg/dl
- Neuro-imaging—abnormal (e.g. mass lesions, hydrocephalus)

**CEP Balamuthia lessons learned**

- More common than previously recognized
- Generally the LP formula is similar to fungal or Mycobacteria tuberculosis
- Most cases fatal, but may be treatable

**Case 2**

Elderly male with fever, emesis, AMS
**Case 2**

- 70 year old male presented with:
  - 4 day history of severe diarrhea
  - 2 day history of AMS and fever
  - Native of India, visiting family in California
  - “Occupational and residential exposure to TB”
- Admitted to ICU

**Case 2**

- Hospital Labs:
  - LP:
    - WBCs 84 cells/mm3 (90% PMNs)
    - Protein 79 mg/dL
    - Glucose 106 mg/dL
  - Brain MRI: Normal
  - EEG: Diffuse slowing

**Case 2 - CEP results**

- CSF PCR
  - Herpes consensus: NEG
  - Mycoplasma: NEG
  - Enterovirus: NEG
  - West Nile: NEG
- Serology
  - WEE, Mycoplasma, Chlamydia, adenovirus, EBV: NEG
- Respiratory PCR:
  - Influenza A/B, adenovirus, Mycoplasma, enterovirus: NEG
  - Human metapneumovirus: NEG

**Case 2 – CEP Results**

- West Nile virus (WNV) testing
  - WNV serology (IgG and IgM): POSITIVE
**WNV Brief Background**

- Emerged in NY in 1999; 1st time in Western hemisphere
- Bird/mosquito cycle
- Most infections asymptomatic
- Symptomatic infections: febrile illness, aseptic meningitis, encephalitis
- Serology cross-reacts with other flaviviruses

**WNV in CEP**

- 54 cases enrolled in CEP
  - Median age: 62 years (range 3 – 90)
  - 63% male
  - 96% with fever
  - Most occur late summer/early fall
  - All had abnormal LP
  - Most positive serology, negative PCR
  
  (Most WNV cases enrolled through WNV Surveillance Project rather than CEP)

**WNV Human Infection “Iceberg”**

- 1 CNS disease case = ~150 total infections
- ~20% “West Nile Fever”
- ~80% Asymptomatic
- ~1% CNS disease
- <1% fatal (~0.1% of total infections)
- Very crude estimates

**CEP WNV lessons learned**

- Serology is best test
- PCR may have a role in immunocompromised hosts
Case 3

Elderly male with fever, confusion, tremors

• 70 year old Asian male admitted with rapidly progressive dementia
  • Family reported cognitive decline for past several weeks
  • On admission he was febrile, complained of blurry vision, very confused, word finding difficulty, tremors

Exposure history
  • No animal contacts
  • No ill contacts
  • Regularly visits the Philippines (including a few weeks prior to onset)
  • No known mosquito bites

Admit labs, neuroimaging
  • Lumbar puncture
    ✓ 12 WBCs/mm³
    ✓ Protein 137 mg/dL
    ✓ Glucose 52 mg/dL
  • MRI: multiple focal lesions in temporal and parietal lobes

Case 3 - CEP results

• CSF PCR
  • HSV1, HSV2: Negative
  • VZ: Negative
  • Mycoplasma: Negative
  • Enterovirus: Negative

• Serology:
  • SLE, WEE, Mycoplasma, Chlamydia, adenovirus, EBV: Not significant

• Respiratory sample-not received
Case 3

- Conclusions: unlikely infectious...CEP testing completed (we thought...)
- But later the hospital contacted us to tell us...

Brain biopsy: “florid meningoencephalitis”

Case 3*

- Paraffin blocks sent to CEP:
  - Special extraction method from paraffin blocks*
  - Positive for enterovirus by PCR
- Also sent to CDC:
  - Positive for enterovirus by PCR

Enterovirus (EV) Brief background

- Leading cause of viral meningitis, especially pediatrics
- Role in encephalitis was previously unknown

  - preferred specimen is frozen tissue, not formalin/paraffin blocks
**CEP Enterovirus Experience**

- EV patients make up 26% of all confirmed or probable cases; ~5% of all CEP cases*
- 98 patients confirmed by PCR in CSF
- 57 considered possible or probable (found outside CNS)
- Median age = 14.0 years (mean=20.5 yrs)
- 63% pediatric, **37% adult**
- CSF EV PCR good, but...not perfect

*Fowikes AL et al., JID, 2008

**CEP Enterovirus Lessons Learned**

- EV usually present in CSF only transiently, therefore time period detected in CSF is brief
- Important to examine for enterovirus in multiple specimens:
  - Brain biopsy if possible
  - Throat/Nasopharyngeal
  - Stool
- Suspect we are missing cases due to unsuitable or incomplete specimens

**Case 4**

Elderly male with stroke vs. encephalitis

- 72 year old male admitted with 5 day history of fever, lethargy, somnolence
- On admission, noted to be very lethargic and had focal R-sided weakness
- Clinician thought most likely a stroke but wanted to r/o encephalitis
**Case 4**

- **Exposure history**
  - Born and raised in Philippines
  - Lived in U.S. for 10 years
  - 10 months PTA visited Philippines
  - No known mosquito bites/animal exposures
- **Admit labs, neuroimaging**
  - Lumbar puncture
    - 10 WBCs/mm³
    - Protein = 172 mg/dL
    - Glucose = 60 mg/dL
  - MRI and CT: mild atrophy

**Case 4 - CEP results**

- **CSF PCR**
  - Herpes consensus: NEG
  - *Mycoplasma*: NEG
  - Enterovirus: NEG
- **Serology**
- **Respiratory PCR:**
  - Influenza A/B, adenovirus, *Mycoplasma*, enterovirus: NEG

**Case 4**

- **Patient died 11 days after hospitalization**
- **No autopsy done**
- **Death certificate: “Cerebral vascular accident”**

**Case 4***

- **CEP:** Deceased core testing includes rabies:
  - Rabies antibody POSITIVE
  - Rabies CSF PCR Negative
  - Respiratory (w/saliva) specimen: PCR+/sequenced as Philippine canine strain

* CDC, MMWR, Human Rabies summary 2010
**Rabies**

**Brief Background**

- Acute, rapidly progressive encephalitis
  - 70% furious form
  - 30% paralytic form (as in this case)
- Variable incubation: few weeks or months – years
- Highest fatality of all infectious diseases
- Worldwide: estimated 100,000 cases/year
- Rare in U.S.: 2-3 cases/year
  - 2000-2009: 30 human cases in the US
  - 8 imported (mostly canine)
  - 22 endemic cases – primarily bat associated
- 30% paralytic form (as in this case)
- Highest fatality of all infectious diseases
- Worldwide: estimated 100,000 cases/year
- Rare in U.S.: 2-3 cases/year
  - 2000-2009: 30 human cases in the US
  - 8 imported (mostly canine)
  - 22 endemic cases – primarily bat associated

**Rabies**

**CEP experience**

- 6 cases total
  - All male
  - Ages:
    - 16 years (2008)
    - 8 years (2006)
    - 66 years (2003)
    - 28 years (2002)
    - 72 years (2001)
    - 49 years (2000)
  - 2 associated with dog strain (Philippines)
  - 4 with bat strain

**Rabies**

**Lessons Learned**

- Rabies testing should be done on all severe, rapidly progressive encephalitis - *even if no animal bite history*
- Ante-mortem testing is possible but tricky
- Multiple assays and multiple specimen types needed
Case 5

Elderly male with headache, confusion, fever

72 year old male presents with 5-day history of headache, confusion, ongoing fevers

- Admission PE: Significant only for intermittent confusion and high blood pressure
- PMHx: Unremarkable except for remote history of anti-tuberculosis therapy for 2 months

Case 5

- LP#1:
  - WBC 106 cells/mm³ (88%L) -
  - Protein 278 mg/dL -
  - Glucose 22 mg/dL -
- 2nd LP 1 week later –very similar to LP#1

Case 5

Other hospital testing:
- Chest X-Ray: Normal
- Cranial CT: Normal
- Cranial MRI: Small lesion suggestive of acute small vessel ischemia
- TB skin test (TST) results 0 mm in duration

Case 5

- CEP testing:
  - Arboviruses, *Mycoplasma* – negative by serology/PCR
  - Herpes viruses (including HSV, VZV, etc.), Enterovirus – negative by PCR
  - *Balamuthia, Acanthamoeba* – negative by serology

Case 5

- Outside testing:
  - CSF TB PCR: NEG
  - CSF MTB smear: NEG
  - Sputum MTB smear: NEG
Case 5

A few weeks later....

- CSF MTB culture: POSITIVE
- Sputum MTB culture: POSITIVE

CNS Mycobacterium tuberculosis

Brief background

- Accounts for 5-6% extrapulmonary TB in U.S.
- Peak age is 6 months - 4 yrs
- CNS TB often described as a meningitis, often with insidious onset
- Clinical symptoms typically include fever, headache, vomiting, neck stiffness, cranial nerve palsy
- MRI/CT characterized by basilar involvement, infarcts, hydrocephalus
  - Thwaites G et al., J Neurol Neurosurg Psych 2000

Tuberculosis CEP Summary

- 34 cases in CEP from 1998-2010
- Present rapidly – median 6 days from onset to presentation (range 0-62 days)
- Median age 29 yrs (8 mon – 77 yrs)
- Predominantly non-White: 50% Hispanic, 38% Asian
  - Christie LJ, Emerg Infect Dis 2008 (first 20 cases)

Tuberculosis CEP summary (n=34 cases)

- Positive CSF TB culture 100%
- Positive CSF TB PCR 25%
- Positive respiratory TB culture 63%
- Abn CXR 45%
- Foreign travel 58%
- Positive PPD 37%
**CEP TB Lessons Learned**

- CNS TB:
  - Found in any age
  - Can be rapidly progressive
  - Encephalopathic changes
- Consider TB if CSF WBC moderately high, protein high, glucose low
  - Calculate CSF/serum ratio for diabetics
- Probe for possible risk factors when taking history (foreign birth, travel, previous TB, TB contacts, etc.)
- Suspect more cases that weren’t diagnosed - how many other MTB cases do we have where the CSF didn’t grow?

**Take home messages**

- Case 1-Balamuthia mandrillis more common than originally thought
- Case 2-West Nile virus-preferred is serology not CSF PCR
- Case 3-Enterovirus is most common in pediatrics but does occur in adults, important not to rely on CSF PCR alone
- Case 4-Rabies is rare however important to consider and can resemble ‘stroke’ syndrome
- Case 5-MTB CNS can be fulminant, don’t rely on PPD or CSF PCR tests

**CEP Overall**

- A negative chest x-ray or TST does not r/o TB
- Consider empiric TB therapy if high suspicion even in face of negative CSF/sputum AFB smear or TB PCR to reduce morbidity/mortality
  - TB PCR is not very sensitive
- Respiratory AFB smears/cultures can be helpful
CEP: Overall Numbers

- 3,466 enrolled (~5000 referred)
- Age, race breakdown
  - Median age: 19 years (range: 6 months – 92 years)
  - 1258 (36%) White
  - 1010 (29%) Hispanic
  - 363 (10%) Asian/Pacific Islander
  - 310 (9%) African American
  - Fairly representative of CA population

CEP Referral and Testing 1998-2010

- Of ~3466 patients
  - 15% with confirmed/probable etiology
  - 15% with possible etiology
  - 12% with non-infectious etiology
  - 58% with unknown etiology

Breakdown of Agents found in CEP Patients

- 59% Viral
- 29% Bacterial
- 6% Bacterial & Viral
- 3% Prion
- 1% Parasitic
- 1% Fungal

Agents Identified in CEP

- Respiratory Viruses 22%
- Enterovirus 25%
- West Nile Virus 5%
- HSV1 11%
- EBV 5%
- E. coli 2%
- RSV 4%
- Coxsackievirus 1%
- Chlamydia 1%
- Salmonella 1%
- Measles 1%
- HHV6 4%
- HSV2 1%
- Measles SSPE 1%
- VZV 1%
- HHV8 1%
- HHV7 1%
- HHV6 1%
- HHV7 1%
- HHV1 1%

Excludes <1% each of Acute HIV, Kayawara, Brucella, Cryptococcus, Cytomegalovirus, Mycoplasmas, EBV, and Epstein-Barr virus.

Respiratory viruses include Rhinovirus, Adenovirus, Parainfluenzas Type 1-4, and coinfections with these agents.
Clinical Profiles

- What about the unknowns?
- Although all cases meet a common case definition, very heterogeneous with respect to presentation, clinical manifestations, outcome
- Subset of cases that look similar are grouped together into “Profiles”

Summary of Profiles

- Profile 1: diffuse generalized edema
- Profile 2: temporal lobe
- Profile 3: intractable seizures (barbiturate coma)
- Profile 4: seizures, rapid recovery
- Profile 5: cerebellar involvement
- Profile 6: movement disorders
- Profile 7: psychiatric component
- Profile 8: recurrent
- Profile 9: miscellaneous
- Profile 10: dementia

Summary

- Diagnosis remains major challenge
- Important to know what assay to do for each disease
- Limitations to CSF/molecular testing
- Important to use combination of assays

- Glaser C, Clin Inf Dis, 2009

Acknowledgements

- Viral and Rickettsial Disease Laboratory Branch
- Centers for Disease Control
- California physicians