SEPSIS AND THE HEMATOLOGY LABORATORY

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Chief medical officer

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PRESENTATION OUTLINE

› Introduction to sepsis epidemiology and formal definitions
› Biological considerations and how they impact the role of the laboratory
› Differential diagnosis to be considered by the hematology laboratory
› Recently proposed markers for sepsis management and their limitations

SEPSIS AND ITS IMPACT TO SOCIETY

Today

Future

Incidence projected to increase by 1.5% per year

>750,000 cases of severe sepsis/year in the U.S.

COMPARABLE GLOBAL EPIDEMIOLOGY

› 95 cases per 100,000
  • 2 week surveillance
  • 206 French ICUs

› 95 cases per 100,000
  • 3 month survey
  • 23 Australian/New Zealand ICUs

› 51 cases per 100,000
  • England, Wales and Northern Ireland

COMPARISON WITH OTHER MAJOR DISEASES

Incidence of Severe Sepsis

Mortality of Severe Sepsis

EMERGENCY DEPARTMENT CRITICAL CARE VOLUME INCREASES

› 102 million national ED visits in 1999
› 17% (17.5 million) "immediately life threatening" 1
› 57 California emergency departments (1990-1999) 2
› 50% (387,616) severe sepsis cases initially go to ED

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LOW INCIDENCE BUT SIGNIFICANT CONSEQUENCES

- 0.5-1% live births
- 50% NICU admissions
- 20-40% mortality rate

CURRENT IMPACT IN THE UNITED STATES

- Up to 50,000 neonates suffering from sepsis
- 10,000-20,000 neonatal deaths
- Incidence and mortality declining in developed nations but not altered in developing world

PATHOGENESIS–EARLY ONSET SEPSIS

- Vertical transmission
  - Vaginal delivery, bacteria colonizing the birth canal
  - Ascending chorioamnionitis prior to delivery
- Chorioamnionitis: 4-fold increase in the risk of neonatal sepsis (1 to 4%)
- Most important pathogen: group B streptococci (GBS)

CLINICAL FINDINGS IN NEONATAL SEPSIS

- Subtle and non-specific, thus requiring a very high level of suspicion for prompt diagnosis and treatment
- Diagnosis should take into account possible risk factors
- Any deviation from an infant’s usual pattern of activity or feeding should raise the suspicion of sepsis

CLINICAL FINDINGS

<table>
<thead>
<tr>
<th>Finding</th>
<th>Frequency</th>
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<tbody>
<tr>
<td>Hyperthermia</td>
<td>+++</td>
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<tr>
<td>Respiratory distress</td>
<td>+</td>
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<td>Anorexia</td>
<td>++</td>
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<td>Vomiting</td>
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<td>Jaundice</td>
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<td>Hepatosplenomegaly</td>
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<td>Lethargy</td>
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<td>Cyanosis</td>
<td>+</td>
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<tr>
<td>Hypothermia</td>
<td>+</td>
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<tr>
<td>Intractability</td>
<td>+</td>
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<tr>
<td>Apnea</td>
<td>+</td>
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<tr>
<td>Abdominal distention</td>
<td>+</td>
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<tr>
<td>Diarrhea</td>
<td>+</td>
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</table>
RISK FACTORS

› Intrapartum maternal temperature ≥38°C (100.4°F)
› Delivery at <37 weeks gestation
› Chorioamnionitis
› Five-minute Apgar score ≤6
› Evidence of fetal distress
› Maternal GBS colonization*
› Membrane rupture ≥18 hours

*Maternal intrapartum antibiotic prophylaxis (IAP) reduces the risk of GBS infection.

LABORATORY EVALUATION

› Blood culture
› CBC with differential
› Lumbar puncture
› Culture of urine and other fluids
› Other inflammatory markers
  • C-reactive protein
  • Cytokines (IL 2, 4, 6, 10, TNF, interferon gamma)
  • Procalcitonin

THE LIMITATIONS OF EACH TEST

› Blood Culture: gold standard, but sensitivity can vary depending on volume of blood and number of bottles
  • Cytokines (IL 2, 4, 6, 10, TNF, interferon gamma)
  • Procalcitonin
› CBC with Differential
  • Poor sensitivity
  • Band counts are very cumbersome for the lab
› Other cultures and lumbar puncture
  • Less than ideal sensitivity

NEW INFLAMMATORY MARKERS FOR SEPSIS

› C-Reactive Protein
  • Poor specificity
› Cytokines (IL 2, 4, 6, 10, TNF, interferon gamma) procalcitonin
  • Both options very expensive and have sub-optimal sensitivity and specificity
› All tests above require additional sample and additional costs

SEPSIS DEFINITIONS

› Sepsis: a clinical syndrome that complicates severe infection...characterized by the cardinal signs of inflammation (vasodilation, leukocyte accumulation and increased microvascular permeability) occurring in tissues that are remote from the infection
› Systemic inflammatory response syndrome (SIRS): an identical clinical syndrome that complicates a noninfectious insult (e.g., acute pancreatitis, pulmonary contusion)
› Both: dysregulation of the inflammatory response with massive and uncontrolled release of proinflammatory mediators initiating a chain of events that lead to widespread tissue injury
SEPSIS DEFINITIONS

› Dysregulation of the inflammatory response with massive and uncontrolled release of pro-inflammatory mediators initiating a chain of events that lead to widespread tissue injury
› Measuring these inflammatory mediators does not provide much help in the discrimination between the two possible causes of this process (infectious sepsis, versus non-infectious SIRS)

USING THE RIGHT DEFINITIONS

› Infection: the invasion of normally sterile tissue by organisms
› Bacteremia: the presence of viable bacteria in the blood
› Systemic inflammatory response syndrome (SIRS): the clinical syndrome that results from a dysregulated inflammatory response to a noninfectious insult, such as an autoimmune disorder, pancreatitis, vasculitis, thromboembolism, burns or surgery

USING THE RIGHT DEFINITIONS

› Sepsis: the clinical syndrome that results from a dysregulated inflammatory response to an infection. Sepsis is defined as the presence (probable or documented) of infection together with systemic manifestations of infection
› Severe sepsis: sepsis-induced tissue hypoperfusion or organ dysfunction due to the infection
› Septic shock: sepsis-induced hypotension persisting despite adequate fluid resuscitation, which may be defined as infusion of 30 mL/kg of crystalloids (a portion of this may be albumin equivalent)

CRITERIA FOR ORGAN DYSFUNCTION NEEDED TO DEFINE SEVERE SEPSIS

› Sepsis-induced hypotension
› Lactate above upper limits of laboratory normal
› Urine output <0.5 mL/kg/hr for more than two hours despite adequate fluid resuscitation
› Acute lung injury with PaO2/FIO2 <250 in the absence of pneumonia as infection source
› Acute lung injury with PaO2/FIO2 <200 in the presence of pneumonia as infection source
› Creatinine >2 mg/dL (176.8 micromol/L)
› Bilirubin >2 mg/dL (34.2 micromol/L)
› Platelet count <100,000 microL–1
› Coagulopathy (INR >1.5)

CRITERIA FOR SYSTEMIC INFLAMATION (EITHER SEPSIS OR SIRS)

› Temperature >38.3 or <36°C
› Heart rate >90 beats/min or more than two standard deviations above the normal value for age
› Tachypnea, respiratory rate >20 breaths/min
› Altered mental status
› Significant edema or positive fluid balance (>20 mL/kg over 24 hours)
› Hyperglycemia (plasma glucose >140 mg/dL or 7.7 mmol/L) in the absence of diabetes
› Arterial hypotension (systolic blood pressure SBP <90 mmHg, MAP <70 mmHg, or an SBP decrease >40 mmHg in adults or less than two standard deviations below normal for age)
THE CLINICAL LABORATORY IN THE MANAGEMENT OF SEPSIS

› Clinical aspects of sepsis care where lab tests may be used
  • Screening
  • Confirming diagnosis
  • Ruling out diagnosis
  • Monitoring therapy
  • Determining prognosis

› New lab tests that address any of these needs will have to be assessed for patient care and for health economics

THE CLINICAL LABORATORY IN THE MANAGEMENT OF SEPSIS

› Key challenges
  • Early diagnosis with better sensitivity than current tests
  • Discrimination between infection and other causes of SIRS
  • Rule out ("EKG for sepsis"), but need ~100% NPV
  • Monitoring antibiotic therapy (procalcitonin)
  • Discriminate bacterial from viral infection

› Lab tests for these uses less likely to impact the current standard of care
  • Confirming diagnosis—treatment starts regardless, gold standard for confirmation is the culture
  • Prognostication—therapy will be aggressive regardless

INFECTION—THE PRESENCE OF VIABLE BACTERIA IN A STERILE BODY SITE

What happens?
› Step 1: Macrophages phagocyte bacteria
› Step 2: Macrophages RELEASE CYTOKINES
› Step 3: Cytokines activate circulating WBC (neutrophils)
› Step 4: Cytokines stimulate bone marrow
› Step 5: Released granulocytes must stay in the circulation and reach the site of infection
› Step 6: Bone marrow releases more granulocytes in blood, some of which are immature (left shift)

STEPS 1 AND 2—MACROPHAGE PHAGOCYTOSIS AND CYTOKINE RELEASE

› STEP 1 TESTS: blood culture and gram stain
› STEP 2 TESTS: CRP, ESR, IL-6

Neutrophil stimulus: toxic granules contain toxic enzymes that kill bacteria opsonized by Ab
Bone marrow stimulus: more granulocytes released in circulation

STEPS 3 AND 4—MORPHOLOGIC CHANGES DETECTABLE AT THIS STAGE—MICROSCOPIC REVIEW
THERE IS A NEED FOR NEW SEPSIS MARKERS

- Current tests routinely ordered in practice, such as the CBC-diff, have very limited sensitivity and specificity
- The changes expected to be seen in sepsis may be absent in a large proportion of cases
- The same changes may be present in a series of other conditions
- The left shift, neutrophilia and leucocytosis expected to be seen in bacterial infection including sepsis, are in fact markers of inflammation
- Other inflammatory conditions, ranging from SIRS to a simple surgery, also lead to these changes

OVER THE LAST DECADE, SEVERAL SEPSIS BIOMARKERS HAVE BEEN PROPOSED

- Review of 3,370 articles discussing 178 biomarkers
- Different clinical applications, including diagnosis, discrimination from SIRS, and prognosis

### Table 1: Cytokine/chemokine biomarkers identified in the literature search (with some selected references)

<table>
<thead>
<tr>
<th>Biomarker</th>
<th>Evaluated in experimental studies</th>
<th>Evaluated in clinical studies</th>
<th>Evaluated as a prognostic factor</th>
<th>Comment</th>
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### Table 2: Cell marker biomarkers identified in the literature search (with some selected references)

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Increased Bands
- This is what we use today to diagnose infection in the CBC-DIFF
- The last steps in the entire process!
Predicted development of septic shock than with vascular endothelial damage biomarkers distinguished between survivors and non-surgical patients. Increased in sepsis compared with healthy controls, correlated with APACHE II score. For neonates, additional blood required. Combinations of several biomarkers, but this requires further evaluation. A single ideal biomarker will ever be found. A combination of several sepsis biomarkers may be more effective, but this requires further evaluation.

All these biological events are associated with a dysregulated inflammatory process. Coagulation biomarkers, vascular endothelial damage biomarkers, vasodilation biomarkers, and organ dysfunction biomarkers are increased in sepsis compared with healthy controls. Acute phase protein biomarkers are increased in sepsis compared with healthy controls, correlated with APACHE II score. For neonates, additional blood required. Combinations of several biomarkers, but this requires further evaluation. A single ideal biomarker will ever be found. A combination of several sepsis biomarkers may be more effective, but this requires further evaluation.

Critical clinical needs a sepsis biomarker should address:

- Earlier diagnosis of sepsis—information would have to be available as part of the routine workup of an ICU or ED patient
- Higher specificity for infection, lower sensitivity to the inflammatory process
- Indication of etiology (gram positive versus gram negative, versus viral or fungal)

Conclusions:

Our literature review indicates that there are many bio-markers that can be used in sepsis, but none has sufficient specificity or sensitivity to be routinely employed in clinical practice. PCT and CRP have been most widely used, but even these have limited abilities to distinguish sepsis from other inflammatory conditions or to predict outcome. In view of the complexity of the sepsis response, it is unlikely that a single ideal biomarker will ever be found. A combination of several sepsis biomarkers may be more effective, but this requires further evaluation.

Key messages:

- More than 170 different biomarkers have been assessed for potential use in sepsis, more for prognosis than for diagnosis
- None has sufficient specificity or sensitivity to be routinely employed in clinical practice
- Combinations of several biomarkers, but this requires further evaluation.

Additional types of sepsis biomarkers:

- Coagulation biomarkers
- Vascular endothelial damage biomarkers
- Vasodilation biomarkers
- Organ dysfunction biomarkers
- Acute phase protein biomarkers

Proposed sepsis biomarkers—practical considerations:

- Added cost to an already constricted health economics environment
- Not part of routine care; thus will only be ordered upon clinical suspicion—diagnosis already made at this point
- For neonates, additional blood required

Critical clinical needs a sepsis biomarker should address:

- Earlier diagnosis of sepsis—information would have to be available as part of the routine workup of an ICU or ED patient
- Higher specificity for infection, lower sensitivity to the inflammatory process
- Indication of etiology (gram positive versus gram negative, versus viral or fungal)
THANK YOU!