In the name of God

Protocol of

the Diagnostic Evaluation of Infection

in Liver Transplant Recipients

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February 2015

Preface

Infections remain a constant threat to all liver transplant recipients, and the incidence of them after liver transplantation (LT) is generally higher than that after other types of solid-organ transplantation. At present, infectious complications are reported in 35% to 68% of liver transplantations. The febrile LT recipient (or even an afebrile patient in whom infection is suspected) presents one of the most challenging diagnostic and management situations facing clinicians. Diagnoses must be made accurately, evaluations must be rapidly performed, and management (often empiric) must be effective. The complexity of host immune and metabolic factors, pharmacokinetic issues (e.g., drug interactions), a broad range of opportunistic pathogens and processes, difficulties in clinical presentations, and uncertainties in evaluation and therapeutic decision-making constitute the major related issues.

Clinicians should adopt a syndromic approach and narrow the differential diagnose. In doing so, the number of causative agents is narrowed, e.g., in nonspecific febrile illnesses, pneumonia, urinary tract and CNS infections.

Taking into account the multi-faceted background , this protocol for the diagnostic evaluation of infections in LT recipients was developed. It is hoped that the protocol is able to address some of these issues and meet the needs of the clinicians involved in managing the LT recipients in transplantation wards of Shiraz University of Medical Sciences. The protocol draws upon the main points from the article entitled “Infections After Solid Organ Transplantation” by Asim A Jani and Ron Shapiro , published by Medscape on December 31, 2014 and is accessible on the following website: http://emedicine.medscape.com/article/430550-overview#aw2aab6b9

I have mainly added the section “Diagnostic evaluation of fever (or suspicious to infection), irrespective of presence or absence of localizing site”. Also, I have updated the content according to “CDC/NHSN Surveillance Definitions for Specific Types of Infections. January 2015. http://www.cdc.gov/nhsn/PDFs/pscManual/17pscNosInfDef_current.pdf.

I acknowledge Dr Bahman Pourabbas, Dr Parisa Badiee, Dr Jalal Mardaneh, Dr Fereshteh Fani, Dr Tayebeh Hashempour and Dr Marziyeh Jamali Doust for their invaluable comments. My thanks also go to Dr Zahra Jafarpour for her help with the organization of the protocol and finally I appreciate Hassan Khajehei for linguistic editing of the content. This guideline was revised and published in Professor Alborzi Clinical Microbiology Research Center in cooperation with Shiraz Transplant Research Center.

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Preface

Introduction

Infections remain a constant threat to all LT recipients, and the incidence of infections after liver transplantation is generally higher than that following other types of solid-organ transplantation (1, 2). This increased incidence is likely related to the technical complexity of the liver-transplantation procedure itself, its performance in a potentially contaminated environment within the abdominal cavity, and the extremely poor medical condition of many recipients, altogether contributing to bacterial infections which remain an important cause of death post liver transplantation (3).

At present, infectious complications have been reported in 35% to 68% of liver transplantations(4-8). Most infections occur within the first 30 days after transplantation and are primarily surgical complications, nosocomial in origin, or, rarely, reactivation of latent infections in the recipients (2).

The febrile LT recipients (or even an afebrile one in whom infection is suspected) present one of the most challenging diagnostic and management situations facing clinicians. Diagnoses must be made accurately, evaluations must be rapidly performed, and management (often empiric) must be effective. The complexity of host immune and metabolic factors, pharmacokinetic issues (e.g., drug interactions), a broad range of opportunistic pathogens and processes, difficulties in clinical presentations, and uncertainties in evaluation and therapeutic decision-making are the major issues. Nevertheless, a logical framework that provides guidance to clinicians is possible (9).

A strategic approach to fever (and associated organ system–related manifestations) in the LT recipient includes the following (9):

1. Vigilance to overall temporal pattern of post-LT infections
2. Careful evaluation of organ-specific considerations
3. Recognition of specific associations (This can be particularly useful in LT recipients who present with nonspecific febrile illnesses).
4. Conducting a thorough clinical assessment using the CREDIT mnemonic, with special attention to the clues in patient's history and physical examination. This framework will hopefully enable the clinician to relate organism category (bacteria, viruses, fungi, parasites) to each of these 6 potential sources of exposure and infection. They include:

   C - Community-acquired

   R - Reactivation

   E - Epidemiologic exposure

   D - Donor-derived infections

   I - Iatrogenic considerations

   T - Travel consideration

5. Following a tailored initial diagnostic evaluation

2. Specific considerations in the evaluation of a febrile LT recipient (9)

-To review the timeframe around specific infections occurring after transplantation.

-To keep in mind that the clock can get reset based on interim rejection episodes or major immunosuppression due to regimen changes .

-To think syndromically and narrow the differential diagnoses:

-To narrow the differential diagnoses of possible organisms that could cause the clinical presentation(s) according to syndromical approach. e.g, nonspecific febrile illness, pneumonia, urinary tract and CNS infections.

-To consider that history-taking in relation to infectious disease has the major significance. All the right questions can be asked according to an initial syndromic differential diagnosis .

-To perform the physical examination as an important tool that should be thorough and timely, and it may be necessary to repeat the examination often, hour by hour or day by day, depending on the pace and severity of illness. This is even more important given the finding that diagnostic errors are associated with length of stays in, for example, the intensive care unit, a setting in which too little attention is often paid to the physical examination. Ideally, information from the examination should be derived directly rather than from other team members. Data suggest that two thirds of missed diagnoses in the ICU in one study were infectious in origin.

- To remain aware of the specific types of infections associated with liver transplantations :

-Surgical wound infections, bloodstream infections, and pneumonia can all result from the usual healthcare-associated pathogens in the first month following transplantation. LT

recipients may also be at risk for anastomotic leaks, wound infections, intra-abdominal abscesses, and bacteremia from a host of enteric organisms in addition to the risks of healthcare-associated infections in the postoperative period .

-To consider several organisms in recipients who present with an occult fever pattern unaccompanied by specific organ-based symptoms. Most significant organisms (common and uncommon) observed in liver transplant recipients could cause presentations including bacterial infections such as *Bartonella*-associated bacillary angiomatosis; endocarditis; tuberculosis, deep-seated *Nocardia* infection; *herpes simplex virus, EBV, CMV, and HHV-6* infections; fungal complications such as *Aspergillus* syndromes; or even parasitic infections including visceral leishmaniasis. Noninfectious conditions such as rejection, PTLD, or drug fever should also be considered.

3- A methodical approach (9)**:**

-Assessment of the vital signs, including observing any pulse-temperature dissociation (indicative of noninfectious processes, endocrine illness, or intracellular pathogens .(

-A complete assessment of mucosal surfaces, including the oral cavity, should be performed. Many oral lesions, including dental and gum health, provide insight into likely etiologies and nutritional status. Inflammatory signs may be blunted given the net state of immunosuppression, as in the case of the neutropenic hosts.

-A search for invasive devices and evidence of recent procedures is advisable .

- The presence or absence of a cardiac murmur and peripheral stigmata of endovascular infection need to be determined .

-The abdominal examination is considered critical; evidence of surgical leaks, intraabdominal processes, and peritonitis can be lifesaving .

- Consider performing pupillary dilation as part of a complete retinal examination, as it may offer clues to a disseminated disease process or increased intracranial pressure associated with a CNS syndrome .

- As in other immunocompromised hosts, the skin examination is paramount, since many infections leave a dermal footprint through many different types of lesions, including macules, papules, erythema patterns, fluctuance, petechiae, and eschars that correspond to primary infections or secondary complications from dissemination .

4- Diagnostic evaluation of infections in LT recipients

4-A. Fever (or suspicious to infection) irrespective of presence or absence of localizing site

1. Urinalysis and urine culture
2. Chest radiography
3. Blood cultures (two sets for confirming bacteremia, one set should be anaerobic and one set from central vein catheter)
4. Peripheral white blood cell count
5. Liver function tests
6. C-reactive protein (CRP)
7. Venous or arterial blood gas, serum lactate (if available)
8. Abdominopelvic ultrasonography
9. Quantitative PCR for CMV
10. Analysis of fluid obtained from intra-abdominal catheter (if present)
    a. Cell count and differential
    b. Glucose, protein and lactate dehydrogenase (LDH)
    c. Gram stain and bacterial cultures(aerobic and anaerobic)

- Direct inoculation of routine blood culture bottles at the bedside with 10 mL of ascitic fluid is recommended

d. KOH test, fungal culture and PCR for *candida*

11. Purified protein derivative (PPD) or QuantiFERON testing using interferon gamma assays if not done before transplantation.

4-B. pulmonary infiltrates (alveolar pattern) (9)

The recommended initial diagnostic evaluation for pulmonary infiltrates (alveolar pattern) is as follows:

1. Sputum Gram stain and culture

    a. Note: Neutrophils and squamous epithelial cells per low power field should be reported. Purulent sputum is defined as secretions from the lungs, bronchi, or trachea that contain ≥25 neutrophils and ≤10 squamous epithelial cells per low power field (x100)

2. Urine Legionella and pneumococcal antigens (if available)

3. Sputum acid-fast bacillus (AFB) smear, culture and PCR for *mycobacteria*

4. Bronchoscopy if fever and infiltrates persist (send specimens as interstitial pattern)

4-C. Pulmonary infiltrates (interstitial pattern)

The recommended initial diagnostic evaluation for pulmonary infiltrates (interstitial pattern) is as follows:

1. Workup for pulmonary infiltrates (alveolar pattern)
2. Bronchoscopy with transbronchial biopsy if fever and infiltrates persist
3. Bronchoalveolar lavage (BAL) fluid for:
    a. Gram stain and Bacterial culture
    b. Viral culture and PCR (if available)
    c. KOH test, fungal culture and PCR for *candida*, *aspergillus*, *mucur* and *cryptosporidium*
    d. AFB stains, mycobacterial culture and PCR for *mycobacteria*
    e. Direct fluorescent antibody (DFA) and culture for Legionella (if available)
    f. DFA or PCR for *Pneumocystis jiroveci*
    g. Quantitative PCR for CMV
    h. Cytology
    i. Modified AFB smear and culture to identify *Nocardia*

4-D. Surgical site and intra-abdominal infections

The recommended initial diagnostic evaluation for surgical site infections (categorized in accordance with the Centers for Disease Control and Prevention criteria including intra-abdominal abscess, peritonitis, cholangitis, and wound infections) is as follows :

1. Flat and upright abdominal film if indicated by surgeon
2. Serum amylase and lipase
3. Surgical drainage or ultrasound-guided aspiration of collection or ascetic fluid, or obtaining bile from T-tube
    - Gram stain and culture for bacteria (aerobic and anaerobic)
    - KOH and fungal culture and PCR for *candida* and *aspergillus*
4. Obtaining specimens from a wound
    - Gram stain and culture for bacteria
    - KOH and fungal culture

Points for specimen collection:

- Perform a surgical approach for wound culture (i.e. superficial material is débrided and a biopsy of the wound base is submitted for culture); swabs generally are the least desirable way to collect a specimen for a wound culture
- Submit the aspirated fluid of wound in a syringe after removing the needle to help ensure the survival of anaerobic bacteria
- Perform gram stain of wound fluid to examine the proportion of squamous epithelial cells and neutrophils, and to determine the usefulness of subsequent culture

4-E. CNS symptoms

The recommended initial diagnostic evaluation for CNS symptoms is as follows:

1. Brain CT scan (with and without contrast) or brain MRI with gadolinium (if no contraindication)
2. Lumbar puncture for CSF analysis:
    a. Cell count and differential
    b. Glucose and protein
    c. Bacterial culture
    d. KOH test and fungal culture
    e. AFB test, adenosine deaminase (ADA) level, mycobacteria culture and PCR for mycobacteria
    f. Cryptococcal antigen detection and PCR for *Cryptococcus*
    g. PCR for HSV and Viral culture (if available)
    h. Cytology
3. Biopsy of mass lesions and/or leptomeninges (specially to identify granulomatous meningitis)

4-F. Diarrhea

The recommended initial diagnostic evaluation for diarrhea is as follows:

1. Stool for WBC and cultures (for enteric bacteria [*Salmonella*, *Shigella*, *Campylobacter*])
2. Stool specimens for *C. difficile* testing
    a. One sample for RT-PCR for toxin B is preferable
    b. At least 2 separate stool specimens for enzyme immunoassay for toxins A and B
3. Three separate stool specimens for ova and parasites
4. If stool studies unrevealing and diarrhea persists, endoscopic evaluation warranted
    a. Mucosal biopsy
    b. Immunohistochemical staining for CMV
    c. KOH test, fungal culture and PCR for candida, aspergillus and mucur
    d. AFB stain and culture and PCR for *mycobacteria*

4-G. Fever without localizing findings

The recommended initial diagnostic evaluation for fever without localizing findings is as follows:

1. Quantitative PCR for *EBV*
2. PCR for *Parvovirus* B19
3. Antigen detection tests available for *adenovirus*, *influenza A*, *respiratory syncytial virus*, and *rotavirus* if available; PCR may also be available

4-H. Lymphadenopathy

The recommended initial diagnostic evaluation for lymphadenopathy is as follows:

1. *T. gondii* serology (IgG with avidity test, IgM)
2. Quantitative PCR for *EBV*
3. Quantitative PCR for *CMV*
4. PCR for *parvovirus B 19*
5. Biopsy of involved lymph node
    a. Histologic examination to look for atypical cells nad granulomata
    b. Aerobic and anaerobic cultures
    c. AFB stain, mycobacterial culture and PCR for mycobacteria
    d. KOH test, fungal culture and PCR for cryptosporidium
    e. Modified AFB smear and culture to identify *Nocardia*
6. CT scanning of neck, chest, abdomen, and pelvis
7. *Bartonella* (cat scratch disease) serology (if available)

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