Hepatitis B: Clear as Mud

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Emory University School of Medicine
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Objectives

1. Distinguish the various stages in the natural history of chronic hepatitis B

2. Know which tests are necessary to monitor hepatitis B activity, and when changes in therapy are indicated

3. Understand the limitations of currently available hepatitis B therapies due to cross-resistance and limited potency
Acute Hepatitis B Virus Infection with Recovery

Typical Serologic Course

Symptoms

HBeAg

anti-HBe

Total anti-HBc

IgM anti-HBc

Window Period
Only core antibody

HBsAg

anti-HBs

Titer

Weeks after Exposure
Progression to Chronic Hepatitis B Virus Infection
Typical Serologic Course

<table>
<thead>
<tr>
<th>Acute (6 months)</th>
<th>Chronic (Years)</th>
</tr>
</thead>
<tbody>
<tr>
<td>HBsAg</td>
<td>anti-HBe</td>
</tr>
<tr>
<td>HBeAg</td>
<td></td>
</tr>
<tr>
<td>Total anti-HBc</td>
<td></td>
</tr>
<tr>
<td>IgM anti-HBc</td>
<td></td>
</tr>
</tbody>
</table>

Weeks after Exposure

CDC
## Hepatitis B Serologic Diagnosis

<table>
<thead>
<tr>
<th></th>
<th>HBsAg</th>
<th>Anti-HBs</th>
<th>HBeAg</th>
<th>Anti-HBe</th>
<th>Anti-HBc</th>
<th>IgM anti-HBc</th>
<th>HBV DNA</th>
<th>ALT</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Acute hepatitis B</strong></td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>High</td>
</tr>
<tr>
<td><strong>Immunity (infection)</strong></td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+/-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>Nml</td>
</tr>
<tr>
<td><strong>Immunity (vaccination)</strong></td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>Nml</td>
</tr>
<tr>
<td><strong>Chronic Hepatitis B</strong></td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+/-</td>
<td>High</td>
</tr>
<tr>
<td><strong>Chronic Infection (Precore Mutant)</strong></td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>High</td>
</tr>
<tr>
<td><strong>Chronic carrier</strong></td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>- or low</td>
<td>Nml</td>
</tr>
</tbody>
</table>
Chronic Hepatitis B (sAg+)

- HBeAg
- Anti-HBe

HBV DNA

ALT

Inactive Carrier

Immune Clearance (eAg+ chronic hepatitis)

Reactivation (eAg- chronic hepatitis)

Adapted from Yim, Hepatology 2006; 43:S173-181
Chronic HBeAg- Disease (Pre-Core/Core Mutant)

- Lower HBV DNA levels but higher risk of cirrhosis (annual 8-10%/year)
- Represents a later phase of chronic hepatitis B
- No strict cutoff to discriminate between chronic carriers and eAg- hepatitis B
Natural History

- sAg clearance occurs at a rate of 0.5-1% per year
  - Can still get HCC even if cleared
- Annual incidence of cirrhosis 2-6% in HBeAg+ disease and 8-10%/year in HBeAg- disease
  - Other risk factors: EtOH, HCV, HIV, high HBV DNA, Genotype C
Alterations in HBV Natural History Among HIV/HBV Coinfected

- More likely to become chronic carrier of HBV (surface antigen positive)
- More likely to be e antigen (HBeAg) positive
- Less likely to be e antibody (anti-HBe) positive
- Less likely to convert HBeAg to anti-HBe
- More likely to go from HBeAg- back to HBeAg+
- Can revert to HBsAg+ from anti-HBs
- Higher levels of HBV DNA

References:

DiMartino, Gastro 2002; 123:1812-1822
Piroth, AIDS 2007; 21:1323-31
Colin, Hepatology 1999; 29:1306-1310
Rouphael, AIDS 2007; 21: 771-4
Benhamou CROI 2005 #933
Gilson, AIDS 1997; 11:597-606
Determining Hepatitis B Status

**Step 1.**
Screening serologies (HBsAg, anti-HBs, anti-HBc)

- HBsAg- anti-HBc+ anti-HBs+
- HBsAg+ Chronic Hepatitis B
- HBsAg- anti-HBc- anti-HBs+

**Step 2.**
Determine stage: HBeAg, anti-HBe, HBV DNA

- HBeAg+ anti-HBe- HBV DNA > 20000 IU/mL
- HBeAg- anti-HBe+/ HBV DNA > 2000 IU/mL
- HBeAg- Anti-HBe+ HBV DNA < 2000 IU/mL

Resolved HBV → Immune

Vaccinated → Immune

Chronic Hepatitis B, eAg+
Chronic Hepatitis B, eAg-
Chronic Hepatitis B carrier
## Monitoring of Chronic Hepatitis B

Regardless of whether treatment is initiated, each patient with hepatitis B should have:

<table>
<thead>
<tr>
<th>Every 3 months:</th>
<th>Every 6 months:</th>
<th>Every 12 months:</th>
</tr>
</thead>
<tbody>
<tr>
<td>ALT</td>
<td>HBV DNA level</td>
<td>HBeAg (if + initially)</td>
</tr>
<tr>
<td>AST</td>
<td></td>
<td>Anti-HBe</td>
</tr>
</tbody>
</table>

DNA levels and HBeAg, anti-HBe should also be checked with any flare of transaminases.
Available HBV Therapies

**Active against HIV and hepatitis B**
- Lamivudine (LAM, 3TC)*
- Emtricitabine (FTC)
- Tenofovir (TDF)*

**Active against hepatitis B only**
- Adefovir (ADV)*
- Telbivudine (LdT)*
- Interferon-α2b*
- Peg-interferon-α2a*

Entecavir (ETV)*

*FDA approved for hepatitis B

L-nucleoside analogues
Acyclic phosphonates
Deoxyguanosine analogues
Consider Potency and Potential for Resistance

Genetic Barrier

Potency

LdT
ETV
TDF
LAM
FTC
ADV
IFN

Adapted from Soriano, AIDS 2008; 22: 1399-1410
Lamivudine Resistance

Year 1 | Year 2 | Year 3 | Year 4
---|---|---|---
HIV- (Leung) | 7% | 39% | 53% | 50%
HIV+ (Benhamou) | 17% | 50% | 75% | 78%
HIV+ (Matthews) | 91% | 94% | 70% | 57%

M204V/I + L180M

Benhamou, Hepatology, 1999, 30:1302-6
Matthews, AIDS 2006; 20:863-870
Adefovir: High rate of nonprimary response

Only 44% achieved <10,000 copies/mL by month 6; very few who failed to response by m6 went on to have further viral suppression.

Reduction in HBV DNA (log copies/mL)

- <3 log
- 3.1-5 log
- >5 log

Month 6 vs Month 12

Fung, J Hepatology 2006; 44:283-290
ADV in HIV: Results at 3 years

![Graph showing percentages of HBV DNA <1000 copies/mL and ALT normalization at baseline and various weeks (48, 96, 144).]

- Baseline: 0% (HBV DNA), 3% (ALT Normalization)
- Week 48: 6% (HBV DNA), 14% (ALT Normalization)
- Week 96: 27% (HBV DNA), 48% (ALT Normalization)
- Week 144: 46% (HBV DNA), 68% (ALT Normalization)

Benhamou, CROI 2004, Abs 835
ACTG 5127: Tenofovir Noninferior to Adefovir for HBV/HIV

Peters, Hepatology 2006; 44:1110-6
<table>
<thead>
<tr>
<th></th>
<th>ADV (n=25)</th>
<th>TDF (n=27)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean change in HBV DNA</td>
<td>-4.03 log</td>
<td>-5.74 log</td>
</tr>
<tr>
<td>from baseline</td>
<td></td>
<td></td>
</tr>
<tr>
<td>HBV DNA&lt;200 at w36</td>
<td>8.6%</td>
<td>5.7%</td>
</tr>
<tr>
<td>HBV DNA&lt;200 at w48</td>
<td>11.4%</td>
<td>20%</td>
</tr>
<tr>
<td>Nml ALT at w48</td>
<td>25%</td>
<td>36%</td>
</tr>
<tr>
<td>HBeAg→anti-HBe</td>
<td>1 pt</td>
<td>0 pts</td>
</tr>
</tbody>
</table>
HBV DNA DAVG\textsubscript{48} (log copies/mL)

Table 3. Serum HBV DNA Decrease in A5127 Study Subjects During Therapy With ADV or TDF

<table>
<thead>
<tr>
<th>HBV DNA drop</th>
<th>w 12</th>
<th>w 24</th>
<th>w 36</th>
<th>w 48</th>
</tr>
</thead>
<tbody>
<tr>
<td>ADV (n = 25)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>&lt;2</td>
<td>3</td>
<td>0</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>2-4</td>
<td>17</td>
<td>15</td>
<td>11</td>
<td>8</td>
</tr>
<tr>
<td>&gt;4</td>
<td>3</td>
<td>5</td>
<td>7</td>
<td>8</td>
</tr>
<tr>
<td>n/a</td>
<td>2</td>
<td>5</td>
<td>7</td>
<td>8</td>
</tr>
<tr>
<td>TDF (n = 27)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>&lt;2</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>2-4</td>
<td>8</td>
<td>4</td>
<td>1</td>
<td>3</td>
</tr>
<tr>
<td>&gt;4</td>
<td>14</td>
<td>13</td>
<td>15</td>
<td>14</td>
</tr>
<tr>
<td>n/a</td>
<td>4</td>
<td>9</td>
<td>10</td>
<td>9</td>
</tr>
</tbody>
</table>

w, Week; n/a: number of subjects for whom data was not available at that time.
## Tenofovir Resistance

### Baseline Characteristics (n=43)

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Median age (years)</td>
<td>41 (IQR 37-42)</td>
</tr>
<tr>
<td>Median CD4 (cells/ul)</td>
<td>378 (IQR 235-475)</td>
</tr>
<tr>
<td>Median plasma HIV RNA (log copies/mL)</td>
<td>2.29 (IQR 1.7-3.4)</td>
</tr>
<tr>
<td>Median ALT (IU/mL)</td>
<td>48 (IQR 31-59)</td>
</tr>
<tr>
<td>Median serum HBV DNA (log copies/mL)</td>
<td>4.6 (IQR 3.0-8.0)</td>
</tr>
<tr>
<td>Mean time on tenofovir (months)</td>
<td>11.2 ± 6.7</td>
</tr>
<tr>
<td>Mean time on lamivudine (months)</td>
<td>35.3 ± 27.5</td>
</tr>
<tr>
<td>HBeAg+ (%)</td>
<td>35 (82%)</td>
</tr>
</tbody>
</table>

Sheldon, Antiviral Ther, 2005; 10:727-34
Novel Mutations detected on TDF Therapy: Patient 1

Sheldon, Antiviral Ther, 2005; 10:727-34
Novel Mutations detected on TDF Therapy: Patient 2

Sheldon, Antiviral Ther, 2005; 10:727-34
# Effect of clinical mutations on HBV susceptibility to TDF *in vitro*

<table>
<thead>
<tr>
<th>Extracellular DNA</th>
<th>IC&lt;sub&gt;50&lt;/sub&gt; (umol/l)</th>
<th>Fold IC&lt;sub&gt;50&lt;/sub&gt;**</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wild-type</td>
<td>12.4</td>
<td>1</td>
</tr>
<tr>
<td>A194T</td>
<td>95</td>
<td>7.6</td>
</tr>
<tr>
<td>L180M+M204V</td>
<td>71</td>
<td>5.7</td>
</tr>
<tr>
<td>L180M + A194T + M204V</td>
<td>&gt;120</td>
<td>&gt;10</td>
</tr>
</tbody>
</table>

** 5-10 fold change = partial resistance; >10 fold confers resistance

Sheldon, Antiviral Ther, 2005; 10:727-34
A194T does not reduce efficacy to TDF

- 10 patients failing TDF with L180M+M204V/I that were found to have A194T
- 6 received TDF+/-FTC as salvage
- All 6 had >3 log decrease in HBV DNA and clinical response to TDF

Fung, AASLD 2008, Abs 880
Tenofovir Resistance

- No HBV polymerase/RT amino acid substitutions associated with TDF resistance were detected through 96 weeks of monotherapy.
- Virologic breakthrough was infrequent and not associated with phenotypic resistance.
  - Most had nonadherence.
Entecavir in HIV/HBV: ETV-038

**DOUBLE-BLIND PHASE**

- ETV 1 mg QD  
  N=51
- Placebo  
  N=17

**OPEN-LABEL PHASE**

- ETV 1 mg QD  
  N=48
- ETV 1 mg QD  
  N=17

Continued LAM as part of HAART

Wk 2  12  24  48

Pessoa, AIDS 2008; 22:1779-1787
Colonno, CROI 2006, #832
# Efficacy Endpoints

<table>
<thead>
<tr>
<th></th>
<th>Entecavir (n=51 w24, n=48 w48)</th>
<th>Placebo (n=17 w24 &amp; w48)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean HBV DNA @ w24</td>
<td>5.52 log copies/mL</td>
<td>9.27 log copies/mL</td>
<td></td>
</tr>
<tr>
<td>Mean HBV DNA @ w48</td>
<td>4.79 log copies/mL</td>
<td>5.63 log copies/mL</td>
<td></td>
</tr>
<tr>
<td>Change in DNA from baseline @ w24</td>
<td>-3.65 log copies/mL</td>
<td>+0.11 log copies/mL</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Change in DNA from baseline @ w48</td>
<td>-4.20 log copies/mL</td>
<td>-3.56 log copies/mL</td>
<td></td>
</tr>
<tr>
<td>HBV DNA&lt;300 copies/mL @ w24</td>
<td>3/51 (6%)</td>
<td>0/17 (0%)</td>
<td></td>
</tr>
<tr>
<td>HBV DNA&lt;300 copies/mL @ w48</td>
<td>4/51 (8%)</td>
<td>0/17 (0%)</td>
<td></td>
</tr>
<tr>
<td>ALT normalization w24</td>
<td>34%</td>
<td>8%</td>
<td>0.08</td>
</tr>
<tr>
<td>ALT normalization w48</td>
<td>37%</td>
<td>46%</td>
<td></td>
</tr>
<tr>
<td>HBeAg loss w48</td>
<td>1 (2%)</td>
<td>0</td>
<td>0.56</td>
</tr>
<tr>
<td>HBeAg seroconv, w24</td>
<td>1 (2%)</td>
<td>0</td>
<td></td>
</tr>
</tbody>
</table>

Pessoa, AIDS 2008; 22:1779-1787
Entecavir Resistance

- Requires “two hits”
- M204V/I +/- L180M as first hit
  - 8 to 10 fold dec in susceptibility to ETV vs wild-type
- Then mutation in I169, T184, S202 or M250
  - These mutations on their own have minimal effect on susceptibility to entecavir
- In presence of M204V/I, one of these leads to 10-250 fold decrease in ETV susc
- M204V/I + 2 mutations → 500-1000 fold decrease in ETV susceptibility

Tenney, AAC, 2004; 48:3498-507
Riddick, J Hep 2008; 48:895-902
Entecavir Resistance

- NA-naïve
  - After 4 years on treatment, a total of 3 patients (<1%) developed ETVr mutations
    - 2 of these had virologic breakthrough
    - Out of 663, 278, 149, 120 tested for resistance in years 1-4 respectively

- LAM-R patients
  - After 4 years on treatment, virologic breakthrough occurred in 1%, 10%, 16%, and 15% in years 1, 2, 3 and 4 respectively

- Cumulative probability of virologic breakthrough through 4 years 0.8% in naïve and 39.5% in LAM-R patients

Colonno, EASL 2008, Abs 781
Development of M184V in HIV

- 4 of 13 patients with >0.5 log decline had HIV rebound after achieving a nadir
  - All ART-experienced
  - 3 had developed M184V at time of HIV RNA rebound after a median of 98 days
  - 2 other ART-experienced patients had M184V before ETV initiation

- 2 ARV-naïve patients developed M184V after median 132 days of ETV
  - 1 more without a baseline geno did as well

Telbivudine in HIV

- No RCT for its use in HIV+
- Cross-resistance limits usefulness in ARV-experienced patients
- AASLD Guidelines currently do not recommend its use in HIV+
- EACS Guidelines present it as alternative to ADV in those not requiring HIV therapy
Telbivudine Resistance

- Most frequent genotypic change M204I
  - Only mutation causally associated with resistance

- Other mutations:
  - L80 (n=26)
  - L180 (n=4)
  - L229 (n=6)

- Single case of M204V/L180M double mutant seen for telbivudine at week 104 in GLOBE

Seifer, DDW 2007; Abs #93
Standring, DDW 2007; Abs. S1781
Liaw, Gastro 2009; 136: 486-495
Telbivudine: HIV Activity?

![Graph showing HIV viral load over days of therapy with comparisons to Adefovir and LdT](image)

- Adefovir
- LdT

Days of Therapy:
- -60
- 0
- 60
- 150
- 180
- 270
- 277
- 284

HIV viral load:
- 0
- 1000
- 2000
- 3000
- 4000
- 5000
- 6000
- 7000
- 8000
- 9000
- 10000

Low AIDS 2009; 23: 546-7
# HBV Resistance Summary

<table>
<thead>
<tr>
<th>Drug</th>
<th>Resistance Changes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lamivudine</td>
<td>M204V/I, L180M</td>
</tr>
<tr>
<td>Adefovir</td>
<td>N236T&lt;sup&gt;a&lt;/sup&gt;, A181V/T, I233V&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>Entecavir</td>
<td>M204V/I&lt;sup&gt;d&lt;/sup&gt; → I169, T184&lt;sup&gt;d&lt;/sup&gt;, S202&lt;sup&gt;d&lt;/sup&gt; or M250</td>
</tr>
<tr>
<td>Telbivudine</td>
<td>M204I</td>
</tr>
<tr>
<td>Tenofovir</td>
<td>A194T</td>
</tr>
</tbody>
</table>

<sup>a</sup> Angus, Gastro 2003; 125:292-97  
<sup>b</sup> Gallego, J Viral Hep 2008; 15:392-98  
<sup>c</sup> Schildgen, NEJM 2006; 354: 1807-12  
<sup>d</sup> Tenney, AAC 2007; 51:902-911
Additional compensatory mutations that restore viral fitness can be seen with the development of drug resistance.

Resistance Summary

Lamivudine
Adefovir
Entecavir (naïve)
Entecavir (LAM-R)
Telbivudine (e+)
Telbivudine (e-)
Tenofovir

Hadziyannis, NEJM 2005; 352: 2673-81
Lok, Hepatology 2007; 45:507-539
Snow-Lampart, AASLD 2008, Abs 977
Lai, NEJM 2007; 257:2576-88
Tenney, EASL 2009, Abs 20
When treatment fails

- Virologic Rebound
- Genotypic Resistance
- ALT flare

HBV DNA viral load vs. Weeks to months (3-7 weeks)

Anecdotal failures of HBV therapy with Truvada

No clear data on what best strategy is

No known mutations for tenofovir though clinical breakthroughs described

If entecavir added, recommend removing lamivudine/emtricitabine from HIV regimen to prevent selective pressure on M204V/I in HBV polymerase
  - Can lead to faster development of ETV resistance
Take home points

- Document more than just “Hepatitis B” in problem list
  - Resolved hepatitis B, Chronic carrier, eAg+ CHB, etc

- The HBV DNA level should be followed regularly, along with ALT and eAg

- Sequencing of antivirals is important: consider prior lamivudine exposure when choosing drugs; avoid entecavir – lamivudine combinations