Phase I Trial of Immunotherapy with BHT-3009 Alone or Combined with Atorvastatin in Patients with Multiple Sclerosis

Recombinant DNA Advisory Committee Meeting
June 8, 2004
BHT-3009 Treatment of Multiple Sclerosis

- Antigen-specific immunosuppression - Well established
  - Pre-clinical models
  - Approved product - Copaxone
  - Multiple products in clinical development

- Plasmids
  - Typically used to enhance immunity
  - Immunosuppressive in correct context

- BHT-3009
  - Strong pre-clinical rationale
  - Good safety profile
  - Phase I trial designed for careful safety monitoring
Multiple Sclerosis (MS)

- Autoimmune disease of nervous system
- Immunological attack on the myelin sheath
- 400,000 patients in US
- Women > men
- Age < 40 years
- Clinical course
  - Relapsing remitting
  - Secondary progressive
  - Primary progressive
  - Progressive relapsing
- Lifelong progression to near total disability

Paralysis, numbness, disequilibrium, incontinence
Multiple Sclerosis

- Treatment Goal: Prevent relapses and progression of neurological disability
- Corticosteroids
  - Accelerate recovery from relapses
- Disease modifying agents
  - Interferon, Copaxone
  - Decrease relapses about 30%
  - Modest effect on disability progression
- Antigen-specific immunotherapy for MS in development
MS Target Antigens

- Known and well characterized self antigens
  - Human immunology
  - Established animal models
- Myelin basic protein (MBP)
- Proteolipid protein (PLP)
- Myelin oligodendrocyte glycoprotein (MOG)
- Myelin associated glycoprotein (MAG)
Trials of MBP-Specific Immunotherapy of MS

- **Copaxone**
  - Random polymers of 4 amino acids derived from immunodominant epitope of myelin basic protein (MBP)
  - Decreased antigen-specific T cell response, Th2 deviation
  - Approved for treatment of MS

- **MBP8289 (17 AA fragment of MBP)**
  - Tolerization and decreased antibody response to MBP
  - In Phase III trial

- **APL (Altered MBP83-99)**
  - Decreased antigen-specific T cell response
  - In phase IIb trial (ITN-sponsored)
MBP-specific Immunotherapy of MS: Safety

- **Worsening MS not observed with Copaxone, oral myelin, MBP8289**

- **APL**
  - Open label single arm trial of high dose APL:
    - 2 of 8 patients had relapses
  - Randomized, double blind trial of APL:
    - 142 patients
    - 26% of placebo patients worsened
    - 18% - 23% of APL-treated patients worsened
  - Currently in 600 patient ITN sponsored Phase IIb trial

- **Hypersensitivity reactions**
  - Observed with Copaxone, APL (& IFNβ)
  - No IgE or anaphylaxis
  - No impact on disease activity
  - Limited relevance to plasmid-based vaccines
  - Atorvastatin
    - No increase in hypersensitivity reactions
    - Concurrent vaccination is safe
BHT-3009: Designed for Immunosuppression

- Encodes DNA for full length hMBP
- No adjuvant
- Immunostimulatory CpG sequences reduced
BHT-3009: Non-Clinical Studies

- **Efficacy studies**
  - Experimental allergic encephalomyelitis
  - Dose and schedule

- **Safety**
  - EAE models
  - Biodistribution
  - Cynomolgus monkey study
EAE Model of MS

- SJL/J mice
- Well characterized model
- Relapsing & remitting course typical of MS
- Immunological mechanisms similar to human MS
- Immunodominant epitope: PLP
BHT-DNA Treats Established EAE

Mean Disease Score

Relapse Rate

BHT-DNA reduces both disease severity and relapse rates in a treatment model

p value = <0.0001 (Mann-Whitney)

*p value = 0.04 (ANOVA-Dunnett Hsu)
Inhibition of Antigen-Specific T-Cells

Reduced Proliferation

Reduced IFN\(\gamma\) Production

Journal of Immunology, 1999
BHT-3009
Dose & Schedule

BAYHILL THERAPEUTICS
DNA: Dose Response

**IFN-gamma ELISPOT**

- **PBS gavage + PBS injection**
- **Atorva 10mg/kg + 2ug PLP**
- **Atorva 10mg/kg + 10ug PLP**
- **Atorva 10mg/kg + 50ug PLP**
- **Atorva 10mg/kg + 200ug PLP**
Treatment Schedule

Every 2 to 4 week treatment decreases IFN-γ production
BHT-3009
Safety - Toxicity
Administration of PLP-Vector to SJL/J Mice Did Not Cause or Worsen EAE

- Administration to naïve animals (4 biweekly injections)
  - No significant anti-PLP antibodies
  - No clinical signs of EAE
  - Histopathology: no lymphocytic infiltration of brain
  - Induction of modest levels of anti-DNA antibodies (3 of 20 mice)

- Administration of PLP-vector to animals either prior to or after induction for EAE with PLP_{139-151} peptide
  - Effective for preventing and treating EAE
  - EAE-related mortality
    - 3 of 232 PLP-Vector treated mice
    - 3 of 208 control mice
<table>
<thead>
<tr>
<th>Biodistribution of BHT-3009 DNA: (150 µg single dose)</th>
<th>Positive Tissues at Each Sacrifice Time-point</th>
</tr>
</thead>
<tbody>
<tr>
<td>Day 3: Blood, Heart, Brain, Kidney, Inguinal LN, Iliac LN, Skin at SOA, injected muscle</td>
<td></td>
</tr>
<tr>
<td>Day 14: Brain, inguinal LN, Iliac LN, Skin at SOA</td>
<td></td>
</tr>
<tr>
<td>Day 42: Heart, skin at SOA</td>
<td></td>
</tr>
<tr>
<td>Day 70: Skin at SOA</td>
<td></td>
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</tbody>
</table>

<table>
<thead>
<tr>
<th>Expression of hMBP RNA (50 µg single dose)</th>
<th>Day 3: Inguinal LN</th>
</tr>
</thead>
<tbody>
<tr>
<td>Day 7: Iliac LN, injected muscle</td>
<td></td>
</tr>
<tr>
<td>Day 28: Skin at SOA</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Expression of hMBP RNA (150 µg single dose)</th>
<th>Day 3: Iliac LN, blood</th>
</tr>
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<tbody>
<tr>
<td>Day 7: Iliac LN, skin at SOA, muscle</td>
<td></td>
</tr>
<tr>
<td>Day 14: Iliac LN</td>
<td></td>
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<tr>
<td>Day 28: Skin at SOA</td>
<td></td>
</tr>
<tr>
<td>Day 42: No tissues expressing hMBP</td>
<td></td>
</tr>
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### GLP Toxicity Study in Cynomolgus Monkeys

#### Species

<table>
<thead>
<tr>
<th>Macaca fascicularis</th>
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#### Dose Groups

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<tr>
<th></th>
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<tbody>
<tr>
<td>A</td>
<td>Vehicle control for BHT-3009 (PBS) &amp; atorvastatin (0.5% CMC/0.1% Tween 80)</td>
</tr>
<tr>
<td>B</td>
<td>150 µg BHT-3009</td>
</tr>
<tr>
<td>C</td>
<td>1500 µg BHT-3009</td>
</tr>
<tr>
<td>D</td>
<td>1500 µg BHT-3009 + atorvastatin at 20 mg/kg/day</td>
</tr>
</tbody>
</table>

#### Treatment

- BHT-3009 (or PBS vehicle) administered bi-weekly (Days 1, 15, 29 and 43)
- Atorvastatin (or vehicle control) administered daily on Days 1 to 49
No pre-terminal mortality
No adverse clinical observations
Mean values for serum and hematology parameters within normal ranges
  - Trends towards a slight increase in ALT and AST in atorvastatin at 20 mg/kg/day group
No gross or organ weight findings in terminal or recovery animals
No histopathologic changes in either terminal or recovery animals
  - Including: cerebrum, thalamus, brain stem, cerebellum, spinal cord, sciatic nerve and bulbus oculi
No effects on percentages of peripheral blood immune cells
No induction of anti-dsDNA, anti-nuclear, or anti-hMBP antibodies
Summary of Pre-clinical Data

- EAE/efficacy data support a DNA dose frequency of biweekly or monthly.
- EAE/efficacy data supports efficacy over a wide range of DNA dose levels.
- No concerning safety or toxicology issues.
Phase I Trial of Immunotherapy with BHT-3009 Alone or Combined with Atorvastatin in Patients with Multiple Sclerosis

IND Cleared 4/04
CTA (Health Canada) Cleared 4/04
2 Directorates: BGTD & TPD
Reduced Antibody Responses

PLP, MBP, MOG & MAG plasmids

Antigens shown are statistically significant for variation of response across each group.

(published in Nature Biotech 2003)

**Immune Activation**

- APC (e.g. Dendritic cell)
- MHC
- TCR
- B7
- CD28
- Activation

**Immune Tolerance**

- APC (e.g. Dendritic cell)
- MHC
- TCR
- B7
- CD28
- Tolerance

Self-antigen Encoding DNA

Lack of Co-stimulation
<table>
<thead>
<tr>
<th>Disease Grade</th>
<th>Clinical Symptoms</th>
</tr>
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<tbody>
<tr>
<td>0</td>
<td>Healthy</td>
</tr>
<tr>
<td>1</td>
<td>Loss of Tail Tone</td>
</tr>
<tr>
<td>2</td>
<td>Hind Limb paraparesis</td>
</tr>
<tr>
<td>3</td>
<td>Hind Limb paralysis</td>
</tr>
<tr>
<td>4</td>
<td>Complete paralysis</td>
</tr>
<tr>
<td>5</td>
<td>Death</td>
</tr>
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MBP-specific Immunotherapy of MS: Safety Issues

- Hypersensitivity reactions/anaphylaxis
  - Observed with Copaxone, APL
  - Risk benefit ratio determines acceptability
  - Limited relevance to plasmid-based vaccines

- Worsening MS
  - Theoretical concern
    - Not observed in Bayhill’s preclinical studies
    - Not supported by clinical data
    - Treatable with high dose steroids
  - Not observed with Copaxone, oral myelin, MBP8289
  - APL
    - Open label single arm trial:
      - 25% worsened (2 of 8 patients)
    - Randomized, double blind trial:
      - 142 patients
      - 26% of placebo patients worsened
      - 18% - 23% of APL-treated patients worsened
Bayhill: Background

▪ **FOUNDERS**

  ▪ Lawrence Steinman, M.D.
    - Chair, Department Immunology
    Stanford University

  ▪ William Robinson, M.D., Ph.D.
    - Assistant Professor of Medicine in
    the Division of Immunology
    and Rheumatology at Stanford
    University

  ▪ PJ Utz, M.D.
    - Assistant Professor of Medicine in
    the Division of Rheumatology
    and Immunology at Stanford
    University

  ▪ Hideki Garren, M.D., Ph.D.
    - Clinical Faculty at Stanford Medical
    School
    - Bayhill: Director, Molecular Biology
    and Immunology

▪ **MANAGEMENT**

  ▪ Mark Schwartz, Ph.D., CEO
    - Calyx, Trega, Synteni, Incyte

  ▪ Frank Valone, M.D., Clinical
    - Dendreon, Titan

  ▪ Hideki Garren, M.D., Ph.D., Research
    - Stanford, Bayhill Founder

  ▪ Stephanie Broome, Ph.D., Regulatory
    - PRA International, SIBIA,
    PowderJect

  ▪ Martin Goldstein, J.D. Corp.
    Development
    - Virologic, Roche, Genentech
Multiple Sclerosis

- Relapsing-remitting course
- Life-long progression to total disability
- Treatment Goal: Prevent relapses and progression of neurological disability
- Corticosteroids – relapses
- Disease modifying agents
  - Interferon
  - Copaxone
  - Mitoxantrone
- Treatment is non-specific immunosuppression
- Antigen-specific immunotherapy for MS in development
Bayhill: Background

- Founded Spring, 2002
- Based in Palo Alto, CA
- 24 employees
- Focus on antigen-specific immunotherapy of autoimmune diseases
- Multiple technology platforms
  - Protein array
  - Plasmid DNA for immunosuppression
    - Multiple sclerosis
    - Type 1 diabetes
    - Rheumatoid arthritis
BHT-DNA reduces both disease incidence and severity in a prevention model.

Journal of Immunology, 1999
<table>
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<tr>
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<th>Mean Disease Score (compared to each other)</th>
<th>Mean Disease Score (compared to respective control)</th>
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<tbody>
<tr>
<td><strong>QW</strong></td>
<td></td>
<td><strong>P=0.22</strong></td>
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<tr>
<td><strong>Q2W</strong></td>
<td>Q2W injections decrease MDS by 0.25</td>
<td><strong>P&lt;0.001</strong></td>
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<td><em>(p=0.01)</em></td>
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<tr>
<td><strong>Q4W</strong></td>
<td>Q4W injections decrease MDS by 0.12</td>
<td><strong>P&lt;0.001</strong></td>
</tr>
<tr>
<td></td>
<td><em>(p=0.01)</em></td>
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