

# **Intra-articular Administration of a Recombinant Adeno-Associated Vector containing a TNF- $\alpha$ Antagonist Gene in Inflammatory Arthritis**

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# Disclosures for P. Mease

- Advisor, clinical investigator
  - Abbott
  - Alder
  - Amgen
  - Biogen IDEC
  - Bristol Myers
  - Centocor
  - Genentech
  - PanGenetics
  - Pfizer
  - Seattle Genetics
  - Targeted Genetics
  - Trubion
  - Wyeth

# Presentation Outline

- Local anti-TNF gene delivery using AAV
- Phase I clinical study
- Phase I/II clinical study
  - Study design and entry criteria
  - Safety
- Study recruitment
- Consent process

# Rationale for Anti-TNF $\alpha$ Therapy

- TNF $\alpha$ : A dominant pro-inflammatory mediator, overproduced in inflammatory arthritis (RA, PsA, AS)
- TNF $\alpha$  blockade ameliorates symptoms and signs of inflammatory arthritis
- Repeated treatment with TNF- $\alpha$  antagonists results in
  - Sustained reduction in symptoms and signs of RA, PsA & AS in majority of patients
  - Protects joints from structural damage

# The State of TNF Inhibitor Therapy

- The majority of patients who receive systemic TNF inhibitors get a significant benefit in both symptoms and signs
- Not everyone treated with a TNF inhibitor improves sufficiently, however

# The State of TNF Inhibitor Therapy

- Suboptimal responses to TNF Inhibitors
  - Approximately 30% RA patients do not achieve ACR 20 response; 50% do not achieve ACR 50 response
  - Persistent disease burden
    - Critical joints for function have persistent synovitis
- Indicators of Suboptimal Response
  - Presence of persistent active synovitis
  - Elevated disease markers (ESR, CRP)
  - Radiologic progression

# rAAV human TNFR:Fc Vector (tgAAC94)

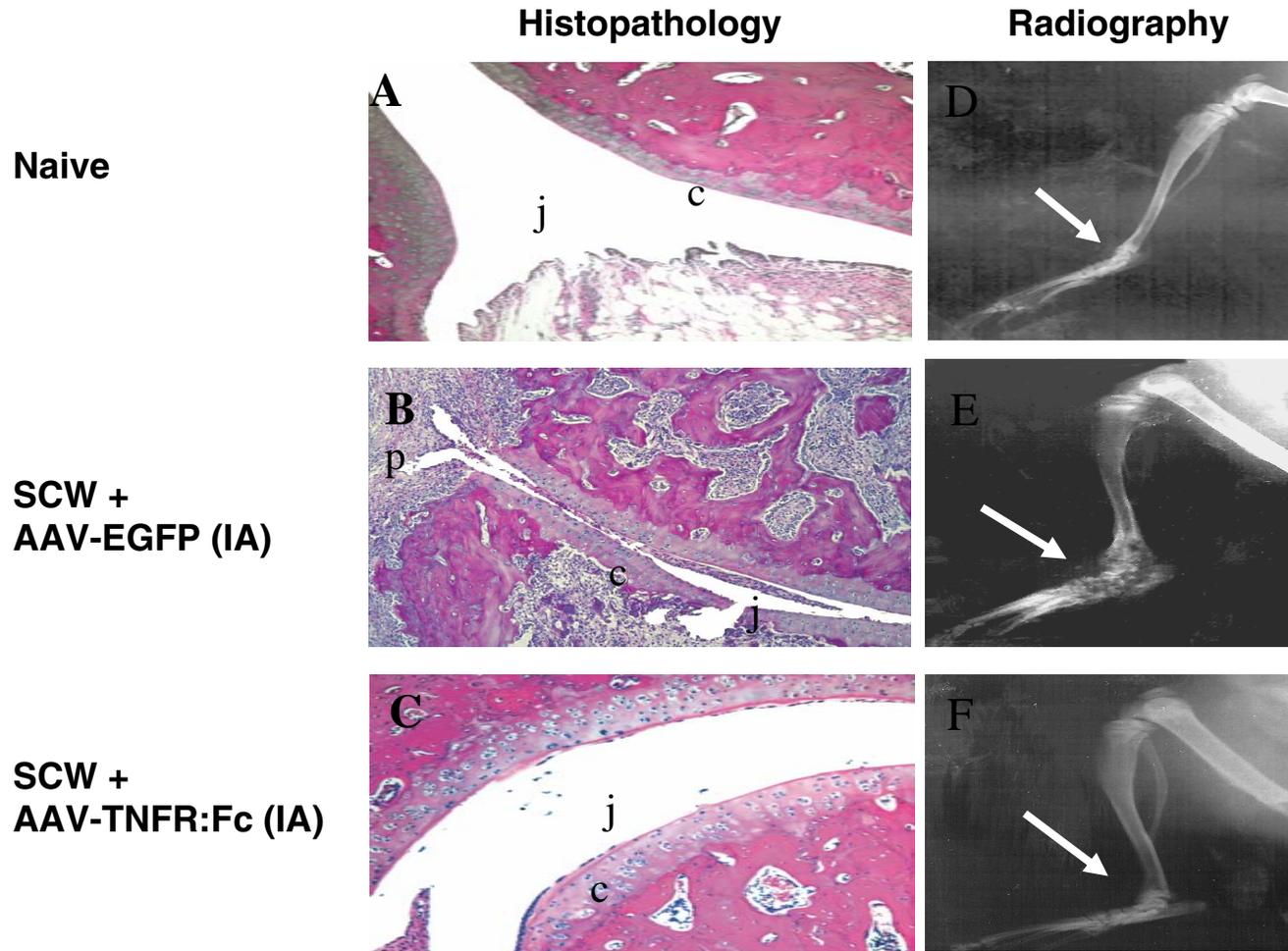


- Single stranded DNA encoding huTNFR:Fc
- huTNFR:Fc cDNA sequence identical to the cDNA used for etanercept production
- Packaged in AAV2 capsid
  - Lacks viral coding sequences
  - Based on wtAAV virus which is considered non-pathogenic
  - Vector non-replicative even in presence of helper virus
  - Persist for life of host cell, primarily as episomal concatemers
- Extensively purified

# Rationale for Targeted Intra-articular Delivery

- Patients with inflammatory arthritis on systemic therapy, including TNF antagonists, with incomplete response
  - Persistent synovitis in critical joints which might benefit from targeted supplemental therapy
- Patients with oligo- or monoarticular inflammatory arthritis not on systemic therapy
  - Targeted, concentrated therapy for active synovitis in patients for whom exposure to systemic therapy might not be essential
- Both groups might benefit from sustained, local anti-TNF therapy provided by gene transfer

# AAV2-rat TNFR:Fc Vector Reduces Severity of Experimental Arthritis



J, joint space; c, cartilage; p, pannus tissue

# Summary: Preclinical Toxicology Studies

## rAAV-TNFR:Fc Therapy

- Safe at doses of up to  $\sim 1 \times 10^{13}$  DRP /mL joint volume in rats
- Limited and transient biodistribution to extra-articular tissues
- Local expression of TNFR:Fc confirmed in the joint
- Circulating TNFR:Fc protein in serum
  - $< 0.0025 \mu\text{g/mL}$  in 98/102 treated animals
  - Detectable levels in 4/102 treated animals
    - Maximum amount detected  $0.01 \mu\text{g/mL}$
  - $C_{\text{max}}$  of etanercept in patient serum is  $3 \mu\text{g/mL}$

# Summary: Non-Human Primate Pharmacology Study of tgAAC94

- AAV-huTNFR:Fc injected IA
  - Doses:  $1 \times 10^{11}$ ,  $1 \times 10^{12}$ ,  $1 \times 10^{13}$  DRP/mL of joint volume
- Amount of vector DNA in injected joint was dose-dependent
  - $< 50$  to  $1 \times 10^9$  copies/ $\mu\text{g}$
- TNFR:Fc mRNA detected in injected joint
- Vector DNA but not TNFR:Fc mRNA detected in spleen and draining LN
- Sporadic huTNFR:Fc protein detected circulating in serum
  - Maximum  $\sim 0.07 \mu\text{g/mL}$  detected over background
  - $C_{\text{max}}$  of etanercept in patient serum is  $3 \mu\text{g/mL}$

# Clinical Studies of AAV2-TNFR:Fc

	Phase I	Phase I/II
Study population	RA, PsA, AS	RA, PsA, AS
Concurrent therapy	Not on TNF antagonists	With or without TNF antagonists
No. of subjects	15	127
RAC Filing	Submitted	Submitted
RAC Review	Public Review Sept 2003	Exempt from public review
Study Status	Completed	Enrolled; On clinical hold

# First Clinical Study: Safety of Targeted, Local AAV2-TNFR:Fc Administration in Subjects NOT on TNF- $\alpha$ Antagonists

- Phase I, dose escalation study
  - 15 subjects with inflammatory arthritis (RA=14; AS=1)  
NOT on TNF- $\alpha$  antagonists
  - At least one problematic joint despite stable regimen
- Single intra-articular injection (knee=14, ankle=1)
  - $1 \times 10^{10}$  DNase resistant particles (DRP) tgAAC94 (n=5)
  - $1 \times 10^{11}$  DRP tgAAC94 (n=6)
  - Placebo (n=4)
- Safety and tolerability
  - No drug-related serious adverse events reported
  - Adverse events considered related by investigator (n=3)
    - Possible: Nasopharyngitis, sinus congestion
    - Probable: Pruritis of injected knee
  - Well-tolerated at doses up to  $5 \times 10^{11}$  DRP (total dose)

# Phase I/II Clinical Study: Safety of Repeat Local AAV2-TNFR:Fc Administration

## *Study Design*

120 subjects in 6 cohorts of 20 subjects each

### Cohorts 1-3: Dose escalation

- 3 dose levels:  $1 \times 10^{11}$ ,  $1 \times 10^{12}$  and  $1 \times 10^{13}$  DRP/mL joint volume

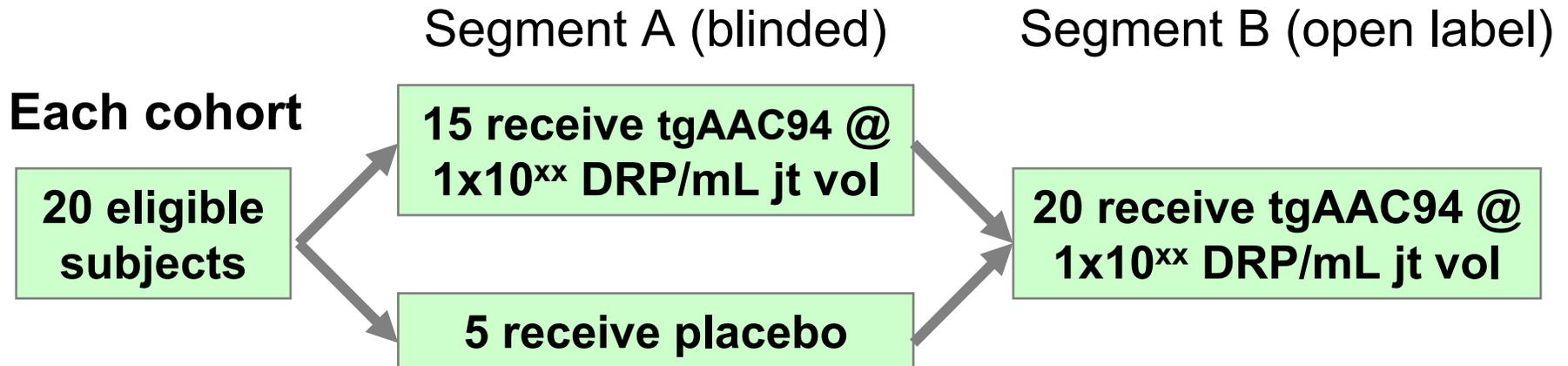
### Cohorts 4-6: Phase 2 expansion to increase subject numbers

- Same three dose levels
- Simultaneous enrollment in all three cohorts
- MRI in subset of subjects to correlate with clinical observations
- Functional assessments

Entry criteria: RA, PsA or AS with at least one problematic joint despite stable regimen; with or without current use of TNF antagonists

# Phase I/II Clinical Study: Safety of Repeat Local AAV2-TNFR:Fc Administration

## *Study Design*



### Two doses

- First dose in Segment A
- Second dose 12 to 30 weeks later, based on predetermined criteria

### Dosing based on joint volume

Knee, 5mL

Ankle, 2mL

Elbow, 1.5 mL

Wrist, 1mL

MCP, 0.5 mL

# Phase I/II Clinical Study

## Study Objectives

- Primary endpoint
  - Safety
- Secondary endpoints
  - Tenderness & swelling of injected joint
  - Time to second injection of study drug
  - Overall disease activity
  - TNFR:Fc protein levels in serum & synovial fluid
  - Serum anti-AAV2 capsid neutralizing titers
- Explore new outcome measures for single joints
  - Patient assessment
  - Functional assessment
  - Joint inflammation and damage on MRI (select subjects)

# Phase I/II Clinical Study

## Inclusion Criteria

- Inflammatory arthritis (RA, PsA, AS)
- Persistent moderate or severe swelling in at least 1 eligible joint despite stable medical regimen
- For subjects with RA, an adequate trial of at least one DMARD
- For subjects on DMARDS, stable regimen x 3 months, with no dose changes x 4 weeks
- Age  $\geq$  18 years and  $<$  75 yrs
- Males and females willing to practice contraception, if of child-bearing capacity
- Able to give written informed consent

# Phase I/II Clinical Study

## Exclusion Criteria

- Disease severe enough to warrant change in regimen for inflammatory arthritis in the next 3 months
- Previous discontinuation of etanercept for safety concerns
- Current use of anakinra or abatacept
- Corticosteroid therapy at doses higher than the equivalent of 10 mg prednisone/day
- Steroid or hyaluronate injection in the target joint, or receipt of an investigational agent less than 4 weeks before screening
- Class IV functional status (Hochberg, et al., 1992)
- Laboratory values outside screening parameters

# Phase I/II Clinical Study

## Exclusion Criteria

- Known HIV or Hepatitis C infection or known positive Hepatitis B surface antigen
- Positive PPD, unless previously treated
- Pregnant or lactating
- Inflammatory bowel disease, such as Crohn's or UC
- Severe pulmonary, liver or kidney disease, uncompensated CHF, myocardial infarction within 6 months, unstable angina, uncontrolled hypertension, uncontrolled asthma, demyelinating neurological disease, history of cancer, insulin-dependent diabetes, recurrent opportunistic infections, active joint infection

# Phase I/II Clinical Study

## Demographics

*Enrollment Completed*

	<b>Placebo</b> N=31	<b>Low Dose</b> 1x10 <sup>11</sup> DRP/mL N=33	<b>Mid Dose</b> 1x10 <sup>12</sup> DRP/mL N=32	<b>High Dose</b> 1x10 <sup>13</sup> DRP/mL N=31
<b>Gender, female</b>	<b>25 (81%)</b>	<b>26 (79%)</b>	<b>24 (75%)</b>	<b>22 (71%)</b>
<b>Race, white</b>	<b>27 (87%)</b>	<b>29 (88%)</b>	<b>29 (91%)</b>	<b>25 (81%)</b>
<b>Age, mean years (range)</b>	<b>55 (22-76)</b>	<b>52 (27-76)</b>	<b>52 (23-75)</b>	<b>54 (27-77)</b>
<b>Arthritis type: RA</b>	<b>25 (81%)</b>	<b>28 (85%)</b>	<b>22 (69%)</b>	<b>26 (84%)</b>
<b>PsA</b>	<b>5 (16%)</b>	<b>3 (9%)</b>	<b>8 (25%)</b>	<b>5 (16%)</b>
<b>AS</b>	<b>1 (3%)</b>	<b>2 (6%)</b>	<b>2 (6%)</b>	<b>0 (0%)</b>
<b>Target joint: Knee</b>	<b>13 (42%)</b>	<b>13 (39%)</b>	<b>15 (47%)</b>	<b>9 (29%)</b>
<b>Ankle</b>	<b>5 (16%)</b>	<b>5 (15%)</b>	<b>8 (25%)</b>	<b>6 (19%)</b>
<b>Wrist</b>	<b>10 (32%)</b>	<b>6 (18%)</b>	<b>5 (16%)</b>	<b>8 (26%)</b>
<b>MCP</b>	<b>3 (10%)</b>	<b>6 (16%)</b>	<b>3 (9%)</b>	<b>3 (10%)</b>
<b>Elbow</b>	<b>0 (0%)</b>	<b>3 (9%)</b>	<b>1 (3%)</b>	<b>5 (16%)</b>

# Phase I/II Clinical Study

## Baseline Status

	<b>Placebo N=31</b>	<b>Low Dose 1x10<sup>11</sup> DRP/mL N=33</b>	<b>Mid Dose 1x10<sup>12</sup> DRP/mL N=32</b>	<b>High Dose 1x10<sup>13</sup> DRP/mL N=31</b>
<b>Prednisone</b>	<b>11 (36%)</b>	<b>15 (46%)</b>	<b>10 (31%)</b>	<b>10 (32%)</b>
<b>Methotrexate</b>	<b>25 (81%)</b>	<b>23 (70%)</b>	<b>20 (63%)</b>	<b>24 (77%)</b>
<b>TNF-<math>\alpha</math> blocker</b>	<b>17 (55%)</b>	<b>20 (61%)</b>	<b>16 (50%)</b>	<b>17 (55%)</b>
<b>Etanercept</b>	<b>11</b>	<b>11</b>	<b>9</b>	<b>8</b>
<b>Infliximab</b>	<b>1</b>	<b>2</b>	<b>3</b>	<b>4</b>
<b>Adalimumab</b>	<b>5</b>	<b>7</b>	<b>4</b>	<b>5</b>
<b>Disease status</b>				
<b>Tender joints (28)</b>	<b>7.4 (0-26)</b>	<b>8.5 (0-25)</b>	<b>7.6 (0-27)</b>	<b>4.4 (0-16)</b>
<b>Swollen joints (28)</b>	<b>6.1 (0-18)</b>	<b>7.5 (0-21)</b>	<b>6.6 (0-23)</b>	<b>5.4 (0-27)</b>
<b>ESR (mm/hr)</b>	<b>29.8 (3-107)</b>	<b>21.4 (1-70)</b>	<b>25.6 (2-103)</b>	<b>25.8 (4-115)</b>

# Phase I/II Clinical Study

## Immunosuppressive Combinations

	<b>Placebo</b> N=31	<b>Low Dose</b> 1x10 <sup>11</sup> N=33	<b>Mid Dose</b> 1x10 <sup>12</sup> N=32	<b>High Dose</b> 1x10 <sup>13</sup> N=31
<b>No immunosuppressives</b>	<b>0</b>	<b>2 (6%)</b>	<b>2 (6%)</b>	<b>1 (3%)</b>
<b>Prednisone only</b>	<b>0</b>	<b>0</b>	<b>1 (3%)</b>	<b>0</b>
<b>Non-TNF DMARD only</b>	<b>10 (32%)</b>	<b>7 (21%)</b>	<b>9 (28%)</b>	<b>9 (29%)</b>
<b>Pred. + non-TNF DMARD</b>	<b>4 (13%)</b>	<b>4 (12%)</b>	<b>4 (13%)</b>	<b>4 (13%)</b>
<b>TNF antagonist alone</b>	<b>4 (13%)</b>	<b>5 (15%)</b>	<b>1 (3%)</b>	<b>3 (10%)</b>
<b>TNF + prednisone</b>	<b>1 (3%)</b>	<b>1 (3%)</b>	<b>2 (6%)</b>	<b>1 (3%)</b>
<b>TNF + non-TNF DMARD</b>	<b>7 (23%)</b>	<b>4 (12%)</b>	<b>10 (31%)</b>	<b>8 (26%)</b>
<b>TNF + non-TNF DMARD + prednisone</b>	<b>5 (16%)</b>	<b>10 (30%)</b>	<b>3 (9%)</b>	<b>5 (16%)</b>

# Phase I/II Clinical Study

## Study Agent Exposure

- **First dose: Blinded study agent – 127 subjects**
- **Second dose: Open label study agent – 74 subjects**
  - One dose of tgAAC94, preceded by placebo – 22 subjects
  - Two doses of tgAAC94 – 52 subjects

	Placebo	Low 1x10 <sup>11</sup> DRP/mL	Mid 1x10 <sup>12</sup> DRP/mL	High 1x10 <sup>13</sup> DRP/mL
<b>Blinded study agent</b>	31	33	32	31
<b>One dose of tgAAC94 preceded by placebo</b>	n/a	8	7	7
<b>Two doses of tgAAC94</b>	n/a	18	17	17

# Phase I/II Clinical Study

## Safety Overview

*Study unblinded for safety analyses*

<b>Adverse Events</b>	<b>Placebo</b>	<b>Low 1x10<sup>11</sup> DRP/mL</b>	<b>Mid 1x10<sup>12</sup> DRP/mL</b>	<b>High 1x10<sup>13</sup> DRP/mL</b>
<b>After 1<sup>st</sup> dose (n=127)</b>	<b>20 (65%)</b>	<b>26 (79%)</b>	<b>20 (63%)</b>	<b>20 (65%)</b>
<b>After 1 active dose preceded by placebo (n=22)</b>	<b>n/a</b>	<b>4 (50%)</b>	<b>6 (86%)</b>	<b>5 (71%)</b>
<b>After 2 active doses (n=52)</b>	<b>n/a</b>	<b>10 (56%)</b>	<b>12 (71%)</b>	<b>15 (88%)</b>

# Phase I/II Study: Serious Adverse Events

Dose Level	Most Recent Injection	Time Since Last Dose	Serious Adverse Event	Relationship per Investigator
1x10 <sup>11</sup>	1 <sup>st</sup>	5 weeks	Infected incision after repair of traumatic ankle fracture (target joint = wrist)	Unlikely
	1 <sup>st</sup>	13 weeks	Abdominal pain from constipation	Unlikely
	2 <sup>nd</sup>	1 week	Cellulitis & ulcer left calf (target joint = wrist)	Unlikely
	2 <sup>nd</sup>	13 weeks	Acute pyelonephritis	Unlikely
1x10 <sup>12</sup>	1 <sup>st</sup>	15 weeks	Septic arthritis	Probably
	2 <sup>nd</sup> (1st dose placebo)	8 weeks	Myocardial infarction	Unlikely
	2 <sup>nd</sup>	4 weeks	Pulmonary emboli	Unlikely
1x10 <sup>13</sup>	1 <sup>st</sup>	10 weeks	Syncope from coronary artery disease	Not related
	2 <sup>nd</sup>	10 days	Disseminated histoplasmosis	Possibly

# Phase I/II Clinical Study

## Rates of Infection

- Includes all types of infection
- Most commonly reported were upper respiratory infection, nasopharyngitis, sinusitis and urinary tract infection

<b>Infections</b>	<b>Placebo</b>	<b>Low 1x10<sup>11</sup> DRP/mL</b>	<b>Mid 1x10<sup>12</sup> DRP/mL</b>	<b>High 1x10<sup>13</sup> DRP/mL</b>
<b>After 1<sup>st</sup> dose (n=127)</b>	<b>11 (36%)</b>	<b>12 (36%)</b>	<b>8 (25%)</b>	<b>5 (16%)</b>
<b>After 1 active dose preceded by placebo (n=22)</b>	<b>n/a</b>	<b>1 (13%)</b>	<b>3 (43%)</b>	<b>3 (43%)</b>
<b>After 2 active doses (n=52)</b>	<b>n/a</b>	<b>6 (33%)</b>	<b>2 (12%)</b>	<b>4 (24%)</b>

# Phase I/II Clinical Study

## Serious Infections

Dose	Most Recent Injection	Time Since Last Dose	Serious Infection	Immuno-suppressives
$1 \times 10^{11}$	1 <sup>st</sup>	5 weeks	Infected incision after repair of traumatic ankle fracture (target joint = wrist)	Azathioprine
	2 <sup>nd</sup>	1 week	Cellulitis & ulcer left calf (target joint = wrist)	Leflunomide, prednisone
	2 <sup>nd</sup>	13 weeks	Acute pyelonephritis	Etanercept, methotrexate, prednisone
$1 \times 10^{12}$	1 <sup>st</sup>	15 weeks	Septic arthritis	Leflunomide
$1 \times 10^{13}$	2 <sup>nd</sup>	10 days	Disseminated histoplasmosis	Adalimumab, methotrexate, prednisone

# Phase I/II Clinical Study

## Elevated Liver Function Tests

*LFTs measured before each injection and 4, 8, 12, 18, 24 & 30 wks after*

- Mild LFT abnormalities
  - All Grade 1 (<2.5 x upper limit)
  - No Grade 2 or greater abnormalities
- ALT elevations in
  - 21/92 (23%) subjects on methotrexate
  - 2/35 (6%) subjects not on methotrexate

ALT Elevations	Placebo	Low 1x10 <sup>11</sup> DRP/mL	Mid 1x10 <sup>12</sup> DRP/mL	High 1x10 <sup>13</sup> DRP/mL
After 1 <sup>st</sup> dose (n=127)	2 (6%)	7 (21%)	3 (9%)	6 (19%)
After 1 active dose preceded by placebo (n=22)	n/a	1 (13%)	1 (14%)	0
After 2 active doses (n=52)	n/a	2 (11%)	2 (12%)	1 (6%)

Normal ranges: ALT 6-34 mg/dL; AST 9-34 mg/dL

# Subjects with LFTs $\geq 1.5 \times$ Upper Limit

Dose Level	Most Recent Injection	ALT AST	Other Meds	Timing / Outcome
Placebo	1 <sup>st</sup>	64 58	MTX	<ul style="list-style-type: none"> <li>• 18 weeks after 1<sup>st</sup> dose</li> <li>• Resolved spontaneously</li> </ul>
1x10 <sup>11</sup>	1 <sup>st</sup>	53 63	MTX	<ul style="list-style-type: none"> <li>• Labs from prior to 1<sup>st</sup> dose</li> <li>• History of episodic increases in LFTs, generally associated with MTX</li> <li>• MTX discontinued with subsequent decrease in LFTs to normal range</li> </ul>
1x10 <sup>12</sup>	2 <sup>nd</sup>	64 62	MTX Statin	<ul style="list-style-type: none"> <li>• 4 weeks after 2<sup>nd</sup> dose</li> <li>• MTX dose decreased; LFTs decreased</li> <li>• Rosuvastatin stopped; LFTs returned to normal</li> </ul>
1x10 <sup>13</sup>	1 <sup>st</sup>	60 39	MTX	<ul style="list-style-type: none"> <li>• 18 weeks after 1<sup>st</sup> dose</li> <li>• Resolved spontaneously</li> </ul>
1x10 <sup>13</sup>	2 <sup>nd</sup>	69 90	MTX	<ul style="list-style-type: none"> <li>• 8 weeks after 2<sup>nd</sup> dose</li> <li>• Resolved spontaneously</li> </ul>

Normal ranges: ALT 6-34 mg/dL; AST 9-34 mg/dL

# Phase I/II Clinical Study

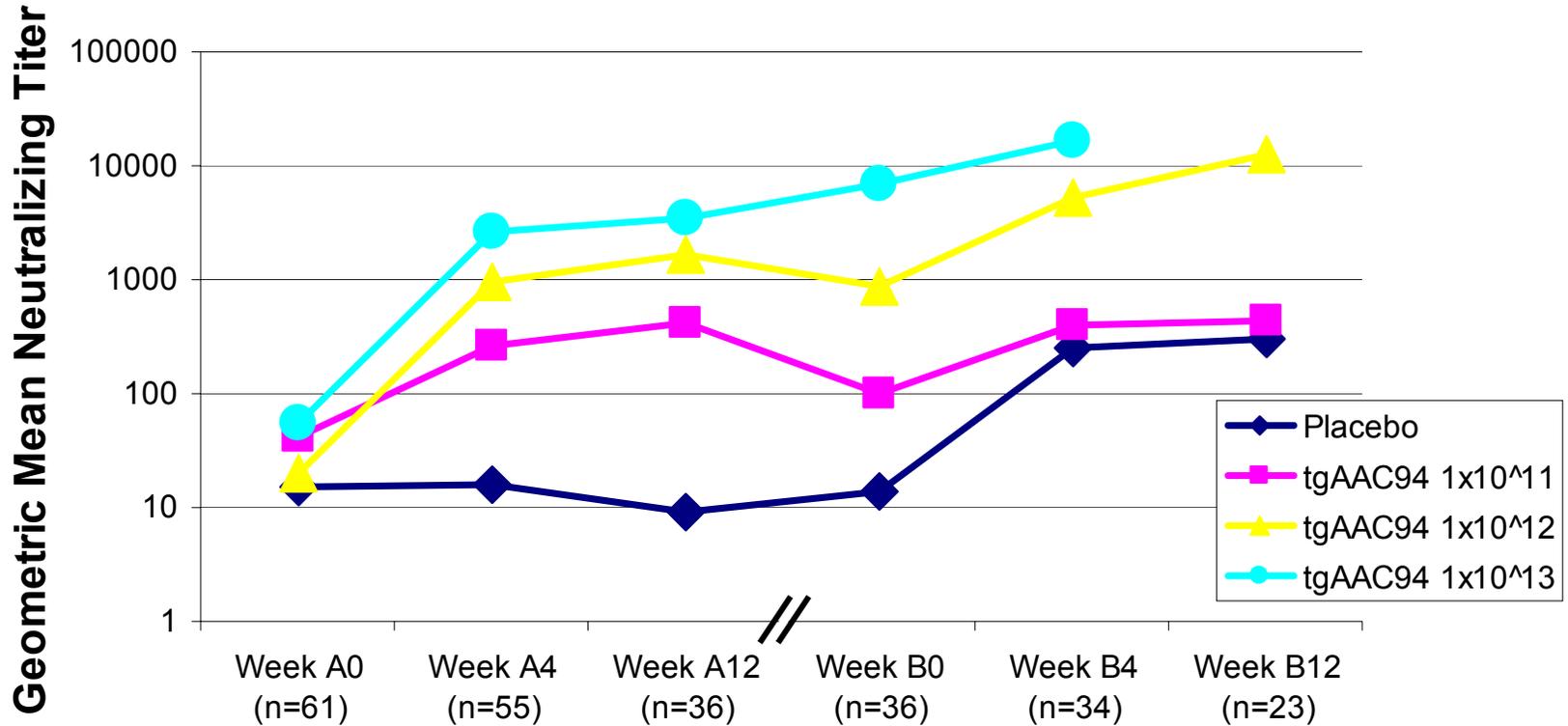
## Administration Site Reactions

- Increases in target joint tenderness & swelling
  - Occurred within 15 days of study agent administration
  - Sometimes accompanied by warmth, redness or itching
- More common after administration of high dose tgAAC94
- Rates varied with the joint injected, ranging from 4.3% (MCP) to 15.9% (wrist)
- Similar incidence in subjects on and off TNF- $\alpha$  antagonists

Administration Site Reactions	Placebo	Low 1x10 <sup>11</sup> DRP/mL	Mid 1x10 <sup>12</sup> DRP/mL	High 1x10 <sup>13</sup> DRP/mL
After 1 <sup>st</sup> dose (n=127)	1 (3%)	2 (9%)	0	6 (19%)
After 1 active dose preceded by placebo (n=22)	n/a	0	0	1 (14%)
After 2 active doses (n=52)	n/a	0	4 (24%)	4 (24%)

# Phase I/II Clinical Study

## Anti-AAV2 Capsid Neutralizing Titers



↑  
1st injection  
(tgAAC94 or placebo)

↑  
2nd injection  
(open label tgAAC94)

**Dose response seen**

<b>Subject 1209 – 1x10<sup>13</sup></b>	
Day A0	1/4
Week A4	1/16
Week A12	1/128
Day B0	1/128

# Vector Biodistribution

- Measured tgAAC94 in blood in
  - 8 subjects/dose level in  $10^{10}$  and  $10^{11}$  cohorts in 1<sup>st</sup> study
  - 10 subjects/dose level in  $10^{12}$  and  $10^{13}$  cohorts in 2<sup>nd</sup> study
- Assay
  - Limit of detection PCR assay for  $10^{10}$  –  $10^{12}$  dose levels
  - Developed quantitative PCR assay for  $10^{13}$  dose level
- Results reported as
  - Detectable = at limit of detection (7.5 copies/ $\mu$ g DNA)
  - Positive = higher than limit of detection ( $>7.5$  copies/ $\mu$ g DNA)
  - Quantifiable = higher than limit of detection and quantified at level indicated

# Vector Biodistribution

- **Quantifiable at low levels after receipt of highest dose of tgAAC94**
  - **Also detectable after receipt of lower doses of tgAAC94**
  - **No vector detected in placebo recipients (data not shown)**

<b>Dose Level (DRP/mL)</b>	<b># Pos or Det / # Actives tested</b>	<b>Results</b>
<b><math>1 \times 10^{10}</math></b>	<b>1/5</b>	<b>1 detectable 3 days after injection</b>
<b><math>1 \times 10^{11}</math></b>	<b>2/6</b>	<b>1 positive &amp; 1 detectable 3 days after injection</b>
<b><math>1 \times 10^{12}</math></b>	<b>6/7</b>	<b>1 positive up to 12 weeks 1 detectable up to 8 weeks 4 detectable at 1 week (n=3) or 12 weeks (n=1)</b>
<b><math>1 \times 10^{13}</math></b>	<b>8/8</b>	<b>4 quantifiable (18-75 copies/<math>\mu</math>g DNA) at 1 week 2 quantifiable (14-17 copies/<math>\mu</math>g DNA) at 4 weeks 1 positive &amp; 2 detectable up to 8 weeks</b>

# TNFR:Fc Expression

Radioimmunoassay at BioMonitor APS (Copenhagen, Denmark)

- Amount of functional TNF antagonist is quantitated by measuring the amount of  $^{125}\text{I}$ -TNFalpha:anti-TNF complexes precipitated after addition of anti-human IgG antibodies
- Standard curve constructed based on specific anti-TNF construct being assayed
- Method does not discriminate between different anti-TNFalpha-IgG constructs
- Limit of detection 0.01  $\mu\text{g}/\text{mL}$

Initial testing in Phase I/II subjects not on TNF antagonists

- **Serum** – NOT DETECTED in 8/8 subjects 4 & 12 weeks after injection with  $1 \times 10^{13}$  DRP/mL
- **Synovial fluid** – NOT DETECTED in 1/1 subjects 13 weeks after injection with  $1 \times 10^{12}$  DRP/mL

***Analysis of additional samples underway***

# Systemic Levels of TNF- $\alpha$ Inhibitors

<b>Systemic Administration of TNF Antagonists in Humans</b>	<b>Mean Steady State Drug Concentration (<math>\mu\text{g/mL}</math>)</b>
<b>Adalimumab</b> , 40 mg SC every other week with concomitant methotrexate	<b>8-9</b>
<b>Infliximab</b> , 3-10 mg/kg IV every 4-8 weeks	<b>0.5-6</b>
<b>Etanercept</b> , 25 mg SC twice a week	<b>2</b>

<b>Intra-articular delivery of tgAAC94</b>	<b>Systemic Levels of TNFR:Fc Protein (<math>\mu\text{g/mL}</math>)</b>
<b><math>1 \times 10^{11}</math>-<math>1 \times 10^{13}</math> DRP/mL in 102 rats</b>	<b>Up to 0.01 in 4 rats</b>
<b><math>1 \times 10^{11}</math>-<math>1 \times 10^{13}</math> DRP/mL in 6 monkeys</b>	<b>Up to ~0.07</b>
<b><math>1 \times 10^{13}</math> DRP/mL in 8 humans</b>	<b>Not detectable &lt; 0.01</b>

# Case Presentation

## Serum Adalimumab Levels

Date of serum collection	Timing	Result ( $\mu\text{g}/\text{mL}$ )
Feb 26, 2007	Prior to 1 <sup>st</sup> injection	5.4
Mar 28, 2007	4 weeks after 1 <sup>st</sup> injection	7.1
May 29, 2007	12 weeks after 1 <sup>st</sup> injection	8.6
Jun 2, 2007	Prior to 2 <sup>nd</sup> injection	8.9

**Expected mean steady state level on Adalimumab 40 mg SC every other week with concomitant methotrexate = 8-9  $\mu\text{g}/\text{mL}$**

# Serious Infection Rate

Biologic	Reference (number of patients)	Event/100 pt yr
None	Doran 2002; (609)	9.5; [odds ratio 1.88 95% CI 1.71-2.07 non RA cohort]
All anti-TNF therapies	Dixon 2006 (British registry); (7664)	5.1 [95% CI 4.5-5.8]
Etanercept	Listing 2005 (German registry); (1529)	6.4 [95% CI 4.5-9.1]
	Salliot 2007 (General practice France); (375)	12.3 ±102
	Wallis 2004 (RCT)	73.5/100,000 patients
Adalimumab	Schiff 2007 (RCT); (10,050)	5.1
	Salliot 2007; (97)	5.3±26
	Bongartz 2006 (RCT)	126/3493 patients [OR 2.01 95%CI 1.31-3.09 placebo]

# Reported Cases of Histoplasmosis in Patients Treated with TNF Antagonists

- Incidence of serious Histoplasmosis infection reported in RCT and post-marketing surveillance
  - Etanercept 3/113,000 patients<sup>1</sup>
  - Infliximab 39/233,000 patients<sup>1</sup>
  - Adalimumab 4/4,870 patients<sup>2</sup>
- Incidence of serious invasive opportunistic infections caused by fungi (e.g. histoplasma, aspergillus, and nocardia)
  - Adalimumab 6 /2334 patients<sup>4</sup>
  - Adalimumab 4 /182 patients<sup>3</sup>
  - Infliximab 7/276 patients<sup>3</sup>
  - Etanercept 7/455 patients<sup>3</sup>

1. Wallis 2004: AERS-FDA Jan 1998- Sept 2002

2. Schiff 2007: Clinical trial safety database (Dec 03-June 05) and post-marketing surveillance (Dec 02-June 06)

3. Salliot 2007: Dec 2004-March 2005 (France)

4. FDA Briefing document 2003 Arthritis Advisory committee meeting

**Intra-articular Administration of a  
Recombinant Adeno-Associated  
Vector containing a  
TNF- $\alpha$  Antagonist Gene in  
Inflammatory Arthritis**

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Austin, Texas

# Disclosures for E. Fudman

- Advisor, clinical investigator
  - Targeted Genetics
  - Abbott
  - Amgen
  - Bristol-Myers Squibb
  - Exagen Diagnostics
  - Genentech
  - Merck
  - Pfizer
  - Rigel
  - Roche
  - Scios
  - TAP
  - Targeted Genetics
  - Trubion
  - Wyeth

# Presentation Outline

- Local anti-TNF gene delivery using AAV
- Phase I clinical study
- Phase I/II clinical study
  - Study design and entry criteria
  - Safety
- Study recruitment
- Consent process

# Phase I/II Clinical Study Study Recruitment

- Tailored to individual site
  - Patient database
  - Referrals
  - IRB-approved advertisements
- Prescreening in compliance with HIPAA
  - Chart review
  - Telephone interview

# Phase I/II Clinical Study Consent Process

Conducted in accordance with

- 21 CFR 50 Protection of Human Subjects
- 21 CFR 312 Responsibilities of Sponsor and Investigators
- ICH Guidelines – Good Clinical Practice
- Guidelines for Research Involving Recombinant DNA Molecules (NIH-RAC Guidelines)
- HIPAA Regulations

# Phase I/II Clinical Study

## Elements of Informed Consent

- Basic elements
  - Description/purpose of the study
  - Risks and benefits
  - Alternative therapy
  - Confidentiality
  - Compensation in event of injury
  - Right to ask questions
  - Voluntary participation
- Additional elements required by RAC
  - Long-term follow-up
  - Autopsy request of family, if subject dies
  - Protection from the media

# Phase I/II Clinical Study Informed Consent Process

- The clinical investigator or designee must provide participant ample time and opportunity to:
  - Review and understand details of trial
  - Ask questions
  - Consult with family or others regarding participation
  - **UNDERSTAND** he/she may get placebo
  - **UNDERSTAND** there may be no benefit
- Use IRB-approved consent form
- Obtain consent prior to conducting any study-related procedures
- Provide subject with copy of signed consent