Global Development of Biosimilar Adalimumab
Zydus has taken a Two Phased Approach to Developing Biologics

Phase 1: Develop Biosimilars – Recombinant Proteins and Monoclonal Antibodies
   - Build Capabilities
   - Build a Development Engine
   - Build a Biosimilars Pipeline

Phase 2: Develop Novel Biologics and Antibody Drug Conjugates
   - Incremental improvement Novel Biologics or Biobetters
   - Novel Targets
   - Mabs Discovery Engine
Capabilities Built in Biologics: Bioreactor Process

Process Iteration  Process Optimisation  Pilot Process

Scale-up  Process Controls

Comparable Toxicity between Zydus Adalimumab and Reference Product in the key comparability toxicity study in rats
Capabilities Built in Biologics: Protein Purification

Purification Systems

Tangential Filtration

Columns

Centrifugation

Cloning  |  Bioreactor Process  |  Protein Purification  |  Characterization  |  Bioassays  |  Toxicity  |  Phase I  |  Phase III
Capabilities Built in Biologics: Characterization

- UPLC/HPLC
- Maldi Tof
- Circular Dichroism
- Gel Electrophoresis
- CZE
Zydus Biologicals Manufacturing Park: Biologics & Vaccines
Manufacturing Facilities
<table>
<thead>
<tr>
<th>Unit I</th>
<th>Unit II</th>
<th>Unit 9</th>
</tr>
</thead>
<tbody>
<tr>
<td>cGMP Drug Substance multi-product facility for Recombinant Proteins</td>
<td>cGMP Drug Substance multi-product facility for Monoclonal Antibodies</td>
<td>cGMP Drug Product multi-product facility for Biologics</td>
</tr>
<tr>
<td>G-CSF, PEGG-CSF, IFN alpha 2b, PEG IFN alpha 2b, PTH and EPO</td>
<td>Adalimumab, Trastuzumab, Bevacizumab and Rituximab etc.</td>
<td>Liquid and lyophilized vials, PFS and Cartridges</td>
</tr>
<tr>
<td>Scale 20L; 400 RBs</td>
<td>Two independent production streams with 2X 5KL and 1X 1KL bioreactors</td>
<td>Liquid/Lyophilized vial line - 1; PFS line - 1; Cartridge line – 1</td>
</tr>
</tbody>
</table>
Adalimumab – A Product for TNF-mediated Disease

- Adalimumab binds TNF-α so that it cannot bind to its receptor.
- Adalimumab is equipped to eliminate cells that produce TNF-α by ADCC/CDC or Apoptosis
Biosimilars: EMA Guidelines

• “A biosimilar is a biological medicinal product that contains a version of the active substance of an already authorised original biological medicinal product (reference medicinal product)”

• “A biosimilar demonstrates similarity to the reference medicinal product in terms of quality characteristics, biological activity, safety and efficacy based on a comprehensive comparability exercise.”

• “…the success of developing a biosimilar will depend on the ability to produce a close copy to the reference medicinal product…”
• “that the biological product is highly similar to the reference product notwithstanding minor differences in clinically inactive components”

• “there are no clinically meaningful differences between the biological product and the reference product in terms of the safety, purity and potency of the product”

• “The result of the comparative analytical characterization may lead to one of four assessments...: Not similar, Similar, Highly similar, or Highly similar with fingerprint-like similarity”

*FDA on Fingerprint like Similarity: “…analytical similarity based on integrated, multi-parameter approaches that are extremely sensitive in identifying analytical differences. ...”*

Adalimumab being a global program, we have designed it with a fingerprint like similarity to the reference product
Adalimumab – Process Adopted for Establishing Biosimilarity

Build a Strong Foundation of Analytical Characterization to Minimize the Dependence on Clinical Data for Establishing Biosimilarity
Demonstration of Biosimilar IDENTITY by Comparing Product Profile
IDENTITY - Polypeptide Profile & Molecular Weight

Polypeptide Profile by SDS-PAGE

Lane 1 – Molecular weight marker
Lane 2 – Reference Product
Lane 3 – Zydus Adalimumab

Profile By SDS-CE

Non-reducing SDS-CE

Zydus Adalimumab

Reference Product

Reducing SDS-CE

Zydus Adalimumab

Reference Product

Molecular Weight by Maldi-Tof

<table>
<thead>
<tr>
<th>Samples</th>
<th>Intact mass (kDa)</th>
<th>Heavy-chain mass (kDa)</th>
<th>Light-chain mass (kDa)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Reference Product</td>
<td>148.3</td>
<td>50.5</td>
<td>23.2</td>
</tr>
<tr>
<td>Zydus Adalimumab</td>
<td>148.3</td>
<td>50.5</td>
<td>23.2</td>
</tr>
</tbody>
</table>

Highly Comparable Polypeptide Profile and Molecular Weight
IDENTITY - Peptide Mapping
IDENTITY - Peptide Mapping

Peptide Mapping by RP-HPLC-UV Analysis of Tryptic Digests

Highly Comparable Tryptic Map & Sequence of the Tryptic Fragments

Zydus Adalimumab

Reference Product
Demonstration of Biosimilar STRUCTURE
Highly Comparable Secondary Structure & Overall Integrity. Further confirmed by Free Thiol and Disulphide Bridge Mapping
Carbohydrate structural elements directly impact Antibody Effector Function
Highly comparable glycan structures and monosaccharide content between Zydus Adalimumab and Reference Product – Comparable effector functions of ADCC & CDC
Highly Comparable EFFECTOR FUNCTIONS – Binding to FcγRIII

Surface Plasmon Resonance by BIACORE

Reference Product
1 – 62.5 nM; 2 – 125 nM; 3 – 250 nM; 4 – 500 nM
5 – 1000 nM; 6 – 2000 nM

Dissociation

Association

Zydus Adalimumab
1 – 62.5 nM; 2 – 125 nM; 3 – 250 nM; 4 – 500 nM
5 – 1000 nM; 6 – 2000 nM

Highly comparable ability to bind NK Cells – Comparable effector function of ADCC

Reference Product – KD 4.47 x 10⁻⁷
Zydus Adalimumab – KD 4.89 x 10⁻⁷
Highly Comparable EFFECTOR FUNCTIONS – Binding to TNF alpha

Highly comparable ability to bind & release TNF α – Comparable Neutralization
Highly Comparable STRUCTURAL COMPONENTS Impacting EFFECTOR FUNCTIONS – Charge Variant Profile

Highly comparable charge variant profile also confirmed by LCMS-MS & cIEF – Comparable Molecular Stability and Bioavailability
Highly Comparable Molecular and Process PURITY

- Highly comparable purity; very low aggregate content – Low Immunogenicity
- High level of process purity and hygiene (low ppm of HCD and low pg/mg of HCD)
Highly Comparable Process PURITY—Virus Free Cell Banks

A total of 18 *in vitro* and *in vivo* tests confirm cell banks to be free of viruses and any other adventitious agents [ICH Q5D and Q5A(R1)]

<table>
<thead>
<tr>
<th>S.No.</th>
<th>Test name</th>
<th>S.No.</th>
<th>Test name</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Sterility</td>
<td>10</td>
<td>In-vitro assay using CHO detector cells</td>
</tr>
<tr>
<td>2</td>
<td>Mycoplasma by culture</td>
<td>11</td>
<td>In- vivo assay using Adult mice</td>
</tr>
<tr>
<td>3</td>
<td>Mycoplasma by PCR</td>
<td>12</td>
<td>In- vivo assay using Suckling mice</td>
</tr>
<tr>
<td>4</td>
<td>Isoenzyme Analysis</td>
<td>13</td>
<td>In-vitro assay for detection of Bovine virus</td>
</tr>
<tr>
<td>5</td>
<td>Transmission Electron Microscopy</td>
<td>14</td>
<td>In-vitro assay for detection of porcine virus</td>
</tr>
<tr>
<td>6</td>
<td>Embryonated egg assay by Allontoic route</td>
<td>15</td>
<td>Infectivity assay for retrovirus (S+L-)</td>
</tr>
<tr>
<td>7</td>
<td>Embryonated egg assay by Yolk sack route</td>
<td>16</td>
<td>Hamster Antibody Production</td>
</tr>
<tr>
<td>8</td>
<td>In- vitro assay using MRC-5 detector cells</td>
<td>17</td>
<td>Gene Sequencing</td>
</tr>
<tr>
<td>9</td>
<td>In- vitro assay using Vero detector cells</td>
<td>18</td>
<td>Gene Copy number</td>
</tr>
</tbody>
</table>

**Table:**

- **Sterility**
- **Mycoplasma by culture**
- **Mycoplasma by PCR**
- **Isoenzyme Analysis**
- **Transmission Electron Microscopy**
- **Embryonated egg assay by Allontoic route**
- **Embryonated egg assay by Yolk sack route**
- **In- vitro assay using MRC-5 detector cells**
- **In- vitro assay using Vero detector cells**
- **In- vitro assay using CHO detector cells**
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- **Gene Sequencing**
- **Gene Copy number**
## Comparable TOXICITY Findings

<table>
<thead>
<tr>
<th>Study Title</th>
<th>Doses</th>
<th>Observations</th>
</tr>
</thead>
<tbody>
<tr>
<td>Repeated Dose Toxicity Study in Rats by Subcutaneous Route</td>
<td>Zydus Adalimumab: 0, 4.1, 20.5 and 41 mg/kg/week</td>
<td>• No mortality and adverse clinical signs of toxicity up to 41 mg/kg&lt;br&gt; • Neurobehavioral &amp; Ophthalmic Examination, Clinical examination, Body Weight &amp; Feed Intake, Hematology, Serum Biochemistry &amp; Urinalysis, Gross Pathology, Organ Weight, Bone marrow examination, Histopathology – All Normal up to the high dose of 41 mg/kg.</td>
</tr>
<tr>
<td></td>
<td>ReferenceProduct: 4.1 mg/kg/week</td>
<td>• Comparable with Reference Product&lt;br&gt; • Immunogenicity - Comparable between Adalimumab treated and reference product treated groups</td>
</tr>
<tr>
<td></td>
<td>Weekly once for four weeks</td>
<td></td>
</tr>
</tbody>
</table>

### Comparable Toxicity between Zydus Adalimumab and Reference Product in the key comparability toxicity study in rats
Comparable TOXICITY Findings

Similar Observations in the remaining Toxicity Studies

1. Acute Toxicity in Mice by Subcutaneous Route
2. Acute Toxicity in Rats by Subcutaneous Route
3. Acute Toxicity in Mice by Intravenous Route
4. Acute Toxicity in Rats by Intravenous Route
5. Skin Sensitization Study in Guinea Pigs
6. Comparability Study - Repeated Dose Toxicity & Immunogenicity in Rabbits by Subcutaneous Route

Zydus Adalimumab found to be safe and similar to the Reference Product
Robust Manufacturing Process is in Place
Robust Manufacturing Process is in Place
SUMMARY & CONCLUSIONS

- World’s First Adalimumab Biosimilar
- Zydus is working with leading CROs to develop Adalimumab as a global product. India is the first country of launch
- Meeting with US FDA planned for Q1 2015
- Zydus Adalimumab is a highly biosimilar product developed to have a fingerprint-like match with reference product - established using a panel of more than 20 analytical assays
- A highly comparable animal toxicity has been established
- A highly comparable clinical profile has been established
- A highly robust manufacturing process has been established
**Abridged Prescribing Information**

**COMPOSITION:** Exemptia™ (Adalimumab) 40 mg /0.8 mL single use pre filled syringe and 20 mg /0.4 mL single use pre filled syringe

**DESCRIPTION:** EXEMPTIATM (Adalimumab) is a recombinant human IgG1 monoclonal antibody specific for human tumor necrosis factor (TNF-α). EXEMPTIATM is supplied as a sterile, preservative-free solution of Adalimumab for subcutaneous administration. The solution of EXEMPTIATM is clear and colorless. **MECHANISM OF ACTION:** Adalimumab binds specifically to TNF-alpha and blocks its interaction with the p55 and p75 cell surface TNF-α receptors. Adalimumab also lyses surface TNF expressing cells in vitro in the presence of complement. Elevated levels of TNF-α is found in the synovial fluid of rheumatoid arthritis, psoriatic arthritis, and ankylosing spondylitis patients and play an important role in both the pathologic inflammation and the joint destruction that are hallmarks of these diseases. **INDICATIONS & DOSAGE:** Rheumatoid Arthritis, Psoriatic Arthritis, and Ankylosing Spondylitis: The recommended dose of EXEMPTIATM for adult patients with rheumatoid arthritis (RA), psoriatic arthritis (PsA), or ankylosing spondylitis (AS) is 40 mg subcutaneously administered every other week. Methotrexate (MTX), other non-biologic DMARDs, glucocorticoids, nonsteroidal anti-inflammatory drugs (NSAIDs), and/or analgesics may be continued during treatment with EXEMPTIATM. Juvenile Idiopathic Arthritis: Exemptia™ dosing in JIA is based on weight; for 10 kg (22 lbs) to <15 kg (33 lbs): 10 mg s.c. every other week. For 15 kg (33 lbs) to < 30 kg (66 lbs): 20 mg s.c. every other week and for ≥ 30 kg (66 lbs): 40 mg s.c. every other week. Plaque Psoriasis or Non-Infectious Uveitis: Initial dose of 80 mg, followed by 40 mg every other week starting from week one after initial dose. Hidradenitis Suppurativa: 160 mg (Day 1) (four 40 mg injections in one day or two 40 mg injections per day for two consecutive days), followed by 80 mg two weeks later (Day 15). Two weeks later (Day 29) begin a maintenance dose of 40 mg every week. Adult Crohn's Disease and Ulcerative Colitis: Initial dose (Day 1): 160 mg s.c. (four 40 mg injections in one day or two 40 mg injections per day for two consecutive days). Second dose two weeks later (Day 15): 80 mg. Two weeks later (Day 29): Begin a maintenance dose of 40 mg s.c. every other week. For patients with Ulcerative Colitis only: Only continue EXEMPTIATM in patients who have shown evidence of clinical remission by eight weeks (Day 57) of therapy. Pediatric Crohn’s Disease: For weight 17 kg (37 lbs) to < 40 kg (88 lbs): Initial dose (Day 1): 80 mg s.c. (two 40 mg injections in one day). Second dose two weeks later (Day 15): 40 mg s.c.. Two weeks later (Day 29): Begin a maintenance dose of 20 mg s.c. every other week. For ≥ 40 kg (88 lbs): Initial dose (Day 1): 160 mg s.c. (four 40 mg injections in one day or two 40 mg injections per day for two consecutive days). Second dose two weeks later (Day 15): 80 mg s.c. (two 40 mg injections in one day). Two weeks later (Day 29): Begin a maintenance dose of 40 mg s.c. every other week. **CONTRAINDICATIONS:** Hypersensitivity to the active substance or to any of the excipients, Moderate to severe heart failure, Active tuberculosis or other severe infections such as sepsis and opportunistic infections. **SPECIAL WARNINGS AND PRECAUTIONS:** Serious and fungal infections: Do not start EXEMPTIATM during an active infection. If an infection develops, monitor carefully, and stop EXEMPTIATM if infection becomes serious. Anaphylaxis or serious allergic reactions may occur. Hepatitis B virus reactivation: Monitor HBV carriers during and several months after therapy. If reactivation occurs, stop EXEMPTIATM and begin antiviral therapy. Demyelinating disease: Exacerbation or new onset, may occur. Heart failure: Worsening or new onset, may occur. Lupus-like syndrome: Stop EXEMPTIATM if syndrome develops **USE IN PREGNANCY AND LACTATION:** Pregnancy Category B: Adequate and well controlled studies with EXEMPTIATM have not been conducted in pregnant women. Adalimumab is an IgG1 monoclonal antibody and IgG1 is actively transferred across the placenta during the third trimester of pregnancy. Lactation: No data is available on the absorption of adalimumab from breast milk in newborns or preterm infants. Caution should be exercised when EXEMPTIATM is administered to a nursing woman. **DRUG INTERACTION:** Biological Products- Concomitant administration of EXEMPTIATM with other biologic DMARDs (e.g., Anakinra and Abatacept) or other TNF blockers is not recommended. Live Vaccines- Avoid the use of live vaccines with EXEMPTIATM. Cytochrome P450 Substrates- The formation of CYP450 enzymes may be suppressed by increased levels of cytokines (e.g., TNFα, IL-6) during chronic inflammation. Upon initiation or discontinuation of EXEMPTIATM in patients being treated with CYP450 substrates with a narrow therapeutic index, monitoring of the effect (e.g., Warfarin) or drug concentration (e.g., Cyclosporine or Theophylline) is recommended and the individual dose of the drug product may be adjusted as needed. **UNDESIRED EFFECTS:** The most serious adverse reactions include the following. Serious Infections- Tuberculosis and Opportunistic Infections- Malignancies. The Clinical experience has reported Upper Respiratory Tract Infection (URTI), Increased creatine phosphokinase, Headache, Rash, Sinusitis, Nausea, Urinary Tract Infection (UTI), Abdominal pain, Flulike syndrome, Hyperlipidemia, Back pain, Hypercholesterolemia, Hematuria, Hypertension, Increased alkaline phosphatase as common side effects. **STORAGE CONDITION:** Store between + 2°C and + 8°C, in the carton to protect from light. Do not freeze Exemptia™. Do not use Exemptia™ if frozen, even if it has been thawed. Keep out of reach of children. **PRESENTATION:** a) Injection: 40 mg /0.8 mL in a single-use prefilled syringe b) Injection: 20 mg /0.4 mL in a single-use prefilled syringe.

Please refer to the full Prescribing Information before starting EXEMPTIATM.
Please consult full Prescribing Information before prescribing.

Zydus Cadila does not recommend the use of any product in any different manner than as described in the prescribing information.

Further information is available on request from:

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Ahmedabad – 382 481
Gujarat, India.
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Thank you

Zydus

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