Application of drugs based on release-active antibodies as immunotherapy agents

OOO “NPF “Materiia Medica Holding”,
Moscow, Russia
Monoclonal antibodies approved for therapeutic use

<table>
<thead>
<tr>
<th>Scientific name</th>
<th>Trade name</th>
<th>Origin and isotype</th>
<th>Target</th>
<th>Licensed indication(s) and year of first approval-region</th>
</tr>
</thead>
<tbody>
<tr>
<td>Capromab pendetide</td>
<td>Prostatecint</td>
<td>Murine IgG1</td>
<td>PSA*</td>
<td>Diagnostic imaging (1996-US)</td>
</tr>
<tr>
<td>Muromonab-CD3</td>
<td>Orthoclone OKT3</td>
<td>Murine IgG2</td>
<td>CD3</td>
<td>Transplant rejection (1992-US)</td>
</tr>
<tr>
<td>Tositumumab iodine 131</td>
<td>Bexxar</td>
<td>Murine IgG2</td>
<td>CD20</td>
<td>NHL® (2003-US)</td>
</tr>
<tr>
<td>Basiliximab</td>
<td>Simulect</td>
<td>Chimeric IgG1</td>
<td>CD25</td>
<td>Prophylaxis for transplant rejection (1998-US)</td>
</tr>
<tr>
<td>Brentuximab vedotin</td>
<td>Adcetris</td>
<td>Chimeric IgG1</td>
<td>CD30 ADC*</td>
<td>ALCL® and Hodgkin lymphoma (2011-US)</td>
</tr>
<tr>
<td>Gemtuzumab</td>
<td>Removab</td>
<td>Chimeric IgG2a/b</td>
<td>CD33, EpCAM*</td>
<td>Malignant ascites (2009-EU)</td>
</tr>
<tr>
<td>Geftinab</td>
<td>Erbitux</td>
<td>Chimeric IgG1</td>
<td>EGFR*</td>
<td>Colonctal, head and neck cancer (2004-US, EU)</td>
</tr>
<tr>
<td>Rituximab</td>
<td>Rituxan, MabThera</td>
<td>Chimeric IgG1</td>
<td>CD20</td>
<td>B-CLL (1997-US, 1998-EU)</td>
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<tr>
<td>Alemtuzumab</td>
<td>Campath</td>
<td>Humanized IgG1</td>
<td>CD52</td>
<td>B-CLL (2001-US)</td>
</tr>
<tr>
<td>Bevacizumab</td>
<td>Avastin</td>
<td>Humanized IgG1</td>
<td>VEGF</td>
<td>Colorectal, lung, breast cancer (2004-US, 2005-EU)</td>
</tr>
<tr>
<td>Elotuzumab</td>
<td>Repliva</td>
<td>Humanized IgG1</td>
<td>CD381a</td>
<td>Myeloma (2011-US, 2012-EU)</td>
</tr>
<tr>
<td>Gantuzumab ozogamycin</td>
<td>Mylotarg</td>
<td>Humanized IgG4</td>
<td>CD33 AAD</td>
<td>Leukemia (2000-US)</td>
</tr>
<tr>
<td>Olizumab</td>
<td>Xolar</td>
<td>Humanized IgG1</td>
<td>IgE</td>
<td>Severe asthma (2003-US, 2005-EU)</td>
</tr>
<tr>
<td>Palivizumab</td>
<td>Synagis</td>
<td>Humanized IgG1</td>
<td>RSV* F protein</td>
<td>Prevention of RSV infection in neonates (1998-US)</td>
</tr>
<tr>
<td>Ramucirumab</td>
<td>Lucentis</td>
<td>Humanized IgG1</td>
<td>VEGF</td>
<td>Micrular degeneration (2006-US, 2007-EU)</td>
</tr>
<tr>
<td>Tecartelumab</td>
<td>Actecra</td>
<td>Humanized IgG1</td>
<td>IL-1R™</td>
<td>Caudal regression syndrome (2010-US, 2009-EU)</td>
</tr>
<tr>
<td>Trastuzumab</td>
<td>Herceptin</td>
<td>Humanized IgG1</td>
<td>HER-2™</td>
<td>HER-2 positive breast cancer (1998-US)</td>
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<tr>
<td>Canakinumab</td>
<td>Ilaris</td>
<td>Human IgG1</td>
<td>IL-1β*</td>
<td>Muckle-Wells syndrome (US, EU-2009)</td>
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<tr>
<td>Denosumab</td>
<td>Prolia, Xgeva</td>
<td>Human IgG2</td>
<td>RANKL*</td>
<td>Osteoporosis, bone metastasis (2009-US, EU)</td>
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<tr>
<td>Golimumab</td>
<td>Simponi</td>
<td>Human IgG1</td>
<td>TNF</td>
<td>RA, psoriatic arthritis, ankylosing spondylitis (2009-US, EU)</td>
</tr>
<tr>
<td>Iplimumab</td>
<td>Yervoy</td>
<td>Human IgG1</td>
<td>CTLA4*</td>
<td>Advanced melanoma (2011-US, EU)</td>
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<tr>
<td>Ottolimumab</td>
<td>Arzerra</td>
<td>Human IgG1</td>
<td>CD20</td>
<td>CLL (2000-US, 2010-EU)</td>
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<tr>
<td>Pantolimus</td>
<td>Vedlizumab</td>
<td>Human IgG2</td>
<td>EGFR</td>
<td>Colonctal cancer (2007-US, EU)</td>
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<tr>
<td>Ustekinumab</td>
<td>Solara</td>
<td>Human IgG1</td>
<td>IL-12p40*</td>
<td>Plaque psoriasis (2009-US, EU)</td>
</tr>
<tr>
<td>Pertuzumab</td>
<td>Perjeta</td>
<td>Humanized IgG1</td>
<td>HER-2™</td>
<td>HER-2 positive breast cancer (2012-US)</td>
</tr>
</tbody>
</table>

Buss NAPS et all// Current Opinion in Pharmacology, 2012
## Limitations in mAbs-based therapies

<table>
<thead>
<tr>
<th>Attributes</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Production</strong></td>
<td>• Safety of biological material used</td>
</tr>
<tr>
<td></td>
<td>• Potential cancer risk in case of EBV introduction</td>
</tr>
<tr>
<td></td>
<td>• Generating stoichiometric amounts of heavy and light chains</td>
</tr>
<tr>
<td></td>
<td>• Glycosylation profile</td>
</tr>
<tr>
<td><strong>Cost</strong></td>
<td>• Production cost is 300-500 million euros</td>
</tr>
<tr>
<td></td>
<td>• Annual cost of therapy is US$100 000 per patient</td>
</tr>
<tr>
<td><strong>Pharmacokinetics</strong></td>
<td>• Rapid elimination of mAbs</td>
</tr>
<tr>
<td></td>
<td>• Non-linear PK and PD profile</td>
</tr>
<tr>
<td></td>
<td>• Effect of demographic variables on mAbs PK</td>
</tr>
<tr>
<td></td>
<td>• Tissue penetration properties</td>
</tr>
<tr>
<td><strong>Route of administration</strong></td>
<td>• Intra-venous infusions</td>
</tr>
<tr>
<td><strong>Safety</strong></td>
<td>• Immunogenicity</td>
</tr>
<tr>
<td></td>
<td>• Side-effects</td>
</tr>
</tbody>
</table>

*H. Samaranayake et al, Annals of Medicine, 2009*
Antibodies engineering to safety and efficacy increase

P. Chames et al, British Journal of Pharmacology, 2009
Release-active antibodies

**Method:** A combination of multiple rounds of consequent decrease in the substance’s initial concentration and physical treatment

**Substance:** Rabbit polyclonal antibodies to different targets

**Level of Abs concentration reduction:** $10^{24}$

The technology was discovered by **Prof. Dr. Oleg Epstein** (Epstein O.I., 1999, Epstein O.I. et al, Usp. Fiziol. Nauk, 2013).
**Ligand-receptor binding**

Regular process

<table>
<thead>
<tr>
<th>Ligand</th>
<th>Receptor</th>
</tr>
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<tbody>
<tr>
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</table>

<table>
<thead>
<tr>
<th>Cell signalling</th>
</tr>
</thead>
</table>

**How monoclonal antibodies work?**

mAbs **neutralize** the targets

<table>
<thead>
<tr>
<th>Antibody</th>
<th>Receptor</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
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</table>

<table>
<thead>
<tr>
<th>X</th>
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</table>

<table>
<thead>
<tr>
<th>Cell signalling prevented</th>
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</thead>
</table>

**How release-active antibodies work?**

RA-Abs **modify** the target

<table>
<thead>
<tr>
<th>RA-Abs</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
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</table>

<table>
<thead>
<tr>
<th>Modified target</th>
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</thead>
</table>

<table>
<thead>
<tr>
<th>Receptor</th>
</tr>
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<tbody>
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</table>

<table>
<thead>
<tr>
<th>Cell signalling</th>
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</table>

<table>
<thead>
<tr>
<th>RA-Abs</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
</tr>
</tbody>
</table>
Study of influence RA of Ab to IFNγ on interferon-gamma by NMR

Preclinical site: Department of Structural Biology University of Pittsburgh School of Medicine (USA)

Design: $^{5}\text{N-}^{1}\text{H-HSQC}$ spectra of IFN-γ in absence (colored in blue) and present of RA of Ab to IFN-γ or placebo (colored in red)

2D HSQC Spectrum of IFNg

IFNg + placebo

IFNg + RA of Ab to IFNg

IFNg + vehicle

IFNg + RA Abs to IFNg

Conclusion: Addition of concentrated RA of Abs to IFNγ causes specific chemical shift changes in two-dimensional $^{1}\text{H,}^{15}\text{N-HSQC}$ spectra of $^{15}\text{N}$-labeled IFNg, located primarily at the IFNg dimer interface
Study of influence RA of Ab to IFNγ on interferon-gamma

**Site:** Department of Structural Biology University of Pittsburgh School of Medicine (USA)

**Conclusion:** The interferon-gamma sites which affected by Anaferon for children were founded
Study of influence RA of Ab to IFNγ on interferon-gamma

**Site:** Euroscreen (Belgium)

**Assay:** Radioligand binding assay

*Increasing of specific binding of \[^{125}\text{I}]\text{IFN}\gamma\text{with IFN}\gamma\text{receptor}

(\% vs control)*

*\(p<0.05\) vs vehicle

**Conclusion:** RA Abs to IFN gamma significantly increase the specific binding of IFNg with IFNg receptor
### List of main Materia Medica drugs based on RA antibodies

<table>
<thead>
<tr>
<th>Product</th>
<th>Lead Indications</th>
<th>Target</th>
</tr>
</thead>
<tbody>
<tr>
<td>Propoten-100</td>
<td>Alcoholism</td>
<td>S-100 protein</td>
</tr>
<tr>
<td>Anaferon</td>
<td>Influenza, upper respiratory infections</td>
<td>IFNγ</td>
</tr>
<tr>
<td>Anaferon for children</td>
<td>Influenza, upper respiratory infections</td>
<td>IFNγ</td>
</tr>
<tr>
<td>Impaza</td>
<td>Erectile Dysfunction</td>
<td>NO synthase</td>
</tr>
<tr>
<td>Arthrofon</td>
<td>Rheumatoid arthritis</td>
<td>TNFα</td>
</tr>
<tr>
<td>Tenoten</td>
<td>Anxiety/Depression, other CNS disorders</td>
<td>S100 protein</td>
</tr>
<tr>
<td>Afala</td>
<td>First and second stage BPH</td>
<td>PSA</td>
</tr>
<tr>
<td>Tenoten for children</td>
<td>Anxiety, Depression, Attention Deficit Disorder</td>
<td>S100 protein</td>
</tr>
<tr>
<td>Dietressa</td>
<td>Obesity</td>
<td>CB1 receptor</td>
</tr>
<tr>
<td>Colofort</td>
<td>Inflammatory Bowel Syndrome</td>
<td>Histamine, S100 protein, TNFα</td>
</tr>
<tr>
<td>Brizantin</td>
<td>Cigarette Smoking</td>
<td>CB1 receptor, S100 protein</td>
</tr>
</tbody>
</table>
AB Biotechnology facility, UK

Materia Medica Holding facility, RF

Manufacture

Antigen production:
Recombinant protein production

Immunization of SPF rabbits and antiserum preparation

Affinity purified antibodies preparation:
Delipidization of antiserum
Viral inactivation stages
Affinity chromatography

Rabbit polyclonal affinity purified antibodies – Drug Substance

Preparation of Drug Substance release-active dilutions

Preparation of Drug Product dosage form

Orodispersible tablets
RA antibodies-based drugs preclinical studies

- Toxicological studies
- Pharmacological activity studies
- Mechanism of action studies

- Well planned and controlled
- Performing in accordance with current requirement
- Performed by world leading research institutions and CRO

Main European collaborators of Materia Medica

- University Paris-Diderot, (France)
- Apcis SA (France)
- Pelvipharm (France)
- Syncosome (France)
- CEREPSA (France)
- Parsolt & Partners Pharmacology (France)
- Urosphere S.A.S (France)
- Eurosreen S.A. (Belgium)
- U-CyTech biosciences (the Netherlands)
- BioMedCode (Greece)
- MD Biosciences (Switzerland)
- Orthotopix Ltd (Finland)
- Pharmahungary 2000 Ltd (Hungary)
- Rheoscience A/S (Denmark)

- University of Pittsburgh (USA)
- Southern Research Institute (USA)
- Bhat Bio-tech India (P) Ltd (India)
- Monash University (Australia)
- PRECOS (UK)
- HD Biosciences Co., Ltd (China)
- Novascreen Biosciences Corporation, (USA)
- Dabur Research Foundation, (India)
- London Imperial College, (UK)
- Ricerca Biosciences, LLC (USA)
- Zen-Bio Inc. (USA)
- University of Tromsø (Norway)
- Utah State University (USA)
RA antibodies-based drugs clinical studies

Clinical sites location:
✓ Russian Federation
✓ Commonwealth of Independent States
✓ Asia

Number of clinical sites:
> 100, one-site and multi-central studies

Design of studies:
Double Blind Randomized Placebo Control or Comparative

Number of patients enrolled:
About 30,000
RA antibodies-based drugs clinical studies

<table>
<thead>
<tr>
<th>Rank</th>
<th>Status</th>
<th>Study</th>
<th>Conditions</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Recruiting</td>
<td>Clinical Trial of Safety and Clinical Efficiency of Ergoferon in Liquid Dosage Form In Treatment of Acute Upper Respiratory Tract Infections In Adult Patients</td>
<td>Acute Upper Respiratory Tract Infections</td>
</tr>
<tr>
<td>2</td>
<td>Active, not recruiting</td>
<td>Clinical Trial of Efficacy of Anferon for Children® in the Treatment of Influenza and Acute Respiratory Viral Infections In Children</td>
<td>Influenza and Acute Respiratory Viral Infections</td>
</tr>
<tr>
<td>3</td>
<td>Recruiting</td>
<td>Clinical Trial of Safety and Clinical Efficiency of Ergoferon in Liquid Dosage Form In Treatment of Acute Upper Respiratory Tract Infections In Children</td>
<td>Acute Upper Respiratory Tract Infections</td>
</tr>
<tr>
<td>4</td>
<td>Recruiting</td>
<td>Clinical Trial of Safety and Efficiency of AlfaLazza in Patients With Symptoms of Benign Prostatic Hyperplasia and Risk of Progression</td>
<td>Benign Prostatic Hyperplasia</td>
</tr>
<tr>
<td>5</td>
<td>Completed</td>
<td>Clinical Trial of Safety and Efficiency of Various Dosage Schedules for Dietessa Drug in Treatment of Obese Patients</td>
<td>Obesity</td>
</tr>
<tr>
<td>6</td>
<td>Recruiting</td>
<td>Clinical Trial of Efficiency and Safety of Subetta In the Combined Treatment of Patients With Type II Diabetes Mellitus</td>
<td>Type II Diabetes Mellitus</td>
</tr>
</tbody>
</table>
Activity of ultra-low doses of a influenza A(H1N1)2009 virus ii

Sergey A. Tarasov1,2, Madison C. Zamboni1,2, Oleg I. Epstein1

1Department of Pediatrics, 2Department of Microbiology and Immunology, and 3Department of Pathology, University of Rochester School of Medicine and Dentistry, Rochester, New York, USA

Research Article
The Novel Oral Drug Subetta Exerts an Antidiabetic Effect in the Diabetic Goto-Kakizaki Rat Model in Comparison with Rosiglitazone

Danielle Balletti1,3, Ewamn Phil Ogbi Epstein,4, and Bernard P. Pralong5

Laboratory of Endocrinology, University of Rochester School of Medicine and Dentistry, Rochester, New York, USA

Abstract
Application of a heterogeneous immunosassay for the quality control testing of release-active forms of diclofenac
Michael Petkovic6, Jelena S. Gavrilovska,7 Sergey A. Tarasov,8 Karin Kopp9,10

Published by De Gruyter, 2015

A RANDOMIZED, OPEN-LABEL, COMPARATIVE, 6-MONTH TRIAL OF ORAL ULTRA-LOW DOSES OF ANTIBODIES TO TUMOR NECROSIS FACTOR- \( \alpha \) AND DICLOFENAC IN RHENUMATOID ARTHRITIS

DUGINA J. L., PETROV V. L., BABAeva A. R., MARTYUSHKOVOKLAD A. V., TOCHEREVOKOVA E. V., EPSTEIN O. I., SERGEeva S. A.

Published by De Gruyter, 2015

Drug Watch

INT. J. TISSUE REACT. VOL. 33(1) 15-21 (2006)

Bakulinov, A. L., Babtysievich, S. I., Stepanov, V. I.

Published by De Gruyter, 2015
RA Abs to be discussed:

**RA Abs to IFN-gamma (IFNg)**

IFN-gamma is a cytokine with antiviral and antiparasitic activities, inhibiting the proliferation of a number of cells.

**RA Abs to S-100 protein (S100)**

S-100 protein is a Ca\(^{2+}\)-binding neuropeptide which play an important role in appearance of depression, anxiety etc.

**RA Abs to endothelial NO synthase (eNOS)**

Endothelial NO synthase is an enzyme catalyzing the production of nitric oxide. This enzyme is one of the responsible for appearance of sexual motivation.
Efficacy of RA of Abs to interferon-gamma (IFNg): Antiviral activity against DNA-viral infections

Preclinical site: Research institute for epidemiology and microbiology (Russia)

Model: genital herpes (herpes simplex virus strain type 2) in guinea pigs

Conclusion: There were decreases of the virus titer and points of a clinical signs in RA Abs to IFNg group in comparison with control and reference standard group

Epstein O.I., Vestnik MAN RS, 2008
Efficacy of RA Abs to IFNg:
antiviral activity against RNA-viral infections

Preclinical site: APCis S.A. (France)
Virus: influenza A/Equine/2/Miami/1/63 (H3N8)

**Conclusion:** Survival rate in RA Abs to IFNg group was comparable to Oseltamivir group

Submitted for publication
Clinical studies of RA Abs to IFNg
Antiviral activity against DNA-viral infection

**Site:** Tomsk Institute of pharmacology (Russia)

**Design:** Randomized double blind placebo-controlled in 236 patients (1-18 y.o.) with chickenpox

**The duration of fever**

![Graph showing the duration of fever for Ra Abs to IFNg and Placebo over 9 days.](image)

**Percent of patients with itch**

![Graph showing the percent of patients with itch for Ra Abs to IFNg and Placebo over 9 days.](image)

**Conclusion:** Administration of Ra Abs to IFNg reduced the duration of fever at 2.7 days, the time of appearance of new lesions - 3.3 days, itch - 4.2 days.

Kudin M.V. et al, CHILDREN INFECTIONS, 2007
Clinical studies of RA Abs to IFNg
Antiviral activity against RNA-viral infections

Site: Influenza institute (Russia); Russian State Medical University (Russia)
Design: Randomized double blind placebo-controlled in 255 patients (0.5 – 14 y.o.)
with upper respiratory viral infections

**Duration of major clinical signs (days)**

> **Conclusion:** Administration of RA Abs to IFNg significantly reduced the duration of clinical signs in comparison with placebo
Efficacy of RA Abs to S-100 protein: anxiolytic activity

**Preclinical site:** Porsolt & Partners Pharmacology (France)

**Model:** Vogel conflict test

**Number of shocks (% vs vehicle)**

- **2.5 ml/kg**
- **5.0 ml/kg**
- **7.5 ml/kg**
- **10.0 ml/kg**

* p<0.05 vs vehicle

**Conclusion:** There are significant increase in a numbers of shocks in RA Abs to S-100 group in comparison with vehicle

*Castagne V., et al //JPP, 2008*
Clinical studies of RA Abs to S-100 protein: anxiolytic activity

**Sites:** Four Russian clinical sites

**Design:** Randomized clinical study in 247 patients (18 – 65 y.o.) with anxiety disorders in comparison with diazepam

Clinical signs assessed by scales

![Bar chart showing clinical signs assessed by scales for Diazepam and RA Abs to S-100 proteins.](chart)

*** Conclusion: Anxiolytic activity of RA Abs to S-100 is close to one of Diazepam***

Efficacy of RA Abs to endothelial NO-synthase (eNOS): sexual activity

Preclinical Site: University of Tromsoe (Norway)
Model: assessment of sexual activity of Fischer 344 rats

Change in preference score on day 28 of treatment (% vs baseline)

Change in intromission ration on day 28 of treatment (% vs baseline)

* p<0.05 vs vehicle

Conclusion: Sexual activity of rats increased after receiving of RA Abs to eNOS in comparison with sildenafil and vehicle group

Chu X and Agmo A,// Pharmacol Biochem Be, 2008
Clinical studies of RA Abs to eNOS: patients with erectile disfunction

**Sites:** Four Russian clinical sites

**Design:** Randomized double blind placebo-controlled clinical trial in 169 patients (18 – 70 y.o.) with erectile disfunction

**Conclusion:** There is an significant activity of RA Abs to eNOS in comparison to placebo

Toxicological studies

**Experimental sites:**
- Research Institute of Pharmacology (Tomsk, Russia),
- Volgograd State Medical University (Volgograd, Russia)

**Studies:**
- Single-dose (acute) toxicity study
- General toxicity study
- Cumulation studies
- Study of mutagenic properties
- Study of allergenic properties
- Study of reproductive toxicity
- Study of postnatal development

**Experimental animals:**
- mice,
- rabbits,
- guinea pigs,
- rats (including sexually immature and infant)

**Route of administration:**
- oral
- intraperitoneal
- subcutaneous

**Conclusion:**
No toxic effects of RA Abs have been revealed exerts positive effect on lactating females (general condition, body weight gain) and postnatal development (facilitates cognition process and memory trace reproduction)
Drugs safety assessment in clinic studies

**Sources of data**

Safety assessment of the drugs during the clinical studies:
1. The assessment in the course of every planned visit via questioning of patients
2. the assessment of vital functions:
3. the assessment of main laboratory rangers:
   - clinical blood analysis
   - clinical urine analysis
   - biochemical markers

Data from regulatory bodies of countries where the drugs have been registered (15 countries) on adverse events associated with the drug use

**Spontaneous reports** on adverse events

Reports in international press on adverse events associated with the drug use

**Conclusion**

✓ The available data indicates no adverse events associated with the drug use
Main advantages of RA Ab as therapeutic agents

Key Advantages
- High safety: has been used for 14 years and in ~100 mln patients
- High efficacy: demonstrated in clinical studies conducted (the same or even better than that of the reference drugs)
- Oral use
- Possibility to combine with other medicinal drugs
- Low price
- Platform to rapidly screen and develop new drugs

Medical Benefits
- Long-term use possible
- Use in pediatric practice
- Use in patients with concomitant diseases
- Use for disease prophylaxis

Future prospects
- Platform to rapidly screen and develop new drugs
- Possibility of testing a plenty of different targets
Thank you for your attention!

Elena Gavrilova

In case of any questions please contact me at:

GavrilovaES@materiamedica.ru