Therapeutic Antibodies and Infectious Diseases

November 20th-22nd 2012,
Tours, *Vinci International Congress Centre,*
Loire Valley, France

ORGANISED BY

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Summary

20th November | Afternoon

13:00 | 14:00 **WELCOME**

14:00 | 15:00 **OPENING CEREMONY**

15:00 | 16:30 **ANTI-BACTERIAL ANTIBODIES**

Chair: Philippe Thullier / Nathalie Heuzé-Vourc'h

- **Development of a recombinant IgG neutralizing the anthrax lethal toxin**
  Thibaut Pelat, Institut de Recherche Biomédicale des Armées, France
- **Development of heavy chain antibodies against the Penton-Valentine leukocidine from *S. aureus***
  Dubravka Drabek, Erasmus Medical Center, Netherlands
- **Human monoclonal antibodies against Staphylococcal enterotoxin B: inhibitors of toxic shock**
  Bradley G. Stiles, Wilson College, USA

16:30 | 17:00 **COFFEE BREAK**

17:00 | 18:30 **ANTI-VIRAL ANTIBODIES**

Chair: Isabelle Dimier-Poisson / Denys Brand

- **Human mAbs against Hendra and Nipah viruses: from discovery in academic setting to administration in humans**
  Dimiter S. Dimitrov, Frederick National Laboratory for Cancer Research, NIH, USA
- **Rewriting history: antibody therapeutics for filoviruses**
  John M. Dye, USAMRIID, USA
- **Isolation and characterization of scFvs targeting filoviruses**
  Jeffrey W. Froude, USAMRMC, USA

21st November | Morning

08:00 | 08:30 **WELCOME**

08:30 | 09:30 **ANTI-MICROBIAL ANTIBODIES AND EFFECTOR FUNCTIONS**

Chair: Dominique Buzoni- Gatel / Gilles Thibault

- **Development of panobacumab, anti-*P. aeruginosa* LPS serotype IATS O11**
  Antonio Perez, Kenta Biotech, Switzerland
- **IgG-FcγR binding and effector functions**
  Pierre Bruhns, Institut Pasteur, France
- **An endogenous human antibody directed against a conserved epitope of M2 confers full protection against various Influenza A strains in an *in vivo* model system**
  Anton Bauer, Intercell, Austria

09:30 | 10:00 **COFFEE BREAK**

10:00 | 11:00 **KEYNOTE LECTURE**

Chair: Mustapha Si-Tahar

- **Broadly neutralising antibodies for therapy and vaccine design**
  Antonio Lanzavecchia, Institute for Research in Biomedicine, Switzerland

11:00 | 12:30 **ANTI-VIRAL ANTIBODIES**

Chair: Francis Barin / Stéphane Paul

- **Broadlly neutralizing antibodies present new prospects to counter HIV infection**
  Pascal Poignard, The Scripps Research Institut, USA
- **Nanobodies based on a dromadary single-domain antibody targeting HIV-1 reverse transcriptase**
  Naima Abidi-Azzouz, Centre de Recherches de Biochimie Macromoléculaire, France

12:30 | 14:00 **LUNCH**

21st November | Afternoon

14:00 | 15:30 **ANTI-VIRAL ANTIBODIES**

Chair: Philippe Roingeard / Valérie Gouilleux-Gruart

- **From polyclonal immunoglobulin to monoclonal antibody: an urgent need for rabies post-exposure prophylaxis**
  Laurent Dacheux, Institut Pasteur, France
- **Broadly neutralizing alpaca nanobody blocks virus entry and cell-cell transmission of hepatitis C virus**
  Pierre Lafaye, Institut Pasteur, France
- **Targeting host entry factors for prevention and treatment of hepatitis C virus infection**
  Mirjam Zeisel, University of Strasbourg, France

15:30 | 16:30 **KEYNOTE LECTURE**

Chair: Hervé Watier

- **Regulatory aspects in the development of monoclonal antibody cocktails**
  Patrick G. Swann, Office of Biotechnology Products, FDA, USA

16:30 | 17:00 **COFFEE BREAK**

17:00 | 18:30 **INVOLVEMENT OF THE FRENCH BIOTECH INDUSTRY IN THE FIELD**

Chair: Jean-Luc Teillaud / André Pèlegrin

- **Development of anti-Shiga toxin mAbs**
  Christian Behrens, LFB Biotechnologies, France
- **The ISAAC and VIVA SCREEN platforms for the generation of human mAbs**
  Majid Mehtali, Vivalis, France
- **Monoclonal antibodies against infectious diseases at SANOFI: where are we?**
  Laurent Fraisse, SANOFI, France

20:00 | **GALA DINNER**

22nd November | Morning

08:30 | 09:00 **WELCOME**

09:00 | 12:00 **THERAPEUTIC ANTIBODIES AND INFECTIOUS DISEASES: A DOUBLE EDGE SWORD?**

Chair: Yvon Lebranchu / Théodora Angoulvant

- **Eculizumab in severe Shiga toxin-associated hemolytic and uremic syndromes (STEC-HUS)**
  Christian Combe, University of Bordeaux Segalen, France
- **Rituximab in mixed cryoglobulinemia secondary to hepatitis C virus infection**
  Marcella Visentini, Sapienza University of Rome, Italy

10:00 | 10:30 **COFFEE BREAK**

- **Tuberculosis and other opportunistic infections during anti-TNF therapy, the French RATIO registry**
  Xavier Mariette, University of Paris-Sud 11, France
- **Natalizumab-associated progressive multifocal leukoencephalopathy (PML)**
  Patrick Vermersch, University of Lille Nord, France
- **CTLA4-Ig and risk of lymphoproliferation after EBV primo-infection**
  Bernard Charpentier, University of Paris-Sud 11, France

12:00 | **LUNCH**

14:00 | **END OF CONGRESS**

Introduction

Dear Friends, Dear Colleagues,

On behalf of the GDR 3260 "Antibodies and therapeutic targeting", the Laboratory of Excellence (LabEx) "MAbImprove", the Cluster de recherche en infectiologie and the Fédération de recherche en infectiologie, I am pleased to announce that the first congress on "Therapeutic Antibodies and Infectious Diseases" will be held in Tours from the 20th to the 22nd of November 2012. Therefore, I would like to take the present opportunity to invite you to join us for that meeting.

We are living an exciting scientific period, with monoclonals antibodies inducing considerable advances in the field of infectious diseases. However, there is a constant need to gather specialists of multiple disciplines to confront what has been learned from clinical experience and from recent pre-clinical and clinical developments.

With the Local Organizing and Scientific Committees, our aim is to build an ambitious and unique program focused on the targeting of microorganisms and their human receptors and the way currently approved monoclonals influence infectious diseases. During this 3 days congress, we will bring together scientific and clinical leaders from all over the world, in order to lead lively and interactive debates. The congress will take place in the modern "Vinci International Congress Centre" strategically located near the TGV fast train station, in the heart of Tours.

In addition to an exciting scientific program, you will benefit from the cultural background and historical landscape that make Tours and its nearest areas (Chenonceau, Villandry, Amboise among many others), one of the most attractive places in France, and this should also contribute towards making this event an unforgettable moment.

I look forward to welcoming and hosting you in Tours in November 2012, and sincerely hope that you will join us to make this endeavour a success.

Pr Hervé Watier

Thibaut Pelat •

Development of a recombinant IgG neutralizing the anthrax lethal toxin

NOTES

• Thibaut Pelat[a], Jean Charles Paucod[a], Christian Behrens[b] and Philippe Thullier[a]

• [a] Unité de biotechnologie des anticorps et des toxines, Département de microbiologie, Institut de Recherche Biomédicale des Armées (IRBA-CRSSA), 38702 La Tronche Cedex, France.
[b] LFB Research Department, 70 rue du Dr Yersin, Bioincubateur Eurasanté, 59120 Loos,France.

Development of a recombinant IgG neutralizing the anthrax lethal toxin

KEYWORDS

Antibody, Anthrax, Protective antigen

ABSTRACT

Bacillus anthracis (B. a.) is a bioweapon of primary importance, which causes anthrax. The pathogenicity of *B. a.* depends mainly on the lethal toxin (LT), composed of the lethal factor (LF) associated with the protective antigen (PA). Human anthrax infections cannot always be treated successfully by antibiotics, as highlighted by the 2001 postal anthrax attacks in the United States. Thus, there is a particular need to develop adjunct therapies to antibiotics, and antibodies (Ab) neutralizing LT are regarded as promising therapeutic molecules. To isolate such an LT-neutralizing Ab, we immunized a macaque (*Macaca fascicularis*) with PA. Using bone marrow, we PCR-amplified specific Fab-encoding genes and cloned them as an immune library. This library was panned by phage display and allowed the isolation of the Fab $35PA_{83}$. It has an affinity equal to 3.4 nM for PA and efficiently neutralizes LT *in vitro* (IC50 = 5.6 nM, corresponding to a [Fab $35PA_{83}$]/[PA] molar ration equal to 1.2), by inhibiting the interaction between PA and its receptors. Computer analysis revealed that the amino acid sequences of the $35PA_{83}$ are 92% identical to their homologs encoded by the most similar human germline sequences, and this high degree of identity is favourable for therapeutic use. Fab $35PA_{83}$ was expressed in fusion with human Fc, as a full-sized IgG. This IgG was successfully evaluated *in vivo* in the A/J mice infected with anthrax Sterne spores and was then further tested in WNZ rabbits infected with a lethal strain (9602) of anthrax. In this model in particular, the injection of IgG $35PA_{83}$ alone (2 mg/kg) 6 hours after the challenge (80 LD50) allowed a survival rate of 100 %. The Fab $35PA_{83}$ was engineered to enhance its affinity, by random mutations in its CDRs followed by selection using the phage technology. This screening allowed to isolate the variant 6.20, which showed an affinity that was enhanced 19-fold (KD = 180 pM) and correspondingly, its IC50 was decreased by 40%. This affinity for PA is the best ever reported and 6.20 has been patented. The clinical development of the IgG deriving from this variant has been launched recently.

Dubravka Drabek •

Development of heavy chain antibodies against the Penton-Valentine leukocidine

NOTES

• Dubravka Drabek[a], Benoit-Joseph Laventie[e], Hendrik Jan Rademaker[a,b], Maher Saleh[e,f], Ernie de Boer[a], Rick Janssensa, Tristan Bourcier[e,f], Audrey Subilia[f], Luc Marcellin[f], Rien van Haperen[a,b], Joyce H.G. Lebbink[d], Tao Chen[a], Gilles Prévost[e], Frank Grosveld[a,b]

• [a] Department of Cell Biology, Erasmus Medical Center, Rotterdam, Netherlands | [b] Harbour Antibodies BV, Rotterdam Netherlands | [d] Department of Genetics and Department of Radiation Oncology, Erasmus Medical Center, PO Box 2040, 3000 CA Rotterdam, The Netherlands | [e] Université de Strasbourg, CHRU Strasbourg, Physiopathologie et Médecine Translationelle, Institut de Bactériologie de la faculté de médecine, 3 rue Koeberlé, 67 000 Strasbourg, France | [f] CHRU Strasbourg, Départements d'Ophtalmologie et d'Anatomie Pathologique, 1 Ave Molière, 67098 Strasbourg Cedex, France.

Development of heavy chain antibodies against the Penton-Valentine leukocidine

KEYWORDS

transgenic mouse, Heavy chain only antibodies (HCAbs), bi-specific antibody, Penton-Valentine leucotoxin, toxin-induced endophtalmitis

ABSTRACT

Penton-Valentine leucotoxin (PVL) is a pore forming toxin associated with current outbreaks of community-associated methicillin-resistant strains and implicated directly in the pathophysiology of *S. aureus* related diseases. The active form of PVL consists of the two components LukS-PV and LukF-PV which induce osmotic lysis following pore formation in host defense cells. Humanized Heavy Chain only Antibodies (HCAb) were generated against the *S. aureus* PVL components from immunized transgenic mice with the aim to neutralize toxin activity. Transgenic mice containing a llama/human hybrid immunoglobulin heavy chain locus was used. One anti-LukS-PV HCAb, three anti-LukF-PV HCAbs with nmolar affinities and one engineered tetravalent bi-specific HCAb were tested *in vitro* and *in vivo*. They all prevent toxin binding and pore formation. Anti-LukS-PV HCAb also binds to gamma hemolysin C (HlgC) and inhibits HlgC-HlgB pore formation. Experiments *in vivo* in a toxin-induced rabbit endophthalmitis model showed these HCAbs inhibit inflammatory reactions and tissue destruction. The tetravalent bi-specific HCAb has the advantage that it is more effective at lower dose and that it consists of a single chain which is easy to produce. These results show the therapeutic potential of HCAbs and in particular bi-specific antibodies.

Bradley G. Stiles •

Human monoclonal antibodies against Staphylococcal enterotoxin B: inhibitors of toxic shock

NOTES

• Stiles BG[a], Larkin EA[b], and Ulrich RG[b]

• [a] Biology Department, Wilson College, Chambersburg, Pennsylvania, USA
[b] US Army Medical Research Institute of Infectious Diseases, Fort Detrick, Maryland, USA

Human monoclonal antibodies against Staphylococcal enterotoxin B: inhibitors of toxic shock

KEYWORDS

Human monoclonal antibody (Mab), Staphylococcal enterotoxin B (SEB), Streptococcal pyrogenic exotoxin (Spe), Cytokines, Shock

ABSTRACT

Staphylococcus aureus produces numerous virulence factors and causes many opportunistic infections around the world. Rising antibiotic resistance amongst *S. aureus*, and other pathogens, leaves fewer alternatives for clinical management. Alternative therapies are necessary for the near and distant future. One family of virulence factors secreted by *S. aurueus* includes the staphylococcal enterotoxins (SEs). These toxins are readily neutralized by human polyclonal antibodies, which can be clinically efficacious against staphylococcal and streptococcal disease. This current study characterized ten human, recombinantly-derived monoclonal antibodies (Mabs) targeting SEB. Mabs were isolated from a naive library panned with a recombinantly-attenuated SEB, previously used as an experimental vaccine. Mabs were examined in various assays as bivalent Fabs and full-length IgGs. By Western blot, eight of ten Fabs recognized low nanogram concentrations of SEB in complex protein mixtures (culture supernatant). The best performing Fabs had nanomolar binding affinities equal to polyclonal IgG, low nanomolar IC_{50}s against SEB in a human peripheral blood mononuclear cell (PBMC) assay, and protected mice from SEB-induced toxic shock. Several Fabs recognized neutralizing epitopes on related staphylococcal toxins (SEC1 and SEC2), as well as *Streptococcus pyogenes* pyrogenic exotoxin (Spe)C, in an ELISA. In contrast, SEA, TSST-1 (staphylococcal toxic shock syndrome toxin 1) and streptococcal SpeA did not bind to any Fab. Four Fabs with the best IC50s were converted into full-length IgGs. Although SEB-binding kinetics were identical between each Fab and its respective full-length IgG, a 250-fold greater inhibition of SEB-induced activation of human PBMCs was observed with two full-length IgGs. Overall, these Mabs possess: 1) high affinity; 2) target specificity; and 3) toxin neutralization qualities essential for any therapeutic agent. As an adjunct with other therapeutics (i.e. antibiotics and/or Mabs against other virulence factors), it is possible that toxin-based Mabs may be useful in combating future staphylococcal and streptococcal infections.

Dimiter S. Dimitrov •

Human mAbs against Hendra and Nipah viruses: from discovery in academic setting to administration in humans

NOTES

• Dimitrov DS

• Frederick National Laboratory for Cancer Research, NIH, Frederick, MD 21702, USA

Human mAbs against Hendra and Nipah viruses: from discovery in academic setting to administration in humans

KEYWORDS

Hendra virus, Nipah virus, therapeutic antibody, ferret model, monkey model

ABSTRACT

Therapeutic monoclonal antibodies (mAbs) have been highly successful in the clinic and more than 30 mAbs have been approved for clinical use in the European Union and the United States. However, there is no a single mAb approved for therapy of any infectious disease (Synagis is the only mAb approved for clinical use against RSV but is for prevention not therapy). In collaboration with C. Broder group we selected from a large naïve human Fab library and extensively characterized a new human mAb, m102, which neutralized both Nipah (NiV) and Hendra (HeV) viruses, and increased its potency by affinity maturation (Zhu et al J Virol 2006, J Inf Dis 2008). The resulting antibody, m102.4, was highly effective in ferrets challenged with NiV (Bossart K et al PLoS Pathogens 2009) and in monkeys challenged with HeV (Bossart K et al Science Translational Med). It was administered to humans in Australia who were exposed to HeV. The crystal structure of a variant of Fab m102.4 complexed with the HeV G protein was solved and suggested a possible mechanism of its cross-reactivity – by mimicking the receptor EphrinB2 (Xu K et al, submitted). The IgG1 m102.4 is currently being developed by the NIAID, NIH, USA, and by the Australian government as a therapeutic. To my knowledge it is the first human mAb administered to humans exposed to henipaviruses, has been used on compassionate basis in Australia, and could be approved for therapy of disease caused by henipaviruses by the USA FDA based on the positive results from two animal models and human safety trials.

Collaborators: Christopher C. Broder (Uniformed Services University Bethesda, MD), Katharine N. Bossart (Boston University and the National Emerging Infectious Diseases Laboratories), Thomas W. Geisbert (Galveston National Laboratory, University of Texas Medical Branch), Heinz Feldmann, Barry Rockx (NIAID, Rocky Mountain Laboratories, Hamilton, Montana), Lin-Fa Wang (CSIRO, Australian Animal Health Laboratory, Geelong, Australia), Antony Dimitrov (Profectus BioSciences, Inc., Baltimore, MD), Dimitar Nikolov (Structural Biology Program Memorial Sloan-Kettering Cancer Center, New York)

John M. Dye •

Rewriting history: antibody therapeutics for filoviruses

NOTES

• Dye, JM

• Virology Division, United States Army Medical Research Institute of Infectious Diseases, 1425 Porter Street, Fort Detrick, Maryland 21702 USA. John.m.dye1@us.army.mil

Rewriting history: antibody therapeutics for filoviruses

KEYWORDS

Filovirus, ebolavirus, passive transfer, and polyclonal antibody

ABSTRACT

Marburg virus (MARV) and Ebola virus (EBOV), from the family Filoviridae, cause acute, and frequently fatal, hemorrhagic fever in humans and are considered one of the most virulent infectious agents known to humans. Because of this high lethality, filoviruses pose significant emerging disease risks in sub-Saharan Africa and are considered potential important biological threats. Currently, there are no preventive vaccines or postexposure treatment options approved for use in humans, making the development of such products a high priority for filovirus researchers worldwide. Antibody therapies for filovirus infection have been disregarded in recent years, in part, due to early negative experimental evidence. We have protected nonhuman primates (NHPs) infected with MARV or EBOV by treating with multiple iterations of concentrated, species-matched, polyclonal IgG antibody administered over the clinical phase of disease. NHPs were exposed to either MARV or EBOV, and treatments were not initiated until 48 hours postexposure with additional treatments on days 4 and 8 postexposure. Even with delayed treatment, complete protection of both MARV and EBOV-challenged animals was achieved. In both of these studies, two out of the three IgG treated animals had no clinical signs of illness with the third animal developing mild and delayed signs of disease followed by full recovery. Further studies using polyclonal antibodies isolated from vaccinated NHPs in passive transfer studies will be presented. These studies point to neutralizing capacity of the treatment antibody as being an essential component in considering therapeutic options. Finally, new efforts just initiated to generate filovirus-specific monoclonal antibodies from human survivors will be discussed. Taken together, these studies clearly demonstrate that postexposureantibody therapies can protect NHPs, opening new avenues for filovirus treatments using established FDA-approved polyclonal or monoclonal antibody technologies.

Jeffrey W. Froude •

Isolation and characterization of scFvs targeting filoviruses

NOTES

• Froude, JW[a±], Pelat, T.[b], Miethe, S.[c], Zak, S.[d], Hust, M.[c], Dye, J.[d] and Thullier, P.[b]

• [a] United States Army Medical Research and Material Command (USAMRMC), Fort Detrick, Maryland, USA
[b] Unite de Biotechnologie des Anticorps, Institute de Recherche Biomedicale des Armees (IRBA-CRSSA), La Tronche, France
[c] Institut fur Biochemie und Biotechnologie, Technische Universitat Braunschweig, Braunschweig, Germany
[d] US Army Medical Research Institute for Infectioius Disease (USAMRIID), Fort Detrick , Maryland, USA
[±] Currently working at b

Isolation and characterization of scFvs targeting filoviruses

KEYWORDS

Filovirus, Marburg, Ebola, Antibody, Therapeutic

ABSTRACT

The limited number of monoclonal antibodies (mAbs) known to neutralize filoviruses have demonstrated an effective neutralization against Ebola viruses but not against the Marburg virus (MARV), and all have yet to show protection in non-human primates (NHP). Recently, macaques were immunized with viral-like particles expressing the glycoproteins (GP) of Ebola virus or MARV at their surface, and polyclonal antibodies (pAbs) were extracted from the sera of these animals. These pAbs were shown to provide post-exposure protection to NHPs challenged by Ebola virus and MARV respectively. Based on those results, a collaboration between the United States (USAMRIID) and French Military (IRBA-CRSSA) medical research communities was initiated to isolate recombinant antibodies from macaques immunized with the same immunogen, utilizing the phage-display technology. An immune library directed against MARV was built after four immunogen injections. It had a size of $1.2x10^{14}$ clones, and was screened against the GP of Marburg Ci67. After screening, 192 clones were sequenced and 18 different scFvs were identified and are being tested for their neutralization properties. Of these, seven neutralizing scFvs have already been identified. Regarding Ebola viruses, the goal was to develop antibodies cross-neutralizing both Ebola-Sudan (SUDV) and Ebola-Zaire (EBOV). The immune library directed against SUDV was built after four injections. It had a size of $1.42x10^{14}$ clones and was first screened against the GP of SUDV, then against the GP of EBOV. After screening, 134 clones were sequenced and 40 different scFvs were identified. Of these, four scFvs have already been identified for their neutralization properties against SUDV and further testing is under way. The amino-acid sequences of these macaque-derived antibody fragments are similar to those of their human germline counterparts, sharing an identity ranging between 62 and 93.6% which is favorable for therapeutic use. These results further demonstrate the ability to isolate antibodies from NHP immune libraries in the perspective of their therapeutic use, to neutralize potential bioweapons.

Development of panobacumab, anti- *P. Aeruginosa* LPS serotype IATS 011

NOTES

• Perez A.
• Chief Medical Officer, Kenta Biotech AG, Wagistrasse 25, CH-8952 Zürich-Schlieren, Switzerland

Development of panobacumab, anti- *P. aeruginosa* LPS serotype IATS O11

KEYWORDS

Pseudomonas, monoclonal antibody, pneumonia, IgM, serotype

ABSTRACT

Nosocomial pneumonia, either hospital acquired or ventilator-associated pneumonia is among the most severe infections in critically ill patients. *Pseudomonas aeruginosa* is responsible for approximately 20% of ventilator-associated pneumonia cases and is one of the most difficult pathogens to treat (overall mortality as high as 70% with directly attributable mortality rates of approximately 40%). The most complete serotyping system for PA, the International Antigenic Scheme (IATS), consists of 20 standard o serotypes. Epidemiology data indicate that of these 20 serotypes, IATS-O1, serogroup 2 (IATS-O2, IATS-O5 and IATS-16), IATS-O6 and IATS-O11 are responsible for 70-75% of *Pseudomonas aeruginosa* infections. The highest mortality rate and lower cure rates have been observed in patients with serotype O1 and O11 infections.

Panobacumab is a specific and fully human IgM/k monoclonal antibody with high avidity (5.81×10^7 M^{-1} $\pm 2.8 \times 10^7$ M^{-1}) for the P. aeruginosa lipopolysaccharide serotype O11, which specifically opsonizes the P. aeruginosa of IATS O11 and mediated complement-dependent phagocytosis.

A double blind, single dose escalation study evaluated the safety and pharmacokinetics of panobacumab in healthy volunteers. Four doses of panobacumab, 0.1, 0.4, 1.2, or 4 mg/kg of body weight or placebo were infused over 2 hr. Plasma concentrations of panobacumab were detected with mean maximum concentrations of drug in plasma of 1.87, 7.57, 24.92, and 83.19 mcg/ml respectively. The mean elimination half-life was between 70 and 95 h. The mean volume of distribution was between 4.76 and 5.47 liters. Clearance ranged between 0.039 and 0.120 liters/hr. No anti-panobacumab antibodies were detected after dosing in any subject. Panobacumab was well tolerated over the entire dose range and no serious adverse events were reported.

A multicentre, open pilot Phase 2a clinical trial (NCT00851435) prospectively evaluated the safety, pharmacokinetics and potential efficacy of three doses of 1.2 mg/kg panobacumab, given every 72 hr, in addition to standard antimicrobial therapy, in 18 patients with nosocomial *P. aeruginosa* serotype O11 pneumonia. In 13 patients receiving the three scheduled doses, the maximal concentration after the third infusion was 33.9 ±8.0 mcg/mL, total area under the serum concentration–time curve was 5.397 ±1.993 h.mcg/mL and elimination half-life was 102.3 ±47.8 hr. Panobacumab was well tolerated, induced no immunogenicity and was detected in respiratory samples. In contrast to Acute Physiology and Chronic Health Evaluation II (APACHE II) predicted mortality of 32%, all 13 patients receiving three doses survived, with a mean clinical resolution in 9.0 ±2.7 days. Two patients suffered a recurrence at days 17 and 20.

The clinical course and outcome of patients with *P. aeruginosa* O11 nosocomial pneumonia treated with panobacumab were compared with patients with *P. aeruginosa* O11 nosocomial pneumonia from a retrospective cohort. Despite a worse predicted prognostic based on risk scores, patients receiving panobacumab had an improved clinical course and outcome. The median time (days) to initial clinical resolution was shorter in the patients treated with panobacumab (8 versus 18.5 days in the retrospective cohort). The initial clinical resolution rate was of 100% versus 64% in the retrospective cohort), the final clinical resolution rate 85% versus 57% in the retrospective cohort, the microbiological resolution rate 31% versus 14% in the retrospective cohort and the survival rate 100% versus 79% in the retrospective cohort, were all numerically superior to those in the cohort study. Adjustment for age and severity scores confirmed these differences.

These data suggest that panobacumab is safe, with a pharmacokinetic profile in patients similar to that in healthy volunteers, and associated with high clinical cure and survival rates in patients developing nosocomial *P. aeruginosa* O11 pneumonia when comparing with those only treated with standard of care. These promising results warrant further trials.

Pierre Bruhns •

IgG-FcγR binding and effector functions

NOTES

• Pierre Bruhns

• Head, Antibody in Therapy & Pathology laboratory, INSERM Unit 760, Immunology Department, Institut Pasteur, Paris, France

IgG-FcγR binding and effector functions

KEYWORDS

FcR, IgG, mouse models inflammation, effector cells

ABSTRACT

Impressive advances in defining the properties of receptors for the Fc portion of immunoglobulins (FcR) have been made over the past several years. Ligand specificities were systematically analyzed for both human and mouse FcRs that revealed novel receptors for specific IgG subclasses. Expression patterns were redefined using novel specific anti-FcR mAbs that revealed major differences between human and mouse systems. The *in vivo* roles of IgG receptors have been addressed using specific FcR knockout mice or in mice expressing a single FcR, and have demonstrated a predominant contribution of mouse activating IgG receptors FcγRIII and FcγRIV to models of inflammation. Novel blocking mAbs specific for these activating IgG receptors have enabled, for the first time, the investigation of their roles *in vivo* in wild-type mice. In parallel, the *in vivo* properties of human FcRs have been reported using transgenic mice and models of inflammation and Ab-mediated therapy, in particular those of human activating IgG receptors FcγRI (CD64) and FcγRIIA (CD32A). Importantly, these studies led to the identification of specific cell populations responsible for the induction of various inflammatory diseases.

Anton Bauer •

An endogenous human antibody directed against a conserved epitope of M2 confers full protection against various Influenza A strains in an *in vivo* model system

NOTES

• Bauer A[a], Bauer M[b] and Beerli RR[b]
• [a] Intercell AG, Campus Vienna Biocenter 3, 1030 Vienna, Austria
[b] Intercell AG, Wagistrasse 25, 8952 Schlieren, Switzerland

An endogenous human antibody directed against a conserved epitope of M2 confers full protection against various Influenza A strains in an *in vivo* model system

KEYWORDS

human monoclonal antibody, Influenza A, ADCC, M2e, mammalian cell display

ABSTRACT

Processes that shape the immune repertoire *in vivo* create endogenous human antibodies with high affinity and low off target reactivity and immunogenicity. Identification of endogenous antibodies directly from the human organism is therefore an attractive option for the search of novel therapeutic or prophylactic mAbs.

We isolated a human mAb directed against the Influenza M2 protein directly from the memory B lymphocytes of human subjects using a proprietary platform technology involving mammalian cell display. This mAb recognizes a conserved M2 epitope present on all Influenza A strains and confers protection upon therapeutic and prophylactic application, respectively, in an *in vivo* mouse model via antibody dependent cellular cytotoxicity.

Antonio Lanzavecchia •

Broadly neutralising antibodies for therapy and vaccine design

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• Antonio Lanzavecchia

Broadly neutralising antibodies for therapy and vaccine design

KEYWORDS

ABSTRACT

Abstract non available

Pascal Poignard •

Broadlly neutralizing antibodies present new prospects to counter HIV infection

NOTES

• Pascal Poignard

• IAVI Neutralizing Antibody Center, Department of Immunology and Microbial Science, The Scripps Research Institute, La Jolla, California, USA

Broadlly neutralizing antibodies present new prospects to counter HIV infection

ABSTRACT

Over the last few years a flurry of HIV-1 broadly neutralizing antibodies have been isolated from infected donors, defining a series of highly conserved vaccine targets on the viral spike. In particular, remarkable breadth and potency has been revealed from antibodies targeting glycan-sensitive epitopes. These advances in the field give hope for the design of new vaccine and therapeutic approaches.

Naima Abidi-Azzouz •

Nanobodies based on a dromadary sigle-domain antibody targeting HIV-1 reverse transcriptase

NOTES

• Naima Abidi-Azzouz[1], Issam HMILA[2], Zakaria BENLASFAR[3], Martine PUGNIERE[4], Yvan BOUBLIK[1], Pascal CLAYETTE[5], Balkiss BOUHAOUALA[2], Gilles DIVITA[1]

• [1] Centre de Recherches de Biochimie Macromoléculaire, UMR-5237 CNRS-UM2-UM1, 1919 Route de Mende, Montpellier 34293 | [2] Laboratoire des Venins et Toxines, Institut Pasteur de Tunis, 13 Place Pasteur, BP-74, 1002 Tunis, Tunisia | [3] Unité des Services Animaliers-Institut Pasteur de Tunis, Tunisia | [4] IRCM (Institut de Recherche en Cancérologie de Montpellier), INSERM U896, Université Montpellier 1, CRLC Val d'Aurelle Paul Lamarque, Montpellier Cedex 5, France | [5] SPI-BIO Commissariat á l'énergie Atomique, Pharmacologie des Rétrovirus, 18 Route du Panorama, BP6, Fontenay aux Roses 9226, France

Nanobodies based on a dromadary sigle-domain antibody targeting HIV-1 reverse transcriptase

KEYWORDS

HIV-1, transcriptase (RT), HCAbs, Nanobodies, cell penetrating peptide

ABSTRACT

The rapid emergence of drug-resistant viruses against all approved clinical drugs against HIV together with inaccessible latent virus reservoirs and side effect of currently used compounds have limited the potency of existing anti-HIV-1 therapeutics. Therefore, there is a critical need for new and safer drugs, active against resistant viral strains, which will be useful for multiple drug combination.The human immunodeficiency virus-1 reverse transcriptase (RT) is a virus-specific protein responsible for the synthesis of double-stranded DNA from the single-strand retroviral RNA genome. Because this enzyme is essential for the retroviral replication, it constitutes a major target for the development of potentially selective antiviral agents.

We have elaborated a new strategy based on short antibody fragments derived from the unique Heavy-chain antibodies (HCAbs) occurring in *Camelidae* called VHHs, that targets RT-activation. Despite these antibodies lack a light chain, and are composed of only a Heavy-chain dimer, they are fully functional in antigen recognition, their antigen-binding domain consists of a single-domain. We have identified a Nanobody that inhibits the DNA-dependent DNA polymerase activity of RT at submicromolar concentrations. NbRT20 binds dimeric RT and stabilizes it in an inactive/non-processive conformation. From a mechanistic point of view, NbRT20 prevents proper binding of primer/template. When associated with the cell penetrating peptide Pep-1, this Nanobody is able to reach intracellular compartments and to inhibit viral proliferation, on PBMC infected with HIV-1_{LAI}, with IC_{50} values in the nM range. Taken together, these results demonstrated that, the nanobody platform may be highly effective at generating extremely potent and selective intrabody to neutralize RT and HIV proliferation.

From polyclonal immunoglobulin to monoclonal antibody: an urgent need for rabies post-exposure prophylaxis

NOTES

• Dacheux L.
• Institut Pasteur, Unit Lyssavirus Dynamics and Host Adaptation, National Reference Centre for Rabies, WHO Collaborating Center for Reference and Research on Rabies, 25 rue du Dr Roux, 75724 Paris Cedex 15, France.

From polyclonal immunoglobulin to monoclonal antibody: an urgent need for rabies post-exposure prophylaxis

KEYWORDS

Rabies, post-exposure prophylaxis, passive immunisation, polyclonal immunoglobulin, monoclonal antibody

ABSTRACT

Rabies remains one of the most ancient and deadly of human infectious diseases, for which no effective curative treatment is available once the first clinical signs appear. This viral zoonosis is transmitted principally by the saliva of infected dogs and still represents a major, but neglected public health problem, with an estimated 55,000 human deaths reported each year, mostly in developping countries in Africa and Asia.

Since the first implementation by Louis Pasteur in 1885 of an efficient post-exposure prophylaxis (PEP), effective protocols and safer products have been developed, providing almost complete protection if given early enough after rabies exposition. Four schedules are approved by the World Health Organization (WHO), all based on wound cleansing and administration of tissue-culture vaccines. A combination with polyclonal rabies immunoglobulin (RIG) of human or equine origin is also given, in case of severe exposure (category III according to WHO). However, only a limited number of individuals exposed to rabies undergo PEP, estimated to 10-16 million people worldwide every year. Among them, nearly 60% do not receive any passive rabies immunotherapy, whereas the category of exposure requires its administration. The adequate supply of PEP is mainly restricted by the high cost of RIG, making them still unaffordable in many parts of the world. Difficulty of production with low interest of manufacturers, short supply (leading to worldwide shortage for some time), limited accessibility and biological risks are other drawbacks of polyclonal serum, especially of human origin.

For these reasons, the development of rabies monoclonal antibodies represents a promising alternative to traditional polyclonal preparations. This way has been explored within the last two decades, and lead to the design of several murin and human monoclonal antibodies, all targeting the glycoprotein (outer protein) of the rabies virus. They are used alone or in cocktails, demonstrated broad *in vitro* neutralisation and/or *in vivo* protection in rodent models. Some of them are undergoing clinical trials, with the assessment of a phase 3 trial for one of the most advanced product.

Massive production of effective rabies monoclonal antibodies could replace classical RIG, so passive immunisation might finally become accessible, affordable and routinely used part of the global health practices for rabies. In combination with an adequate supply of modern tissue-culture vaccines, but also with information actions for healthcare personnel on medical practices related to PEP, and implementation of measures for the control of dog rabies, the global burden of human rabies will be significantly reduced.

Pierre Lafaye •

Broadly neutralizing alpaca nanobody blocks virus entry and cell-cell transmission of hepatitis C virus

NOTES

• Pierre Lafaye[1], Alexander W. Tarr[2] , Laurence Damier-Piolle[3,4], Luke Meredith[5], Richard A Urbanowicz[1], Annalisa Meola[3,4], Jean-Luc Jestin[3,4], Matthijs Backx[1], Jane A. McKeating[5], Felix A. Rey[3,4], Jonathan K Ball[1], Thomas Krey[3,4]

• [1] Institut Pasteur, Departement de Biologie Structurale et Chimie, Plateforme de Production de proteines recombinantes, 75724 Paris Cedex 15, France | [2] School of Molecular Medical Sciences, University of Nottingham, Queen's Medical Centre, Nottingham, United Kingdom | [3] Institut Pasteur, Unité de Virologie Structurale, 75724 Paris Cedex 15, France | [4] CNRS URA 3015, 75724 Paris Cedex 15, France | [5] Hepatitis C Research Group, Institute For Biomedical Research, University of Birmingham, Birmingham B15 2TT, United Kingdom.

Broadly neutralizing alpaca nanobody blocks virus entry and cell-cell transmission of hepatitis C virus

KEYWORDS

Hepatitis C virus, broadly neutralizing antibodies, immunotherapy, VHH, llama

ABSTRACT

While conventional immunoglobulins are heterotetramers composed of two heavy and two light chains, with a molecular weight of about 150 KD, in camelids a significant proportion of serum antibodies are homodimeric IgGs with a molecular weight of about 80 kD. Their heavy chains contain three domains instead of four in conventional antibodies. The variable domain of these heavy-chain only antibodies is referred to as VHH. Recombinant VHHs (12-14 kD in size) constitute intact antigen-binding domains and exhibit a broad antigen-binding repertoire. Their hypervariable regions are expanded and exhibit special characteristics that allow VHHs to recognize unique epitopes that are poorly immunogenic for conventional antibodies.

We immunized a llama with the soluble ectodomain of HCV E2 and we isolated four heavy-chain-only antibodies in that specifically recognize HCV E2. We expressed the corresponding recombinant VHH domains and demonstrated that two of them neutralize Hepatitis C virus (HCV) infection. This was shown using both retroviral particles pseudotyped with HCV glycoproteins (HCVpp) and cell-culture derived HCV particles (HCVcc). One of the neutralizing VHHs potently neutralized a wide range of Hepatitis C virus (HCV) strains and isolates, rendering it a potential tool for HCV immunotherapy. The neutralizing efficiency of this monovalent VHH is comparable to that of a potent, well-characterized broadly neutralizing bivalent human mAb.

VHHs targeting antigens from numerous viruses have been reported, among them HIV, influenza A virus, poliovirus, foot and mouth disease virus and rotaviruses. Notably, for numerous virus systems an improvement of the biological activity by up to 500 fold has been observed upon dimerization of neutralizing VHHs, either by fusing two identical VHHs (monospecific) or two different VHHs (bispecific).

Importantly, most of the broadly neutralizing monoclonal antibodies against HCV E2 (human and murine) identified so far do not inhibit cell-cell spread of HCV, a route of virus transmission that is believed to play an important role *in vivo* and likely represents an important evasion mechanism from the humoral immune response of the patient. In contrast to the non-effective human and mouse mAbs, the more potent VHH inhibits cell-cell spread, suggesting an important advantage over other broadly neutralizing antibodies that have been put forward into potential therapeutic applications so far.

NOTES

• Zeisel M.B.[a]
• [a] Inserm, U748, Université de Strasbourg, Strasbourg, France.

Targeting host entry factors for prevention and treatment of hepatitis C virus infection

KEYWORDS

Hepatitis C virus, antiviral, entry inhibitor, scavenger receptor class B type I, claudin-1

ABSTRACT

With an estimated 170 million infected individuals, hepatitis C virus (HCV) has a major impact on public health. HCV infection is major causes of chronic liver disease and hepatocellular carcinoma worldwide. Furthermore, virus-induced liver disease is a major indication of liver transplantation. In the past years, direct-acting antivirals (DAAs) targeting HCV enzymes have been developed. DAAs increase the virologic response to anti-HCV therapy (interferon alfa and ribavirin) but may lead to selection of drug-resistant variants and treatment failure. To date, strategies to prevent HCV infection are still lacking and antiviral therapy in difficult-to-treat patients remains limited. Alternative or complementary approaches addressing the limitations of current antiviral therapies are to boost the host's innate immunity or interfere with host factors required for pathogenesis. Attachment of the virus to the host cell followed by viral entry is the first step in a cascade of interactions between the virus and the target cell required for successful initiation of infection. The development of novel HCV model systems allowed to increase the understanding of the complex viral entry process thereby offering new therapeutic targets to prevent the virus to reach its site of replication. Both virus and host cell components involved in virus entry may serve as targets for the development of HCV entry inhibitors. Major progress has been made over the past years in the characterization of host cell factors involved in virus entry and the sequence of events ultimately leading to viral replication. HCV entry is believed to be a multistep process involving a concerted interplay between the virus and several host cell factors such as highly sulfated heparan sulfate, CD81, the scavenger receptor class B type I (SR-BI), claudin-1 (CLDN1) and occludin (OCLN). Moreover, HCV entry is regulated by different host cells kinases among which epidermal growth factor receptor (EGFR). Studies from our laboratory have shown that antibodies targeting the extracellular domains of SR-BI and CLDN1 provide useful tools for the prevention of HCV infection. Indeed, antibodies specific for SR-BI and CLDN1 target HCV entry during post-binding steps and thereby prevent initiation of cell-free infection with all major HCV genotypes as well as highly infectious HCV escape variants re-infecting the liver graft. Moreover, these antibodies also allow to inhibit direct HCV cell-to-cell transmission, an alternative entry route that enables the virus to escape most of the neutralizing anti-HCV antibodies, and viral spread when added post-infection. Thus, targeting host entry factors with monoclonal antibodies is an efficient strategy to both prevent initiation and maintenance of infection in a genotype independent manner. Moreover, combination of anti-SR-BI or anti-CLDN1 antibodies with neutralizing anti-HCV envelope antibodies, interferon alfa, DAAs or other host-targeting agents (HTAs) resulted in a marked and synergistic inhibition of HCV infection with undetectable toxicity. Our results provide the rationale for the development of antiviral strategies combining entry inhibitors with interferon alfa, DAAs or HTAs by taking advantage of synergy. The uncovered combinations may increase the efficacy of the current interferon alfa-based regimen, open novel perspectives for interferon alfa-free regimens and provide efficient strategies to prevent liver graft infection. Taken together, HCV entry inhibitors, such as antibodies specific for host entry factors, provide an interesting perspective for novel antiviral strategies against HCV since they have (i) a high genetic barrier to resistance (ii) a pan-genotypic antiviral activity and (iii) complementary mechanisms of action to DAAs and may therefore act in a synergistic manner with current standard of care or DAAs in clinical development.

Patrick G. Swann •

Regulatory aspects in the development of monoclonal antibody cocktails

NOTES

• Patrick G. Swann

• Division of Monoclonal Antibodies, Office of Biotechnology Products, Center for Drug Evaluation and Research, Food and Drug Administration, Bethesda, MD, United States of America.

Regulatory aspects in the development of monoclonal antibody cocktails

KEYWORDS

Monoclonal antibody, drug development, infectious diseases, investigational new drug application, antibody cocktails

ABSTRACT

Despite a slow beginning, monoclonal antibodies (mAbs) have had many successes over the past 15 years and are the best-selling class of biologics. The diversity of constructs is also increasing and includes antibody conjugates, bispecific mAbs, antibody fragments, Fc-fusion proteins and antibody cocktails. This presentation will focus on regulatory aspects in the development of antibody cocktails. General results of a survey of active Investigational New Drug Applications to the U.S. FDA will be presented highlighting the number of applications utilizing antibody cocktails and their general clinical indications. Available guidance and regulatory considerations that can facilitate development of antibody cocktails will be discussed with an emphasis on the proposed utilization of antibody cocktails for the treatment of infectious diseases.

Christian K. Behrens •

Development of anti-Shiga toxin mAbs

NOTES

• Behrens CK[a], Reymond D[b], and the SHIGATEC clinical trial network

• [a] LFB Biotechnologies, Les Ulis, France.
[b] Thallion Pharmaceuticals Inc, Montreal, Canada.

Development of anti-Shiga toxin mAbs

KEYWORDS

Infectious disease, monoclonal antibody, Shiga toxin-producing E. coli (STEC), drug development, clinical trial

ABSTRACT

Infection with Shiga toxin (Stx)-producing *E. coli* (STEC) can lead to severe complications including haemolytic uremic syndrome (HUS), neurological complications and death. It is a leading cause of acute renal failure in children and continues to be a major public health concern. This is highlighted by the German EHEC outbreak in 2011 in which more than 3800 infected individuals, 855 HUS cases and 53 deaths were reported. Today, there are no approved therapies to prevent the development of HUS or its associated complications.

Shigamabs is comprised of two chimeric monoclonal antibodies (cαStx1 and cαStx2) against the Shiga toxins Stx 1 and 2, respectively, that has been shown to be active *in vitro* and protective in animal models. Shigamabs was shown to be safe and well tolerated in four Phase I trials in healthy adults.

A randomized, double-blind, placebo controlled phase II trial was conducted in Argentina, Chile and Peru in children aged 6 months to 18 years, presenting with bloody diarrhoea and testing positive for Stx and/or the O157 strain of *E. coli*. Patients were randomized 2:1 to two arms: A) standard of care + Shigamabs, or B) standard of care + placebo. Two doses (1 mg/kg and 3 mg/kg of each antibody) were evaluated in two sequential cohorts. Primary endpoints were safety and tolerability; secondary endpoints were efficacy and pharmacokinetics.

A total of 45 patients aged 7 months to 11 years (median age = 20 months) were enrolled, 15 to each study group. Of the STEC strains isolated, 82% produced Stx2, 14% produced Stx1, and 4% produced both Stx1 + Stx2. No drug-related adverse events were reported. Pharmacokinetic profiles showed a faster elimination of cαStx1 than cαStx2. One of 30 patients (3%) having received product, developed an asymptomatic immune response to cαStx2. In terms of efficacy, two patients progressed to HUS; one in the placebo group requiring 6 days of peritoneal dialysis, and one in the 3 mg/kg treated group requiring no dialysis.

In conclusion, the study demonstrated the ability to enrol STEC-infected children at the stage of early bloody diarrhoea. Shigamabs appeared safe and well-tolerated in this paediatric population. Given the small sample size, there were no significant trends in terms of efficacy.

Majid Mehtali •

VIVA|SCREEN™ Technology for the high throughput discovery of rare native human therapeutic monoclonal antibodies

NOTES

• Majid Mehtali

• Vivalis, 6 rue Alain Bombard 44800 Saint-Herblain, France

VIVA I SCREEN™ Technology for the high throughput discovery of rare native human therapeutic monoclonal antibodies

KEYWORDS

Native human antibodies, Microarray chips, infectious diseases

ABSTRACT

VIVA I SCREEN is a breakthrough technology that allows the efficient and rapid isolation of highly potent fully human native monoclonal antibodies without being hampered by the antigen-specific but non-biologically active antibodies that often represent over 95% of all antibodies discovered against a defined target.

Three specific achievements were instrumental to the successful development of this technology:

- The access to a very large population of human donors, healthy or diseased, through a network of agreements with blood transfusion centers and hospitals.
- The optimization of the culture and expansion of human primary memory B cells, to isolate in two weeks specific subpopulations of B lymphocytes secreting antigen-specific antibodies confirmed to display potent biological activities.
- The use of specifically-designed microarray chips that contain 62,500 wells with size and shape optimized for a single human B lymphocyte per well, enabling the rapid automated analysis of the primary B lymphocytes at the single cell level. Using the principle of ELISA, individual B lymphocytes secreting the biologically active antigen-specific antibodies are automatically retrieved, used for RNA isolation, antibody gene cloning and production of recombinant antibodies. Such recombinant antibodies are then produced at mg to gram scales for further detailed *in vitro* and *in vivo* analysis to select the best antibody candidates for development.

This technology was successfully applied for a series of infectious and non infectious targets and allowed the discovery of a large number of highly potent native human antibodies displaying affinities to the targets in the picoMolar range, including antibodies produced by B lymphocytes present only at extremely low frequency in circulating human PBMCs (<1/100,000,000). Examples of antibody discovery program for infectious diseases will be presented and discussed.

Laurent Fraisse •

Monoclonal antibodies against infectious diseases at Sanofi: where are we?

NOTES

• Laurent Fraisse

• SANOFI, France

Monoclonal antibodies against infectious diseases at Sanofi: where are we?

ABSTRACT

Despite global efforts to halt the increase and spread of antimicrobial resistance, bacteria continue to become less and less susceptible to antimicrobial drugs, while rates of discovery for new antibiotics are declining. Consequently, multidrug-resistant bacteria are becoming a severe threat to public health and therefore it is critical to identify new strategies in an attempt to combat infectious diseases. Besides a significant investment in antibiotic research and development Sanofi is also exploring the potential of alternative therapies, inc luding monoclonal antibodies for the prevention and treatment of severe bacterial infections. SAR279356 was in-licensed from Alopexx (F598) in December-2009. SAR279356 is an intact, fully human monoclonal antibody (mAb) that binds to the conserved bacterial surface polysaccharide poly-N-acetylglucosamine (PNAG) and is able to induce opsonophagocytosis. SAR279356 is currently in Phase II clinical trial, which investigates the pharmacokinetics, pharmacodynamics and safety of a single intravenous dose of SAR279356 in intensive care unit (ICU) patients with mechanical ventilation. PNAG is expressed by many pathogens (*including among others Staphylococcus aureus, Streptococcus pneumoniae, Escherichia coli, Klebsiella pneumoniae, and Acinetobacter baumannii*). SAR279356 is developed for the prevention of nosocomial infections in ICU patients at risk of infections caused by pathogens that express PNAG.

Sanofi Pasteur has partnered with KaloBios Pharmaceuticals in 2010 to develop KB001-A, an anti-PcrV (component of the virulence factor Type III Secretion system) PEGylated monoclonal antibody fragment ('Fab) for the prevention of serious *Pseudomonas aeruginosa* infections in mechanically ventilated patients. A functionally equivalent pre-cursor 'Fab (KB001) had previously been evaluated and was well-tolerated in a Phase II study of mechanically ventilated patients. KB001-A is an optimized compound anticipated to resume clinical studies soon.

These two monoclonal antibodies both address important medical needs and are complimentary in their spectrum of activity against nosocomial bacterial pathogens.

Eculizumab in severe Shiga toxin-associated hemolytic and uremic syndromes (STEC-HUS)

NOTES

• Combe C[a,b] and Delmas Y[a]
• [a] Service de Néphrologie Transplantation Dialyse, Centre Hospitalier Universitaire, Bordeaux, France.
[b] Unité INSERM U1026, Université Bordeaux Segalen, Bordeaux, France.

Eculizumab in severe Shiga toxin-associated hemolytic and uremic syndromes (STEC-HUS)

KEYWORDS

Hemolytic Uraemic Syndrome, Shiga Toxin, Low Platelets, Complement, Eculizumab

ABSTRACT

Thrombotic microangiopathies (TMA) are microvascular occlusive diseases with platelets aggregation leading to peripheral thrombocytopenia and mechanical destruction of red blood cells. Typical hemolytic uremic syndrome (HUS) occurs in the setting of gastroenteritis caused mostly in children by shiga-toxin producing Escherichia coli (STEC) of O157:H7 serotype 2. There are no established guidelines for STEC-HUS treatment: the effect of plasma exchange (PE) is not proven in adults; in children, the use of PE is usually limited to HUS with neurologic involvement, in addition to supportive therapy.

During May and June 2011, an outbreak of STEC-HUS due to E. coli O104:H4 hit North. Previously, E. coli O104:H4 related STEC-HUS had been reported in a sporadic case in South Korea. Various treatments have been used during the German outbreak, including PE, immunoadsorption and eculizumab, a humanized monoclonal antibody that inhibits C5 terminal complement common pathway that has recently been approved in atypical HUS.

In June 2011, an outbreak of HUS occurred in the town of Bègles near Bordeaux, France, due to E. coli O104:H4, with characteristics close to the German. Amongst 24 patients with O104:H4 STEC infection, 7 patients presented with HUS after the contaminating meal; 2 patients later had household contamination. All STEC-HUS patients were admitted to our department:

HUS developed 6 days (median; range 3-12) after digestive symptoms: platelets count was 46 G/l (24-109), hemoglobin 11.8 g/dL (10.1-13.9), LDH 901 IU/mL (453-2568), creatinine 152 µmol/l (48-288). All pts had extrarenal complications (liver 9, pancreas 5, brain 3, heart 3). Two pts were dialyzed, 1 pt was ventilated.

After failure of plasma exchange to increase platelets in the first 3 pts, eculizumab was administered as in atypical HUS in all 9 pts, 0 to 4 days after HUS diagnosis (median 1 day). One pt with very severe neurological HUS received immunoadsorption.

Outcome was favorable in all pts, with rapid normalization of hemoglobin, platelets, LDH levels, renal function, and neurological improvement. There were no deaths and no serious adverse events related to eculizumab.

In our experience, early treatment of O104:H4 STEC HUS by eculizumab was associated to rapid and efficient recovery.

However, reports from Germany question the efficiency of eculizumab and of plasmapheresis with or without glucocorticoids.

Further evaluation of the role of complement and the effect of eculizumab in STEC HUS is warranted.

NOTES

• Visentini M., Fiorilli M. and Casato M.
• Department of Clinical Immunology, Sapienza University of Rome, Viale dell'Università 37, 00185 Rome, Italy

Rituximab in mixed cryoglobulinemia secondary to hepatitis C virus infection

KEYWORDS

hepatitis C virus, mixed cryoglobulinemia, vasculitis, rituximab, dose-effectiveness

ABSTRACT

Chronic hepatitis C virus (HCV) infection is responsible for extrahepatic manifestations including mixed cryoglobulinemia (MC), a benign lymphoproliferative disorder associated with systemic vasculitis caused by immune complexes formed by monoclonal IgM rheumatoid factor and IgG. The clinical manifestations of MC range from a mild form with purpura, arthralgia and fatigue to a severe form with progressive renal disease, skin ulcers and debilitating peripheral neuropathy. About 10% of MC patients develop over time non-Hodgkin's lymphoma (NHL). The treatment of choice for MC is eradication of HCV with pegylated interferon alpha (PEG-IFN) and ribavirin, but almost half of the patients are refractory to antiviral therapy. Treatment with immunosuppressive drugs (glucocorticoids, azathioprine or cyclophosphamide) is scarcely effective, bears significant side effects and may accelerate liver disease; plasmapheresis is effective for disease flares but is inadequate for long term treatment.

Over the last decade, several off-label uncontrolled studies suggested the efficacy and safety of anti-B-cell therapy with rituximab for refractory HCV-associated MC (reviewed in [1]). Rituximab has been also used in conjunction with antiviral therapy with remarkable results in severe MC. The rituximab dosage used in nearly all published reports was 375 mg/m^2 given four times, the treatment schedule used for B cell NHL, and the response rate (complete plus partial responses) was 86% with a mean time to relapse of 6.7 months. A recently published[2] randomized controlled trial demonstrated the superiority of rituximab monotherapy to standard immunosuppressive therapy; the dosage used in this study was 1000 mg given twice, with a response rate of 80% and a surprisingly long mean time to relapse of 18 months. We recently sought to demonstrate the non-inferiority of a low-dose regimen (250 mg/m^2 given twice) of rituximab monotherapy for refractory HCV-related MC in a phase 2 single-arm two-stage study trial. Preliminary data in 27 patients indicated that this dosage could be as efficacious as the 375 mg/m^2 x 4 regimen, with a response rate of 79% and a mean time to relapse of 6.5 months[3]. B-cell depletion ("4 cells/μL) was achieved in all but one patient, but did not apparently correlate with clinical response or with cryocrit decrease at any time point over a 12-month follow-up. Circulating B cells bearing t(14;18)bcl-2/IgH, a translocation suspected to play a role in MC-associated lymphomagenesis, remained undetectable after rituximab in all patients initially positive even after B-cell repopulation. Side effects were comparable to those seen in patients treated with high-dose rituximab, and increase of HCV viral load, reported in some studies, was not observed. Our current results in 41 treated patients support the published preliminary findings. Collectively, the available data indicate that low-dose rituximab may provide a more cost/effective alternative for treating refractory HCV-associated mixed cryoglobulinemia.

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Xavier Mariette •

Tuberculosis and other opportunistic infections during anti-TNF therapy, the French RATIO registry

NOTES

• Xavier Mariette

• Université Paris-Sud, Hôpitaux Universitaires Paris-Sud, INSERM U1012.

Tuberculosis and other opportunistic infections during anti-TNF therapy, the French RATIO registry

KEYWORDS

anti-TNF therapy, monoclonal antibody, soluble receptor, tuberculosis, opportunistic infections

ABSTRACT

In most of metaanalysis of randomized control trials and in most of registries, but not in all, anti-TNF therapy increases the risk of common serious infections. Actually, this increase risk is probably present only at the beginning of the treatment since after time, improvement of disease activity and decrease of associated immuno-suppression (steroide) may counterbalance the increased risk induce by anti-TNF.

However an increased risk of a small subset of serious infections: tuberculosis and opportunistic infections persists in patients treated with anti-TNF therapy. But the absolute risk of these infections remains much lower than the risk of most common serious infection like pneumonia.

These very rare side effects are not well captured by randomized control trials, metaanalysis or registries. For this reason, we set up an original approach in France by collecting prospectively all the cases of TB and opportunistic infections occurring in patients treated with anti-TNF for three years.

This re-enforced dedicated pharmacovigilance network included the official pharmacovigilance units of the French agency of drug, the pharmacovigilance units of the companies, and the scientific societies of rheumatology, gastro-enterology, dermatology, pneumology and infectious diseases.

Through this network we could collect 69 cases of TB and 43 cases of non-TB opportunistic infections in patients treated with anti-TNF for three years. It is much more than in most of the metaanalysis or in registries.

Thereafter, we could calculate the standardized incidence RATIO (SIR) of TB compared with the general population and we also performed a case control study with 2 or 3 age- and sex-matched patients treated with anti-TNF without any side effect for each case of TB or non-TB opportunistic infections.

We confirmed that the risk of TB was 12 times more frequent in patients treated with anti-TNF than into the general population. We found a clear difference of risk of TB and of non-TB opportunistic infections between the monoclonal antibodies and the soluble receptor.

Different hypothesis are made for explaining this difference of mechanism of action between monoclonal antibodies and soluble receptor which can explain this differential risk of these very rare severe adverse events but also the differential efficacy in granulomatous diseases as Crohn's disease.

Patrick Vermersch •

Natalizumab-associated progressive multifocal encephalopthy

NOTES

• Patrick Vermersch

• University of Lille Nord de France, Department of Neurology, 59037 Lille cedex.

Natalizumab-associated progressive multifocal encephalopthy

KEYWORDS

multiple sclerosis, natalizumab, progressive multifocal encephalopathy, JC virus

ABSTRACT

A challenge for the clinician treating patients with multiple sclerosis (MS) is to determine the most effective treatment while weighing the benefits and risks. Results of the phase 2 and phase 3 studies on natalizumab were received with great interest, in part due to the improved risk reduction for relapse rate, disease progression, and MRI metrics observed in comparison to results in trials of beta-interferon and glatiramer acetate. However, except for very active relapsing patients, natalizumab is used as a second line treatment, because of safety issues. Indeed, natalizumab is associated with a substantial risk of progressive multifocal leukoencephalopathy (N-PML), due to CNS infection by the opportunistic JC virus (JCV). Its physiopathology is not fully understood. An N-linked glycoprotein, present on many human cells, is one of the cellular receptors for the JC virus. Additionally, the JC virus can bind to the serotoninergic 5-HT2a receptor to infect astroglial cells in culture. This receptor is present in several cell types, including kidney epithelial cells, B lymphocytes, platelets, glial cells, and neurons. After asymptomatic primary infection, which occurs usually in childhood, the virus remains quiescent in the kidneys, bone marrow, and lymphoid tissue. It is suggested that during immunsuppression, a reactivation of the virus occurs, probably related to rearranged regulatory regions of the DNA virus and are necessary for the reactivation of the JC virus that causes PML. As of june 2012, 267 confirmed cases of N-PML have been observed, giving rise to an overall risk of approximately 0.25%. N-PML pathogenesis remains partially elusive although risk factors have now been clearly delineated. In patients with prior JCV infection detected by serum anti-JCV antibodies, duration of therapy and prior use of immunosuppressants (IS) increase the risk of N-PML. No N-PML have been developped in JCV seronegative patients. The clinical outcome of patients with MS who developed N-PML was highly variable, ranging from asymptomatic cases to varying degrees of neurologic disability or even death. It was also observed in real-life setting that the earlier N-PML was diagnosed and treated, the better was the clinical outcome. Clinical vigilance is now considered as the established cornerstone of PML risk-management algorithm. However periodic brain MRI scans, particularly in high-risk situations, are likely to provide earlier detection of N-PML and better outcomes. In seronegative patients, JCV status is controlled every 6 months for detect a seroconversion. The use of natalizumab requires a perfect collaboration between neurologists, radiologists and biologists.

CTLA4-Ig and risk of lymphoproliferation after EBV primo-infection

NOTES

• **Charpentier B**, François H, Beaudreuil S and Dürrbach A
• Dept of Nephrology, University Hospital of Bicêtre, 94275 Kremlin-Bicêtre; UMR 1014 INSERM/Université Paris-Sud 11, Villejuif, France

CTLA4-Ig and risk of lymphoproliferation after EBV primo-infection

KEYWORDS

CTLA4-Ig, EBV, B lymphocytes, proliferation, safety

ABSTRACT

Long-term graft and patient survival remain significant challenges in kidney transplantation, and new therapies are needed to improve long-term outcomes. Belatacept, a first-in-class selective costimulation blocker, has been approved for prophylaxis of organ rejection in kidney transplant recipients who are positive for Epstein-Barr virus. In phase 3 trials, belatacept demonstrated superior preservation of renal function and comparable patient/graft survival when compared with cyclosporine, while avoiding the renal toxicities and other adverse events associated with the use of a calcineurin inhibitor. The most serious adverse events reported with belatacept are posttransplant lymphoproliferative disorder (PTLD), other malignancies, and serious infections, including progressive multifocal leukoencephalopathy (PML) and polyomavirus nephropathy. Analysis of data from the long-term extension of the phase 2 study pooled with 2-year data from the phase 3 trials revealed that 16 cases of PTLD occurred overall: 8 in belatacept MI patients, 6 in belatacept LI patients, and 2 in cyclosporine patients. Nine of these cases (6 in belatacept MI and 3 in belatacept LI) involved the CNS; of these, frequency was highest among EBV(-) patients, occurring in 5 of 96 (5%) of EBV(-) patients and in 4 of 805 (0.5%) of EBV (+) patients. As EBV seronegativity was found to be associated with an increased risk of CNS PTLD, belatacept is contraindicated in patients who are EBV seronegative or with unknown EBV serostatus. between the LI and cyclosporine treatment groups and was higher in the belatacept MI group. CNS infections, while uncommon, also occurred with similar frequency in the LI and cyclosporine treatment groups, and with higher frequency in the MI treatment group. These outcomes were consistent with findings from an exposure-response analysis that determined that safety events, such as serious infections and CNS events, appear to be associated with higher average belatacept concentrations, as the belatacept MI regimen provided overall higher exposure than the LI regimen as designed (~2-fold that of the LI regimen between Months 2 through 7 in the phase 3 trials).

Despite the increased incidence of PTLD, the LI regimen was associated with fewer deaths and serious infections than cyclosporine-based treatment. The overall frequency of malignancies was similar. The improved renal benefit with belatacept may translate into improvements in long-term graft and patient outcomes. Targeting T-cell costimulation is an important new option for maintenance immunosuppression in kidney transplant recipients.

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