Clinical & Experimental Immunology
The Journal of Translational Immunology
British Society for immunology
doi:10.1111/cei.12197
TRANSLATIONAL NEUROIMMUNOLOGY REVIEW SERIES
Clinical and Experimental Immunology
Series originators and editors: Olaf Stüve and Uwe Zettl

Monoclonal antibodies in treatment of multiple sclerosis

P. S. Rommer,*‡ A. Dudesek,* O. Stüve§¶† and U.K. Zettl*
**Clinic and Policlinic of Neurology, University of Rostock, Rostock, †Department of Neurology, Klinikum rechts der Isar, Technische Universität München, Munich, Germany, ‡Department of Neurology, Medical University of Vienna, Vienna, Austria, §Department of Neurology, University of Texas Southwestern Medical Center, and ¶Neurology Section, VA North Texas Health Care System, Dallas VA Medical Center, Dallas, TX, USA*

Accepted for publication 29 August 2013
Correspondence: P. S. Rommer, Medical University of Vienna, Vienna 1090, Austria.
E-mail: paulus.rommer@meduniwien.ac.at

Summary

Monoclonal antibodies (mAbs) are used as therapeutics in a number of disciplines in medicine, such as oncology, rheumatology, gastroenterology, dermatology and transplant rejection prevention. Since the introduction and reintroduction of the anti-alpha4-integrin mAb natalizumab in 2004 and 2006, mAbs have gained relevance in the treatment of multiple sclerosis (MS). At present, numerous mAbs have been tested in clinical trials in relapsing–remitting MS, and in progressive forms of MS. One of the agents that might soon be approved for very active forms of relapsing–remitting MS is alemtuzumab, a humanized mAb against CD52. This review provides insights into clinical studies with the mAbs natalizumab, alemtuzumab, daclizumab, rituximab, ocrelizumab and ofatumumab.

Keywords: alemtuzumab, daclizumab, multiple sclerosis, natalizumab, rituximab

Introduction

Based on the work of Köhler and Milstein, who produced the first monoclonal antibody (mAb) in 1975 [1], numerous mAbs have been approved for treatment of various diseases by the US Food and Drug Administration (FDA) in oncology (alemtuzumab, rituximab, ofatumumab, bevacizumab), rheumatology (tocilizumab, adalimumab, golimumab), gastroenterology (infliximab, certolizumab pegol), dermatology (efalizumab, ustekinumab) and transplant rejection prevention (daclizumab, basiliximab). Natalizumab was the first mAb to gain approval for a neurological disease, multiple sclerosis (MS) [2]. Alemtuzumab (Lemtrada®) might be the second mAb to be approved for MS. On 27 June 2013, the Committee for Medicinal Products for Human Use (CHMP) of the European Medicines Agency (EMA) adopted a positive opinion for approval of alemtuzumab for active relapsing–remitting MS (RRMS) [3].

Currently, several mAbs are being tested in clinical studies. This review provides information on the different mAbs, summarizes the results of clinical trials and outlines potential side effects of each agent.

Natalizumab (Tysabri®)

Background

Natalizumab is a humanized mAb immunoglobulin (Ig)G4 antibody targeting the α4-chain of α4β1 integrin and other α4-integrin-containing adhesion molecules. The α4β1 integrin heterodimer is also known as very late activating antigen-4 (VLA-4). VLA-4 is expressed on the surface on leucocytes. Upon binding of natalizumab, VLA-4 is impaired

in its ability to bind to vascular cell adhesion molecule-1 (VCAM-1) and its other ligands, including fibronectin. As a consequence, leucocytes are not able to adhere to the inner lining of cerebral vascular walls, and to migrate subsequently through the blood–brain barrier (BBB) into the central nervous system (CNS) [4]. In 2004, natalizumab received accelerated approval by the FDA on basis of the interim results of two ongoing Phase III clinical trials [5,6]. However, the occurrence of three progressive multifocal leucoencephalopathy (PML) cases in patients with MS and Crohn's disease led to the voluntary withdrawal of natalizumab by its manufacturers in 2005. Following an evaluation period, natalizumab was reintroduced in 2006.

Clinical trials

Natalizumab showed superior efficacy over placebo in two Phase III trials in patients with RRMS. The first trial (AFFIRM) tested natalizumab *versus* placebo in patients with RRMS. The relapse rate was reduced in the natalizumab group by 68% ($P < 0{\cdot}001$) compared with the placebo group at year 1, and the sustained progression as measured by the Kurtzke Expanded Disability Scoring Scale (EDSS) was lower by 42% ($P < 0{\cdot}001$) in the treatment group over 2 years. Natalizumab led to a reduction of 83% in new or enlarging hyperintense lesions [5]. In patients with insufficient response to treatment with immunomodulatory therapy with interferon (IFN)-β1a, natalizumab or placebo was given as an add-on to IFN-β-1a (SENTINEL). Combination therapy led to a lower annualized relapse rate (ARR) over 2 years compared with IFN-β and placebo (0·34 *versus* 0·75, $P < 0{\cdot}001$). The natalizumab group showed a relative reduction in disease progression risk by 24% ($P = 0{\cdot}02$) compared to the IFN-β plus placebo group [6].

Potential side effects

Currently, more than 300 cases of PML have been reported [7]. Risk evaluation tools have been introduced to determine the individual risk of developing PML. PML is caused by the human polyoma virus JC that becomes latent in the bone marrow or kidney after exposure, and that appears to reactivate during prolonged and severe immunosuppression. The manufacturer offers testing for the anti-JCV antibody status to determine prior exposure to JC virus. Depending on exposure status, risk stratification can be made. Three risk factors appear to be relevant: besides the positive anti-JCV antibody status, the duration of treatment with natalizumab (more than 24 monthly infusions) and prior treatment with immunosuppressants such as mitoxantrone or cyclophosphamide increases the risk of PML [8]. The risk differs from <1/10 000 in patients with negative testing for JCV, no prior immunosuppressant treatment and treatment duration lower than 24 months, to up to 11/1000 in patients with positive JCV status, prior treatment with immunosuppressants and treatment duration longer than 24 months [9]. In case of any suspicion of PML, treatment with natalizumab has to be terminated immediately. The clinical picture suspicious for PML has to be supported by brain imaging and should be confirmed by cerebrospinal fluid (CSF) polymerase chain reaction (PCR) testing for JC. Plasma exchange is performed to accelerate the elimination of natalizumab. A clinical deterioration is often reported after termination of natalizumab. The reason for this is the immune reconstitution inflammatory syndrome (IRIS). IRIS has been reported in patients with AIDS and PML. Glucocorticosteroids are given in such cases [10].

Beside PML, adverse effects may be headache, fatigue, arthralgia, infections of the urinary and respiratory tract, diarrhoea, rash, vaginitis and abdominal pain. Hepatoxicity may occur and liver enzymes have to be tested. Anaphylaxis may occur in response to mAb treatment. There are no well-controlled trials in pregnant women. In animal studies, adverse effects on the fetus were reported. In case of pregnancy natalizumab should be given only if the potential benefit justifies/outweighs the potential risk on the fetus. Moreover, neutralizing antibodies (nAb) in response to natalizumab may occur. In such cases nAbs may prevent natalizumab from efficacy. Before treatment starts, infection with HIV has to be excluded. It is contraindicated in cases of malignomas or of concomitant immunosuppressants. It should not be applied in cases with acute herpes infections or other active or chronic infections [9].

Outlook

In conclusion, natalizumab is a highly potent drug. The patients have to be informed about the risk of PML. Risk stratification as mentioned above is compulsory. After 24 months of treatment, re-evaluation of treatment options must be performed. Treatment duration of more than 24 months has to be considered. Depending on the other risk factors for PML, the decision has to be made as to whether to continue with treatment with natalizumab or to terminate treatment with it. After cessation of natalizumab, re-occurrence of disease activity has been observed in up to one-third of the patients [11–16]. The results of clinical trials on how to de-escalate from natalizumab are, in part, inconsistent. A trial testing glatiramer acetate in 40 patients with prior treatment with natalizumab showed that almost two-thirds of the patients were relapse-free over the follow-up period of 12 months. Furthermore, the ARR was significantly lower than prior to treatment with natalizumab. The patients remained stable on EDSS for a follow-up of 12 months. Magnetic resonance imaging (MRI) did not reveal a rebound of disease activity [17]. Another trial included patients who were either treated with glatiramer acetate or remained off therapy. In both groups

recurrence of disease activity was observed (five of seven patients on glatiramer acetate and all six patients on treatment). In this trial no differences could be seen with regard to relapse rate, MRI parameters or EDSS between both groups [11]. One-third of patients treated with pulsed glucocorticosteroids in combination with glatiramer acetate reported new relapses within 6 months after cessation of natalizumab [18].

Switching to fingolimod after a washout period of 3 months led to reported disease progression in up to 50% of the patients [19]. The washout period of 3 months might represent a risk factor for recurrence of disease activity [20]. Sequential glucocorticosteroid treatment during the washout period did not have a significant influence on the outcome [21]. After switching to fingolimod, PML has been diagnosed in a patient with prior treatment with natalizumab for 3·5 years and positive JC-virus status [22]. Nevertheless, fingolimod seems to reduce the risk of recurrence of disease activity when compared with no treatment. Furthermore, initiation of treatment with fingolimod within 3 months after cessation of natalizumab seems to be associated with lower risk of relapses when compared to fingolimod started after 3 months [23].

Currently, switching from natalizumab to other therapeutics is not a routine procedure. Larger trials are lacking. Clinical factors such as relapses prior to treatment with natalizumab, efficacy of treatment and risk attitude of the patients may help in making the decision. If treatment continues with natalizumab, the patients must be informed about the higher risk of PML [24,25]. The washout period (2–3 months) in switching to fingolimod or mitoxantrone after cessation of natalizumab represents a risk factor for disease activity. Determination of the immune status should be carried out, and MRI scans should be performed for providing disease status after treatment with natalizumab. At present, no fixed guidelines are available; an individual decision has to be made between the clinician and the patient.

Alemtuzumab (Campath®, MabCampath®, Lemtrada®)

Background

Alemtuzumab is a humanized IgG1 kappa mAb targeting CD52-antigen on the surface on mainly mature lymphocytes, monocytes, dendritic cells and granulocytes, but not on stem cells. Antibody-mediated cell cytoysis (ADCC), complement-dependent cytolysis (CDC) and apoptosis are the main effects of alemtuzumab [26–28]. Originally, alemtuzumab was developed as a rat mAb to reduce the number of circulating T cells in order to prevent graft-*versus*-host disease in bone marrow transplantation [29]. It was developed by researchers in the department of pathology at the University of Cambridge, from which its name is derived: Cam(bridge)path(ology). To reduce allergic reactions and to improve tolerability the rodent parts were decreased and the first humanized mAb was developed: Campath-1H [30]. In 2001, alemtuzumab (Campath®, MabCampath®) was approved by the FDA and the EMA for treatment of B cell chronic lymphocytic leukaemia (B-CLL) that is treatment-resistant to standard medications, including alkylating agents and fludarabine therapy [31,32].

Clinical trials

Alemtuzumab was tested in several small trials in MS. Seven patients with either secondary progressive MS (SPMS) or primary progressive MS (PPMS) were given alemtuzumab. The treatment led to a significant reduction ($P < 0{\cdot}001$) in new lesions as measured by MRI [33]. Coles and colleagues tested alemtuzumab in 27 patients with SPMS. Enhancing lesions on MRI were suppressed over the follow-up period of 18 months between 66 and 90%. However, disability as measured by EDSS progressed in more than half of the patients [34]. A subset of patients suffering from a progressive course showed a reduction in gadolinium (Gd)-enhancing lesions, but disability proceeded in these patients and increased brain and spinal cord atrophy was observed [35].

Based on the results of the small trials, a Phase II trial testing alemtuzumab against IFN-β-1a in patients with active MS (at least two relapses over the last 12 months before screening), short disease duration (not longer than 3 years) and disability not more than 3·0 on the EDSS score was assessed. Alemtuzumab led to a decrease in the EDSS score (–0·39), whereas disability increased under therapy with IFN-β-1a (+0·38, $P < 0{\cdot}001$). The ARR was significantly lower in the alemtuzumab group (0·10 *versus* 0·36; $P < 0{\cdot}001$). Furthermore, the lesion burden was significantly lower in the alemtuzumab group compared to the IFN-β group ($P = 0{\cdot}005$) [36]. The results of the trial suggest that patients with short disease duration and highly active MS may profit from treatment with alemtuzumab. The 5-year follow-up showed a sustained efficacy in comparison to IFN-β-1a [37].

Two Phase III trials (MS care I and II) confirmed the efficacy of alemtuzumab in relapsing MS patients. One trial tested alemtuzumab in treatment-naive patients who were given either alemtuzumab intravenously (i.v.) 12 mg per day on 5 consecutive days and again after 12 months for 3 days, or IFN-β-1a 44 μg subcutaneously (s.c.) three times a week. Alemtuzumab led to a 54·9% improvement in relapse rate ($P < 0{\cdot}0001$). More patients were relapse-free in the alemtuzumab group compared to the IFN-β group (78 *versus* 59%, $P < 0{\cdot}0001$) over the trial period of 2 years [38]. The second trial enrolled RRMS patients with at least one relapse over the last 12 months under treatment with IFN-β or glatiramer acetate. The enrolled patients were given IFN-β-1a 44 μg three times a week, or alemtuzumab 12 mg on 5 consecutive days and then again on 3 consecutive days at month 12. Relapses were fewer in the patients treated with

alemtuzumab, and more patients were relapse-free in the alemtuzumab group (65 *versus* 47%, $P < 0{\cdot}0001$) at 2 years. The sustained disability accumulation was higher in the IFN group compared to the alemtuzumab group (20 *versus* 13%, $P = 0{\cdot}008$) [39].

Potential side effects

Immune thrombocytopenic purpura (ITP) was reported in three patients, one of whom died in the alemtuzumab group in a Phase 2 trial [36]. As a consequence, alemtuzumab was suspended in trials in 2005. Afterwards, monitoring was required for monthly complete blood counts for detecting abnormalities as early as possible, and for monthly side contacts with the patient [40]. Besides ITP, herpes infections, thyroid-associated diseases and infections were more common in the alemtuzumab group [38]. Infusion-associated effects occurred more often in the alemtuzumab group. Up to 18% of all treated patients with alemtuzumab developed thyroid-associated disorders in Phase III trials [38,39].

Infusion-related side effects are relatively common [39]. During the first infusion, urticaria, pyrexia and rigor may occur. Neurological symptoms will last for several hours [34]. Concomitant medication with glucocorticosteroids, antihistamines and antibiotics is recommended [41]. The most important adverse effects are secondary humoral autoimmunity that may result from reconstitution of B cells prior to regulatory T cells [29], and may occur up to 5 years after therapy. Autoimmunity is described in about one-fifth of all treated patients [42–44]. Thyroid-associated autoimmune diseases are most common, but also haematological, renal and dermatological autoimmune diseases may occur, most of them in the second year of treatment [45,46]. Drug monitoring (infections, autoimmunity inclusive ITP) is essential in patients treated with alemtuzumab.

Outlook

Alemtuzumab seems to be a candidate for induction therapy, meaning that treatment is initiated with one drug and maintenance treatment is performed with another drug. This approach has been shown to be successful in treatment with mitoxantrone, with short- and long-lasting benefits. Despite the risks for autoimmune disease, alemtuzumab may be an ideal candidate for patients with active MS [47]. There are reports of PML under alemtuzumab in patients treated for CLL or non-Hodgkin lymphoma [48]. Currently, there are no cases of PML in patients with MS. The clinician should be aware the risk of PML, and we would recommend testing for JC virus status. Depending on the result, individual decision-making has to consider the risk of PML.

On 14 August 2012 the marketing authorization holder withdrew MabCampath® (alemtuzumab) voluntarily from market authorization. Patients with a B-CLL and who need treatment with MabCampath® will receive it through patient access programmes [49]. Recently a positive opinion was adopted, and the granting of market authorization for Lemtrada® (alemtuzumab) was recommended by the EMA [3].

Daclizumab (Zenapax®)

Background

Daclizumab is a humanized IgG1 mAb and binds to the alpha subunit of the human high-affinity IL-2 receptor (CD25) on activated lymphocytes and inhibits binding of IL-2. As a consequence, the receptors are saturated and T cell activation is prevented. It is approved by the FDA for prophylaxis of acute organ rejection in patients with renal transplantation [50]. It has been withdrawn in Europe for commercial reasons [51]. Mechanisms of action are not fully understood. Interestingly, therapy response correlated with expansion of $CD56^{bright}$ natural killer (NK) cells [52]. Expansion of $CD56^{bright}$ NK cells may lead to down-regulated T cell responses. The increase of $CD56^{bright}$ NK cells correlated with therapy response [53]. This phenomenon could also be detected in the CSF [54]. NK cells play an incompletely understood role in the pathophysiology of MS, but it seems that they are involved in the immunoregulation of MS. In animal studies, NK cell depletion led to exacerbation in experimental autoimmune encephalomyelitis (EAE). In clinical trials it could be shown that IFN-β led to an expansion of immunoregulatory $CD56^{bright}$ and to a decrease in cytotoxic $CD56^{dim}$ NK cells [55]. Expansion of $CD56^{bright}$ NK cells may serve as biomarker for therapy response in individual patients [56].

Clinical trials

Daclizumab has been tested in several small trials in patients with MS. Bielekova *et al.* tested daclizumab in patients with incomplete response to IFN-β treatment. Eleven patients were included in this Phase II open-label trial. Daclizumab administered 1 mg/kg i.v. every second week in the first month and thereafter every 4 weeks (seven total infusions) led to a decrease in new Gd-enhancing lesions of 78% and the total burden of Gd-enhancing lesions decreased by 70% [57]. A small trial tested daclizumab in nine patients with RRMS and incomplete response to IFN-β. Again, a significant reduction in Gd-enhancing lesions could be shown in comparison to prior treatment with IFN-β alone. The positive effect of daclizumab on clinical outcomes could be seen in combination with IFN-β and with daclizumab alone [58]. Further trials in RRMS patients led to an improvement in EDSS by

2·2 points in comparison to EDSS prior to treatment with daclizumab ($P < 0{\cdot}0001$), and ARR decreased from 2·15 to 0·33 ($P < 0{\cdot}0001$) [59]. A retrospective analysis of patients receiving daclizumab was performed in comparison with patients off treatment with daclizumab. More than 1300 MRIs were analysed from 70 patients. The decline in brain volume was significantly lower in the daclizumab group than in the control group [60].

A Phase II randomized, double-blind, placebo-controlled trial tested daclizumab in active RRMS patients on therapy with IFN-β. Daclizumab was given s.c. in low (1 mg/kg every 4 weeks) or high doses (2 mg/kg every 2 weeks) as an add-on to IFN-β over 24 weeks. The placebo group was given IFN-β and placebo. A total of 230 patients were enrolled into the trial and subdivided into three groups. The mean number of Gd-enhanced lesions differed significantly between the placebo and the high-dose group in favour of the high-dose group (4·75 *versus* 1·32, $P = 0{\cdot}004$) [52].

Recently, the results of a Phase II trial testing daclizumab s.c. *versus* placebo in RRMS patients have been published. Daclizumab was administered every 4 weeks with a 52-week follow-up. Daclizumab was tested in two doses: low-dose daclizumab 150 mg s.c. and high-dose daclizumab 300 mg s.c. The ARR was significantly lower in both daclizumab groups compared to the placebo group, more patients were relapse-free in the low-dose daclizumab group (81%) and in the high-dose daclizumab group (80%) compared to placebo (64%, $P < 0{\cdot}0001$ and $P = 0{\cdot}0003$, respectively). Serious adverse events were more common in the daclizumab groups (9% in the high-dose and 7% in the low-dose groups) than in the placebo group (6%) [61]. Currently, a Phase III trial testing daclizumab against IFN-β is ongoing (Efficacy and Safety of Daclizumab High Yield Process Versus Interferon β 1a in Patients with Relapsing–Remitting Multiple Sclerosis (DECIDE). Recruitment has been terminated [62].

Potential side effects

The most common side effects are rash, lymphadenopathy and fever. Other side effects include headache, paraesthesia, fatigue, constipation and transient liver enzyme increase [59], as well as a higher rate of infections and depression [57]. Side effects from studies in patients with kidney transplant when daclizumab is given as prophylaxis for rejection include nausea, diarrhoea, dyspepsia, vomiting, abdominal pain, oedema extremities, tremor, headache, dizziness, fever, pain, chest pain, oliguria, dysuria, pulmonary oedema, insomnia and tachycardia. More rare are blurred vision, pruritus, arthralgia and myalgia. In these trials daclizumab or placebo were given as add-on to cyclosporin or corticosteroids. Anaphylactic reactions may occur in response to daclizumab. Daclizumab should not be used in pregnancy, as well-controlled trials are lacking [63].

Outlook

Daclizumab seems to be more effective in RRMS than approved standard therapies, and seems to be effective when administered either i.v. or s.c. Results of large Phase III trials are pending.

Anti CD20 therapeutics

The role of B cells in the pathophysiology in MS has been long debated. There are hints – the presence of B cells, plasmablasts and immunoglobulins in MS lesions, as well as immunoglobulins and oligoclonal band in the CSF – that B cells at least contribute to the pathogenesis of MS [64]. Furthermore, B cells seem to be involved in T cell activation, cytokine production and demyelination and remyelination [65]. This has been the rationale for the use of antibodies targeting CD20 on B cells. The first anti-CD20 mAb was rituximab. Currently, trials are testing newer CD20 mAbs (ocrelizumab, ofatumumab).

Rituximab

Background

Rituximab is a chimeric IgG1 mAb targeting CD20 on cells of the B cell lineage. Its effect is born by apoptosis, ADCC and CDC [66,67]. Originally, it was developed for the treatment of B cell lymphoma [68,69]. It is approved by the FDA under certain circumstances for treatment of non-Hodgkin lymphoma (NHL), CLL, rheumatoid arthritis and granulomatosis with polyangiitis and microscopic polyangiitis [70]. It is approved by the EMA for NHL, CLL and rheumatoid arthritis [71]. Although it is not approved for MS, it is quite often used off-label [72].

Clinical trials

Hauser *et al.* tested rituximab in a Phase II trial in patients with RRMS. One hundred and four patients were either assigned to receive rituximab 1000 mg on day 1 and 15 i.v. or placebo. The primary end-point was the number of Gd-enhancing lesions at weeks 12, 16, 20, 24 and 48. Further assessments included ARR, safety issues and the portion of patients with relapses. Clinical and imaging outcomes showed significantly better results for the rituximab group, sustained over the study period of 48 weeks [73]. A trial tested rituximab in 32 patients with incomplete response to disease-modifying therapy (DMT). The patients received 375 mg/m^2 rituximab as an add-on to DMT weekly for 4 consecutive weeks. Gd-enhancing lesions decreased significantly to time before rituximab treatment [74].

Rituximab has been tested in progressive [75] and relapsing MS. A trial including 439 patients with primary progressive MS (PPMS) administered either rituximab or placebo twice every 24 weeks over 96 weeks. The primary end-point – time to confirmed progression – did not reach significance (rituximab group 30·2 *versus* 38·5% in the placebo group; $P = 0{\cdot}14$). Although patients receiving rituximab had fewer new T2 lesions ($P < 0{\cdot}001$), brain volume change was similar in both groups. Nevertheless, a subgroup of patients (age <51 years, and/or patients with Gd-enhancing lesions) showed a significant delay in time to confirmed disease progression [76]. Similarly, a small case-series tested rituximab in secondary progressive MS (SPMS) patients. All patients reported relapses or Gd-enhancing lesions before study entry. After therapy with rituximab, patients remained stable on EDSS score, no relapses occurred and no Gd-enhancing lesions were shown on the MRI [77]. Both results suggest that as long as inflammatory processes are ongoing, rituximab may be effective.

Potential side effects

Side effects are mainly infusion-associated. They are common and may occur in every fourth patient. In the majority of cases they are reported during the first infusion. Premedication with glucocorticosteroids, slowing down the rate of infusion and administration of histamine receptor antagonists may be the proper means of dealing with this [78]. Side effects include nausea, headache, urinary and respiratory tract infections [73,79], as well as fever, chills and muscle spasm during infusion [74]. A cytokine release syndrome may occur, and thus the treating clinician has to be aware of this complication [80].

A retrospective analysis of patients with different autoimmune diseases and rituximab treatment showed that infections may occur in up to 13% of the patients. The incidence of infections differed between the various diseases and was most common in myasthenia gravis. Allergic reactions were reported in almost every tenth patient [72]. Although this analysis included different diseases, it allows a brief insight into possible side effects. It should be noted that PML has occurred in patients with rheumatoid arthritis treated with rituximab [81]. Patients should be informed about the risk of PML.

Outlook

The results of clinical trials suggest that rituximab is effective in RRMS. The results of the OLYMPUS trial suggest that even a subset of patients with progressive MS may benefit from therapy with rituximab. As possible biomarkers, Gd-enhancing lesions representing inflammatory processes may serve. Newer anti-CD20 antibodies will supersede the use of rituximab.

Ocrelizumab

Background

Like rituximab, ocrelizumab targets CD20 cells on the B cell lineage. In contrast to rituximab, ocrelizumab is a humanized IgG1 mAb. Thus, ocrelizumab seems to lead to fewer allergic reactions and anti-idiotypic antibodies. In contrast to rituximab, its efficacy appears to be mediated more by ADCC than CDC [82].

Clinical trials

A Phase II, randomized, placebo-controlled, multi-centre trial tested ocrelizumab in RRMS patients. The included patients were assigned to receive placebo, low-dose ocrelizumab (600 mg) on days 1 and 15, high-dose ocrelizumab (2000 mg) on days 1 and 15 or IFN-β-1a once a week. At week 24 patients received either 600 mg ocrelizumab (the former placebo, low-dose ocrelizumab and IFN groups) or 1000 mg (the former high-dose ocrelizumab group). The primary end-point was the number of Gd-enhancing lesions. At week 24 the number of Gd-enhancing lesions was decreased by 89% in the low-dose and by 96% in the high-dose ocrelizumab groups. Both groups showed better results than the IFN-β group. Furthermore, relapses were significantly lower in both groups (the low-dose ocrelizumab group led to a reduction in relapses of 80% and the high-dose ocrelizumab group to a reduction of 73%) compared with the other groups [83]. The extended follow-up revealed that no patients had Gd-enhancing lesions at week 96 [84].

Potential side effects

Initial trials in patients with rheumatoid arthritis were suspended due to fatal outcomes [85]. Side effects in trials in MS were associated with the first infusion. Severe side effects did not show significant differences between the groups. The most common side effects were headache, infections and chills [83]. However, one patient in the ocrelizumab group died from a systemic inflammatory response syndrome. It is not clear if it was related to the ocrelizumab exposure.

Outlook

First results show very good efficacy in RRMS. At present some Phase III trials in RRMS and PPMS are ongoing [62].

Ofatumumab (Arzerra®)

Background

Ofatumumab targets CD20 on cells of the B cell lineage. Originally, ofatumumab was generated from transgenic

mice. Using genetic engineering techniques, heavy and light chain genes from humans were used and transfected in a murine myeloma cell line. It is a fully human IgG1 mAb. It seems to mediate its B cell-depleting effects more through CDC than rituximab. Furthermore, although rituximab and ofatumumab bind the same target, ofatumumab seems to dissociate from it at a slower rate and binds to an additional epitope [86]. In conclusion, it differs from rituximab with regard to pharmacodynamics. Ofatumumab (Arzerra®) is approved as a second-line therapeutic treatment in CCL in Europe [87] and by the FDA [88].

Clinical trials

Ofatumumab was tested in RRMS patients. Thirty-eight patients were included in a Phase II trial. Patients either received 100 , 300 or 700 mg ofatumumab, or placebo twice. After 24 weeks patients who had received placebo were given ofatumumab, and the other group received placebo. MRI showed a sustained reduction in brain lesions [89,90].

Potential side effects

No severe side effects were reported [90].

Outlook

At present, a Phase II trial is testing ofatumumab s.c. in RRMS patients [62].

Tocilizumab and eculizumab

Recently reports have been published about tocilizumab and eculizumab in the treatment of neuromyelitis optica (NMO) patients [91–94].

Tocilizumab

Background

Tocilizumab is a humanized IgG1 mAb targeting the IL-6 receptor. It is approved by the FDA as a second-line therapeutic treatment in rheumatoid arthritis, for the treatment of active systemic juvenile idiopathic arthritis and for children with polyarticular juvenile idiopathic arthritis [95].

For several years, it has been debated whether NMO is a distinct disease entity or a subtype of MS. The detection of NMO-IgG – targeting aquaporin-4 channels (anti-Aqp-4 antibody) – in 2004 [96] favours the first variant. Thus, humoral immune response seems to play a major role in pathogenesis of NMO. Consequently, the role of complement activation is of great importance. Anti-Aqp-4 antibody is predominantly IgG1, and activates complement. Activated complement amplifies the inflammatory processes. As a final step, the membrane attack complex lyses plasma membranes on astrocytic endfeet [94,97]. As part of the humoral immune response B cells seem, at least partly, to be responsible for the pathogenesis of NMO. However, some patients fail to respond to rituximab. Whereas CD20 is expressed on B cells, it is not expressed on plasmablasts secreting antibodies after terminal differentiation. Thus, rituximab may not destroy plasmablasts.

IL-6 promotes production of anti-Aqp-4 antibodies from plasmablasts *in vitro*. Consequently, a IL-6-dependent B cell subpopulation is at least partly responsible for disease activity in NMO [98]. Increased levels of IL-6 could be found in the CSF of NMO patients, underlining the critical role of IL-6 in NMO [99]. Besides increased levels of IL-6, increased memory Th17 cells could also be detected in patients with NMO and MS. High levels of Th17 cells were associated with clinical features and decreased after glucocorticosteroid treatment [100]. In addition, both NMO and MS patients showed an expanded T cell receptor repertoire representing an up-regulated T cell activity [101]. The importance of T helper type 17 (Th17) and IL-17 in autoimmune processes has been discussed for years [100]. In addition to its effects on plasmablasts, IL-6 has important functions in the differentiation of Th17 cells [102,103].

Clinical trials

In case reports and small case-series, tocilizumab has been used in patients with NMO. Araki and colleagues reported a patient who experienced eight relapses over the last 18 months under treatment with azathioprine. Tocilizumab was administered for 6 months, with monthly doses of 8 mg/kg i.v. Four days after the first infusion the patient relapsed. During the follow-up, no further relapses occurred. The EDSS decreased from 3·5 to 2·0. Azathioprine could be tapered from 50 mg daily to 50 mg weekly [91]. Three patients without an adequate response to other immunomodulating/immunosuppressive treatments, including azathioprine, mitoxantrone or rituximab, were administered tocilizumab. Despite CD20 depletion, the patients relapsed and worsened on the EDSS score. The mean ARR in the patients was 2·6 (range 1·7–2·7). Mean EDSS was 5·0. Tocilizumab was administered at doses of 6 mg/kg every 4 (one patient) or 6 weeks (two patients). Two minor relapses occurred with remission after glucocorticosteroid treatment. ARR decreased to 0·6 [92]. Kieseier *et al.* reported a patient with severe relapses under treatment with rituximab, mitoxantrone and alemtuzumab. Despite the absence of CD3, CD4, CD8, CD19 and CD20 cells in CSF and blood relapses occurred. Glucocorticosteroids were administered and plasma exchange was performed during acute relapses. The patients were given tocilizumab 8 mg/kg i.v. every 4 weeks. EDSS decreased from 6·5 prior to tocilizumab to 2·5. No Gd-enhancing lesions could be detected on MRI [93].

Table 1. Monoclonal antibodies currently used/tested in multiple sclerosis.

mAb	subunits	IgG-subclass	clinical situation	Indication
Natalizumab	Humanized	IgG4	Approved	Active RRMS
Alemtuzumab	Humanized	IgG1 kappa	Phase III*	RRMS
Daclizumab	Humanized	IgG1	Off-label, Phase III	RRMS
Rituximab	Chimeric	IgG1	Off-label, Phases II/III	RRMS, SPMS, PPMS,
Ocrelizumab	Humanized	IgG1	Phase III	RRMS, PPMS
Oftamumab	Human	IgG1	Phase II	RRMS

*Positive opinion for approval of alemtuzumab (Lemtrada®) for active relapsing–remitting multiple sclerosis (RRMS) by the European Medicines Agency(EMA). SPMS: secondary progressive MS; PPMS: primary progressive MS.

Potential side effects

A decline in systolic blood pressure was reported in one patient after the first infusion. Infections such as enteritis and upper respiratory tract infections may occur under tocilizumab. Currently, no severe side affects have been reported [91–93]. Upper respiratory tract infections and hypercholesterolaemia have been reported from clinical trials in rheumatoid arthritis. Headache and reduced white blood cell count are common side effects in juvenile idiopathic polyarthritis and in systemic juvenile idiopathic arthritis [104].

Outlook

Tocilizumab leads to an improvement of the course of disease in NMO patients. It was given in patients with severe disease course, and no adequate response to other treatment. Tocilizumab targets the IL-6 receptor; consequently, the survival of plasmablasts is influenced. Plasmablasts do not express the CD20 marker, and cannot be depleted by rituximab.

The role of Th17 cells has been reported in MS [100]. The humoral immune response seems to be at least partly involved and may play an independent role in the pathogenesis of MS. The role of B cells in MS gained attention after positive results of treatment with rituximab were reported in MS patients [73]. Furthermore, the role of B cells is clear, as in almost every MS patient oligoclonal bands can be seen in the CSF. B cells seem to play an important role in mediating T cell responses [64,65,105]. Thus, from a scientific viewpoint, tocilizumab may be an interesting candidate for treatment in MS.

Eculizumab

Background

Eculizumab is a humanized IgG2 and IgG4 kappa mAb targeting the C5 complement protein. Consequently, the cleavage to C5a and C5b is inhibited, and generation of the membrane attack complex C5b-9 is prevented. It is approved by the EMA for patients with paroxysmal nocturnal haemoglobinuria and atypical haemolytic uraemic syndrome [28,106,107].

Clinical trials

Eculizumab has been tested in a trial with NMO-IgG-positive patients. All patients had an active disease with at least two relapses in the last 6 months or three relapses in the last 12 months. Nine patients were on immunosuppressive treatment with no adequate treatment response prior to inclusion. Patients received 600 mg eculizumab i.v. weekly for the first month, and afterwards 900 mg i.v. every 2 weeks for a period of 48 weeks. Fourteen patients were enrolled; five were treatment-naive. Relapses decreased tremendously during the 12 months period under eculizumab. Twelve patients were relapse-free, whereas in two patients treatment for possible relapses was performed with glucocorticosteroids. No patient worsened on disability outcome scales. Mean EDSS improved from 4·3 to 3·5 [94].

Potential side effects

Headache, dizziness, nausea and diarrhoea were the most frequent side effects. One patient suffered from meningococcal sepsis and sterile meningitis, from which he recovered. One patient suffered a transient ischaemic attack. Rheumatoid arthritis was diagnosed in one patient, as swelling and pain of the hands was reported. The patient had had a similar event 3 years previously [94].

Outlook

To what extent eculizumab will be an option for treatment in MS is difficult to predict. The role of B cells in MS and the humoral immune response is discussed above.

Conclusion

Table 1 gives a summary of mAbs either approved or currently in clinical trials.

MAbs have changed the treatment of multiple sclerosis tremendously. Natalizumab has proved its efficacy, but it also has shown the potential risks and dangers of very potent modern therapeutics. Alemtuzumab might become the second mAb to enter the approved-level stage of therapeutics. The potential risks concern autoimmunity, and have to weighed against its benefits. Management of patients will probably be required for regular monitoring of biological markers to assure patient safety.

Several other mAbs may eventually gain approval in MS, including daclizumab, ocrelizumab and ofatumumab. The ultimate challenge for physicians will be to identify the right drug for the right patient.

Disclosure

Dr Rommer served as consultant to Bayer Pharma AG and received speaker honorary from Shire. Dr Rommer is an employee of Novartis. Dr Dudesek has nothing to disclose. Dr Stüve has received research funding from Teva Pharmaceuticals. He is on the editorial boards of *JAMA Neurology*, *Multiple Sclerosis*, and *Therapeutic Advances in Neurological Disorders*. Dr Zettl received speaker honorary from Bayer Pharma AG, Novartis Pharma AG, Teva Pharma AG, Biogen IDEC, Merck Serono GmbH, Sanofi-Aventis GmbH.

References

1 Köhler G, Milstein C. Continuous cultures of fused cells secreting antibody of predefined specificity. Nature 1975; **256**:495–7.
2 Reichert JM. Monoclonal antibodies as innovative therapeutics. Curr Pharm Biotechnol 2008; **9**:423–30.
3 European Medicines Agency (EMA). Available at: http://www.ema.europa.eu/ema/index.jsp?curl=pages/medicines/human/medicines/003718/smops/Positive/human_smop_000544 (accessed 4 July 2013).
4 Engelhardt B, Kappos L. Natalizumab: targeting alpha4-integrins in multiple sclerosis. Neurodegener Dis 2008; **5**:16–22.
5 Polman CH, O'Connor PW, Havrdova E *et al*. A randomized, placebo-controlled trial of natalizumab for relapsing multiple sclerosis. N Engl J Med 2006; **354**:899–910.
6 Rudick RA, Stuart WH, Calabresi PA *et al*. Natalizumab plus interferon beta-1a for relapsing multiple sclerosis. N Engl J Med 2006; **354**:911–23.
7 Dahlhaus S, Hoepner R, Chan A *et al*. Disease course and outcome of 15 monocentrically treated natalizumab-associated progressive multifocal leukoencephalopathy patients. J Neurol Neurosurg Psychiatry 2013; **84**:1068–74.
8 Bloomgreen S, Richman S, Hotermans C. Risk of natalizumab-associated progressive multifocal leukoencephalopathy. N Engl J Med. 2012; **366**:1870–80.
9 US Food and Drug Administration. Highlights of prescribing information. Available at: http://www.accessdata.fda.gov/drugsatfda_docs/label/2013/125104s813lbl.pdf (accessed 5 July 2013).
10 Kleinschmidt-DeMasters BK, Miravalle A, Schowinsky J *et al*. Update on PML and PML-IRIS occurring in multiple sclerosis patients treated with natalizumab. J Neuropathol Exp Neurol 2012; **71**:604–17.
11 Havla J, Gerdes LA, Meinl I *et al*. De-escalation from natalizumab in multiple sclerosis: recurrence of disease activity despite switching to glatiramer acetate. J Neurol 2011; **258**:1665–9.
12 Vellinga MM, Castelijns JA, Barkhof F, Uitdehaag BM, Polman CH. Postwithdrawal rebound increase in T2 lesional activity in natalizumab-treated MS patients. Neurology 2008; **70**:1150–1.
13 Kerbrat A, Le Page E, Leray E *et al*. Natalizumab and drug holiday in clinical practice: an observational study in very active relapsing remitting multiple sclerosis patients. J Neurol Sci 2011; **308**:98–102.
14 West TW, Cree BA. Natalizumab dosage suspension: are we helping or hurting? Ann Neurol 2010; **68**:395–9.
15 Killestein J, Vennegoor A, Strijbis EM *et al*. Natalizumab drug holiday in multiple sclerosis: poorly tolerated. Ann Neurol 2010; **68**:392–5.
16 O'Connor PW, Goodman A, Kappos L *et al*. Disease activity return during natalizumab treatment interruption in patients with multiple sclerosis. Neurology 2011; **76**:1858–65.
17 Rossi S, Motta C, Studer V *et al*. Effect of glatiramer acetate on disease reactivation in MS patients discontinuing natalizumab. Eur J Neurol 2013; **20**:87–94.
18 Magraner MJ, Coret F, Navarré A *et al*. Pulsed steroids followed by glatiramer acetate to prevent inflammatory activity after cessation of natalizumab therapy: a prospective, 6-month observational study. J Neurol 2011; **258**:1805–11.
19 Rinaldi F, Seppi D, Calabrese M, Perini P, Gallo P. Switching therapy from natalizumab to fingolimod in relapsing–remitting multiple sclerosis: clinical and magnetic resonance imaging findings. Mult Scler 2012; **18**:1640–3.
20 Daelman L, Maitrot A, Maarouf A, Chaunu MP, Papeix C, Tourbah A. Severe multiple sclerosis reactivation under fingolimod 3 months after natalizumab withdrawal. Mult Scler 2012; **18**:1647–9.
21 Cohen M, Maillart E, Papeix C *et al*. ENIGM: A French Observational Study about Switching from Natalizumab to Fingolimod in Multiple Sclerosis. Meeting abstract. Presented at the AAN in San Diego, CA, USA, 12 February 2013.
22 Novartis reports case of PML in Gilenya-treated patient who received prior therapy with Biogen Idec, Elan's Tysabri. Available at: http://www.verusmed.com/articles/view/68696/ (accessed 31 July 2013).
23 Tackenberg B, Hellwig K, Meinl I *et al*. Fingolimod reduces recurrence of disease activity after natalizumab withdrawal in multiple sclerosis. J Neurol 2013; **260**:1382–7.
24 Rommer PS, Zettl UK, Kieseier B *et al*. Requirement for safety monitoring for approved multiple sclerosis therapies: an overview. Clin Exp Immunol 2014; **175**:397–407.
25 Winkelmann A, Loebermann M, Reisinger EC, Zettl UK. Multiple sclerosis treatment and infectious issues: update 2013. Clin Exp Immunol 2014; **175**:425–38.
26 Crowe JS, Hall VS, Smith MA, Cooper HJ, Tite JP. Humanized monoclonal antibody CAMPATH-1H: myeloma cell expression genomic constructs, nucleotide sequence of cDNA constructs and comparison of effector mechanisms of myeloma and Chinese hamster ovary cell-derived material. Clin Exp Immunol 1992; **87**:105–10.
27 Rommer PS, Stüve O, Goertsches R, Mix E, Zettl UK. Monoclonal antibodies in the therapy of multiple sclerosis: an overview. J Neurol 2008; **255** (Suppl. 6):28–35.

28 Rommer PS, Patejdl R, Zettl UK. Monoclonal antibodies in the treatment of neuroimmunological diseases. Curr Pharm Des 2012; **18**:4498–507.
29 Hale G, Bright S, Chumbley G *et al*. Removal of T cells from bone marrow for transplantation: a monoclonal antilymphocyte antibody that fixes human complement. Blood 1983; **62**:873–82.
30 Riechmann L, Clark M, Waldmann H, Winter G. Reshaping human antibodies for therapy. Nature 1988; **332**:323–7.
31 US Food and Drug Administration. Product Approval Information – Licensing Action. Available at: http://www.accessdata.fda.gov/drugsatfda_docs/appletter/2001/alemmil050701L.htm (accessed 5 July 2013).
32 European Medicines Agency (EMA). EPAR summary for the public. Available at: http://www.ema.europa.eu/docs/en_GB/document_library/EPAR_-_Summary_for_the_public/human/000353/WC500025261.pdf (accessed 5 July 2013).
33 Moreau T, Thorpe J, Miller D *et al*. Preliminary evidence from magnetic resonance imaging for reduction in disease activity after lymphocyte depletion in multiple sclerosis. Lancet 1994; **344**:298–301.
34 Coles AJ, Wing MG, Molyneux P *et al*. Monoclonal antibody treatment exposes three mechanisms underlying the clinical course of multiple sclerosis. Ann Neurol 1999; **46**:296–304.
35 Paolillo A, Coles AJ, Molyneux PD *et al*. Quantitative MRI in patients with secondary progressive MS treated with monoclonal antibody Campath 1H. Neurology 1999; **53**:751–7.
36 Coles AJ, Compston DA, Selmaj KW *et al*. CAMMS223 Trial Investigators. Alemtuzumab vs. interferon beta-1a in early multiple sclerosis. N Engl J Med 2008; **359**:1786–801.
37 Coles AJ, Fox E, Vladic A *et al*. Alemtuzumab more effective than interferon β-1a at 5-year follow-up of CAMMS223 clinical trial. Neurology 2012; **78**:1069–78.
38 Cohen JA, Coles AJ, Arnold DL *et al*. CARE-MS I investigators. Alemtuzumab versus interferon beta 1a as first-line treatment for patients with relapsing-remitting multiple sclerosis: a randomised controlled phase 3 trial. Lancet 2012; **380**:1819–28.
39 Coles AJ, Twyman CL, Arnold DL *et al*. CARE-MS II investigators. Alemtuzumab for patients with relapsing multiple sclerosis after disease-modifying therapy: a randomised controlled phase 3 trial. Lancet 2012; **380**:1829–39.
40 Minagar A, Alexander JS, Sahraian MA, Zivadinov R. Alemtuzumab and multiple sclerosis: therapeutic application [Review]. Expert Opin Biol Ther 2010; **10**:421–9.
41 European Medicines Agency (EMA). MabCampath. Available at: http://www.ema.europa.eu/docs/en_GB/document_library/EPAR_-_Summary_for_the_public/human/000353/WC500025261.pdf (accessed 10 July 2013).
42 Clatworthy MR, Wallin EF, Jayne DR. Anti-glomerular basement membrane disease after alemtuzumab. N Engl J Med 2008; **359**:768–9.
43 Ontaneda D, Cohen JA. The benefits and risks of alemtuzumab in multiple sclerosis. Expert Rev Clin Immunol 2013; **9**: 189–91.
44 Jones JL, Phuah CL, Cox AL *et al*. IL-21 drives secondary autoimmunity in patients with multiple sclerosis, following therapeutic lymphocyte depletion with alemtuzumab (Campath-1H). J Clin Invest 2009; **119**:2052–61.
45 Cossburn M, Pace AA, Jones J *et al*. Autoimmune disease after alemtuzumab treatment for multiple sclerosis in a multicenter cohort. Neurology 2011; **77**:573–9.
46 Weetman A. Immune reconstitution syndrome and the thyroid. Best Pract Res Clin Endocrinol Metab 2009; **23**:693–702.
47 Edan G, Le Page E. Induction therapy for patients with multiple sclerosis: why? when? how? CNS Drugs 2013; **27**:403–9.
48 Keene DL, Legare C, Taylor E, Gallivan J, Cawthorn GM, Vu D. Monoclonal antibodies and progressive multifocal leukoencephalopathy. Can J Neurol Sci 2011; **38**:565–71.
49 European Medicines Agency (EMA). MabCampath (alemtuzumab). Withdrawal of the marketing authorisation in the European Union. Available at: http://www.ema.europa.eu/docs/en_GB/document_library/Public_statement/2012/08/WC500130945.pdf (accessed 5 July 2013).
50 Roche. Sterile concentration for injection. Available at: http://www.accessdata.fda.gov/drugsatfda_docs/label/2002/daclhof072902LB.pdf (accessed 5 July 2013).
51 European Medicines Agency (EMA). Public statement on Zenapax (daclizumab). Available at: http://www.ema.europa.eu/docs/en_GB/document_library/Public_statement/2009/11/WC500011995.pdf (accessed 5 July 2013).
52 Wynn D, Kaufman M, Montalban X *et al*., CHOICE investigators. Daclizumab in active relapsing multiple sclerosis (CHOICE study): a phase 2, randomised, double-blind, placebo-controlled, add-on trial with interferon beta. Lancet Neurol 2010; **9**:381–90.
53 Bielekova B, Howard T, Packer AN *et al*. Effect of anti-CD25 antibody daclizumab in the inhibition of inflammation and stabilization of disease progression in multiple sclerosis. Arch Neurol 2009; **66**:483–9.
54 Bielekova B, Richert N, Herman ML *et al*. Intrathecal effects of daclizumab treatment of multiple sclerosis. Neurology 2011; **77**:1877–86.
55 Chanvillard C, Jacolik RF, Infante-Duarte C, Nayak RC. The role of natural killer cells in multiple sclerosis and their therapeutic implications. Front Immunol 2013; **4**:63.
56 Nicholas J, Morgan-Followell B, Pitt D, Racke MK, Boster A. New and emerging disease-modifying therapies for relapsing-remitting multiple sclerosis: what is new and what is to come. J Cent Nerv Syst Dis 2012; **16**:81–103. doi: 10.4137/JCNSD.S6692.
57 Bielekova B, Richert N, Howard T *et al*. Humanized anti-CD25 (daclizumab) inhibits disease activity in multiple sclerosis patients failing to respond to interferon beta. Proc Natl Acad Sci USA 2004; **101**:8705–8.
58 Rose JW, Burns JB, Bjorklund J, Klein J, Watt HE, Carlson NG. Daclizumab phase II trial in relapsing and remitting multiple sclerosis: MRI and clinical results. Neurology 2007; **69**:785–9.
59 Rojas MA, Carlson NG, Miller TL, Rose JW. Long-term daclizumab therapy in relapsing–remitting multiple sclerosis. Ther Adv Neurol Disord. 2009; **2**:291–7.
60 Borges IT, Shea CD, Ohayon J *et al*. The effect of daclizumab on brain atrophy in relapsing–remitting multiple sclerosis. Mult Scler Relat Disord. 2013; **2**:133–40.
61 Gold R, Giovannoni G, Selmaj K *et al*. SELECT study investigators. Daclizumab high-yield process in relapsing–remitting multiple sclerosis (SELECT): a randomised, double-blind, placebo-controlled trial. Lancet 2013; **381**:2167–75.
62 Available at: http://clinicaltrials.gov (accessed 7 July 2013).
63 Roche. Sterile concentrate for injection. Available at: http://www.accessdata.fda.gov/drugsatfda_docs/label/2002/daclhof072902LB.pdf (accessed 7 July 2013).

64 Cross AH, Waubant E. MS and the B cell controversy. Biochim Biophys Acta 2011; **1812**:231–8.
65 Boster A, Ankeny DP, Racke MK. The potential role of B cell-targeted therapies in multiple sclerosis. Drugs 2010; **70**:2343–56.
66 Grillo-López AJ. Rituximab: an insider's historical perspective. Semin Oncol 2000; **27** (Suppl. 12):9–16.
67 Waubant E. Spotlight on anti-CD20. Int MS J 2008; **15**: 19–25.
68 Reff ME, Carner K, Chambers KS *et al*. Depletion of B cells *in vivo* by a chimeric mouse human monoclonal antibody to CD20. Blood 1994; **83**:435–45.
69 Maloney DG, Grillo-López AJ, White CA *et al*. DEC-C2B8 (Rituximab) anti-CD20 monoclonal antibody therapy in patients with relapsed low-grade non-Hodgkin's lymphoma. Blood 1997; **90**:2188–95.
70 US Food and Drug Administration. Rituximab (marketed as Rituxan) information. Available at: http://www.fda.gov/Drugs/DrugSafety/PostmarketDrugSafetyInformationforPatientsandProviders/ucm109106.htm (accessed 7 July 2013).
71 European Medicines Agency (EMA). MabThera. Available at: http://www.ema.europa.eu/ema/index.jsp?curl=pages/medicines/human/medicines/000165/human_med_000897.jsp&mid=WC0b01ac058001d124 (accessed 7 July 2013).
72 Tony HP, Burmester G, Schulze-Koops H *et al*., GRAID investigators. Safety and clinical outcomes of rituximab therapy in patients with different autoimmune diseases: experience from a national registry (GRAID). Arthritis Res Ther 2011; **13**:R75.
73 Hauser SL, Waubant E, Arnold DL *et al*., HERMES Trial Group. B-cell depletion with rituximab in relapsing–remitting multiple sclerosis. N Engl J Med 2008; **358**:676–88.
74 Naismith RT, Piccio L, Lyons JA *et al*. Rituximab add-on therapy for breakthrough relapsing multiple sclerosis: a 52-week phase II trial. Neurology 2010; **74**:1860–7.
75 Monson NL, Cravens PD, Frohman EM, Hawker K, Racke MK. Effect of rituximab on the peripheral blood and cerebrospinal fluid B cells in patients with primary progressive multiple sclerosis. Arch Neurol 2005; **62**:258–64.
76 Hawker K, O'Connor P, Freedman MS *et al*., OLYMPUS trial group. Rituximab in patients with primary progressive multiple sclerosis: results of a randomized double-blind placebo-controlled multicenter trial. Ann Neurol 2009; **66**:460–71.
77 Rommer PS, Patejdl R, Winkelmann A, Benecke R, Zettl UK. Rituximab for secondary progressive multiple sclerosis: a case series. CNS Drugs 2011; **25**:607–13.
78 Brown BA, Torabi M. Incidence of infusion-associated reactions with rituximab for treating multiple sclerosis: a retrospective analysis of patients treated at a US centre. Drug Saf 2011; **34**:117–23.
79 Bar-Or A, Calabresi PA, Arnold D *et al*. Rituximab in relapsing–remitting multiple sclerosis: a 72-week, open-label, phase I trial. Ann Neurol 2008; **63**:395–400.
80 Cree B. Emerging monoclonal antibody therapies for multiple sclerosis. Neurologist 2006; **12**:171–8.
81 Palazzo E, Yahia SA. Progressive multifocal leukoencephalopathy in autoimmune diseases. Joint Bone Spine 2012; **79**:351–5.
82 Kausar F, Mustafa K, Sweis G *et al*. Ocrelizumab: a step forward in the evolution of B-cell therapy. Expert Opin Biol Ther 2009; **9**:889–95.
83 Kappos L, Li D, Calabresi PA *et al*. Ocrelizumab in relapsing–remitting multiple sclerosis: a phase 2, randomised, placebo-controlled, multicentre trial. Lancet 2011; **378**:1779–87.
84 Hughes S. Ocrelizumab in MS: Encouraging Long-term Data. 29 March 2013. Available at: http://www.medscape.com/viewarticle/781671 (accessed 8 July 2013).
85 Reid K. UPDATE 4-Roche, Biogen suspends arthritis drug after deaths. 8 March 2010. Available at: http://www.reuters.com/article/2010/03/08/roche-idUSLDE62705720100308 (accessed 8 July 2013).
86 Zhang B. Ofatumumab. MAbs 2009; **1**:326–31.
87 European Medicines Agency (EMA). Arzerra. Available at: http://www.ema.europa.eu/docs/en_GB/document_library/EPAR_-_Summary_for_the_public/human/001131/WC500093092.pdf (accessed 8 July 2013).
88 US Food and Drug Administration. Arzerra™ (Ofatumumab) injection for intravenous use. Available at: http://www.fda.gov/ohrms/dockets/ac/09/briefing/2009-4444b1-02-GSK.pdf (accessed 8 July 2013).
89 Genmab press release. 10 September 2010; Available at: http://ir.genmab.com/releasedetail.cfm?ReleaseID=639964 (accessed 8 July 2013).
90 Soelberg Sorensen P, Drulovic J, Havrdova E, Lisby S, Graff O, Shackelford S. 2010. Magnetic resonance imaging (MRI) efficacy of ofatumumab in relapsing remitting multiple sclerosis (RRMS) – 24-week results of a phase II study. Presented at: ECTRIMS; October 13–16, 2010; Gothenburg, Sweden, P136.
91 Araki M, Aranami T, Matsuoka T, Nakamura M, Miyake S, Yamamura T. Clinical improvement in a patient with neuromyelitis optica following therapy with the anti-IL-6 receptor monoclonal antibody tocilizumab. Mod Rheumatol 2013 **23**:827–31.
92 Ayzenberg I, Kleiter I, Schröder A *et al*. Interleukin 6 receptor blockade in patients with neuromyelitis optica nonresponsive to anti-CD20 therapy. JAMA Neurol 2013; **70**:394–7.
93 Kieseier BC, Stüve O, Dehmel T *et al*. Disease amelioration with tocilizumab in a treatment-resistant patient with neuromyelitis optica: implication for cellular immune responses. JAMA Neurol 2013; **70**:390–3.
94 Pittock SJ, Lennon VA, McKeon A *et al*. Eculizumab in AQP4-IgG-positive relapsing neuromyelitis optica spectrum disorders: an open-label pilot study. Lancet Neurol 2013; **12**: 554–62.
95 US Food and Drug Administration. Medication Guide. Available at: http://www.fda.gov/downloads/Drugs/DrugSafety/UCM197463.pdf (accessed 8 July 2013).
96 Lennon VA, Wingerchuk DM, Kryzer TJ *et al*. A serum autoantibody marker of neuromyelitis optica: distinction from multiple sclerosis. Lancet 2004; **364**:2106–12.
97 Chihara N, Aranami T, Sato W *et al*. Interleukin 6 signaling promotes anti-aquaporin 4 autoantibody production from plasmablasts in neuromyelitis optica. Proc Natl Acad Sci USA 2011; **108**:3701–6.
98 Mitsdoerffer M, Kuchroo V, Korn T. Immunology of neuromyelitis optica: a T cell–B cell collaboration. Ann NY Acad Sci 2013; **1283**:57–66.
99 Li Y, Wang H, Long Y, Lu Z, Hu X. Increased memory Th17 cells in patients with neuromyelitis optica and multiple sclerosis. J Neuroimmunol 2011; **234**:155–60.

100 Warabi Y, Yagi K, Hayashi H, Matsumoto Y. Characterization of the T cell receptor repertoire in the Japanese neuromyelitis optica: T cell activity is up-regulated compared to multiple sclerosis. J Neurol Sci 2006; **249**:145–52.

101 European Medicines Agency (EMA). RoActemra. Available at: http://www.ema.europa.eu/ema/index.jsp?curl=pages/medicines/human/medicines/000955/human_med_001042.jsp&mid=WC0b01ac058001d124 (accessed 6 August 2013).

102 Zhou L, Ivanov II, Spolski R *et al*. IL-6 programs T(H)-17 cell differentiation by promoting sequential engagement of the IL-21 and IL-23 pathways. Nat Immunol 2007; **8**:967–74.

103 Torchinsky MB, Blander JM. T helper 17 cells: discovery, function, and physiological trigger. Cell Mol Life Sci 2010; **67**:1407–21.

104 Dobson R, Meier UC, Giovannoni G. More to come: humoral immune responses in MS. J Neuroimmunol 2011; **15**: 13–21.

105 European Medicines Agency (EMA). Soliris. Available at: http://www.ema.europa.eu/ema/index.jsp?curl=pages/medicines/human/medicines/000791/human_med_001055.jsp&mid=WC0b01ac058001d124 (accessed 8 July 2013).

106 US Food and Drug Administration. Highlights of prescribing information. Available at: http://www.accessdata.fda.gov/drugsatfda_docs/label/2011/125166s172lbl.pdf (accessed 8 July 2013).

107 Hinson SR, Pittock SJ, Lucchinetti CF *et al*. Pathogenic potential of IgG binding to water channel extracellular domain in neuromyelitis optica. Neurology 2007; **69**:2221–31.