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Use of non-human primate disease models

Non-human primates are essential models in the translational research of multiple sclerosis

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Ageing Western societies are facing an increasing prevalence of chronic inflammatory and degenerative diseases for which no effective treatments exist. The pressure on the drug development industry to develop such treatments creates a need for translationally relevant animal models, which faithfully replicate essential pathogenic mechanisms of the human disease. In this Short Review, we discuss the essential role of the non-human primate (NHP) in the translational research into the pathogenesis and treatment of the autoimmune neurological disease multiple sclerosis (MS).

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Introduction

Multiple sclerosis (MS) is an autoimmune neurological disease characterized clinically by the accumulation of neurological deficits, including sensory and motor functions, and pathologically by the presence of inflammatory/demyelinated lesions in the brain and spinal cord [1]. Lesions are usually well-defined areas of inflammation and tissue injury, which can be visualized with magnetic resonance imaging (MRI). Fig. 1a shows the clinical presentation of MS in the majority of patients (±85%) with relapse-onset disease: Pre-symptomatic MS is rarely diagnosed as it remains largely subclinical, but lesions can be detected as hyper-intense foci on brain MRI scans, such as depicted in Fig. 1b. This is followed by a period of variable length, ranging from a few to many years, where episodes of neurological disability (relapses) alternate with intermittent recovery (remission). Commonly observed focal neurological deficits include loss of sensation, visual symptoms, motor paralysis, as well as bowel and bladder dysfunction. Fatigue, cognitive disturbance, and neuropathic pain are also very common. In approximately two thirds of patients, relapsing remitting (RR) disease evolves into secondary progressive (SP) disease, in which there is progressive worsening of symptoms that become independent of relapses. By contrast, approximately 15% of the patients experience primary progressive disease, in which symptoms are slowly progressive from the start (primary progressive MS, PPMS).

Fig. 1b shows the main pathological hallmarks of MS, namely infiltration of blood-borne immune cells (inflammation), destruction of myelin sheaths around axons (demyelination) and degeneration of neurons and axons (neurodegeneration), which culminates in substantial reduction of brain mass (atrophy) (for review: Ref. [2]). In

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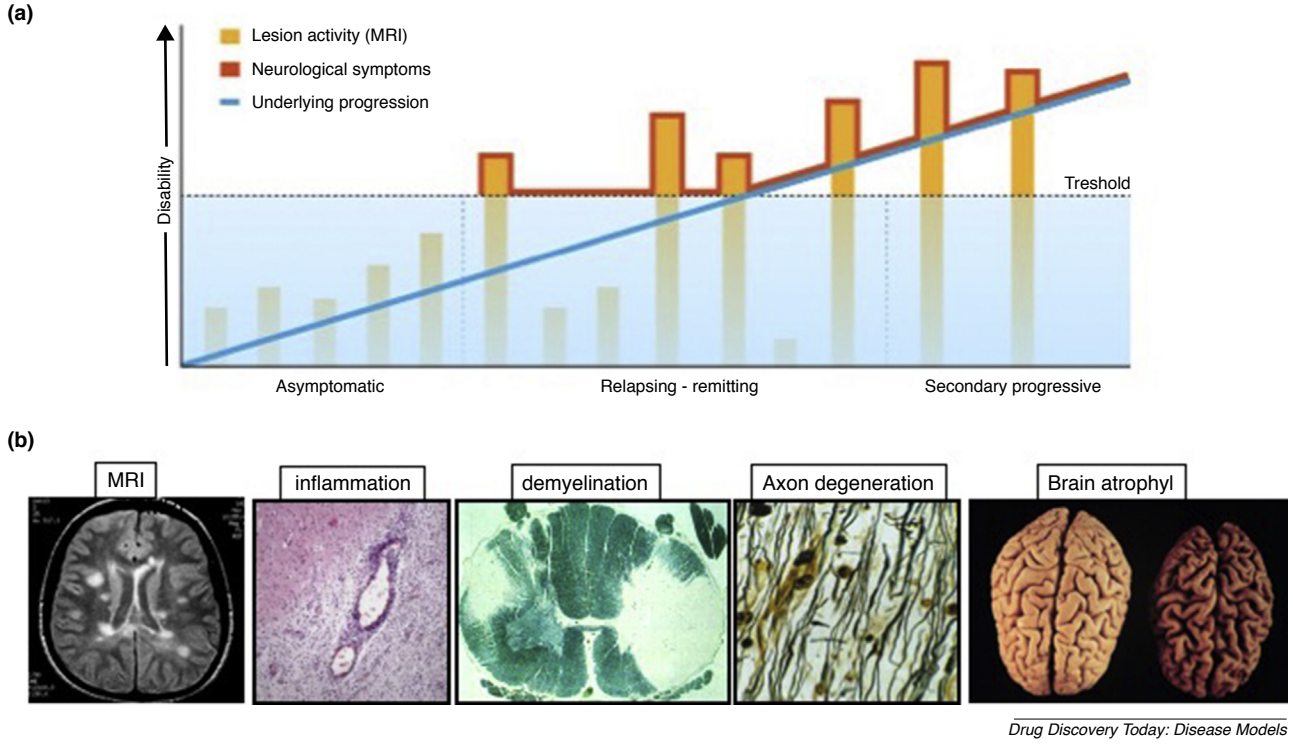

Fig. 1. Clinical (a) and pathological (b) aspects of multiple sclerosis.
(a) is a graphical representation of the relapsing-remitting (RR)/secondary progressive (SP) disease course that is found in the majority of the patients. We posit that this type of MS is caused by degenerative pathology, affecting neurons and oligodendrocytes (blue line), on which discrete episodes of autoimmune-driven inflammatory activity (relapse; orange bars) are superimposed. The ensuing disease activity is indicated by the red line. Primary progressive (PP) MS is caused by the degenerative pathology without the superimposed relapses (blue line only).

presymptomatic MS, lesions are thought to be mainly inflammatory with limited demyelination, while lesions in RRMS display self-limiting inflammation and demyelination with potential repair by new myelin formation (remyelination) as oligodendrocytes are spared. In progressive MS damage becomes permanent as degeneration of neurons and oligodendrocytes becomes more irreversible.

For the treatment of RRMS a number of reasonably effective disease modifying immunotherapies, ranging from low to medium and high efficacy, are available for clinical use with a tendency for more potent disease-modifying drug (DMD) to be associated with more significant side effects and risks of opportunistic infections. With regards to progressive MS, only one DMD has most recently been approved by the American Food and Drug Administration (ocrelizumab, for PPMS), while the European Medicines Agency and other agencies are still assessing it [3]. Despite the successes in experimental medicine, a considerable number of innovative treatments obtained in animal models, failed to reproduce promising effects when tested in patients and sometimes exerted detrimental effects. Such experiences indicate that the disease models currently used in preclinical research, do not always include essential aspects of the human disease or adequately represent pathological aspects of MS. Accumulating evidence reviewed in this publication indicates that the non-human primate (NHP) models of MS may help bridge the translational gap between currently used rodent disease models and the patient.

Modeling MS in animals

Although the cause of MS is unknown, genomic and epidemiological studies indicate that the initiation and progression of MS involves autoimmune reactions elicited by the interaction of genetic and environmental factors. Of the ±200 genes now found associated with the risk of developing MS, the vast majority has a function in the immune system [4]. Moreover, the strongest environmental risk factors – late infection with Epstein Barr Virus, vitamin D insufficiency and smoking – have been associated with modulation of immunological functions [5]. These observations have identified the immune system as the main culprit in MS and therefore the most relevant target of intervention therapy.

The possibility to directly investigate the disease process in patients is limited by ethical reasons and by the inaccessibility of the target organs, being brain and spinal cord, collectively indicated as CNS. Hence, animal models are indispensable for the translational research of pathogenic

mechanisms and therapy development. Nowadays, MS researchers can choose from many animal models, ranging from *Caenorhabditis elegans* worms and Drosophila flies to vertebrate animals, such as zebrafish, mice, rats and monkeys. The cumulative contribution of each of these models to our current understanding of immunological and neurodegenerative processes has been immense. Nevertheless, translation of scientific discoveries into effective therapies for the MS patient has been notoriously difficult. The two main causes of this attrition, lack of efficacy and unforeseen toxicity, indicate that a promising clinical effect of a new treatment in currently used animal models has insufficient predictive value for clinical success [6].

The validity of an animal model for the understanding of a human disease is based on at least 4 criteria: 1. **Face validity**, representing the phenomenological and pathophysiological similarity; 2. **Predictive validity**, representing the ability of a model to correctly predict the efficacy of a treatment; 3. **Construct validity**, representing the degree of similarity in the pathophysiological mechanisms and symptoms; 4. **External validity**, representing the extent to which the observed effect of a treatment can be generalized to the diverse MS patient population [7]. In addition to these basic validity criteria, several more practical criteria are used such as reproducibility, background knowledge, amenability to experimental manipulation, ethics and costs.

The lowest laboratory animal species with a human-like immune system with regard to basic design and function is the mouse. By far the largest proportion of current preclinical research into MS is based on a limited number of genetically homogeneous (inbred) mouse strains, which are bred and raised under very clean, specific pathogen-free (SPF) conditions. Experimental manipulations eliciting relevant clinical and/or pathological aspects of MS in these mice include: 1. Genetic modification, 2. Active disease induction by the injection of CNS homogenate or purified CNS proteins formulated with (an) immune potentiating adjuvant(s) and 3. Passive disease induction by transfer of immune cells or molecules from a diseased animal to a suitable healthy recipient. The current discussion will be limited to the actively induced model, called Experimental Autoimmune Encephalomyelitis (EAE).

Experimental autoimmune encephalomyelitis (EAE)

EAE is by far the most frequently used MS animal model. However, it is pertinent to emphasize here that many differences exist between EAE and MS (Refs. [8–10] and Table 1), which may explain the high failure of new treatments in the translation from EAE to MS. Nevertheless, we believe that despite shortcomings of the model, a well-designed study in an optimal EAE model can provide relevant information on the clinical relevance of a new treatment [11].

Mouse EAE

Active EAE is induced by the combined activation of adaptive and innate immune mechanisms in genetically susceptible mouse strains, such as C57BL/6, SJL/J and Biozzi ABH, via inoculation of antigen/adjuvant emulsion [9]. The antigen required for the reproducible induction of robust EAE depends on the genetic background of the mice [11]. The most frequently used and best characterized mouse models in studies on MS pathogenesis and screening of drug candidates are RR EAE induced with proteolipid protein peptide (PLP) 139–151 in SJL/J mice and progressive EAE induced with myelin oligodendrocyte glycoprotein (MOG) peptide 35–55 in C57BL/6 mice. Immunization of Biozzi ABH mice with MOG peptide 8–21 elicits an elegant, albeit less frequently used, RR/SP MS like-disease model [12]. There are also several important EAE models in rats, but with the current focus on the mouse these are much less used than until a decade ago.

Mouse EAE is initiated by CNS infiltration of CD4+ T cells, which upon transmigration of the blood brain barrier (BBB) and interaction with local antigen presenting cells (APC), such as perivascular macrophages and microglia cells, release pro-inflammatory factors within the CNS parenchyma. These enhance permeability of the BBB for macrophages and B cells and for serum factors such as antibodies and complement. Collectively, these factors undertake the autoimmune attack on the myelinated axons (Fig. 2). Debris from the injured myelin sheaths are removed from the CNS by myeloid cells, which drain to cervical and lumbar lymph nodes [13]. The observation that surgical removal of these draining lymph nodes abrogates the characteristic chronic relapsing EAE course in Biozzi ABH mice suggests that new T cell specificities are activated there, which drive EAE chronicity [14].

This cascade of pathophysiological reactions has been the template for therapy development in MS. However, the failure of almost all therapies targeting CD4+ T cells in the translation from EAE model to MS patient has shed doubt on the construct validity of the mouse-EAE based CD4-dominated disease concept [15]. The question is therefore warranted whether EAE is an inadequate model for MS, or whether inbred/SPF mice are imperfect models for the human autoimmune disease.

Non-human primates (NHP) EAE

The close evolutionary proximity of human and non-human primates is reflected by their close genetic and immunological proximity. For the NHP species used for EAE modeling, *Macaca mulatt*a (rhesus monkey), *Macaca fascicularis* (cynomolgus monkey) and *Callithrix jacchus* (common marmoset), the evolutionary distances have been estimated at 25, 25 and 35 million years. Different from SPF-bred mice but just like humans, NHP are genetically outbred and have been freely exposed from birth to environmental microbes, which shape

Table 1. Cininal, pathological and immunological aspects of mouse and primate EAE models, compared with MS

	Mouse EAE	Old World primate EAE	New World primate EAE	MS
General	Max lifespan ±2 years Closed/clean environment Standard diet	Max lifespan ±20 years Open/dirty environment Varied diet	Max lifespan ±15 years Open/dirty environment Varied diet	Max lifespan >90 years Open/dirty environment Varied diet
Induction				
- Active immunization	Myelin/MBP/PLP/MOG + CFA + *B. pertussis* (toxin)	Myelin/MBP/MOG + CFA or IFA	Myelin/MBP/PLP/MOG + CFA or IFA	N.A. (spontaneous)
- Passive transfer	Yes, within inbred strain	Only autologous	Autologous + between twins	N.A.
Genetics				
-Status	Inbred	Outbred	Outbred	Outbred
- Susceptibility	MHC II	MHC II	MHC I and II	MHC I and II + >200 genes
Disease course				
- Hyperacute onset	Common	Common	Uncommon	Rare (ADEM)
- Relapsing-remitting	Model-dependent (SJL/PLP)	Uncommon	Common	Common
- Progressive	Model-dependent (B6/MOG)	Never	Common	Common
Pathology				
White matter				
- Inflammation	CD4+ T cell/macrophage	T cell/neutrophil	T cells/macrophage/μglia	Macrophage/μglia/T&B cells
- Demyelination	Primary demyel	Primary demyel + necrosis	Primary demyel	Primary demyel
- Remyelination	Rare	Rare	Present	Present
- (Neuro)degeneration	Absent	Absent	Absent	Present
Grey matter				
- Inflammation	Rare	Absent	Meningeal	Meningeal
- Demyelination	Rare	Absent	Subpial/intracort/leukocort	Subpial/intracort/leukocort
- Remyelination	NA	NA	Present	Present
- (Neuro)degeneration	NA	NA	Present	Present
Immunology				
- CD4+ T cell	Proven pathogenic role Th1/17	Proven pathogenic role Th1	Proven pathogenic role Th1	Early-stage pathogenic role?
- CD8+ T cell	Pathogenic role uncertain	Pathogenic role uncertain	Proven pathogenic role	Late-stage? pathogenic role?
- B cell	Depletion improves disease	Not tested	Depletion improves disease	Depletion improves disease
- Antibody	Facilitates ADCC/CDC	Not tested	Facilitates ADCC/CDC	Involvement in type II lesions
- T reg cells	Protective role	Not tested	Not tested	Unclear role

their immune system and underlie the remarkable susceptibility to EAE [16].

During the past 20 years, an in-depth analysis has been carried out in marmosets of the immune reactions elicited by injection with human CNS myelin. As the details of these studies have been reviewed elsewhere [17–19], we will only summarize the most salient findings here.

- Immunization of marmosets with myelin isolated from an MS patient brain elicited a chronic progressive neurological disease that approximates MS in clinical and pathological presentation [20]. A combined radiological (MRI) and neuropathological analysis showed that formation of MS-like lesions occurs disseminated in time and space, just like in the human disease. Subsequent analyses showed that lesions are present in the white as well as grey matter of brain and spinal cord [21].
- Disease progression in mouse EAE models is associated with diversification of the T cell and antibody response, a phenomenon known as epitope spreading, but the nature of the response does not essentially change. This is different in marmoset EAE. After the observation that autoimmunity against MOG is essential for chronic EAE development [22], two pathogenically relevant autoimmune pathways were identified triggered by (recombinant) human MOG. These pathways have been extensively reviewed elsewhere [11,18,23]. In brief, one pathway replicates autoimmune mechanisms and pathology observed in mouse EAE models and involves a synergistic attack on CNS myelin of pro-inflammatory T cells and myelin binding autoantibodies. A second pathway, which has no known correlate in mouse EAE models, involves autoaggressive cytotoxic T cells (CTL), which seem to drive EAE progression and have the capacity to kill myelin forming oligodendrocytes.
- The marmoset EAE model displays a clear pathogenic role of simian herpesviruses related to those associated with MS, namely cytomegalovirus (CMV) [24] and Epstein Barr Virus (EBV) [19]. The available evidence indicates that the auto-

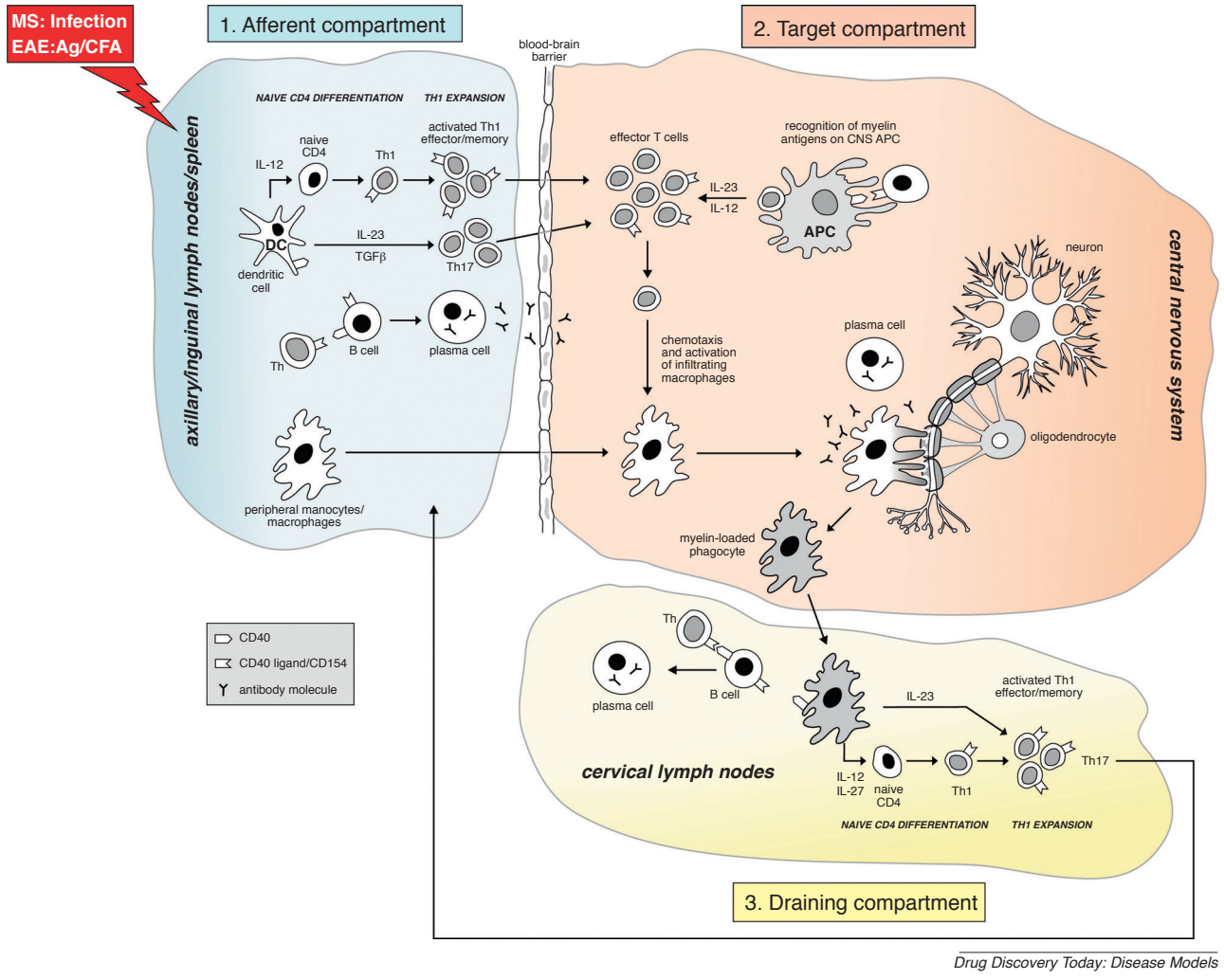

Fig. 2. Three-compartment model for the EAE immunopathogenesis (from Ref. [45]).
MS is thought to start by an infection that activates autoreactive T and B lymphocytes in the peripheral immune compartment (afferent compartment; grey). In the EAE model this is achieved by the injection of antigen (Ag)-adjuvant (CFA) emulsion. The activated immune cells are released into the blood, from where they transmigrate the blood–brain-barrier and infiltrate the CNS. Within this target compartment reactivation occurs via interaction with resident antigen presenting cells (APC), eliciting a cascade of pathophysiological events culminating in an attack on axon-myelin units. Debris are removed from the CNS either by passive drainage (not shown) or by phagocytes. These debris can be found within phagocytic cells in the CNS-draining cervical and lumbar lymph nodes. Conceptually, within this draining compartment (yellow), new T and B cell specificities are activated, which are seeded over the afferent compartment.

aggressive CTL may originate from a repertoire of effector memory T cells, generated for keeping chronic CMV expression quiescent. In addition, infection with EBV endows B lymphocytes with the capacity to activate these T cells. This concept has been highlighted as a novel association between infection and autoimmunity [25].

- Autoreactive T cells present in the pathogen-educated marmoset immune system can be directly activated by injection of a synthetic peptide, representing residues 34–56 of human MOG, adjuvated with the mineral oil IFA [26]. The absence of danger signals in this formulation may explain why SPF-bred Biozzi ABH and C57BL/6 mice fail to develop a reaction against this formulation [26]. Recent data show that this novel pathogenic mechanism elicits neuropathological aspects of progressive MS, including cortical grey matter demyelination, activation of oxidative injury mechanisms, redistribution of iron and damage to mitochondria [27]. We can therefore speculate that the transition of the mouse EAE-like pathogenic pathway 1 to the more MS-like pathogenic pathway 2 represents the transition from RRMS to SPMS.

Corroborating the validity of marmoset EAE for MS

The remarkable neuropathological similarities between the marmoset EAE model and MS (**face validity**) does not necessarily imply that the underlying pathogenic mechanisms are relevant for MS (**construct validity**). To assess

the **construct and predictive validities** of marmoset EAE as preclinical MS model, the effects of therapeutic mAbs that survived or failed in the translation from mouse EAE to MS were assessed.

Pathway 1-CD4+ Th cells

In mouse EAE models, two pathogenic Th subsets were defined, namely Th1 and Th17, which differentiate from Th0 progenitor cells under the influence of the IL-12/IL-23 cytokine axis [28]. Experiments with a mAb generated against the shared p40 subunit of both cytokines (ustekinumab; IL-12p40) showed protection of marmosets against EAE when treatment was started at the time of EAE induction [29]. In contrast, late treatment exerted only a moderate clinical effect, although the MRI-detectable activity and enlargement of brain lesions were suppressed [30]. The same antibody exerted no significant beneficial effect in a RRMS clinical trial [31]. We also tested an anti-IL-17A mAb in the marmoset model; this mAb also failed to show a beneficial clinical effect [32]. Of note, the anti-IL-17A mAb secukinumab exerted only a moderate beneficial effect in RRMS on MRI-detectable lesion activity [33]. Interestingly, both ustekinumab and secukinumab show satisfactory clinical effects in psoriasis patients.

An explanation for the discrepancies between EAE and RRMS may be that the pathogenic pathway 1 mechanism may represent only the biological onset of MS, which probably occurs long before the diagnosis RRMS is made.

B cells

A recent publication describes the remarkable history of B cell depletion as exciting new treatment for MS [34]. The original thought behind this treatment was to get rid of autoantibodies that upon binding myelin activate myelin destruction mechanisms. Contrary to expectations, treatment of RRMS patients with a mAb directed against CD20, a broadly expressed surface marker in the B cell lineage, exerted a dramatic and long-lasting clinical effect, associated with dramatic and almost immediate reduction of inflammatory lesion activity [35]. The observation that antibody levels were not altered was remarkable, although may be explained by the lack of CD20 expression on plasma cells. Another type of treatment aiming at the depletion of B cells works by capturing factors that B cells need for survival and differentiation, such as ''B lymphocyte stimulator'' (BlyS) and 'a proliferation inducting ligand' (APRIL) [36]. This was achieved with atacicept, a soluble fusion protein combining the joint receptor of BlyS and APRIL on B cells (TACI) with the Fc part of human IgG. This construct showed promising clinical effects in SLE patients [37], but worsened RRMS [38]. Replication of these two treatment concepts in marmoset EAE showed that in both scenarios circulating B cells were depleted, but that the anti-CD20 mAb exerted a superior clinical effect. The explanation found was the differential depletion of CalHV3, the EBV-related lymphocryptovirus (LCV) of marmosets, from the immune repertoire paralleling the discrepant clinical effect. These findings led us to posit a core pathogenic role of LCV-infected B cells in the pathogenic process [39].

CD8+ T cells in pathway 2

It is well recognized that the T cell infiltrate in the MS lesion is dominated by CD8+ T cells [40]. This dominance is not commonly reflected in EAE models, although it was found in virus-induced models of MS [41]. However, the latter models are beyond the scope of this article as equivalents have not been established in NHP. We are not aware of ongoing clinical trials testing the efficacy of CD8+ T cell antagonists in RRMS. However, we observed a profound clinical effect of anti-human CD20 mAb in the MOG34-56/IFA induced marmoset EAE model, which is driven by autoaggressive CD8+ CTL.

Concluding remarks

Despite the dramatic progress made in the molecular analysis of MS pathogenesis, translation of data from biomedical research into clinical applications remains a challenge. The problems encountered with technology transfer from bench to bedside are not new and are not confined to MS, but affects almost all clinical disciplines. It is also not a European science problem, but a world-wide concern. Within the European Union, research programs such as Horizon 2020 and Innovative Medicine Initiative have been set up to fund the building of bridges across this ''valley of death''.

The necessity to improve the predictive validity of the animal models used in preclinical research is clear [42]. We believe that lessons should be learned from a detailed analysis of the reasons why translation failed and this knowledge can be used for adjustment of the used animal model. As explained elsewhere, advantage can be taken from the two dimensions of the NHP EAE model for this reverse translation exercise as these create a useful bridge between the rodent EAE model and the MS patient [43]. The relevance of the NHP in drug development exceeds the advantage of immunological cross-reactivity for therapeutic mAb, as the disease models provide also important information on the MS pathogenesis that cannot be obtained in mouse EAE. In this respect, the NHP is an equally important pillar under the bridge that connects preclinical and clinical research, as molecular cell biology and mouse disease models (Fig. 3).

Scientists using NHP for their preclinical research need to take the concerns in society and politics seriously and invest where possible in alternatives for research in the living primate as defined in the 3R principles: Replacement, Reduction and Refinement. However, it is pertinent to emphasize here that these principles were formulated with the discomfort to the animals in mind, not the clinical relevance of a disease

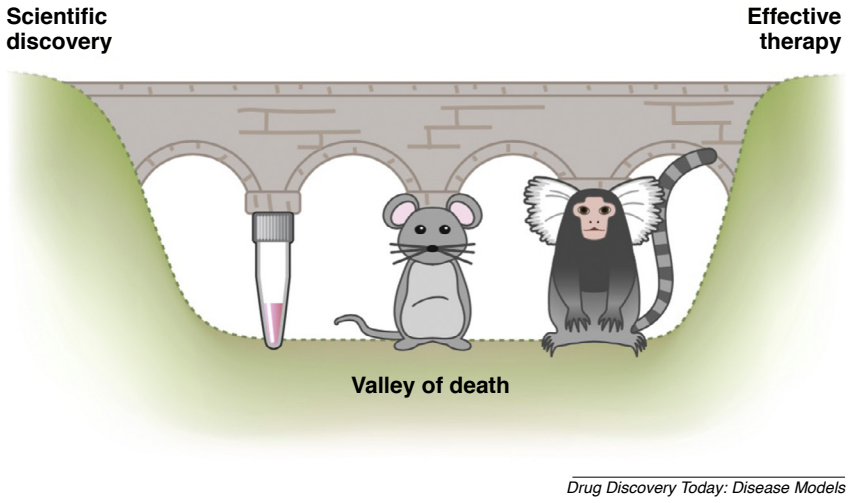

Fig. 3. Bridging the valley of death in MS therapy development: three pillars.
Classical translational research into the pathogenesis and treatment of MS is based on discovery research in cell or tissue cultures (pillar 1). Proof that a new scientific concept is valid for the patient is usually tested in standard rodent EAE models (pillar 2). When safety data have been obtained in relevant animal models, which in the case of biologicals can involve non-human primates, clinical evaluation is started. We posit here that a third pillar is missing, namely preclinical efficacy tests in non-human primate EAE models to reduce the failure rate of new therapies in the clinic because of the lack of efficacy.

model for drug development. This is illustrated by the experience that an investment in Clinical Relevance and Refinement can create conflicts with the Reduction principle, which hampers their application [44].

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References

[1] Dendrou CA, et al. Immunopathology of multiple sclerosis. Nat Rev Immunol 2015;15(9):545–58.
[2] Lassmann H. Pathology and disease mechanisms in different stages of multiple sclerosis. J Neurol Sci 2013;333(1–2):1–4.
[3] Ransohoff RM, et al. Multiple sclerosis—a quiet revolution. Nat Rev Neurol 2015;11(3):134–42.
[4] Sawcer S, et al. Multiple sclerosis genetics. Lancet Neurol 2014;13(7):700–9.
[5] Ascherio A, et al. The initiation and prevention of multiple sclerosis. Nat Rev Neurol 2012;8(11):602–12.
[6] Kola I, Landis J. Can the pharmaceutical industry reduce attrition rates? Nat Rev Drug Discov 2004;3(8):711–5.
[7] Vervliet B, Raes F. Criteria of validity in experimental psychopathology: application to models of anxiety and depression. Psychol Med 2013;43(11):2241–4.
[8] Friese MA, et al. The value of animal models for drug development in multiple sclerosis. Brain 2006;129(Pt. 8):1940–52.
[9] Mix E, et al. Animal models of multiple sclerosis for the development and validation of novel therapies — potential and limitations. J Neurol 2008;255(Suppl. 6):7–14.
[10] Mix E, et al. Animal models of multiple sclerosis–potentials and limitations. Prog Neurobiol 2010;92(3):386–404.
[11] 't Hart BA, et al. EAE: imperfect but useful models of multiple sclerosis. Trends Mol Med 2011;17(3):119–25.
[12] Amor S, et al. Biozzi mice: of mice and human neurological diseases. J Neuroimmunol 2005;165(1–2):1–10.
[13] Laman JD, Weller RO. Drainage of cells and soluble antigen from the CNS to regional lymph nodes. J Neuroimmune Pharmacol 2013;8(4):840–56.
[14] van Zwam M, et al. Surgical excision of CNS-draining lymph nodes reduces relapse severity in chronic-relapsing experimental autoimmune encephalomyelitis. J Pathol 2009;217(4):543–51.
[15] Ransohoff RM. EAE: pitfalls outweigh virtues of screening potential treatments for multiple sclerosis. Trends Immunol 2006;27(4):167–8.
[16] Haanstra KG, et al. Induction of experimental autoimmune encephalomyelitis with recombinant human myelin oligodendrocyte glycoprotein in incomplete Freund's adjuvant in three non-human primate species. J Neuroimmune Pharmacol 2013;8(5):1251–64.
[17] 't Hart BA, et al. Modelling of multiple sclerosis: lessons learned in a non-human primate. Lancet Neurol 2004;3:589–97.
[18] 't Hart BA, et al. The primate autoimmune encephalomyelitis model; a bridge between mouse and man. Ann Clin Transl Neurol 2015;2(5):581–93.
[19] 't Hart BA, et al. EBV infection and multiple sclerosis: lessons from a marmoset model. Trends Mol Med 2016;22(12):1012–24.
[20] 't Hart BA, et al. Histopathological characterization of magnetic resonance imaging-detectable brain white matter lesions in a primate model of multiple sclerosis: a correlative study in the experimental autoimmune encephalomyelitis model in common marmosets (*Callithrix jacchus*). Am J Pathol 1998;153(2):649–63.
[21] Kap YS, et al. The common marmoset as an indispensable animal model for immunotherapy development in multiple sclerosis. Drug Discov Today 2016;21(8):1200–5.
[22] Jagessar SA, et al. Autoimmunity against myelin oligodendrocyte glycoprotein is dispensable for the initiation although essential for the progression of chronic encephalomyelitis in common marmosets. J Neuropathol Exp Neurol 2008;67(4):326–40.
[23] 't Hart BA, et al. A B cell-driven autoimmune pathway leading to pathological hallmarks of progressive multiple sclerosis in the marmoset experimental autoimmune encephalomyelitis model. Front Immunol 2017;8:804.
[24] Vanheusden M, et al. Cytomegalovirus: a culprit or protector in multiple sclerosis? Trends Mol Med 2015;21(1):16–23.

[25] Moore DJ. Viral infection crosses up antigen presentation to drive autoimmunity. Sci Transl Med 2016;8(349). 349ec120.
[26] Jagessar SA, et al. Induction of progressive demyelinating autoimmune encephalomyelitis in common marmoset monkeys using MOG34-56 peptide in incomplete freund adjuvant. J Neuropathol Exp Neurol 2010;69(4):372–85.
[27] Dunham JD, et al. Oxidative injury and iron redistribution are pathological hallmarks of marmoset experimental autoimmune encephalomyelitis. J Neuropathol Exp Neurol 2017;76(6):467–78.
[28] Gran B, et al. Role of the IL-12/IL-23 system in the regulation of T-cell responses in central nervous system inflammatory demyelination. Crit Rev Immunol 2004;24(2):111–28.
[29] Brok HP, et al. Prevention of experimental autoimmune encephalomyelitis in common marmosets using an anti-IL-12p40 monoclonal antibody. J Immunol 2002;169(11):6554–63.
[30] 't Hart BA, et al. Suppression of ongoing disease in a nonhuman primate model of multiple sclerosis by a human-anti-human IL-12p40 antibody. J Immunol 2005;175(7):4761–8.
[31] Segal BM, et al. Repeated subcutaneous injections of IL12/23 p40 neutralising antibody, ustekinumab, in patients with relapsing-remitting multiple sclerosis: a phase II, double-blind, placebo-controlled, randomised, dose-ranging study. Lancet Neurol 2008;7(9):796–804.
[32] Kap YS, et al. Effects of early IL-17A neutralization on disease induction in a primate model of experimental autoimmune encephalomyelitis. J Neuroimmune Pharmacol 2011;6(3):341–53.
[33] Havrdova E, et al. Activity of secukinumab, an anti-IL-17A antibody, on brain lesions in RRMS: results from a randomized, proof-of-concept study. J Neurol 2016;263(7):1287–95.
[34] Hauser SL. The Charcot Lecture | beating MS: a story of B cells, with twists and turns. Mult Scler 2015;21(1):8–21.
[35] Hauser SL, et al. B-cell depletion with rituximab in relapsing-remitting multiple sclerosis. N Engl J Med 2008;358(7):676–88.
[36] Cancro MP. The BLyS family of ligands and receptors: an archetype for niche-specific homeostatic regulation. Immunol Rev 2004;202:237–49.
[37] Cogollo E, et al. Profile of atacicept and its potential in the treatment of systemic lupus erythematosus. Drug Des Dev Ther 2015;9:1331–9.
[38] Kappos L, et al. Atacicept in multiple sclerosis (ATAMS): a randomised, placebo-controlled, double-blind, phase 2 trial. Lancet Neurol 2014;13 (4):353–63.
[39] 't Hart BA, et al. The primate EAE model points at EBV-Infected B cells as a preferential therapy target in multiple sclerosis. Front Immunol 2013;4:145.
[40] Babbe H, et al. Clonal expansions of CD8(+) T cells dominate the T cell infiltrate in active multiple sclerosis lesions as shown by micromanipulation and single cell polymerase chain reaction. J Exp Med 2000;192(3):393–404.
[41] Lassmann H, Bradl M. Multiple sclerosis: experimental models and reality. Acta Neuropathol 2017;133(2):223–44.
[42] Schafer S, Kolkhof P. Failure is an option: learning from unsuccessful proof-of-concept trials. Drug Discov Today 2008;13(21–22):913–6.
[43] 't Hart BA, et al. Improvement of preclinical animal models for autoimmune-mediated disorders via reverse translation of failed therapies. Drug Discov Today 2014;19(9):1394–401.
[44] 't Hart BA. Primate autoimmune disease models; lost for translation? Clin Translat Immunol 2016;5:e122.
[45] 't Hart BA, et al. Multiple sclerosis — a response-to-damage model. Trends Mol Med 2009;15(6):235–44.