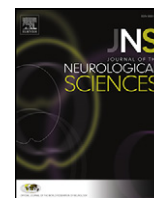


Contents lists available at [ScienceDirect](http://ScienceDirect.com)

## Journal of the Neurological Sciences

journal homepage: [www.elsevier.com/locate/jns](http://www.elsevier.com/locate/jns)

## Alemtuzumab improves neurological functional systems in treatment-naïve relapsing-remitting multiple sclerosis patients

Edward J. Fox<sup>a,\*</sup>, Daniel Wynn<sup>b</sup>, Alasdair J. Coles<sup>c</sup>, Jeffrey Palmer<sup>d</sup>, David H. Margolin<sup>d</sup>,  
on behalf of CAMMS223 Investigators<sup>a</sup> Central Texas Neurology Consultants, MS Clinic of Central Texas, Round Rock, TX, USA<sup>b</sup> Consultants in Neurology MS Center, Northbrook, IL, USA<sup>c</sup> Department of Clinical Neurosciences, University of Cambridge, Cambridge, UK<sup>d</sup> Sanofi Genzyme, Cambridge, MA, USA

## ARTICLE INFO

## Article history:

Received 30 June 2015

Received in revised form 19 January 2016

Accepted 11 February 2016

Available online 12 February 2016

## Keywords:

Alemtuzumab

Disability

Disease-modifying therapy

Expanded Disability Status Scale

Functional systems

Multiple sclerosis

## ABSTRACT

**Background:** Individual functional system scores (FSS) of the Expanded Disability Status Scale (EDSS) play a central role in determining the overall EDSS score in patients with early-stage multiple sclerosis (MS). Alemtuzumab treatment improves preexisting disability for many patients; however, it is unknown whether improvement is specific to certain functional systems.

**Objective:** We assessed the effect of alemtuzumab on individual FSS of the EDSS.

**Methods:** CAMMS223 was a 36-month, rater-blinded, phase 2 trial; treatment-naïve patients with active relapsing-remitting MS, EDSS  $\leq 3$ , and symptom onset within 3 years were randomized to annual courses of alemtuzumab or subcutaneous interferon beta-1a (SC IFNB-1a) 44  $\mu$ g three times weekly.

**Results:** Alemtuzumab-treated patients had improved outcomes versus SC IFNB-1a patients on most FSS at Month 36; the greatest effect occurred for sensory, pyramidal, and cerebellar FSS. Among patients who experienced 6-month sustained accumulation of disability, clinical worsening occurred most frequently in the brainstem and sensory systems. For patients with 6-month sustained reduction in preexisting disability, pyramidal and sensory systems contributed most frequently to clinical improvement.

**Conclusions:** Alemtuzumab demonstrated a broad treatment effect in improving preexisting disability. These findings may influence treatment decisions in patients with early, active relapsing-remitting MS displaying neurological deficits. [ClinicalTrials.gov](http://ClinicalTrials.gov) Identifier NCT00050778.

© 2016 The Authors. Published by Elsevier B.V. This is an open access article under the CC BY-NC-ND license (<http://creativecommons.org/licenses/by-nc-nd/4.0/>).

## 1. Introduction

Alemtuzumab (LEMTRADA™) is a humanized monoclonal antibody that selectively targets CD52 to deplete circulating T and B lymphocytes, thought to be critical mediators of multiple sclerosis (MS) inflammatory processes [1,2]. After lymphocyte depletion, a distinctive pattern of T- and B-cell repopulation begins within weeks, leading to a rebalanced immune system [3,4]. Alemtuzumab is approved in many countries as treatment for adult patients with relapsing-remitting MS (RRMS) who also meet country-specific selection criteria. Alemtuzumab has been evaluated in one 3-year phase 2 trial (CAMMS223, [ClinicalTrials.gov](http://ClinicalTrials.gov) identifier NCT00050778) [5] and two 2-year phase 3 studies (Comparison of Alemtuzumab and Rebif® Efficacy in Multiple Sclerosis I [CARE-MS I, NCT00530348] and CARE-MS II [NCT00548405]) [6,7]. Alemtuzumab significantly reduced the risk of relapse and showed

significant benefit on many radiological outcomes versus subcutaneous interferon beta-1a (SC IFNB-1a, Rebif®; EMD Serono, Inc., Rockland, MA) in active RRMS patients who were treatment-naïve (CAMMS223 and CARE-MS I) [5,6] or who had inadequate efficacy response, defined as at least one relapse, to prior therapy (CARE-MS II) [7]. Safety findings with alemtuzumab were consistent across trials, with the most common adverse events being infusion-associated reactions; other notable adverse events were infections and autoimmune adverse events [5–8].

Disability in patients with MS is commonly assessed using the Expanded Disability Status Scale (EDSS), which is a 10-point scale with 0.5-point steps based on seven functional systems [9]. These functional systems measure different aspects of disability, i.e., impairments in bowel/bladder, brainstem, cerebellar, cerebral, pyramidal, sensory, and visual functioning. In the early stages of MS, individual functional systems of the EDSS play a greater role than ambulatory impairment in determining the overall EDSS score, and patients are more likely to experience deficits in the functional systems of pyramidal, cerebellar, brainstem, and sensory than in the systems of visual, bowel/bladder, and cerebral [10].

\* Corresponding author at: MS Clinic of Central Texas, Central Texas Neurology Consultants, 16040 Park Valley Dr, Bldg B, Suite 100, Round Rock, TX 78681, USA.

E-mail addresses: [foxtexms@gmail.com](mailto:foxtexms@gmail.com) (E.J. Fox), [dwynnmd@gmail.com](mailto:dwynnmd@gmail.com) (D. Wynn), [ajc1020@medschl.cam.ac.uk](mailto:ajc1020@medschl.cam.ac.uk) (A.J. Coles), [david.margolin@genzyme.com](mailto:david.margolin@genzyme.com) (D.H. Margolin).

At enrollment, patients in CAMMS223 had low levels of physical disability (defined as EDSS score  $\leq 3$ ), as is the case with many MS patients in clinical trials [11–14]. In this trial, the rate of sustained accumulation of disability (SAD; defined as an increase from baseline in EDSS score of  $\geq 1.0$  [ $\geq 1.5$  in patients with a baseline EDSS score of 0]) confirmed over 6 months was reduced by 71% with alemtuzumab versus SC IFNB-1a ( $p < 0.001$ ). Disability measured by mean EDSS score over time also decreased significantly with alemtuzumab, but increased with SC IFNB-1a. Furthermore, sustained reduction in preexisting disability (SRD; a decrease from baseline by  $\geq 1$  EDSS point in patients with baseline EDSS scores  $\geq 2.0$ ) confirmed over 6 months was significantly more likely to be observed in alemtuzumab patients [8]. More recently, similar results for these disability endpoints were reported for patients enrolled in CARE-MS II [7].

In the present analysis of CAMMS223, we examined the treatment effect of alemtuzumab compared with SC IFNB-1a on each of the individual EDSS functional systems to determine if alemtuzumab's beneficial treatment effect is broadly based or specific to certain functional systems. Additionally, we examined the relationship between overall EDSS and the functional systems to understand which systems contribute to SAD and to SRD among alemtuzumab-treated patients.

## 2. Methods

Detailed methods for CAMMS223 have previously been published [5]; a brief summary is provided here.

### 2.1. Patients

Treatment-naïve patients with RRMS with EDSS scores of  $\leq 3.0$ , MS symptom onset within 3 years, at least two relapses in the previous 2 years, and evidence of at least one gadolinium-enhancing lesion on any of up to four monthly screening MRI scans were randomized to the study. Due to a misdiagnosis of MS, one patient was not included in the efficacy analyses. Ethics board approvals were obtained from all sites, and all patients provided written informed consent.

### 2.2. Study design

CAMMS223 was a 3-year, rater-blinded, active-controlled, head-to-head, phase 2 trial. Patients were randomly allocated in a 1:1 ratio to receive alemtuzumab 12 mg or SC IFNB-1a (44  $\mu\text{g}$  three times per week). Other patients were randomized to receive alemtuzumab 24 mg. However, since the 24-mg dose level is not approved for use in treatment of MS in any jurisdiction and is no longer being studied, and given the robust effects observed at the 12-mg dose, analyses of the 24-mg group are omitted from this report. Alemtuzumab was administered via intravenous infusions on 5 consecutive days at baseline and on 3 consecutive days 12 months and 24 months later (the latter course was optional and at the discretion of the treating physician if the  $\text{CD4}^+$  T-cell count was  $\geq 100 \times 10^6$  cells/L). In September 2005, alemtuzumab dosing within the trial was suspended after immune thrombocytopenia developed in three patients (including the fatal index case). All safety and efficacy assessments proceeded as planned, and patients in the SC IFNB-1a group continued to receive medication [5]. The alemtuzumab dosing suspension was lifted in April 2008 [15]. As a result of the suspension, 75% of patients were precluded from receiving the optional third course of alemtuzumab at Month 24.

### 2.3. Study outcomes

The results of this study represent Class I evidence due to several characteristics of the study design. Those assessments in the study pertaining to the key efficacy endpoints (e.g., EDSS for disability and relapse) used masked raters. The study was randomized to ensure

that the baseline characteristics were comparable between treatment arms.

### 2.4. Disability assessment

The EDSS score was assessed at baseline and quarterly using the “Neurostatus” training and scoring system [16] by a neurologist blinded to treatment arms. The functional systems assessed in the present analysis were bowel/bladder, brainstem, cerebellar, cerebral, pyramidal, sensory, and visual.

### 2.5. Statistical analysis

Summary statistics of observed values and change from baseline were calculated for each functional system at each time point. For functional system scores (FSS) that included a letter for subclass identification (such as 1X, 2A, and 3B), only the numeric portion was used for the statistical analysis. A significance level of 0.05 was used for these post hoc analyses, and no adjustment was made for multiple hypothesis testing. All FSS were evaluated equally throughout this analysis. All statistical tests were two-sided. Change from baseline for each functional system was categorized as improved (decreased FSS of at least 1.0 point), no change (stable FSS  $\pm 0.5$  point), or worsened (increased FSS of at least 1.0 point). The overall treatment effect across all time points through Month 36 was tested with a repeated measures proportional odds model. The above analysis was also conducted using a subset of patients who had received only two annual courses of alemtuzumab.

Kaplan-Meier and Cox proportional hazards model analyses for each functional system were conducted for time to sustained ( $\geq 6$  months) clinical worsening of functional system and time to sustained clinical improvement of functional system. Clinical worsening was defined as an on-treatment FSS increase of  $\geq 2.0$  points for patients with a baseline score  $\leq 1.0$ , or any increase in score from a baseline FSS of  $\geq 2.0$  points. Clinical improvement was defined as a  $\geq 1.0$ -point decrease in FSS for the subset of patients with baseline FSS  $\geq 1.0$ . Analyses of visual and bowel/bladder functional systems used actual, not converted scores. Spearman correlation coefficients were used to compare FSS changes from baseline at Months 6 and 36. For patients with 6-month SAD in EDSS, the number of patients with worsening in each functional system at the time of SAD was counted. For patients with 6-month SRD in EDSS, the number of patients with improvement in each functional system at the time of SRD was counted.

## 3. Results

### 3.1. Patients

Complete demographic and baseline clinical characteristics and patient disposition have been previously reported [5]. Baseline characteristics were well-balanced between the SC IFNB-1a ( $N = 111$ ) and alemtuzumab 12-mg ( $N = 112$ ) treatment groups. The mean age of the total population was 32.3 years, 64.3% were female, and 90.1% were Caucasian. The mean EDSS score was 1.9, 94.9% of patients had an EDSS score  $\geq 1$ , and the mean time since first relapse was 1.5 years. Of the patients randomized to receive alemtuzumab 12 mg, 6 patients received only one course, 78 received two courses, and 24 received three courses during the core 36-month study.

### 3.2. Functional systems

All FSS were balanced between treatment groups at baseline. Within the alemtuzumab treatment group, the largest mean reductions from baseline to Month 36 occurred in the sensory ( $-0.28$  points), pyramidal ( $-0.17$  points), and cerebellar FSS ( $-0.15$  points). Compared with patients receiving SC IFNB-1a, patients in the alemtuzumab group

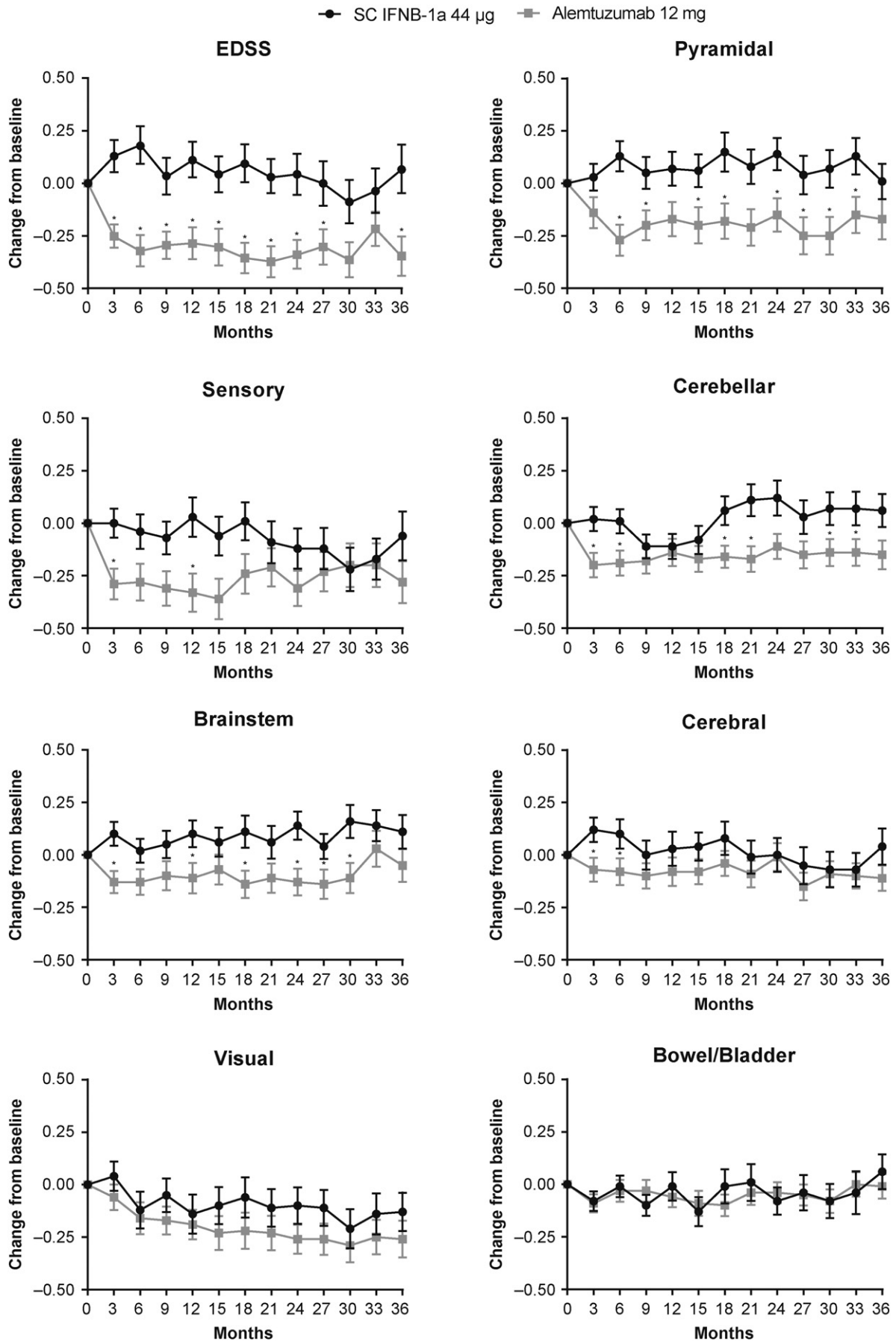
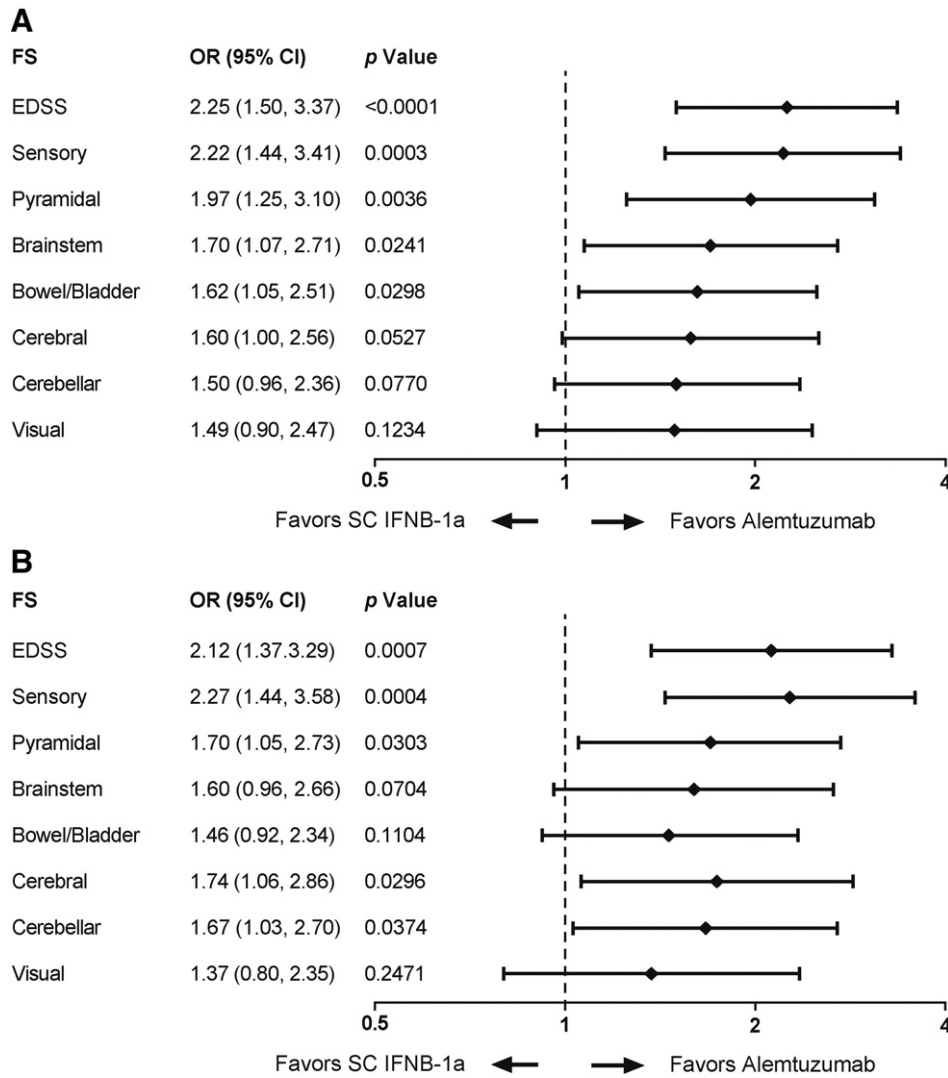


Fig. 1. Mean changes from baseline for the Expanded Disability Status Scale (EDSS) and functional system scores. Abbreviations: IFNB-1a, interferon beta-1a; SC, subcutaneous. \* $p < 0.05$ , alemtuzumab 12 mg vs SC IFNB-1a.



**Fig. 2.** Odds ratios (ORs) for Expanded Disability Status Scale (EDSS) and functional systems (FS) score improvements over 36 months, alemtuzumab 12 mg vs subcutaneous interferon beta-1a (SC IFNB-1a) 44 µg: (A) overall patient cohort and (B) patients receiving only two alemtuzumab 12-mg courses.

had lower mean observed FSS values at Month 36 and larger mean FSS reductions from baseline through Month 36, with the sensory, pyramidal, cerebellar, and brainstem FSS showing the greatest treatment effect (Fig. 1). Changes from baseline versus SC IFNB-1a were statistically significant ( $p < 0.05$ ) in the alemtuzumab treatment group for the brainstem, pyramidal, and cerebellar FSS for at least half of the time points assessed. Repeated measures proportional odds analyses through Month 36 indicated that treatment with alemtuzumab led to a consistently better outcome on each functional system compared with SC IFNB-1a (Fig. 2). For the alemtuzumab group, the effect was statistically significant versus SC IFNB-1a for the sensory, pyramidal, brainstem, and bowel/bladder functional systems. Although numerically favorable for alemtuzumab, results for the cerebral, cerebellar, and visual functional systems did not achieve statistical significance (Fig. 2A). Results were similar to the above when comparing FSS changes for alemtuzumab-treated patients who received only two courses of alemtuzumab with those for SC IFNB-1a patients (Fig. 2B).

Patients treated with alemtuzumab were also less likely to experience sustained clinical worsening of FSS and more likely to experience sustained clinical improvement of FSS in most of the functional systems compared with patients treated with SC IFNB-1a. In patients treated with alemtuzumab, the beneficial treatment effect versus SC IFNB-1a

was significant for sustained clinical worsening in the pyramidal functional system, and for sustained clinical improvement in the pyramidal and sensory functional systems (Table 1).

### 3.3. Relationship of functional system changes to changes in EDSS scores in alemtuzumab-treated patients

The relationship between functional systems and sustained changes in EDSS was examined. Among the 18 alemtuzumab patients who experienced 6-month SAD in EDSS, the associated worsening occurred most often in the brainstem and sensory functional systems (both 38.9% of patients who experienced SAD) and least often in the visual functional system (5.6%) (data not shown). For the 66 alemtuzumab patients with 6-month SRD in EDSS, the pyramidal functional system contributed most often (53.0% of SRD events) to that clinical improvement, whereas the bowel/bladder functional system contributed least often (19.7%) (data not shown).

## 4. Discussion

The involvement of a greater number of functional systems early in MS disease predicts the accumulation of future disability [17,18], so it is important to understand the number and identity of the

functional systems impacted by disease-modifying therapy to inform treatment choices. Although few data exist to describe which functional systems are most impacted by disease-modifying therapies, evidence from a previous study demonstrated that therapies with benefits on disability outcomes do not necessarily impact all affected functional systems equally. In that analysis, intramuscular IFNB-1a was found to have a statistically significant positive effect compared with placebo on scores in the pyramidal system only. This was despite the observation that the study population had pyramidal (37%), cerebellar (38%), and sensory (34%) involvement at baseline [19].

As previously reported, alemtuzumab treatment led to a significant and sustained improvement in preexisting MS-related disability compared with SC IFNB-1a [5,8]. Alemtuzumab's greater suppression of MS relapse and new brain lesion formation does not fully account for the differential effect on disability as measured by EDSS [20]. The present report extends these findings with the observation that alemtuzumab's positive effects on disability, as assessed by the overall EDSS score, were similarly observed in each of the individual functional systems. Patients receiving alemtuzumab had a consistently better outcome for all functional systems compared with patients receiving SC IFNB-1a, with the strongest treatment effects

being observed for the sensory, pyramidal, and cerebellar functional systems.

These treatment effects were durable, being sustained through Month 36 even though most patients had last received alemtuzumab at Month 12. Indeed, results from the subgroup of patients who received only two courses of alemtuzumab, and received no therapy for the final 24 months of follow-up, were comparable to those in the overall alemtuzumab group. Furthermore, similar findings were reported in an analysis of EDSS FSS in CARE-MS II, in which all alemtuzumab patients were randomized to receive 2 courses in the core study [21].

Changes in any FSS can impact the overall EDSS score, by definition, but some functional systems are more often involved in MS relapses and consequently are more likely to change. Furthermore, all FSS changes may not be equally likely to persist and to lead to sustained clinical worsening of FS or sustained clinical improvement of FS. In our study, onset of SAD was most often linked with worsening in the sensory and brainstem functional systems and least frequently with visual functional system change. Among patients who experienced SRD, the pyramidal functional system contributed most frequently to that clinical improvement, whereas the bowel/bladder functional system contributed the least. This apparent driving of EDSS changes by more

**Table 1**

Kaplan-Meier analyses for 6-month sustained clinical worsening and 6-month sustained clinical improvement for functional systems over 36 months (full analysis set).

Functional system	SC IFNB-1a 44 µg (N = 111)	Alemtuzumab 12 mg (N = 112)	SC IFNB-1a 44 µg (N = 111)	Alemtuzumab 12 mg (N = 112)
	6-Month sustained clinical worsening		6-Month sustained clinical improvement	
<b>Visual</b>				
Patients with outcome – n	3	2	24	34
KM estimate of event (95% CI)	0.03 (0.010, 0.089)	0.02 (0.005, 0.078)	0.26 (0.184, 0.370)	0.33 (0.248, 0.433)
Hazard ratio (95% CI)		0.67 (0.110, 4.072)		1.35 (0.796, 2.274)
Treatment effect – %		33.0		34.5
p value		0.6640		0.2686
<b>Brainstem</b>				
Patients with outcome – n	3	4	13	23
KM estimate of event (95% CI)	0.04 (0.012, 0.107)	0.05 (0.017, 0.119)	0.14 (0.081, 0.224)	0.22 (0.151, 0.310)
Hazard ratio (95% CI)		1.21 (0.270, 5.434)		1.84 (0.930, 3.639)
Treatment effect – %		21.1		84.0
p value		0.8022		0.0799
<b>Pyramidal</b>				
Patients with outcome – n	8	1	12	34
KM estimate of event (95% CI)	0.09 (0.047, 0.176)	0.01 (0.001, 0.068)	0.13 (0.077, 0.225)	0.33 (0.246, 0.428)
Hazard ratio (95% CI)		0.12 (0.015, 0.960)		2.88 (1.485, 5.583)
Treatment effect – %		88.0		187.9
p value		0.0457		0.0018
<b>Cerebellar</b>				
Patients with outcome – n	5	4	17	27
KM estimate of event (95% CI)	0.06 (0.027, 0.148)	0.04 (0.015, 0.101)	0.18 (0.116, 0.276)	0.26 (0.182, 0.350)
Hazard ratio (95% CI)		0.68 (0.182, 2.543)		1.68 (0.913, 3.085)
Treatment effect – %		31.9		67.8
p value		0.5679		0.0954
<b>Sensory</b>				
Patients with outcome – n	5	2	24	38
KM estimate of event (95% CI)	0.06 (0.025, 0.139)	0.02 (0.005, 0.081)	0.26 (0.181, 0.366)	0.36 (0.277, 0.460)
Hazard ratio (95% CI)		0.26 (0.048, 1.346)		1.70 (1.016, 2.839)
Treatment effect – %		74.4		69.8
p value		0.1076		0.0433
<b>Bowel/bladder</b>				
Patients with outcome – n	3	1	19	19
KM estimate of event (95% CI)	0.03 (0.011, 0.107)	0.01 (0.001, 0.064)	0.21 (0.137, 0.308)	0.18 (0.120, 0.271)
Hazard ratio (95% CI)		0.34 (0.035, 3.268)		0.88 (0.466, 1.668)
Treatment effect – %		66.1		11.9
p value		0.3496		0.6980
<b>Cerebral</b>				
Patients with outcome – n	3	1	15	22
KM estimate of event (95% CI)	0.03 (0.010, 0.092)	0.01 (0.001, 0.069)	0.16 (0.100, 0.255)	0.21 (0.145, 0.304)
Hazard ratio (95% CI)		0.30 (0.031, 2.902)		1.44 (0.748, 2.789)
Treatment effect – %		69.9		44.4
p value		0.2988		0.2734

Abbreviations: CI, confidence interval; IFNB-1a, interferon beta-1a; SC, subcutaneous.

objective functional system endpoints, such as the pyramidal and brainstem FSS, minimizes the likelihood of the EDSS findings being influenced by potential bias resulting from patient blinding not being possible in this study. The relationship between functional systems and EDSS was similar to that reported by Scott et al., who identified the pyramidal and cerebellar functional systems as the systems most responsive to MS disease-modifying therapy at multiple levels of the EDSS [19].

The observation that improvements with alemtuzumab over SC IFNB-1a in our study were seen across all of the functional systems indicates potential benefits in multiple aspects of patients' quality of life (QoL). Indeed, our results are consistent with QoL analyses of phase 3 trial data showing improvement in a variety of QoL measures with alemtuzumab over time, as well as in comparison with SC IFNB-1a [22]. Furthermore, the particularly strong effects of alemtuzumab on the pyramidal functional systems in our present study are in agreement with the observation that the most profound QoL benefits with alemtuzumab were seen in mobility and other physical components of the QoL scales assessed [22].

Limitations of this study include the modest degree of change in FSS that is observed in the early stages of MS (all patients had a baseline EDSS  $\leq 3$  in this study). At a higher baseline EDSS, the contribution of individual FSS to the overall EDSS score may have been different. Additionally, differential dropout of patients treated with SC IFNB-1a may have biased results at later time points in the study. Per protocol, patients who experienced an SAD event in the study discontinued from the trial. As more patients treated with SC IFNB-1a had SAD early in the course of the trial, the discontinuation rate in this treatment arm was higher than in the alemtuzumab treatment arm. However, this limitation would tend to bias the results against alemtuzumab, since discontinuation of SC IFNB-1a patients with SAD removed their worsening values from the SC IFNB-1a group's mean at subsequent time points. Thus, our findings may underestimate alemtuzumab's relative treatment effect, but that would not alter the conclusions of this study.

In conclusion, alemtuzumab's greater efficacy compared with SC IFNB-1a, an active comparator, was observed consistently across all functional systems, indicating a broad treatment effect. Whether neurological impairments residual from prior MS attacks involved motor control or sensation or sphincter control, they were more likely to improve with alemtuzumab treatment than with SC IFNB-1a. Since alemtuzumab's effect was most evident in the functional systems most often affected and most strongly driving SAD in early RRMS, these findings are likely to have clinical relevance.

### Conflict of interest

Dr Fox reports receiving consultancy fees, honoraria, travel, and research support from Acorda, Bayer, Biogen, Chugai, Eli Lilly, EMD Serono, Novartis, Ono, Opexa Therapeutics, Roche, Sanofi Genzyme, and Teva. Dr Wynn reports receiving research support and/or consulting fees from Acorda Therapeutics, Adamas, Avanir Pharmaceuticals, Chugai, EMD Serono GlaxoSmithKline, Hoffman LaRoche/Genentech, Novartis, Ono Pharmaceutical, Opexa Therapeutics, Osmotica, Pfizer, Questcor, Receptos/Celgene, Sanofi-Aventis/Genzyme, SanBio, Sunovion, Teva, TG Therapeutics and XenPort. Dr Coles reports receiving consulting fees, lecture fees, and institutional grant support from Sanofi Genzyme. Mr Palmer was employed at Sanofi Genzyme during the development of the manuscript. Dr Margolin is an employee of Sanofi Genzyme.

### Funding/support

Funding was provided by Sanofi Genzyme and Bayer Healthcare Pharmaceuticals.

### Acknowledgements

The authors would like to thank Marco Rizzo and Isabel Firmino for reviewing and providing input on the manuscript; Isabel Firmino is an employee of Sanofi Genzyme; Marco Rizzo was an employee of Sanofi Genzyme at the time the work was conducted. Data analysis was carried out by Linda Kasten, PROMETRIKA, LLC, Cambridge, MA, USA, which was supported by Sanofi Genzyme. Editorial support for this manuscript was provided by Fiona Nitsche, PhD, and Susan M Kaup, PhD, which was funded by Sanofi Genzyme. Fiona Nitsche is an employee of Evidence Scientific Solutions; Susan M Kaup was an employee of Evidence Scientific Solutions at the time the work was conducted.

### References

- [1] Y. Hu, M.J. Turner, J. Shields, M.S. Gale, E. Hutto, B.L. Roberts, et al., Investigation of the mechanism of action of alemtuzumab in a human CD52 transgenic mouse model, *Immunology* 128 (2009) 260–270, <http://dx.doi.org/10.1111/j.1365-2567.2009.03115.x>.
- [2] M.S. Weber, B. Hemmer, Cooperation of B cells and T cells in the pathogenesis of multiple sclerosis, *Results Probl. Cell Differ.* 51 (2010) 115–126, [http://dx.doi.org/10.1007/400\\_2009\\_21](http://dx.doi.org/10.1007/400_2009_21).
- [3] A.L. Cox, S.A. Thompson, J.L. Jones, V.H. Robertson, G. Hale, H. Waldmann, et al., Lymphocyte homeostasis following therapeutic lymphocyte depletion in multiple sclerosis, *Eur. J. Immunol.* 35 (2005) 3332–3342, <http://dx.doi.org/10.1002/eji.200535075>.
- [4] E. Havari, M.J. Turner, J. Campos-Rivera, S. Shankara, T.H. Nguyen, B. Roberts, et al., Impact of alemtuzumab treatment on the survival and function of human regulatory T cells in vitro, *Immunology* 141 (2014) 123–131, <http://dx.doi.org/10.1111/imm.12178>.
- [5] CAMMS223 Trial Investigators, A.J. Coles, D.A. Compston, K.W. Selmaj, S.L. Lake, S. Moran, et al., Alemtuzumab vs. interferon beta-1a in early multiple sclerosis, *N. Engl. J. Med.* 359 (2008) 1786–1801, <http://dx.doi.org/10.1056/NEJMoa0802670>.
- [6] J.A. Cohen, A.J. Coles, D.L. Arnold, C. Confavreux, E.J. Fox, H.P. Hartung, et al., Alemtuzumab versus interferon beta 1a as first-line treatment for patients with relapsing-remitting multiple sclerosis: a randomised controlled phase 3 trial, *Lancet* 380 (2012) 1819–1828, [http://dx.doi.org/10.1016/S0140-6736\(12\)61769-3](http://dx.doi.org/10.1016/S0140-6736(12)61769-3).
- [7] A.J. Coles, C.L. Twyman, D.L. Arnold, J.A. Cohen, C. Confavreux, E.J. Fox, et al., Alemtuzumab for patients with relapsing multiple sclerosis after disease-modifying therapy: a randomised controlled phase 3 trial, *Lancet* 380 (2012) 1829–1839, [http://dx.doi.org/10.1016/S0140-6736\(12\)61768-1](http://dx.doi.org/10.1016/S0140-6736(12)61768-1).
- [8] A.J. Coles, E. Fox, A. Vladic, S.K. Gazda, V. Brinar, K.W. Selmaj, et al., Alemtuzumab versus interferon beta-1a in early relapsing-remitting multiple sclerosis: post-hoc and subset analyses of clinical efficacy outcomes, *Lancet Neurol.* 10 (2011) 338–348, [http://dx.doi.org/10.1016/S1474-4422\(11\)70020-5](http://dx.doi.org/10.1016/S1474-4422(11)70020-5).
- [9] J.F. Kurtzke, Rating neurologic impairment in multiple sclerosis: an expanded disability status scale (EDSS), *Neurology* 33 (1983) 1444–1452.
- [10] J.F. Kurtzke, Natural history and clinical outcome measures for multiple sclerosis studies. Why at the present time does EDSS scale remain a preferred outcome measure to evaluate disease evolution? *Neurol. Sci.* 21 (2000) 339–341.
- [11] R.J. Fox, D.H. Miller, J.T. Phillips, M. Hutchinson, E. Havrdova, M. Kita, et al., Placebo-controlled phase 3 study of oral BG-12 or glatiramer in multiple sclerosis, *N. Engl. J. Med.* 367 (2012) 1087–1097, <http://dx.doi.org/10.1056/NEJMoa1206328>.
- [12] G. Giovannoni, G. Comi, S. Cook, K. Rammohan, P. Rieckmann, P. Soelberg Sørensen, et al., A placebo-controlled trial of oral cladribine for relapsing multiple sclerosis, *N. Engl. J. Med.* 362 (2010) 416–426, <http://dx.doi.org/10.1056/NEJMoa0902533>.
- [13] L. Kappos, E.W. Radue, P. O'Connor, C. Polman, R. Hohlfeld, P. Calabresi, et al., A placebo-controlled trial of oral fingolimod in relapsing multiple sclerosis, *N. Engl. J. Med.* 362 (2010) 387–401, <http://dx.doi.org/10.1056/NEJMoa0909494>.
- [14] C.H. Polman, P.W. O'Connor, E. Havrdova, M. Hutchinson, L. Kappos, D.H. Miller, et al., A randomized, placebo-controlled trial of natalizumab for relapsing multiple sclerosis, *N. Engl. J. Med.* 354 (2006) 899–910, <http://dx.doi.org/10.1056/NEJMoa044397>.
- [15] A.J. Coles, E. Fox, A. Vladic, S.K. Gazda, V. Brinar, K.W. Selmaj, et al., Alemtuzumab more effective than interferon beta-1a at 5-year follow-up of CAMMS223 clinical trial, *Neurology* 78 (2012) 1069–1078, <http://dx.doi.org/10.1212/WNL.0b013e31824e8ee7>.
- [16] L. Kappos, J. Lechner-Scott, C. Lienert, Interactive Training DVD-ROM for a Standardised, Quantified Neurological Examination and Assessment of Kurtzke's Functional Systems and Expanded Disability Status Scale in Multiple Sclerosis. Version 01/07 Ed, Point de Vue Audiovisuelle Produktionen, Basel, Switzerland, 2007.
- [17] M.P. Amato, G. Ponziani, M.L. Bartolozzi, G. Siracusa, A prospective study on the natural history of multiple sclerosis: clues to the conduct and interpretation of clinical trials, *J. Neurol. Sci.* 168 (1999) 96–106.
- [18] C. Hirst, G. Ingram, R. Swingler, D.A. Compston, T. Pickersgill, N.P. Robertson, Change in disability in patients with multiple sclerosis: a 20-year prospective population-based analysis, *J. Neurol. Neurosurg. Psychiatry* 79 (2008) 1137–1143, <http://dx.doi.org/10.1136/jnnp.2007.133785>.

- [19] T.F. Scott, X. You, P. Foulds, Functional system scores provide a window into disease activity occurring during a multiple sclerosis treatment trial, *Neurol. Res.* 33 (2011) 549–552, <http://dx.doi.org/10.1179/1743132810Y.0000000017>.
- [20] J.L. Jones, J.M. Anderson, C.L. Phuah, E.J. Fox, K. Selmaj, D. Margolin, et al., Improvement in disability after alemtuzumab treatment of multiple sclerosis is associated with neuroprotective autoimmunity, *Brain* 133 (2010) 2232–2247, <http://dx.doi.org/10.1093/brain/awq176>.
- [21] V. Brinar, D.L. Arnold, A.J. Coles, H.P. Hartung, E. Havrdova, K.W. Selmaj, et al., Alemtuzumab improves EDSS via effects on functional systems – CARE-MS II, *Mult. Scler.* 19 (2013) P649.
- [22] T. Moreau, E. Havrdova, G. Giovannoni, D.H. Margolin, L. Kasten, B. Singer, Alemtuzumab improves quality of life in relapsing-remitting multiple sclerosis patients who relapsed on prior therapy: Three-year follow-up of CARE-MS II, *Mult. Scler.* 20 (2014) P044.