

NEUROLOGY

Results of the Avonex Combination Trial (ACT) in relapsing-remitting MS
J. A. Cohen, P. B. Imrey, P. A. Calabresi, K. R. Edwards, T. Eickenhorst, W. L. Felton,
III, E. Fisher, R. J. Fox, A. D. Goodman, C. Hara-Cleaver, G. J. Hutton, B. F. Mandell,
T. F. Scott, H. Zhang, C. Apperson-Hansen, G. J. Beck, P. L. Houghtaling, M. T.
Karafa, M. Stadtler and For the ACT Investigators
Neurology 2009;72;535-541
DOI: 10.1212/01.wnl.0000341934.12142.74

This information is current as of July 14, 2009

The online version of this article, along with updated information and services, is
located on the World Wide Web at:

<http://www.neurology.org/cgi/content/full/72/6/535>

Neurology® is the official journal of the American Academy of Neurology. Published continuously since 1951, it is now a weekly with 48 issues per year. Copyright © 2009 by AAN Enterprises, Inc. All rights reserved. Print ISSN: 0028-3878. Online ISSN: 1526-632X.



Results of the Avonex Combination Trial (ACT) in relapsing-remitting MS

J.A. Cohen, MD
P.B. Imrey, PhD
P.A. Calabresi, MD
K.R. Edwards, MD
T. Eickenhorst, MD, PhD
W.L. Felton III, MD
E. Fisher, PhD
R.J. Fox, MD
A.D. Goodman, MD
C. Hara-Cleaver, RN,
MSN
G.J. Hutton, MD
B.F. Mandell, MD, PhD
T.F. Scott, MD
H. Zhang, PhD
C. Apperson-Hansen,
MStat
G.J. Beck, PhD
P.L. Houghtaling, MS
M.T. Karafa, PhD
M. Stadler
For the ACT
Investigators*

Address correspondence and
reprint requests to Dr. Jeffrey A.
Cohen, Mellen Center U-10,
Cleveland Clinic Foundation,
9500 Euclid Avenue, Cleveland,
OH 44195
cohenj@ccf.org

Supplemental data at
www.neurology.org

ABSTRACT

Objective: To assess the safety, tolerability, and efficacy of interferon beta-1a (IFN β -1a) combined with methotrexate (MTX), IV methylprednisolone (IVMP), or both in patients with relapsing-remitting multiple sclerosis (RRMS) with continued disease activity on IFN β -1a monotherapy.

Methods: Eligibility criteria included RRMS, Expanded Disability Status Scale score 0–5.5, and ≥ 1 relapse or gadolinium-enhancing MRI lesion in the prior year on IFN β -1a monotherapy. Participants continued weekly IFN β -1a 30 μ g IM and were randomized in a 2 \times 2 factorial design to adjunctive weekly placebo or MTX 20 mg PO, with or without bimonthly IVMP 1,000 mg/day for 3 days. The primary endpoint was new or enlarged T2 lesion number at month 12 vs baseline. The study was industry-supported, collaboratively designed, and governed by an investigator Steering Committee with independent Advisory and Data Safety Monitoring committees. Study operations, MRI analyses, and aggregated data were managed by an academic coordinating center.

Results: The 313 participants had clinical and MRI characteristics typical of RRMS. Combinations of IFN β -1a with MTX or IVMP were generally safe and well tolerated. Although trends suggesting modest benefit were seen for some outcomes for IVMP, the results did not demonstrate significant benefit for either adjunctive therapy. The data suggested IVMP reduced anti-IFN β neutralizing antibody titers.

Conclusions: This trial did not demonstrate benefit of adding low-dose oral methotrexate or every other month IV methylprednisolone to interferon beta-1a in relapsing-remitting multiple sclerosis.

Neurology[®] 2009;72:535–541

GLOSSARY

ACT = Avonex Combination Trial; **BPF** = brain parenchymal fraction; **DEXA** = dual energy X-ray absorptiometry; **EDSS** = Expanded Disability Status Scale; **GdE** = gadolinium-enhancing; **IFN β -1a** = interferon beta-1a; **IVMP** = IV methylprednisolone; **MSFC** = MS Functional Composite; **MTX** = methotrexate; **N/E** = new or enlarged; **NAb** = neutralizing antibody; **OR** = odds ratio; **RRMS** = relapsing-remitting multiple sclerosis; **SENTINEL** = Safety and Efficacy of Natalizumab in Combination with Interferon β -1a in Patients with Relapsing-Remitting MS.

Several lines of evidence support the rationale for combination therapy for patients with relapsing-remitting multiple sclerosis (RRMS) with continued disease activity despite disease-modifying monotherapy. MS pathogenesis has numerous potential mechanisms, and may be heterogeneous across patients and within patients over time. Partially effective agents could work additively or synergistically. Agents in combination might allow lower dosing and side effects. Combination therapies are useful for other immune-mediated diseases, hypertension, infections, and cancers.

The Avonex Combination Trial (ACT) assessed the safety, tolerability, and efficacy of interferon beta-1a (IFN β -1a) combined with low-dose oral methotrexate (MTX), IV methylpred-

*The ACT study sites and committee members are listed in appendix e-1 on the *Neurology*[®] Web site at www.neurology.org.

From the Cleveland Clinic (J.A.C., P.B.I., E.F., R.J.F., C.H.-C., B.F.M., G.J.B., P.L.H., M.T.K., M.S.), OH; Johns Hopkins University (P.A.C.), Baltimore, MD; MS Center of Southern Vermont (K.R.E.), Bennington; Biogen Idec (T.E., H.Z.), Cambridge, MA; Virginia Commonwealth University Medical Center (W.L.F.), Richmond; University of Rochester (A.D.G.), NY; Baylor College of Medicine (G.J.H.), Houston, TX; Drexel College of Medicine (T.F.S.), Pittsburgh, PA; and Case Western Reserve University (C.A.-H.), Cleveland, OH.

Biogen Idec, Inc. funded this study. Pfizer, Inc. provided methylprednisolone.

Preliminary results presented at the annual meeting of the American Academy of Neurology, Boston, MA, May 2007.

Disclosure: Author disclosures are provided at the end of the article.

nisolone (IVMP), or both in participants with active RRMS on IFN β -1a alone. Although clinical experience and preliminary studies supported the safety and benefit of these combinations, additional data were needed to justify widespread use.

METHODS ClinicalTrials.gov identifier: NCT00112034.

Study design. ACT was a randomized, multicenter, investigator-run 2 \times 2 factorial clinical trial. The rationale and design were reported previously.¹ Seventy-two centers participated (appendix e-1 on the *Neurology*[®] Web site at www.neurology.org), including 28 university clinics, 7 multispecialty groups, and 37 general neurology practices. A treating neurologist and treating nurse supervised medical management. An examining neurologist performed a standardized neurologic examination, calculated Functional System and Expanded Disability Status Scale (EDSS)^{2,3} scores at scheduled and unscheduled visits, and confirmed protocol-defined relapses. An examining technician performed the MS Functional Composite (MSFC)⁴ at scheduled and unscheduled visits. Both were trained and certified at prestudy investigators' meetings.

Participants. Protocol and consent documents were approved by local institutional review boards. Participants provided written informed consent before any study-related procedures, and were re-consented after major protocol revisions. Key inclusion criteria included age 18–55, MS diagnosis,⁵ RR course,⁶ EDSS 0.0–5.5,^{2,3} IFN β -1a treatment for ≥ 6 months, active disease in prior 12 months (≥ 1 relapse or ≥ 1 gadolinium-enhancing [GdE] cranial/spinal MRI lesion ≥ 6 months after initiating IFN β -1a), MRI demonstrating T2-hyperintense lesions (T2 lesions) consistent with MS, and ability to perform the MSFC.⁴ Key exclusion criteria included medical conditions or laboratory abnormalities contraindicating study medications, relapse within 60 days, inability to undergo MRI with Gd, MS treatments (specified washouts), and prior anti-IFN β neutralizing antibody (NAb) titer $\geq 1:5$.

Treatment. Eligible participants continued weekly IFN β -1a 30 μ g IM and were randomized with equal probabilities to the following groups: group 1 weekly oral placebo, group 2 weekly oral MTX 20 mg, group 3 weekly oral placebo and bimonthly IVMP 1,000 mg/day 3 consecutive days, and group 4 weekly MTX and bimonthly IVMP. All participants received daily oral folic acid 1 mg. MTX/placebo was uptitrated from 10 to 20 mg over 2 months. The dose could be divided, reduced, or temporarily discontinued according to a prespecified algorithm for laboratory abnormalities or side effects. Participants, study personnel, and steering committee including study statistician were blinded to MTX/placebo assignment. Clinical and MRI evaluators and study statistician were blinded to IVMP assignment.

Outcome measures. The primary endpoint was new or enlarged (N/E) T2 lesion number at month 12 vs baseline. Secondary outcomes were GdE lesion number, relapse rate, MSFC change, and brain parenchymal fraction (BPF) percent change. Analyses of T2- and T1-hypointense (T1) lesion volumes, time to first relapse, relapse-free proportion, and EDSS change were preplanned. Participants were classified on a composite outcome defined as any combination of EDSS worsening (increase by ≥ 1.5 from 0 or ≥ 1.0 from ≥ 1.0), relapse within 365 days, or GdE or N/E T2 lesion on follow-up MRI.

Screening, baseline, and follow-up assessments. Follow-up was 12 months postrandomization. Screening visit included vital signs, physical examination, EDSS, three MSFC practice sessions, blood chemistry, complete blood count, hepatitis screen, HIV antibody screen, serum pregnancy test, urinalysis, chest X-ray, and brain MRI. Baseline visit, within 30 days, included vital signs, EDSS, MSFC, blood chemistry, complete blood count, urinalysis, and anti-IFN β NAb. Participants were randomized by interactive voice response system. Participants randomized to IVMP underwent dual energy X-ray absorptiometry (DEXA) scanning of nondominant or bilateral total hip and femoral neck and anteroposterior lumbar spine before treatment.

Safety and laboratory evaluations were scheduled at months 1, 2, 3, 4, 6, 8, 10, 12, and 1 month after completion of treatment, or at early termination. EDSS, MSFC, and anti-IFN β NAb were repeated at months 6 and 12 or at early termination. Cranial MRI and DEXA scan were repeated at month 12 (2 months after the last scheduled IVMP course) or early termination.

On-study relapse was defined as new/recurrent neurologic symptoms developing ≥ 30 days after onset of a previous confirmed relapse; evolution < 3 months; duration ≥ 24 hours; with corresponding change on examination, and without fever or intercurrent illness. An unscheduled visit was conducted within 72 hours of site notification. Relapses confirmed by the examining neurologist were treated, if needed, with IVMP 1,000 mg/day for 3–5 days without oral taper.

MRI acquisition and analyses. Magnetic resonance images were acquired at 1.5 T per standardized protocol,¹ and shipped electronically to the MRI Analysis Center. Before enrolling participants, sites submitted an MS patient test scan for approval of scanning and data transfer techniques. Scans underwent sub-voxel registration to the baseline scan, and N/E T2 lesion number, T2 and T1 lesion volumes, GdE lesion number and volume, and BPF were analyzed using automated software.^{1,7,8} Enlarged T2 lesions had volume increase $\geq 20\%$.

Anti-IFN β NAb. Serum anti-IFN β NAb were measured at Biogen Idec using a two-step assay.⁹ Samples with titer ≥ 20 were classified as positive.

Study governance and management. The steering committee of investigators plus representatives of Cleveland Clinic MS Academic Coordinating Center and sponsor (appendix e-1) held monthly teleconferences for protocol development, study oversight, analysis plan approval, and results interpretation. An independent data safety monitoring committee met semiannually to review trial conduct and unblinded interim safety data. The data safety monitoring committee provided input concerning study design, analysis, and interpretation. An advisory committee provided additional clinical, imaging, and statistical input. MS academic coordinating center responsibilities included site management, administrative support of study committees, MRI analysis center, medication and visit tracking, methylprednisolone distribution, medication and adverse event coding, data management, and statistical analyses.¹

Statistical considerations. ACT used permuted-blocks randomization with blocks of four and eight, start based on anticipated site enrollment, and restrictions limiting intrasite confounding. Power against the primary endpoint was estimated using month 12–24 data from 260 participants (32.4% of 802) at both doses in the European Study Group on Interferon-beta-1a in MS Dose Comparison Study (IFN β -1a dose-comparison study),¹⁰ whose baseline-month 18 disease activity mimicked ACT entry criteria. Power for either treatment assum-

ing inefficacy of the other was estimated at 89% against a beneficial cumulative odds ratio (OR) = 0.45 in a proportional odds model, and 81% against OR = 0.50. Power estimates declined to 81% and 72% with the other therapy assumed highly effective (OR = 0.20). Re-estimates based on ACT data suggest these values exceeded achieved power by an absolute 10%.

MRI lesions were analyzed by proportional odds models¹¹ for counts and rank-based nonparametric analysis of variance or covariance^{12,13} for volume changes. Relapse rates were analyzed by negative binomial regression,¹⁴ time to first relapse by Cox proportional hazards regression, relapse-free proportions by logistic regression, and individual annualized relapse rates by proportional odds model for five categories. A proportional odds model was fit to trichotomized EDSS change: worsening (increase by ≥ 1.5 from 0 or ≥ 1.0 from ≥ 1.0), improved (decrease ≥ 1.0), or stable. MSFC change was evaluated by factorial analysis of variance, and relative BPF change by Gaussian generalized linear model with log link.¹⁵ A composite freedom-from-MS activity dichotomy was analyzed by logistic regression. Categorical data and rank analyses were used for skewed outcomes to avoid heavy influences by participants exhibiting unusually large values on analyses of means or totals. Most variables were screened for MTX \times IVMP interactions using the interaction test for a factorial model of indicated type, at prespecified 1% level. Lesion volume variables were screened for interaction using stratified exact tests on residuals from main effects models. Absent significant interaction, treatment effects were tested at prespecified 5% level in a main effects model. Analyses were adjusted for prespecified baseline covariates selected blind to ACT outcome data based on analyses of the IFN β -1a dose-comparison study¹⁰ and the placebo group of Safety and Efficacy of Natalizumab in Combination with Interferon β -1a in Patients with Relapsing-Remitting MS (SENTINEL).¹⁶

Reported analyses followed strict intention-to-treat. N/E T2 lesion and relapse rate analyses used multiple imputation by predictive mean matching for participants not undergoing month 12 MRI scans, and to project data forward for early dropouts. Imputations used treatment-group-specific models, including prespecified covariates, time-dependent variables identifying cessation of MTX or IVMP, and, for relapses, on-study relapses prior to withdrawal and 6-month MSFC if available. For other outcomes except MSFC, missing data were imputed by last observation carried forward or, when no follow-up observation was obtained, averaging the four treatment group means of study completers. MSFC was calculated by averaging Timed 25-Foot Walk, Nine-Hole Peg Test, and 3-second Paced Auditory Serial Addition Test Z-scores, using pooled baseline data as reference population.¹⁷ Missing MSFC values were imputed using standard conventions.¹⁸

Adverse events were coded using MedDRA (version 6.1) preferred terms and systems, and analyzed using 2 \times 2 factorial and main effects logistic regression models, with exact tests and mid- p values for events with cell expected values < 4 . Generalized additive modeling¹⁹ was used to study vital sign trends. Treatment effects on anti-IFN β NAb titer were tested by exact stratified Wilcoxon test.²⁰ Appendix e-2 provides additional statistical details.

RESULTS Enrollment and follow-up. ACT initiated enrollment in June 2003 with a 900-participant target. Because of slow enrollment, the protocol was revised in January 2005, reducing target enrollment to 300–350, changing the primary endpoints from relapse rate for MTX and BPF change for IVMP to N/E T2 lesions for both, and shortening follow-up

from 24 to 12 months. The protocol revision process was detailed previously.¹ Enrollment closed in May 2005 with 313 participants. The figure summarizes follow-up. Discontinuation numbers, timing, and reasons were comparable across treatments (table e-1).

Baseline characteristics. Baseline demographic, clinical, and MRI characteristics were typical of RRMS and generally comparable across groups (table e-2). Participants had relatively mild neurologic impairment. The majority (86.5%) qualified for enrollment by clinical relapse in the preceding year. Most participants qualifying by MRI only had brain activity. Despite recently active disease and significant MRI lesion burden, only 22.4% had baseline GdE lesions.

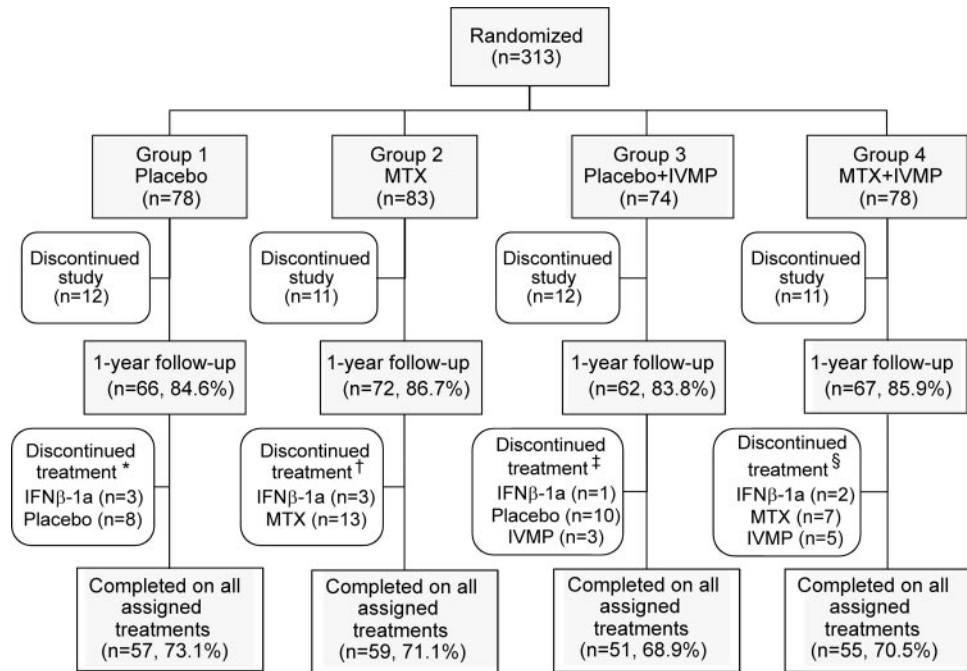
Efficacy results. Table 1 shows the distribution by treatment of the primary endpoint, N/E T2 lesion number. Sixty-six to eighty percent of patients had ≤ 1 lesion for all groups. Although the group 4 mean was half that of group 1, this was largely due to the distribution across treatments of 7 participants (2.2%) with 12–22 lesions, including 5 controls. Without these participants, the control group mean was the lowest of the four treatment groups. Overall, 216/442 (49.0%) of the N/E lesions were enlarged and 58/442 (13.1%) were also GdE. Conversely, 58/77 (75.3%) of GdE lesions at month 12 were N/E.

Table 2 and table e-3 summarize data on secondary outcomes. Table 3 and table e-4 report main effects of both therapies for study outcomes. No MTX \times IVMP interactions were significant. Main effects were small to moderate. Specifically, for lesion volume measures, the mean rank differences of 1.6 to 10.6 were small relative to the possible maximum with strong treatment effect of 78.25.

Modest trends favoring MTX were observed for several outcomes; no effect approached significance. Trends with IVMP were favorable for the primary outcome of N/E T2 lesions (OR = 0.74), the secondary outcomes GdE lesions (OR = 0.69) and relapse rate (RR = 0.70), other relapse-derived measures, EDSS change (OR = 0.76), MSFC change (difference in means = 0.02), the composite disease activity outcome (OR = 0.78), and T2 lesion volumes (for which trends were marginally significant: $p = 0.07$ for absolute and $p = 0.04$ for relative changes). However, p values for the primary and four secondary outcomes were 0.12 or higher. Trends with GdE and T1 lesion volumes were negligible, and BPF showed an unfavorable trend (change ratio = 1.08).

Safety and tolerability. Combination regimens were generally well-tolerated. While participants remained on treatment, 97.1% of IFN β -1a, 89.6% of MTX, and 94.2% of IVMP doses were full doses. There were 3,413 total AEs including all follow-up (a minor

Figure Study summary



*Two participants discontinued interferon beta-1a (IFNβ-1a) and placebo. †Three participants discontinued IFNβ-1a and methotrexate (MTX). ‡One participant discontinued IFNβ-1a and placebo, and two participants discontinued placebo and IV methylprednisolone (IVMP). §Two participants discontinued IFNβ-1a and MTX.

ity of participants were followed >1 year under the original 24-month protocol). There were no malignant neoplasms, opportunistic infections, or unanticipated toxicities (tables e-5 and e-6). Forty-one serious adverse events occurred in 36 participants, including one sudden death assessed as probably due to myocardial infarction, and considered unrelated to study medications by the investigator. No autopsy was performed. Bradycar-

dia in group 2 and spontaneous abortion in group 4 were assessed as related to MTX, which was permanently stopped. Pneumonia in group 4 was assessed as likely related to oral study medication and IVMP, neither of which were stopped.

The only significant time trend in vital signs was respiratory rate decline (approximately 1 breath per minute) over 1 year in Group 3 ($p = 0.0002$), with a nonsignificant trend in group 4. There were moderate overall decreases in hemoglobin and hematocrit and increased mean corpuscular volume with MTX. No clinically significant effects of IVMP on hematologic parameters were seen. Clinically significant effects of MTX on alanine transaminase and aspartate transaminase were infrequent and generally mild. Mean transaminase levels increased modestly over time in IVMP-treated participants, with very few clinically significant abnormal values. There were no clinically important treatment differences in frequencies of urinalysis abnormalities. DEXA results will be reported separately, but this IVMP regimen appeared not to accelerate bone mineral density loss.

Anti-IFNβ neutralizing antibodies. Overall, 38/808 (4.7%) of assays in 18/313 (2.6%) of participants were positive. The data suggested IVMP inhibited serum anti-IFNβ NAB at months 6 ($p = 0.015$) and 12 ($p = 0.018$) (table 4).

DISCUSSION The safety of ACT combination therapies was consistent with known effects of MTX

Table 1 Frequency distribution of the primary endpoint: new or enlarged T2-hyperintense lesions

No. of new or enlarged T2 lesions	Group 1: Placebo (n = 70)	Group 2: MTX (n = 76)	Group 3: Placebo + IVMP (n = 66)	Group 4: MTX + IVMP (n = 74)
0	57.1	43.4	56.1	60.8
1	20.0	22.4	16.7	18.9
2	2.9	9.2	4.5	10.8
3	7.1	9.2	6.1	2.7
4	2.9	5.3	4.5	2.7
5	1.4	—	1.5	—
6	—	3.9	4.5	2.7
7	—	—	3.0	—
8	—	2.6	1.5	—
9	1.4	2.6	—	1.4
12+	7.1	1.3	1.5	—
Maximum	20	22	16	9
Mean	1.8	1.9	1.5	0.9

MTX = methotrexate; IVMP = IV methylprednisolone.

Endpoint	Group 1: Placebo	Group 2: MTX	Group 3: Placebo + IVMP	Group 4: MTX + IVMP
GdE lesion no., n (%)				
0*	68 (87.2)	72 (86.7)	66 (89.2)	72 (92.3)
1-2	7 (9.0)	10 (12.0)	6 (8.1)	5 (6.4)
3-4	3 (3.8)	0 (0.0)	1 (1.4)	1 (1.3)
≥5	0 (0.0)	1 (1.2)	1 (1.4)	0 (0.0)
Mean	0.27	0.23	0.36	0.10
Relapses per patient year, mean*	0.53	0.40	0.40	0.28
MSFC absolute change, mean†	-0.084	0.020	0.148	-0.030
BPF relative decline, mean %‡	0.56	0.37	0.45	0.54

*Using last observation carried forward data. Based on 70, 76, 65, and 74 patients in the respective treatment groups with GdE data from follow-up scans.

†Adjudicated relapses within 1-year follow-up, based on all follow-up of all patients.

‡Negative values indicate decline and positive values indicate improved function. Based on 74, 79, 70, and 74 patients in the respective treatment groups with follow-up MSFC.

§Using last observation carried forward data. Based on 70, 76, 66, and 74 patients in the respective treatment groups with BPF data from follow-up scans.

MTX = methotrexate; IVMP = IV methylprednisolone; GdE = gadolinium-enhancing; MSFC = Multiple Sclerosis Functional Composite; BPF = brain parenchymal fraction.

and IVMP and raised no new concerns when combined with IFN β -1a. Trends favorable to IVMP were seen for several clinical and MRI endpoints, but these were not significant. The data did not suggest benefit of MTX combined with IFN β -1a.

Previous publications discussed the rationale for combination therapy in MS, selection of MTX and IVMP as adjunctive therapies, and the dose regimens.^{1,21} Numerous trials of MS combination ther-

apies have been conducted, almost all pilot studies.²¹ SENTINEL, the largest combination therapy trial to date, demonstrated the tolerability and efficacy of natalizumab combined with IM IFN β -1a compared to IM IFN β -1a.¹⁶ The design precluded comparison of the combination to natalizumab, and illustrated the potential for unanticipated toxicity (progressive multifocal leukoencephalopathy).^{22,23} Despite a seemingly strong rationale for combination MS ther-

Endpoints	Effect measure	Adjuvant treatment					
		MTX			IVMP		
		Adjusted effect size	95% CI	Wald p value	Adjusted effect size	95% CI	Wald p value
Primary							
N/E T2 lesions*	Odds ratio	0.98	0.63-1.54	0.93	0.74	0.47-1.15	0.18
Secondary							
GdE lesion no.†	Odds ratio	0.69	0.33-1.44	0.32	0.69	0.33-1.45	0.33
Relapse rate‡	Rate ratio	0.77	0.51-1.15	0.20	0.71	0.46-1.09	0.12
MSFC change§	Difference in means	0.03	-0.07-0.13	0.52	0.02	-0.08-0.12	0.65
BPF relative change¶	Change ratio	0.88	0.65-1.19	0.41	1.08	0.80-1.46	0.60

*Main effects ordinal regression (proportional cumulative odds) covariance model of categories (0, [1], 5+), with prespecified adjustment for baseline age and GdE lesion number (0, [1], 6+).

†Main effects ordinal regression (proportional cumulative odds) covariance model of categories (0, 1, 2+), with prespecified adjustment for baseline GdE lesion number (0, [1], 6+).

‡Main effects negative binomial analysis of covariance, with prespecified adjustment for baseline age, GdE lesion number (0, [1], 6+), relapses in the prior 3 years, and MSFC.

§Main effects analysis of variance model.

¶Main effects Gaussian generalized linear covariance model with log link function and prespecified adjustment for baseline GdE lesion number (0, [1], 6+).

MTX = methotrexate; IVMP = IV methylprednisolone; CI = confidence interval; N/E = new or enlarged; GdE = gadolinium-enhancing; MSFC = Multiple Sclerosis Functional Composite; BPF = brain parenchymal fraction.

Table 4 Changes in serum anti-interferon beta-1a neutralizing antibody titer from baseline to month 6 and month 12

Baseline to:	Group 1: Placebo			Group 2: MTX			Group 3: Placebo + IVMP			Group 4: MTX + IVMP		
	↑	↔	↓	↑	↔	↓	↑	↔	↓	↑	↔	↓
Month 6	4	0	1	1	1	2	0	0	2	0	0	4
Month 12	2	0	1	3	0	1	0	0	2	0	0	4

MTX = methotrexate; IVMP = IV methylprednisolone; ↑ = increase in titer; ↔ = no change in titer; ↓ = decrease in titer.

apy, no large-scale study has yet definitively shown an advantage over both platform and adjunctive agents.

ACT had several noteworthy features in design, conduct, and analysis.¹ Most important, although ACT was a large-scale trial funded by a pharmaceutical company, scientific governance and study management were investigator-run. It was hoped this would foster innovation in design and conduct, promote analyses of interest to the field, and contribute to acceptance of results. In general, administrative aspects of ACT functioned well.

Also of interest was the 2 × 2 factorial design, the advantages of which are illustrated by comparison to alternative two- and three-group designs, assuming 300 total available participants. The factorial approach allows evaluation of each adjunct by comparing 150 patients allocated to it with all remaining patients as controls. Moreover, the MTX + IVMP group provides data on use of both adjuncts. Separate two-group studies with roughly 75 participants per group would have lower statistical power and provide no data on combined adjunct use. A three-group study allows comparison of placebo vs MTX vs IVMP adjunct therapies in groups of about 100 patients each, but again joint use is not tested.

The 2 × 2 factorial design retains much of the flexibility of separate two-group designs, for example allowing distinct endpoints with different levels of masking for the two agents, as in the original ACT design. This is advantageous when distinct mechanisms of action targeting different disease manifestations are hypothesized. The sample size advantage of the factorial approach, though, assumes that a second adjunct leaves the statistical effect of the first unchanged. ACT data were consistent with this assumption, showing no significant MTX × IVMP interactions.

ACT had several shortcomings. First, although the MTX comparison was placebo-controlled, IVMP was compared to no treatment. IV placebo was considered, but ethical and practical issues, including difficulty of blinding due to infusion-related adverse effects, were felt to outweigh advantages. To minimize potential bias, clinical and MRI evaluators (including the evaluating neurologist who adjudicated

potential relapses) and the study statistician were blinded to IVMP assignment during the trial. Second, largely due to financial constraints, DEXA scans were performed only on participants receiving bi-monthly IVMP, precluding isolating IVMP's bone mineral density effects from those of MS or other causes. Third, sample size was limited by unexpectedly slow enrollment. The factorial approach requires participants accept randomization to both test agents. Some potential participants who might have enrolled in an MTX trial were deterred by potential randomization to IVMP.¹ Finally, event rates in ACT were lower than previous studies. For example, mean N/E T2 lesions were 1.8 in ACT group 1 (IFNβ-1a alone) vs 2.5 in the IFNβ-1a dose-comparison study¹⁰ 30 μg group at month 24 vs 12 and 2.4 in the SENTINEL¹⁶ IFNβ-1a alone group at year 1 vs baseline. In consequence, achieved power was modestly reduced, but remained substantial against effects half or less those of natalizumab.

ACT outcomes, particularly MRI measures, were highly skewed. This renders analyses based on average lesion counts or volumes vulnerable to distortion by uncommon very high observations. Hence, ordinal (rank-based) statistical methods were chosen for most analyses, including for the primary endpoint. The nonsignificant benefit with IVMP observed largely in the upper tail of N/E T2 lesions may represent a substantial treatment effect limited to a small subpopulation, a widespread but small benefit missed by chance, or a chance effect of randomization. ACT data do not distinguish these possibilities.

Overall, 5.1% of participants had a positive baseline IFNβ NAb titer, a slightly higher proportion than in other studies of IM IFNβ-1a.¹⁰ The increased prevalence might reflect selection of participants with active disease on IFNβ-1a therapy. Although the number of participants with positive baseline NAb titers was small, ACT data suggest that IVMP might lower NAb titers, which deserves further study.

AUTHOR CONTRIBUTIONS

The study statistician was P.B.I.

ACKNOWLEDGMENT

The authors thank John Barnard, PhD, for consultations on multiple imputation methods in general and their approach in particular.

DISCLOSURE

Dr. Cohen has received personal compensation as a consultant or speaker from Biogen Idec, Eisai, Eli Lilly, Genentech, Genzyme, Glaxo Smith Kline, IMPAX, Incyte, Novartis, Schering Plough, Serono, Teva, and Wyeth. He has received research support paid to his institution from Artielle, Biogen Idec, BioMS, Genzyme, Orchestra Therapeutics, Novartis, and Teva. Dr. Imrey holds stock in Sepracor. Dr. Calabresi has received personal compensation as a consultant for Amgen, Biogen Idec, Eisai, Genentech, Millenium, Novartis, Serono, Teva, and Vertex. He has received research support paid to his institution from Biogen Idec, Eisai, Genentech, Millenium, Novartis, Serono, and Teva. Dr. Edwards has received personal compensation as a consultant or speaker from Biogen Idec, Novartis, Pfizer, Serono, and Teva. Dr. Felton has received personal compensation as a speaker from Biogen Idec, Bristol Myers Squibb, Pfizer, Sanofi-Aventis, Serono, and Teva. He has received research support paid to his institution from NIH, Merck, Novartis, Eli Lilly, Centocor, NMT Medical, Boehringer Ingelheim, Ono Pharma, Biogen Idec, Sanofi-Aventis, and Taisho. Dr. Fisher has received personal compensation as a consultant or speaker from Biogen Idec and Millenium. She has received research support paid to her institution from Biogen Idec and Millenium. Dr. Fox has received personal compensation as a consultant or speaker from Biogen Idec, Genentech, Novartis, and Teva. He has received research support paid to his institution from Biogen Idec and Genentech. Dr. Goodman has received personal compensation as a consultant or speaker from Alza, Berlex, Biogen Idec, Genentech, Serono-Pfizer, and Teva. He received personal compensation for serving on the editorial board of *Reviews in Neurological Disease*. He received research support paid to his institution from Acorda, Berlex, Biogen Idec/Elan, Serono, and Teva. Dr. Hutton has received personal compensation as a consultant from Bayer, Biogen Idec, and EMD Serono. He has received research support paid to his institution from Biogen Idec, Genzyme, Genentech, Opexa, and Novartis. Dr. Mandell has received personal compensation as a consultant from Pfizer, Sanofi, and Savient. He is Editor-in-Chief of *Cleveland Clinic Journal of Medicine*. Dr. Scott has received personal compensation as a consultant or speaker from Biogen Idec, Cephalon, Pfizer, and Teva. Drs. Eickenhorst and Zhang are full-time employees of Biogen Idec. Ms. Hara-Cleaver, Dr. Apperson-Hansen, Dr. Beck, Dr. Houghtaling, Dr. Karafa, and Ms. Stadler have nothing to disclose.

Received June 3, 2008. Accepted in final form October 16, 2008.

REFERENCES

1. Cohen JA, Calabresi PA, Chakraborty S, et al. Avonex Combination Trial (ACT) in relapsing-remitting MS: rationale, design, and baseline data. *Mult Scler* 2008;14:370–382.
2. Kappos L. Neurostatus. Basel, Switzerland: Department of Neurology, University Hospital, CH4031; 2002.
3. Kurtzke JF. Rating neurologic impairment in multiple sclerosis: an expanded disability status scale (EDSS). *Neurology* 1983;33:1444–1452.
4. Rudick R, Antel J, Confavreux C, et al. Recommendations from the National Multiple Sclerosis Society Clinical Outcomes Assessment Task Force. *Ann Neurol* 1997;42:379–382.
5. McDonald WI, Compston DA, Edan G, et al. Recommended diagnostic criteria for multiple sclerosis: guidelines from the International Panel on the Diagnosis of Multiple Sclerosis. *Ann Neurol* 2001;50:121–127.
6. Lublin FD, Reingold SC. Defining the clinical course of multiple sclerosis: results of an international survey. *Neurology* 1996;46:907–911.

7. Fisher E, Cothren RM, Tkach JA, Masaryk TJ, Cornhill JF. Knowledge-based 3D segmentation of MR images for quantitative MS lesion tracking. *SPIE Medical Imaging* 1997;3034:19–25.
8. Rudick RA, Fisher E, Lee J-C, Simon J, Jacobs L, the Multiple Sclerosis Collaborative Research Group. Use of the brain parenchymal fraction to measure whole brain atrophy in relapsing-remitting MS. *Neurology* 1999;53:1698–1704.
9. Rudick RA, Simonian NA, Alam JA, et al. Incidence and significance of neutralizing antibodies to interferon beta-1a in multiple sclerosis. *Neurology* 1998;50:1266–1272.
10. Clanet M, Radue EW, Kappos L, et al. A randomized, double-blind, dose-comparison study of weekly interferon β -1a (Avonex) in relapsing MS. *Neurology* 2002;59:1507–1517.
11. Ananth CV, Kleinbaum DG. Regression models for ordinal responses: a review of methods and applications. *Int J Epidemiol* 1997;26:1323–1333.
12. Koch GG, Tangen CM, Jung J-W, Amara IA. Issues for covariance analysis of dichotomous and ordered categorical data from randomized trials and non-parametric strategies for addressing them. *Stat Med* 1998;17:1863–1892.
13. Tangen CM, Koch GG. Nonparametric analysis of covariance for hypothesis testing with logrank and Wilcoxon scores and survival-rate estimation in a randomized clinical trial. *J Biopharm Stat* 1999;9:307–338.
14. Gardner W, Mulvey EP, Shaw EC. Regression analyses of counts and rates: Poisson, over-dispersed Poisson, and negative binomial models. *Psychol Bull* 1995;118:392–404.
15. Khuri AI, Mukherjee B, Sinha BK, Ghosh M. Design issues for generalized linear models: a review. *Stat Sci* 2006; 21:376–399.
16. Rudick RA, Stuart WH, Calabresi PA, et al. Natalizumab plus interferon beta-1a for relapsing multiple sclerosis. *N Engl J Med* 2006;354:911–923.
17. Fox RJ, Lee J-C, Rudick RA. Optimal reference population for the Multiple Sclerosis Functional Composite. *Mult Scler* 2007;13:909–914.
18. Fischer JS, Jak AJ, Knicker JE, Rudick RA, Cutter G. Multiple Sclerosis Functional Composite (MSFC). Administration and Scoring Manual: Unitech. National Multiple Sclerosis Society; 2001.
19. Wood SN. Thin plate regression splines. *JR Stat Soc B* 2003;65:95–114.
20. La Vange LM, Koch GG. Rank score tests. *Circulation* 2006;114:2528–2533.
21. Cohen JA, Confavreux C. Combination therapy in multiple sclerosis. In: Cohen JA, Rudick RA, eds. *Multiple Sclerosis Therapeutics*, 3rd ed. London: Informa Healthcare; 2007:681–697.
22. Kleinschmidt-Demasters BK, Tyler KL. Progressive multifocal leukoencephalopathy complicating treatment with natalizumab and interferon-beta-1a for multiple sclerosis. *N Engl J Med* 2005;353:369–374.
23. Langer-Gould A, Atlas SW, Bollen AW, Pelletier D. Progressive multifocal leukoencephalopathy in a person treated with natalizumab. *N Engl J Med* 2005;353:375–381.

Results of the Avonex Combination Trial (ACT) in relapsing-remitting MS
J. A. Cohen, P. B. Imrey, P. A. Calabresi, K. R. Edwards, T. Eickenhorst, W. L. Felton,
III, E. Fisher, R. J. Fox, A. D. Goodman, C. Hara-Cleaver, G. J. Hutton, B. F. Mandell,
T. F. Scott, H. Zhang, C. Apperson-Hansen, G. J. Beck, P. L. Houghtaling, M. T.
Karafa, M. Stadler and For the ACT Investigators

Neurology 2009;72;535-541

DOI: 10.1212/01.wnl.0000341934.12142.74

This information is current as of July 14, 2009

Updated Information & Services	including high-resolution figures, can be found at: http://www.neurology.org/cgi/content/full/72/6/535
Supplementary Material	Supplementary material can be found at: http://www.neurology.org/cgi/content/full/72/6/535/DC1
Subspecialty Collections	This article, along with others on similar topics, appears in the following collection(s): MRI http://www.neurology.org/cgi/collection/mri Clinical trials Randomized controlled (CONSORT agreement) http://www.neurology.org/cgi/collection/clinical_trials_randomized_controlled_consort_agreement Multiple sclerosis http://www.neurology.org/cgi/collection/multiple_sclerosis
Permissions & Licensing	Information about reproducing this article in parts (figures, tables) or in its entirety can be found online at: http://www.neurology.org/misc/Permissions.shtml
Reprints	Information about ordering reprints can be found online: http://www.neurology.org/misc/reprints.shtml

