# A Double-Blind, Placebo-Controlled, Randomized Trial of Cladribine in Relapsing-Remitting Multiple Sclerosis

**John S. Romine,* Jack C. Sipe,* James A. Koziol,‡ Jack Zyroff,† and Ernest Beutler‡**

*Division of Neurology and †Department of Radiology, Scripps Clinic, and ‡Department of Molecular and Experimental Medicine, The Scripps Research Institute, La Jolla, CA

We conducted an 18-month, placebo-controlled, double-blind study to evaluate cladribine in the treatment of 52 patients with relapsing-remitting multiple sclerosis. Patients received either placebo or cladribine 0.07 mg/kg/day by subcutaneous injection for 5 consecutive days as six monthly courses for a total cumulative dose of 2.1 mg/kg. Analysis of results revealed a statistically significant favorable effect of cladribine on the joint frequency and severity of relapses and magnetic resonance imaging (MRI) findings. MRI-enhancing lesions were completely suppressed in the cladribine patients by the sixth month of treatment. Mild segmental herpes zoster occurred in two cladribine-treated patients and one patient receiving placebo. Otherwise, there were no side effects or adverse events. We conclude that cladribine shows promise as a treatment for relapsing-remitting multiple sclerosis.

Although the cause and cure of multiple sclerosis (MS) remains unknown, it is generally accepted that immunological mechanisms play an important role in the inflammatory demyelination of the central nervous system that is the pathological hallmark of the disease. For this reason, current approaches to the treatment of MS patients are focused on modulation or suppression of the immune system.

The drug cladribine [2-chlorodeoxyadenosine (2-CdA), Mylinax, Ortho McNeil, Raritan, NJ] is a selective immunosuppressive molecule synthesized by Carson to mimic the immunodeficiency state seen in hereditary adenosine deaminase deficiency (1). It is a purine nucleoside with chlorine substituted for hydrogen at the 2 position of the purine ring. This makes the molecule resistant to adenosine deaminase, leading to accumulation of deoxynucleotides and selective killing of lymphocytes with relatively little toxicity in other tissues (2). The drug is used widely as a treatment for lymphoid malignancies and is especially effective in hairy cell leukemia (3,4). Because of positive results from our therapeutic trial of cladribine in progressive MS (5,6), we undertook this study to evaluate cladribine in patients with the relapsing-remitting form of MS.

**Key words:** immunosuppression; 2-chlorodeoxyadenosine.

Address correspondence and reprint requests to: John S. Romine, M.D., Division of Neurology, MS313, Scripps Clinic, 10666 North Torrey Pines Road, La Jolla, CA 92037.

Received 18 September 1998; Accepted 23 September 1998.

## METHODS

### Patient Selection

The study subjects were 52 patients with clinically definite relapsing-remitting MS for at least 1 year. All patients had a history of two or more relapses in the previous 2 years and Extended Disability Status Score (EDSS) scores of 6.5 or less at time of study entry. The majority of patients had been followed at Scripps Clinic. Exclusion criteria included: 1) prior treatment with an immunosuppressive drug within 3 months; 2) a serum creatinine of $>1.5$ mg/dl; 3) serum glutamic-oxaloacetic transaminase/serum glutamic-pyruvic transaminase or alkaline phosphatase elevated to twice the upper limit of normal; 4) baseline neutrophil counts of $<1600/\mu l$ or platelet counts of $<130{,}000/\mu l$; and 5) previous total lymphoid irradiation or prior extensive myelosuppressive chemotherapy.

The study plan, risks, and potential benefits were explained to each patient in detail. All patients gave informed consent to participate in the study.

### Study Design

An 18-month, randomized, placebo-controlled, double-blind study was conducted in the facilities of the General Clinical Research Center (GCRC) of Scripps Clinic. After completion of screening evaluations, 52 patients were stratified according to gender, age (in 10-year intervals), and degree of disability as mea-

sured by Scripps Neurological Rating Scale (SNRS; 7) (in 10-point intervals). The stratified groups were then randomized in blocks of four to either the placebo arm or the cladribine arm. In all, 27 patients were randomized onto the cladribine arm and 25 onto the placebo arm. Throughout the study, patients, neurologists, nurses, and the neuroradiologist remained blinded to treatment assignment. A pharmacist was informed of patient assignment by code in order to dispense placebo or the appropriate dose of cladribine to each patient.

In all patients, clinical neurological exams plus SNRS and EDSS rating scales were performed at baseline and repeated by the same neurologist every month for the first year, every 3 months for the second year, and within 48 hr or less of report by a patient of a relapse. A clinical relapse was defined as the appearance of new symptoms or worsening of an existing symptom, attributable to MS and accompanied by objective worsening of neurological findings. To be scored as a relapse the alterations must have been preceded by disease stability or improvement lasting for at least 30 days, and the worsening must have lasted at least 24 hr and occur in the absence of fever. Relapse severity was rated as follows: 1) mild relapse—decrease in SNRS of 1–7; 2) moderate relapse—decrease in SNRS of 8–14; or 3) severe relapse—decrease in SNRS of 15 or greater.

Magnetic resonance imaging (MRI) of the brain was performed on a 1.5 T Signa scanner (General Electric, Milwaukee, WI) for each patient at baseline, and then monthly for the first year and every 6 months the second year. T1-weighted scans were obtained in the sagittal and axial planes. Axial scans of 3 mm thickness and zero interslice gaps were done about 10 min after the intravenous injection of gadopentetate dimeglumine (Magnevist, Berlex Laboratories). Special attention was given to careful repositioning of patients to guarantee reproducible slice positions. The regions of contrast enhancement on T1-weighted scans were outlined by hand on filmed images. All scans were interpreted and marked by the same neuroradiologist (J.Z.), who had no knowledge of patient treatment assignment. These were then duplicated by a technologist using the taped raw data and a computer workstation [ANALYZE (8), Rochester, MN]. Quantitation of MRI findings involved the determination of lesion areas on the consecutive sections of the T1-weighted scans as interpreted by one of two skilled technologists, then calculation of volumes by assuming homogeneity of lesions across the sections. Initially, the taped raw data from the individual scans were read into a volume-rendering software program, ANALYZE, running on a Hewlett-Packard 712/60 workstation. Our methodology for lesion area determination is a semiautomated quantitative technique adapted from Wicks et al. (9) and Filippi et al. (10).

## Drug Administration

In contrast to our earlier study of intravenous cladribine in progressive MS (5), the drug was administered subcutaneously because of greater ease of administration and because it has now been established that the pharmacological properties and response rates of cladribine in lymphoproliferative diseases are the same if the drug is given either intravenously or subcutaneously (11). Each patient received a course of five consecutive daily subcutaneous injections of cladribine, 0.07 mg/kg/day or an equivalent volume of saline placebo, fractionated into two or three injection sites, and given monthly for 6 months for a total cumulative dose of 2.1 mg/kg of cladribine. A complete blood count was obtained before each monthly course of treatment and reviewed by the pharmacist, and the next dose of cladribine was given only if blood count safety criteria were met according to an algorithm designed for this purpose by one of us (E.B.; Table 1). If these criteria were not met, a placebo dose was substituted. The study design included eight monthly courses. The last two courses ordinarily consisted of placebo, but if a drug dose had been omitted because of blood count inadequacy, then active drug could be given at month 7 or 8 instead of placebo.

## Statistical Considerations

Two primary outcome measures were identified: 1) the joint frequency and severity of clinical relapses as judged by neurological examination; and 2) the numbers of enhancing lesions on T1-weighted MRI brain

**Table 1. Pretreatment safety criteria for monthly courses of cladribine in multiple sclerosis**

1. Platelet count must be:
   a. 200,000 or higher, or
   b. Between 150,000 and 200,000 and represent more than 50% of previous pretreatment platelet count, or
   c. Between 125,000 and 150,000 and represent at least 80% of previous pretreatment platelet count
2. Absolute granulocyte count must be greater than 1000
3. Hemoglobin level must not have declined:
   a. More than 1.5 g/dl from previous monthly pretreatment level, or
   b. 3 g/dl or more from baseline

scans. Outcomes were to be assessed at 1 year. A sample size of 25 patients per group would be sufficient to detect a decline in the annual rate of exacerbations, from 1 in the placebo group to 0.5 in the cladribine group, with a two-sided Poisson test at alpha level 0.05 (12). Similarly, on the basis of findings from our chronic progressive MS trial, we postulated that the frequency of enhancing lesions in the placebo group would remain at 50% throughout the course of this study, whereas the frequency of enhancing lesions in the cladribine-treated group would decline from 50% to <10% at 1 year. A sample size of 25 patients per treatment group would be sufficient to detect a difference of 50% versus 10% with a power of 0.90, using a two-sided binomial test at alpha level 0.05.

Our analyses were intent-to-treat, in that all data from every patient initially randomized either to placebo or to cladribine are included and reported for that initial treatment group. Blinded observations were undertaken up to 18 months from baseline (trial entry); hence, information is reported out to this period. We present primary analyses—that is, analyses of the primary outcomes at 1 year—and identify other analyses as secondary. We did not impute data values for any patient not observed out to 18 months.

Comparison of the joint frequency and severity of relapses between the two treatment groups was undertaken using Mantel's (13) extension of the Mantel-Haenszel procedure, here denoted $Q_M$. (Mantel's procedure can incorporate arbitrary scores for the degree of relapse, but we chose to score objectively by means of ranks based on drop in SNRS score, separately in each monthly summary table of relapses, as cross-classified by treatment group.) A stratified version of Mantel's test and a general linear model with Poisson link (that is, a Poisson regression model; 14) were also used to evaluate the significance of covariate information as predictors of clinical relapse. Confidence intervals for relapse rates were calculated under the assumption that the numbers of events followed a Poisson distribution in each treatment group. Comparison of the frequency of enhancing lesions on T1-weighted MRI scans over time was done with McNemar's test for paired data (within treatment groups) and Fisher exact test (between treatment groups); logistic regression was also used to assess the significance of covariate information. A nonparametric repeated measures analysis of variance procedure (15) was used to compare neurological performance scores (EDSS and SNRS) between the two treatment groups over the course of the study. The EDSS and SNRS scores were considered to be secondary outcome measures since little change might be expected between the two groups over the relatively short time frame of the study. Two-sided p values relative to the null distributions of the observed test statistics are reported.

Intrarater reliability of the determination of presence of enhancing lesions on T1-weighted MRI scans was assessed by means of a test-retest of 20 scans by the examining neuroradiologist (J.Z.). Discrepancies between the two independent evaluations were 15% (3/20). In a similar spirit, both examining neurologists (J.R. and J.S.) participated in a study of inter-rater and intrarater reliability, with regard to the neurological rating scales. Twenty patients (J.R., 10; J.S., 10) were assessed by the same examiner twice on the same day, the period between examinations ranging from 135 min to 240 min. Intrarater agreement for one examiner (J.S.) on the EDSS was perfect; the weighted $\kappa$ coefficient of agreement (16) for the other examiner was 0.997. The weighted $\kappa$ coefficients of agreement between the paired SNRS scores were 0.999 for both examiners. Separately, 20 patients were independently assessed by each examiner on the same day. Inter-rater agreement was high: the weighted $\kappa$ coefficient of association was 0.990 for the EDSS and 0.957 for the SNRS. Inter-rater agreement on the EDSS was 100% for all sets of examinations when agreement was defined as a difference of less than or equal to 1.0, and 95% when agreement was defined as a difference of less than or equal to 0.5. Inter-rater agreement on the SNRS was 95% when agreement was defined as a difference of no more than 10 points, and 90% when agreement was defined as a difference of no more than 5 points.

## RESULTS

### Trial Considerations

There was one withdrawal on the placebo arm at 3 months (conversion disorder complicating assessment of underlying MS), and one withdrawal on the cladribine arm at 4 months (patient moved out of state); all of the remaining patients received standard intervention without deviations, as specified in the protocol. Thus, 26 cladribine patients and 24 placebo patients were available for evaluation at 12 months. During the period from 12 to 18 months, five patients on placebo withdrew: two patients moved out of state, two withdrew for unspecified reasons, and one withdrew because of worsening MS. One patient receiving cladribine also withdrew because of worsening MS. Thus, 25 cladribine patients and 19 placebo patients were available for evaluation the entire 18-month period. Figure 1 depicts the trial profile. During the period from 12 to 18 months, the blinding was removed from two cladribine patients and two placebo patients.

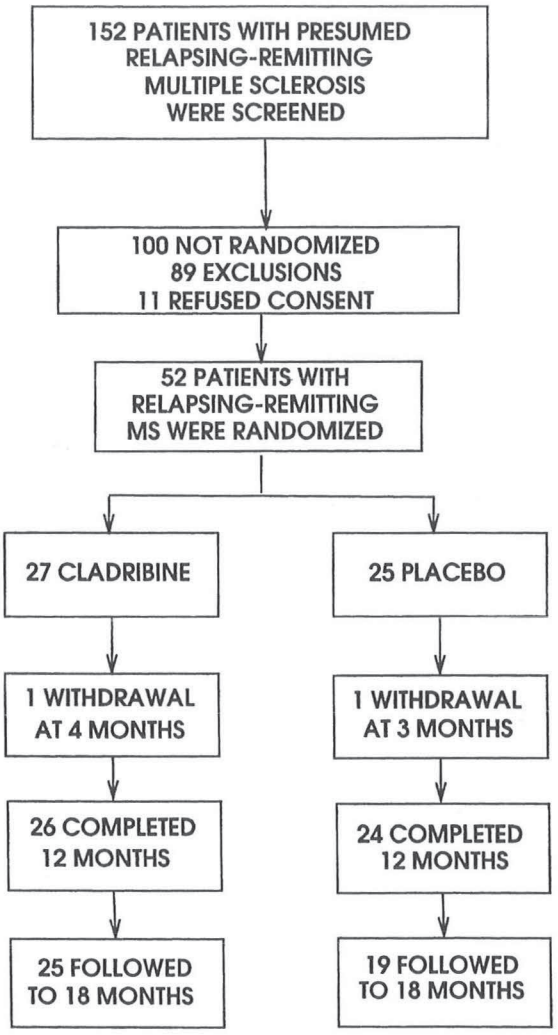


**Figure 1. Trial profile of the cladribine relapsing-remitting MS clinical trial.**

Because of potential bias, information from these patients concerning their frequency and severity of exacerbations subsequent to the point of unblinding is not used in the calculation and comparison of exacerbation rates between the two treatment groups.

### Demographic and Baseline Characteristics in the Two Treatment Groups after Randomization

The two groups were similar in terms of baseline clinical characteristics (Table 2). Each group had an approximate 2:1 female-to-male preponderance and comparable mean age, disease duration, and baseline EDSS. Patients randomized to cladribine therapy averaged a slightly greater number of exacerbations in the 12 months prior to study entry than patients randomized to placebo.

**Table 2. Baseline demographic and clinical characteristics**

| | Placebo (n = 25) | Cladribine (n = 27) |
|---|---|---|
| Sex | | |
| Male | 7 | 9 |
| Female | 18 | 18 |
| Race | | |
| White | 25 | 24 |
| Other | 0 | 3 |
| Age (years) | | |
| Mean | 39.8 | 43.4 |
| 25th percentile | 36.5 | 38.5 |
| 50th pecentile | 41 | 44.5 |
| 75th percentile | 44 | 49.5 |
| Range | 31–52 | 30–52 |
| Years with symptoms | | |
| Mean | 9.1 | 10.2 |
| 25th percentile | 3.5 | 4.5 |
| 50th pecentile | 9 | 8 |
| 75th percentile | 12.5 | 12.5 |
| Range | 1–25 | 1–29 |
| Number of exacerbations in previous year | | |
| 1 | 13 | 5 |
| 2 | 5 | 16 |
| 3 or 4 | 7 | 6 |
| Baseline EDSS | | |
| Mean | 3.8 | 3.9 |
| 25th percentile | 2.5 | 2.3 |
| 50th pecentile | 3.5 | 5.5 |
| 75th percentile | 5.3 | 5.5 |
| Range | 2–6.5 | 2–6.5 |
| Baseline SNRS | | |
| Mean | 75.8 | 76.1 |
| 25th percentile | 67 | 66 |
| 50th pecentile | 75.5 | 78.5 |
| 75th percentile | 86 | 86.5 |
| Range | 54–98 | 41–93 |

EDSS, Extended Disability Status Score. SNRS, Scripps Neurological Rating Scale.

### Effect of Cladribine on Outcome Measures

Figure 2 depicts the frequency and severity of exacerbations for all patients enrolled in the study. We examined the joint distribution of frequency and severity over months 7 through 12 for treatment comparisons: on the basis of our prior experience (17), we expected the maximum immunosuppression on cladribine therapy would not be achieved prior to month 7. Using the extended Mantel-Haenszel procedure, we found that there is a statistically significant reduction in the frequency and severity of exacerbations in the cladribine group compared to the placebo group over months 7 through 12 ($Q_M = 2.30$, $2p = .021$). Over this period, the relapse rate in the cladribine group

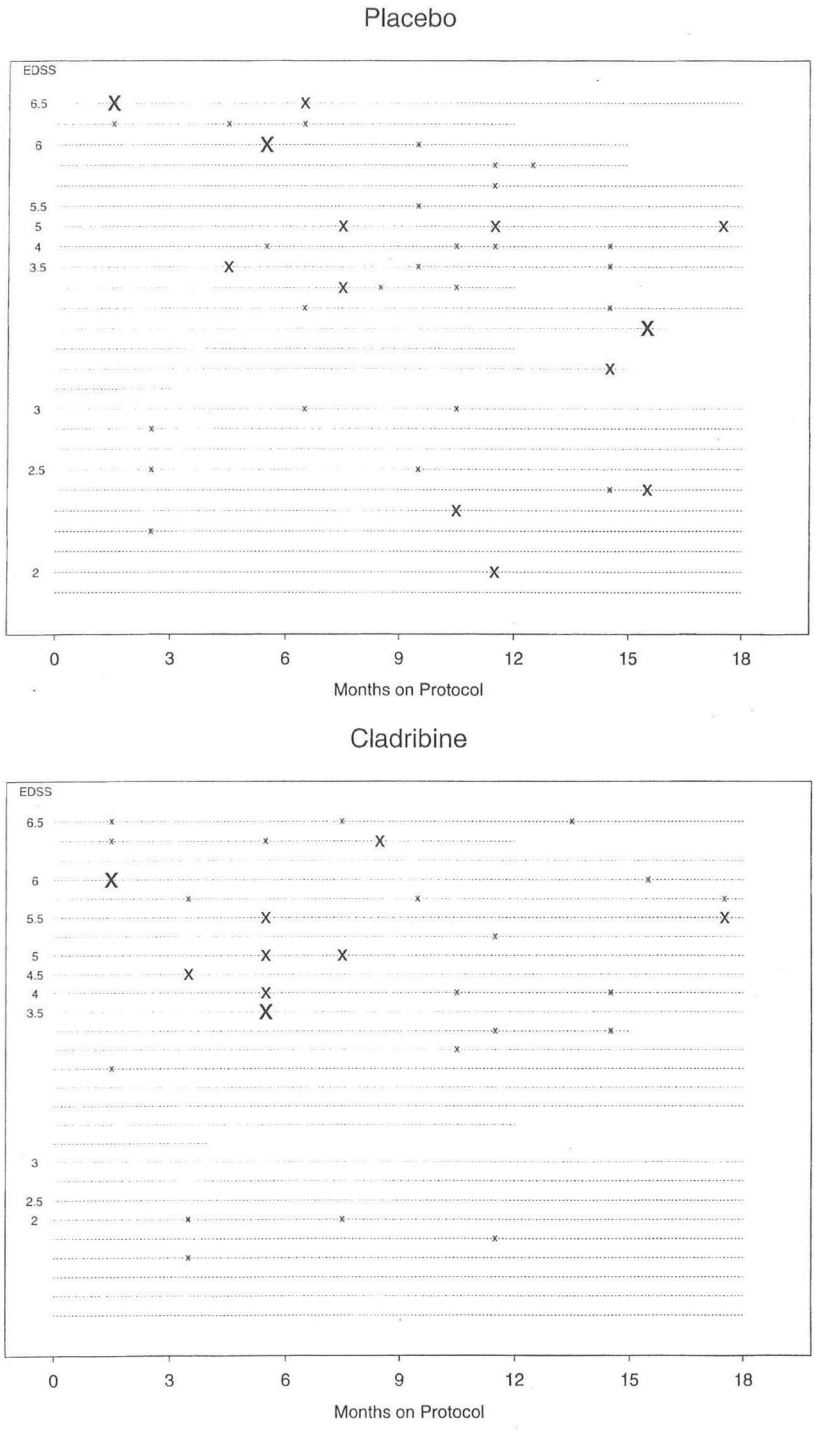


**Figure 2. Event charts for frequency and severity of exacerbations over the course of the study in placebo-treated and cladribine-treated patients.**
Within treatment groups, patients are ordered in terms of decreasing EDSS scores at baseline; patients with identical EDSS scores at baseline are ordered by numbers of new exacerbations. Mild, moderate, or severe exacerbations are denoted by progressively heavier X's.

was 0.77 per year [95% confidence interval (CI), 0.37–1.41], compared to 1.67 per year in the placebo group (95% CI, 1.02–2.57). With Poisson regression, we identified treatment along with two other covariates, baseline EDSS and number of exacerbations in the year prior to start of treatment, as significant predictors of relapse over months 7 through 12 (with fewer relapses being associated with cladribine therapy, lower EDSS scores at baseline, and fewer exacerbations in the year prior to start of treatment). In secondary analyses, we found that the reduction in the distribution of frequency and severity of exacerbations in the cladribine group relative to the placebo group is sustained at 18 months: $Q_M = 2.59$, $2p = .010$ over

DOCKET ALARM

# Explore Litigation Insights

Docket Alarm provides insights to develop a more informed litigation strategy and the peace of mind of knowing you're on top of things.

## Real-Time Litigation Alerts

Keep your litigation team up-to-date with **real-time alerts** and advanced team management tools built for the enterprise, all while greatly reducing PACER spend.

Our comprehensive service means we can handle Federal, State, and Administrative courts across the country.

## Advanced Docket Research

With over 230 million records, Docket Alarm's cloud-native docket research platform finds what other services can't. Coverage includes Federal, State, plus PTAB, TTAB, ITC and NLRB decisions, all in one place.

Identify arguments that have been successful in the past with full text, pinpoint searching. Link to case law cited within any court document via Fastcase.

## Analytics At Your Fingertips

Learn what happened the last time a particular judge, opposing counsel or company faced cases similar to yours.

Advanced out-of-the-box PTAB and TTAB analytics are always at your fingertips.

## API

Docket Alarm offers a powerful API (application programming interface) to developers that want to integrate case filings into their apps.

**LAW FIRMS**
Build custom dashboards for your attorneys and clients with live data direct from the court.

Automate many repetitive legal tasks like conflict checks, document management, and marketing.

**FINANCIAL INSTITUTIONS**
Litigation and bankruptcy checks for companies and debtors.

**E-DISCOVERY AND LEGAL VENDORS**
Sync your system to PACER to automate legal marketing.