sis that shell-breaking predators became more important as agents of selection among gastropods in the middle Mesozoic. The fossil record further shows that shell-breaking fish and crustaceans appeared in the Devonian but did not diversify on a large scale until the Jurassic (*1, 2, 7*). These trends through time were accompanied by other important changes in shallow-water marine communities, including increased disturbance of soft sediments by burrowing animals (*8*) and increased destruction of rocks by boring and grazing organisms (*9*). The biological component of selection has evidently undergone substantial change over the course of Earth history.

GEERAT J. VERMEIJ
Department of Zoology, University of Maryland, College Park 20742

DAVID E. SCHINDEL
Department of Geology and Geophysics, Yale University, New Haven, Connecticut 06520

EDITH ZIPSER
Department of Zoology, University of Maryland

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5. Material was obtained from several localities in seven stratigraphic intervals in Middle and Upper Pennsylvanian strata of north-central Texas (Strawn Group: Grindstone Creek, East Mountain, and Palo Pinto shales; Canyon Group: Wolf Mountain and Colony Creek shales; Cisco Group: Finis and Wayland shales; collections in Yale Peabody Museum), from the St. Cassian Group of the Upper Triassic of northern Italy (Museo de Cortina d'Ampezzo), from the Ripley Formation in the Upper Cretaceous of the southeastern United States (U.S. National Museum of Natural History), from the Lower Gatun Formation in the Upper Miocene of Panama (U.S. National Museum of Natural History and Vermeij collections), and from the Recent (ten localities in the tropical Western and Eastern Pacific; Vermeij collection). All measured samples contained at least ten well-preserved shells. Median frequencies of repair (Table 1) illustrate the observed trend but were not used in the statistical treatment of the data. Further details of the method are given in (*3, 4*).
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Intrathecal Interferon Reduces Exacerbations of Multiple Sclerosis

Abstract. *Ten patients with multiple sclerosis who were treated with human fibroblast interferon (IFN-B) for 6 months showed a significant reduction in their exacerbation rates compared with their rates before treatment ($P < .01$). The IFN-B was administered intrathecally by serial lumbar punctures. There was no significant change in the exacerbation rates of ten multiple sclerosis control patients before and during the period of observation. The IFN-B recipients have now been on the study a mean of 1.5 years, the controls, 1.2 years. The clinical condition of five of the IFN-B recipients and one of the control patients has improved, whereas the condition of five of the controls and one of the IFN-B recipients has deteriorated ($P < .036$). These findings warrant cautious optimism about the efficacy of intrathecal IFN-B in altering the course of multiple sclerosis and support concepts of a viral or dysimmune etiology of the disease.*

There is evidence that multiple sclerosis (MS) is caused (at least partially) by a viral infection of the central nervous system (CNS) that acts as a "trigger" for repeated exacerbations of neurologic symptoms characteristic of the disease (*1, 2*). Interferon (IFN) is a naturally occurring biologic product with potent antiviral activities (*3*). It does not cross the blood-brain barrier in significant quantity when administered systemically, but can be safely administered intrathecally (*4*). For these reasons we conducted a randomized controlled study of the effects of intrathecally administered human fibroblast interferon (IFN-B) in a series of MS patients.

We included in this study 20 patients (15 women and 5 men aged 15 to 40 years, mean age 30.7 years) who fulfilled clinical and laboratory criteria (*5, 6*) for making the diagnosis of MS with certainty (Table 1). We obtained the informed consent of each patient. The patients were randomly assigned to a group of ten IFN-B recipients or ten controls. The study was not blinded; serial lumbar punctures to inject placebo in the controls were not justified. There were seven women and three men (aged 20 to 40 years, mean age 30.2 years) in the recipient group and eight women and two men (aged 15 to 39 years, mean age 31.1 years) in the control group. Duration of disease prior to entering the study was 1.0 to 19.4 years (mean 8.0 years) in the recipient group and 2.8 to 20.5 years (mean 8.5 years) in the controls. The types of diseases were: exacerbating-remitting with residua (ER-R) in two IFN-B recipients and two controls; exacerbating-remitting, progressive (ER-P) in three IFN-B recipients and one control; and stable with residua (S-R) in five IFN-B recipients and seven controls. The basis for this classification has been described elsewhere (*6, 7*).

All IFN-B recipients had received adrenocorticotropin (ACTH) intravenously or intramuscularly or oral steroids during exacerbations prior to the study; they agreed that they would not receive these therapies should they exacerbate for at least the first year of the study. Should they desire such treatments for relapses after the first year, then ACTH or oral prednisone could be administered without withdrawing them from the study. Controls received these steroids for exacerbations as required during the study. Two controls (see Table 1, controls 1 and 9) also took oral prednisone (10 to 30 mg) on alternate days during the study (as they had done for the previous 1 to 2 years). Initially there were 12 patients in each group. One recipient and two controls withdrew shortly after entering the study; one recipient died during the first month of the study but his death was not related to IFN-B administration.

Each patient underwent a complete neurologic examination at the beginning of the study and at least monthly after entering the study. The recipients were also assessed semiweekly for the first month of the study (when they were receiving IFN-B twice per week). The severity of the patients' symptoms and signs were scored by disability status according to the method described by Kurtzke (*8*). On the basis of the severity of the signs, an overall assessment of the patients' clinical condition (improved, unchanged, worsened) was made at the time of each reevaluation.

An exacerbation was defined as a separate episode or period of development of new symptoms or a group of symptoms when the clinical course had been stationary or improving during the previous month (*6, 9*). The bout had to last longer than 24 hours and there had to be objective neurologic changes confirming the deterioration on examination. If symptoms progressed after a stationary

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or improving period of less than 1 month, then that time was included as part of a single exacerbation. The duration of an exacerbation was considered as the time elapsing from the onset of symptoms until the new or worsened symptoms and signs had reached their maximum (*6, 9*). For each patient the frequencies of exacerbations prior to entering the study were determined and exacerbation rates (exacerbations per year) for the duration of disease prior to entering the study were calculated. New exacerbation rates were calculated for each patient monthly during the study and compared with the rates before the study (two-tailed *t*-test). The number of exacerbations to be expected or predicted during the study [exacerbation rates before the study multiplied by time (years) on the study] was determined monthly for each patient. The predictions were compared with the actual number of exacerbations that occurred during the study (two-tailed *t*-test).

The IFN-B was prepared at Roswell Park Memorial Institute in the laboratory of J. Horoszewicz by superinduction of human fibroblasts (*10*). Purification by affinity chromatography (*11*) yielded a product with a specific activity of 10^7 interferon reference units (IRU) of IFN-B per milligram of protein. The purified IFN-B was stabilized by addition of human albumin, lyophilized, and kept at 4°C at 2×10^6 or 3×10^6 IRU per vial.

The IFN-B was administered by serial lumbar punctures performed semiweekly for the first 4 weeks and then once per month for the next 5 months of the study. At the time of each lumbar puncture, 5 to 15 ml of cerebrospinal fluid (CSF) was withdrawn for analysis prior to intrathecal injection of the IFN-B. The dosage to each recipient was 1×10^6 IRU/m^2 at the time of each lumbar puncture. Recipients underwent a lumbar puncture 1 year after entering the study so that CSF could be obtained for analysis. Controls underwent lumbar punctures upon entering the study and after 1 year in the study for CSF analysis.

The IFN-B recipients were hospitalized for at least 1 week at the start of the study and were observed for toxic reactions. Vital signs were recorded at least every 8 hours and more frequently if indicated. Complete blood counts including platelet and reticulocyte counts, blood chemistries (electrolytes, liver, and renal screens), and urinalyses were performed at least twice while they were in the hospital and more often if indicated. If the recipients developed severe headaches or other significant side effects after the first two lumbar punctures they remained in the hospital for 2 to 3 weeks and were observed closely. Subsequent doses of IFN-B were administered in an outpatient treatment room.

The CSF was analyzed for cells, glucose, and total protein after each lumbar puncture and for immunoglobulin and myelin basic protein at the time of lumbar punctures 1, 12, and 14 in the IFN-B recipient group and at the time of the two lumbar punctures in the controls. We analyzed the CSF and serum for the presence of IFN at the time of the lumbar punctures in both groups (*12*).

In Table 1 we summarize the pertinent clinical data on these patients. As of August 1981 the IFN-B recipients had been followed for 1.4 to 1.6 years (mean 1.5 years) since entering the study and the controls for 1.2 to 1.3 years (mean 1.2 years).

Two of the IFN-B recipients had a total of four exacerbations since their first IFN-B injections. Six of the controls had a total of ten exacerbations since entering the study. The recipient group had significantly fewer exacerbations during the study than predicted by their pre-study exacerbation rates ($P < .01$), but the controls exacerbated as predicted. In seven of the recipients, one to six exacerbations were predicted, but none occurred. Although exacerbations did occur in recipients 3 and 5, the numbers of exacerbations were far less than predicted. No exacerbations occurred in recipient 9 during the study, in agreement with the prediction. Four controls

Table 1. Effects of intrathecally administered IFN-B on patients with MS.

Patient	Age (years)	Sex	Type of disease*	Duration of disease before study (years)	Exacerbations before study		Years on study	Exacerbations on study		Match‡	Exacerbation rate on study	Clinical assessment
					Number	Rate		Expected†	Actual number			
							Recipients					
1	20	F	ER-R	1	4	4	1.5	6.0	0	No (less)	0	Better
2	25	F	ER-P	6.2	14	2.3	1.6	3.6	0	No (less)	0	Better
3	29	F	ER-P	9.1	26	2.9	1.6	4.6	1	No (less)	0.6	Unchanged
4	40	F	S-R	8.1	10	1.2	1.5	1.9	0	No (less)	0	Better
5	26	F	ER-P	6.3	21	3.3	1.6	5.3	3	No (less)	1.9	Worse
6	29	M	S-R	10.3	6	0.6	1.6	1.0	0	No (less)	0	Better
7	29	F	ER-R	2.5	4	1.6	1.4	2.1	0	No (less)	0	Better
8	26	F	S-R	7.0	5	0.9	1.4	1.1	0	No (less)	0	Unchanged
9	39	M	S-R	19.4	8	0.4	1.4	0.6	0	Yes	0	Unchanged
10	39	F	S-R	10.3	8	0.8	1.4	1.1	0	No (less)	0	Unchanged
							Controls					
1	15	F	ER-P	3.2	4	1.3	1.2	1.5	3	No (more)	2.6	Worse
2	26	F	ER-R	2.8	3	1.1	1.2	1.3	1	Yes	0.9	Worse
3	39	F	S-R	5.5	3	0.7	1.2	0.8	1	Yes	0.9	Better
4	30	F	S-R	4.5	3	0.9	1.2	1.0	2	No (more)	1.7	Unchanged
5	39	M	S-R	20.5	4	0.2	1.2	0.2	0	Yes	0	Unchanged
6	31	F	ER-R	10.5	5	0.5	1.3	0.6	0	Yes	0	Worse
7	34	M	S-R	14.5	5	0.3	1.2	0.4	2	No (more)	1.7	Unchanged
8	26	F	S-R	6.5	5	0.8	1.3	1.0	0	No (less)	0	Worse
9	33	F	S-R	9.8	3	0.3	1.2	0.4	1	No (more)	0.9	Worse
10	38	F	S-R	7.7	6	0.8	1.1	0.9	0	Yes	0	Unchanged

*Type of disease when entering study: ER-R, exacerbating-remitting with residua; ER-P, exacerbating, progressive; S-R, stable with residua. †Determined by multiplying the exacerbation rate before the study by the number of years on the study. ‡Determined by comparing the expected exacerbation rate with the actual number of exacerbations on the study: "No (less)" indicates disagreement, actual number less than expected; "No (more)" indicates disagreement, actual number more than expected; "Yes" indicates agreement.

had more exacerbations during the study than predicted; one had fewer, and the other five controls exacerbated as predicted (Table 1). The overall exacerbation rates for the IFN-B recipients decreased during the study (mean rate before study, 1.8, as opposed to 0.25 on the study, $P < .01$), whereas the controls' rates increased overall (mean rate before study, 0.69, compared to 0.83 on the study; not significant). The recipients had higher exacerbation rates before ($P < .02$) and lower exacerbation rates during the study ($P < .05$) than the controls.

In recipients 1, 2, and 4 who had no exacerbations during the study, the exacerbation-free intervals since being on the study (1.5 to 1.6 years, mean 1.5 years) are the longest such intervals they ever experienced since disease onset (1.7 to 4.5, mean 3.1 times longer than longest remission before the study).

The clinical disability status scores were not significantly different in the two groups (recipients, 2 to 8, mean 5.3; controls, 3 to 7, mean 3.9) but the IFN-B group differed from the controls at the beginning of the study in having had more (2.6 times) exacerbations and containing 20 percent more patients with exacerbating-remitting diseases. The clinical impressions indicated that in five recipients and one control the disease had improved, in four recipients and four controls the disease was unchanged, and in one recipient and five controls the disease was worse ($P < .036$ for improvement in recipients, Mann-Whitney).

There were no exacerbations during the study in the five improved IFN-B recipients. One unchanged recipient (No. 3) had one exacerbation and the recipient in whom the disease had worsened (No. 5) had three exacerbations during the study. Exacerbations occurred in all three types of clinical respondents in the controls. The improved control (No. 3) had one exacerbation, two of the unchanged controls (Nos. 4 and 7) had two exacerbations each, and three of the controls in whom the disease had worsened had a total of five exacerbations during the study.

Headache was the most common side effect (experienced by all IFN-B recipients on at least one occasion) of the treatment, and usually began 6 to 12 hours after a lumbar puncture; however, sometimes a headache began immediately and sometimes up to 24 hours after the lumbar puncture. The usual duration of headache was 24 hours. Low-grade fevers (maximum 101°F) occurred in six IFN-B treated patients and began, on the average, 6 hours after lumbar punctures and lasted 6 to 12 hours. All IFN-B recipients developed transient CSF pleocytosis and increases in total protein, but there was no relation between the timing of maximum pleocytosis and the occurrence of systemic toxicity symptoms. Five recipients had detectable IFN-B (3 to 23 IRU) in the CSF on one to three occasions during the first month of treatment. There was no relation between the detection of IFN-B in the CSF and the clinical responses. No IFN was detected in the CSF or serum of the controls.

The IFN-B recipients showed a significant decrease in exacerbation rates during the study compared with their rates prior to the study, but the control group had statistically the same exacerbation rates before and during the study. The alterations in the IFN-B–treated patients' disease courses is further evidenced by the three in whom the remission periods since entering the study are 1.7 to 4.5 times longer than any prestudy remissions.

Our observations on the exacerbations in the IFN-B recipients are consistent with the idea that a virus present in the CNS of MS patients acts as a trigger for repeated exacerbations and that this trigger was suppressed or eradicated by the antiviral effects of the IFN-B (*1, 2*). Our observations are also consistent with a dysimmune etiology for MS such that overall augmentation of the immune system conferred by IFN-B stabilized a previously "oscillating" immune system that had been producing exacerbations (*2, 3, 13*). In this regard it has been demonstrated that a proportion of MS patients produce defective IFN responses to viruses and T-cell mitogens (*14*).

Most of the IFN-B recipients whose condition improved had exacerbating-remitting diseases for shorter durations with fewer exacerbations as well as lower disability status scores before the study than those recipients whose condition was unimproved or worse. Thus, it may be important to begin IFN-B treatment of MS patients as soon as possible after the disease is diagnosed while they have still had few exacerbations and their clinical disabilities remain relatively mild. The transient toxic reactions to IFN-B administered intrathecally were similar to those observed when it was administered systemically (*3*) and seem acceptable for the benefits achieved (*15*).

It could be argued that, for scientific reasons, the control patients should have received intrathecally administered foreign protein. We considered that the toxic effects of such administration were not justifiable for the purposes of this study. The possibility of a placebo effect in the IFN-B recipients is minimized by the consistency of the responses and the length of the period of observation.

LAWRENCE JACOBS
Dent Neurologic Institute, Millard Fillmore Hospital, Buffalo, New York 14209, and *Division of Neurobiology, Department of Physiology, State University of New York, School of Medicine, Buffalo 14214*
JUDITH O'MALLEY
ARNOLD FREEMAN
Roswell Park Memorial Institute, Buffalo, New York 14203
ROSLYN EKES
Dent Neurologic Institute, Millard Fillmore Hospital

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