Experimental therapies for multiple sclerosis: current status

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■ Multiple sclerosis is the most common cause of nontraumatic disability affecting young adults in the United States. Its etiology remains unclear, but evidence points to alteration of normal immune system function in the pathogenesis of this illness. This article reviews the results of clinical trials of experimental therapeutic agents in multiple sclerosis. The background and action of each treatment are described, and the clinical experience with each agent is reviewed. Obstacles to evaluating the effectiveness of treatment methods in MS and challenges to the design of future clinical trials and the interpretation of trial outcomes are discussed.

□ INDEX TERMS: MULTIPLE SCLEROSIS; AZATHIOPRINE; POLYMERS; CYCLOPHOSPHAMIDE; CYCLOSPORINS; INTERFERONS; PLASMAPHERESIS; LYMPHATIC IRRADIATION □ CLEVE CLIN J MED 1992; 59:63–74

Multiple sclerosis (MS) is the most common cause of nontraumatic disability affecting young adults in the United States. There is still no cure for this illness and current treatments for disease progression remain unsatisfactory. There are approximately 250,000 existing cases, and nearly 8,800 new cases are diagnosed annually. MS is characterized by a variable and unpredictable course and by a multiplicity of clinical manifestations. Only 15% to 20% of MS patients followed longitudinally will experience an exacerbation each year. After 2 years, approximately 50% of patients who were initially defined as chronic progressive will spontaneously stabilize, as will 20% of patients initially defined as relapsing.[1]

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Although the etiology of MS remains unclear, at least seven separate lines of evidence point to changes in normal immune system function.[2] These include: (1) intrathecal synthesis of immunoglobulins of restricted heterogeneity; (2) characteristic distribution of interleukins, interferons, tumor necrosis factor, and T cell subsets within active MS plaques; (3) association of disease frequency with certain human leukocyte antigen complex haplotypes; (4) coexistent systemic immune abnormalities consistent with immune activation; (5) decrease in suppressor cell numbers and function; (6) transient benefit seen with immunomodulatory therapy; and (7) clinical worsening seen with interferon gamma (IgG).

This article, the first of two on the current status of MS therapy, reviews the results of clinical trials of experimental therapeutic agents in MS. The treatments are presented one by one, in alphabetical order. For each treatment, the background and possible mechanisms of action are described, and the clinical experience with the agent is reviewed. This article

concludes with a discussion of obstacles to evaluating the effectiveness of treatment methods in MS and challenges to the design of future clinical trials and the interpretation of trial outcomes.*

A second article, to be published in an upcoming issue of this journal, focuses on pharmacotherapeutic agents for the day-to-day care of symptomatic problems experienced by MS patients.

AZATHIOPRINE TRIALS

Background

Azathioprine (AZA) is an orally administered purine analogue which, in the intestinal wall, is rapidly converted to 6-mercaptopurine, and subsequently into 6-thioinosinic acid. The latter compound is believed to be the active form; it selectively affects rapidly replicating cells by decreasing the rate of cell division. Full immunosuppressive action usually is not achieved before 3 to 6 months. AZA can react with sulfhydryl compounds such as glutathione and, thus, serves as a "pro drug," permitting the slow liberation of mercaptopurine in tissues. AZA achieves superior immunosuppression compared with mercaptopurine.[3] Both AZA and mercaptopurine are metabolized rapidly by oxidation or methylation in erythrocytes and the liver, and are excreted by the kidney.

The precise mechanism of the immunomodulatory action of AZA is uncertain. Several studies have shown that it suppresses cell-mediated hypersensitivity reactions and produces alterations in antibody production. AZA is also lympholytic and reduces the number of circulating natural killer cells. Studies of immune function in MS patients receiving AZA monotherapy have shown reduced pokeweed-mitogen-driven IgG secretion in vitro, reduced Con A suppressor activity in AZA-treated vs untreated MS patients, and reduction of the natural killer cell population and killer-cell-mediated antibody-dependent cellular cytotoxicity.[4]

Clinical experience

The first experience with AZA in MS was reported in 1969.[5] Based on its beneficial effect in renal homograft (allograft) recipients,[6] the rationale for using the drug was that the clinical course, the pathological pattern of the acute foci, and the finding of abnormal gamma globulins seemed similar to the rejection phenomenon seen in renal homotransplantation. In this initial study, six MS patients with relapsing progressive disease previously unresponsive to steroids were treated with oral AZA, 1.4 to 2.2 mg/kg/day. All patients experienced stabilization or slight improvement in neurologic status. Although the study design was inadequate by today's standards, it sparked interest in AZA: 21 clinical investigations of this drug were reported between 1971 and 1990.[7] Ten of the studies used properly matched controls, but only 4 were double-blind studies. The trials showed varied therapeutic efficacy, but all reported only limited toxicity. Early concerns that increased rates of malignancy are associated with AZA have not been confirmed, with the exception of a very slight increase in non-Hodgkin's lymphoma.[8]

Three randomized, double-blind, controlled trials of AZA were reported between 1988 and 1990. The largest of these was undertaken by the British and Dutch Multiple Sclerosis Azathioprine Trial Group.[9] In this study, 354 clinically definite relapsing and chronic progressive MS patients were randomized to receive either AZA 2.5 mg/kg/day or placebo in a double-blind trial. During a 3-year follow-up period, only small differences were seen between the groups, using widely accepted measures of disability. These measures included the Kurtzke Disability Status Scale[10] (DSS), a global measure of neurologic disability ranging from 0 (normal) to 10 (dead); and the Hauser Ambulation Index[11] (AI), a measure of ambulatory capability ranging from 0 (normal) to 9 (restricted to wheelchair and incapable of independent transfers). Though trends in deterioration which favored AZA were seen using the DSS, AI, and exacerbation rate (number of exacerbations per year), none were statistically significant. The differences became more noticeable with passing time, and the difference in mean deterioration seen with the AI became significant after 3 years. The authors concluded that the marginal benefits observed with AZA did not warrant routine use of the drug in MS, even though the observed toxicity was limited.

The second trial, by Ellison et al[12] in 1989, randomized 98 clinically definite chronic progressive MS patients to receive AZA treatment with methylprednisolone, AZA with placebo, or placebo and placebo. They were then evaluated longitudinally for 3 years in double-blind fashion. The initial AZA dose was 2.2 mg/kg/day, thereafter adjusted to maintain the white blood cell (WBC) count between 3,000 and 4,000 cells/μL. Methylprednisolone was administered in 1-g

*The reader may wish to consult *Treatment of Multiple Sclerosis: Trial Design, Results and Future Perspectives*, edited by Rudick and Goodkin (Springer-Verlag, in press).

pulses in 50 mL 5% dextrose in water over 30 minutes, and was given in the morning on the first 3 days of each of the first 3 months of treatment. Starting at the same time, a single oral morning dose of 96 mg methylprednisolone was administered on alternate days. During the second month, this was decreased to 72 mg every other day and, in the third month, to 48 mg every other day. Thereafter, methylprednisolone was decreased by 4 mg every 2 weeks and was stopped after a total of 36 weeks of treatment.

A statistically significant reduction in relapse rate was seen for all AZA patients who were followed for 3 years (AZA with methylprednisolone vs placebo $P = 0.03$, and AZA alone vs placebo $P = 0.04$). Progression of disability, as measured by the sum of Standardized Neurological Examination scores, was reduced in those patients in the group using AZA with methylprednisolone who completed the protocol exactly as intended ($P < 0.05$). Outcome assessments by examining and treating physicians and patients favored a therapeutic benefit for AZA compared with placebo. Though trends favoring AZA compared with placebo were seen for all patients followed for 3 years, the differences were not significant using the Standardized Neurological Examination, DSS, or Mickey's Illness Severity Scores[13] (a less widely accepted measure of neurologic disability in MS patients than the DSS). Significant hematologic and hepatic abnormalities were associated with AZA, but serious non-MS abnormalities were uncommon and were equally distributed among the three groups. The authors concluded that the benefits of AZA with or without steroids did not outweigh the risks, and so did not recommend AZA for patients with chronic progressive MS.

The most recent trial of AZA was reported by Goodkin et al in 1990.[14] Fifty-nine patients with clinically definite relapsing remitting MS were randomized to treatment with AZA 3.0 mg/kg/day vs placebo and then evaluated longitudinally for 2 years in double-blind fashion. AZA dose was adjusted to maintain the WBC count between 3,500 and 4,000 cells/μL. The primary outcome measures in this study were change in mean Kurtzke Expanded DSS[15] (EDSS) and change in exacerbation rate. A nonsignificant trend favoring AZA was seen for change in the mean EDSS score. A reduction in exacerbation rate was seen in the AZA group during the first ($P = 0.16$) and second years ($P = 0.05$) of the study. All nine secondary outcome measures showed a trend favoring AZA ($P = 0.02$). Two of these secondary measures showed statistically significant differences favoring AZA: time to deterioration in EDSS ($P = 0.04$) and AI ($P = 0.03$). Effective blinding of patients and physicians in this study was achieved. No severe or lasting toxicity was noted. The authors concluded that a modest therapeutic benefit was observed with AZA, and that the use of this drug in relapsing or chronic progressive MS patients should be decided by patient and physician on an individual basis after considering the risks and clarifying expectations for therapeutic benefit.

COPOLYMER 1 TRIALS

Background

Although the antigens responsible for inducing the production of autoreactive T cells in MS remain unidentified, experimental allergic encephalomyelitis (the animal model for MS) can be induced by immunization using myelin basic protein. It was therefore theorized that a polypeptide of similar structure might inhibit the immune response to myelin basic protein, and thus block its encephalitogenic action. In 1967, work began at The Weizmann Institute on a series of seven synthetic polypeptides which were created specifically to mimic myelin basic protein. One of these, Copolymer 1 (COP 1), was created by random polymerization of L-alanine, L-glutamic acid, L-lysine, and L-tyrosine in the ratio of 6.0 to 1.9 to 4.7 to 1.0.

COP 1 was not found to be encephalitogenic, and it actually suppressed experimental allergic encephalomyelitis in rabbits, guinea pigs, mice, and nonhuman primates without evident toxicity.[16] However, immunologic studies have not yet demonstrated cross-reactivity between COP 1 and myelin basic protein in humans.[17] Nonetheless, based upon preliminary work, a trial of COP 1 was organized using MS patients.

Clinical experience

The initial experience with COP 1 was reported by Bornstein et al.[18] They compared 25 patients with early exacerbating-remitting disease who were treated with COP 1 for 2 years against 23 control patients. The COP 1 patients had fewer relapses than did the controls, and some had less progressive disability. Though the decrease in relapse rate is desirable, this measure can be misleading since relapse rates in MS patients do not correlate strongly with accumulating disability (for example, patients can deteriorate gradually without exacerbations). The positive effects of COP 1 in terms of disability were restricted to patients with initial DSS of 0 to 2, ie, those with minimal impairment on

neurologic examinations. No positive effect on disability was seen for patients with an initial DSS ≥ 3.0, and the benefit was marginal when all patients were analyzed as a group. Additionally, the study may not have been completely blinded, since many of the COP 1 patients experienced erythema or soreness at their injection sites.

COP 1 has also been used in patients with chronic progressive MS. In a randomized, double-blind, placebo-controlled trial involving 106 patients with chronic progressive MS who were followed for 2 years,[19] a trend favoring COP 1 in time to progression of at least 1 point on the EDSS was evident, but it was not statistically significant. The percentages of patients in each group who worsened by at least one EDSS point likewise did not differ significantly. A multicenter trial of COP 1 in relapsing MS is scheduled to begin in September 1990 and results should be available in 1992.

CYCLOPHOSPHAMIDE TRIALS

Background

Cyclophosphamide (CTX) is an alkylating agent that possesses both cytotoxic and immunosuppressive properties. It was developed as a result of a specific effort to create an agent with greater neoplastic tissue selectivity than mechlorethamine, which was the first of the nitrogen mustards.[20] Its cytotoxic activity stems from its ability to disrupt fundamental mechanisms of cell growth, including mitotic activity and differentiation. Its immunosuppressive action is due to the preferential sensitivity of lymphocytes to this drug.

CTX is well absorbed orally and can also be administered intravenously (IV), intramuscularly (IM), intrapleurally, and intraperitoneally. In the liver, CTX drug is broken down to active metabolites, which are then transported to target sites by the circulatory system.

The rationale for using an immunosuppressive such as CTX in MS therapy is based in part on the perception that MS patients have abnormally high levels of immune system activation, arising perhaps from antigenic stimulation or from loss of normal immune system suppression. This hypothesis is supported by the finding that untreated MS patients have decreased numbers of CD4+CD45R+ (suppressor inducer) T cells and increased numbers of activated T cells in the blood, spinal fluid, and brain; increased numbers of CD4+CDw29+ (helper inducer) T cells in the spinal fluid and brain; and increased oligoclonal IgG production in the spinal fluid.[2]

CTX can normalize some of these findings in MS patients. For example, monthly intravenous CTX administration in doses ranging from 1,000 to 2,000 mg/m^2 results in a marked reduction of T helper/inducer cells and less striking decrease in suppressor/cytotoxic cells in MS patients.[21] These and other potentially beneficial effects of CTX last long enough to suggest practical therapeutic utility. Monthly pulses of CTX IV for 1 year induced reductions in numbers of suppressor/cytotoxic cells and associated natural killer cells, and lowered antibody-dependent cellular cytotoxicity functions. These effects lasted 1 to 2 months after cessation of therapy. Additionally, reduced numbers of B cells and FcR+ cells in these patients recovered to baseline abnormal values in 2 to 4 months after stopping therapy, and the return to abnormal values of helper cell subsets and total T cell numbers, helper/suppressor ratio, and proliferative responses to mitogens (phytohemagglutinin) took more than 4 months.[22] Others have found that decreased helper cell subset populations can still be found up to 13.5 years after discontinuation of IV CTX treatment (8 grams in 20 days).[23]

Clinical experience

In 1966, Aimard et al[24] described the first use of CTX in a single case of MS. The therapeutic benefit seen in this study led to an open uncontrolled trial in which 30 MS patients were treated with CTX 200 mg/day IV for 4 to 6 weeks. At the end of 2 years, 50% of the patients were either improved or stable.[25]

Several additional uncontrolled trials between 1968 and 1975 which used CTX with or without steroids had varying results. In 1975 Hommes et al[26] reported their experience with administration of a short intensive course of CTX (100 mg IV qid×20 days) combined with oral prednisone (50 mg bid × 20 days, followed by a 3 week taper) to 32 patients with chronic progressive MS. Patients were examined before and immediately after treatment, then again 6 to 33 months following treatment. Of 24 patients followed 6 months or more, 16 were improved, 7 unchanged, and 1 was worse, using a standardized neurological examination. The authors interpreted their results cautiously and concluded that only a double-blind trial of CTX and prednisone would determine whether a beneficial effect is seen in patients with chronic progressive MS.

In 1977 Gonsette et al[27] treated 110 relapsing MS patients with a short intensive course of CTX IV (sufficient to maintain a leukopenia of 2,000 cells/μL for 2 to 3 weeks: total dose 1 to 12 g). The study was not

controlled, randomized, or blinded. A reduction to the relapse rate recorded 2 years before treatment was reported. Two thirds of the patients had stabilization of their disability for 2 or more years. The benefits were confined to patients whose disease duration was less than 10 years.

In 1981 Theys et al reported a nonrandomized study[28] in which 21 CTX-treated MS patients with moderately advanced disability showed no difference in clinical course or disability after 2 years, compared to a retrospectively matched group of 21 untreated MS patients.

In 1983 Hauser et al[11] reported the results of a randomized, unblinded, controlled trial comparing various administrations of CTX and adrenocorticotropic hormone (ACTH) in chronic progressive MS patients. Three treatment groups were used: ACTH with a short, intensive course of CTX (400 to 500 mg/day IV in 4 divided doses until the WBC count fell to 4000 cells/μL); low dose oral CTX (2 mg/kg/day for 8 weeks) with ACTH and six plasma exchanges; and ACTH alone. Patients in each treatment group received ACTH, 25 units IV for 3 days followed by a reduction of 5 units every 3 days until day 16, at which time the dose was switched to 40 units IM tapered by 20 units every 3 days to 0 on day 22).

The group receiving the short, intensive course of CTX with ACTH sustained fewer treatment failures (worsening of disability) than either the group receiving low dose CTX with ACTH and plasma exchange ($P = 0.087$) or the group receiving ACTH alone ($P = 0.0004$). The authors noted that the benefits of this therapy lasted only 12 months, and thereafter 11 of the 16 stabilized patients began to regress. In addition to the expected hematologic toxicity, all patients treated with intravenous CTX experienced alopecia, and one third experienced severe nausea. The authors concluded that short-course intensive immunosuppression with intravenous CTX favorably influenced the course of chronic progressive MS.

The apparent limited duration of efficacy of CTX in chronic progressive MS led investigators to explore the possibility that maintenance boosters of CTX given subsequent to the above intensive induction program might confer longer-lasting benefit. In 1987, Goodkin et al[29] confirmed a significant benefit in chronic progressive MS patients 12 and 18 months following CTX induction, compared with untreated nonrandomized demographically similar controls. Following the induction period, patients were randomized to treatment with bimonthly maintenance boosters (700 mg/m^2 for 2 years) or no further CTX treatment. Patients receiving CTX maintenance boosters demonstrated a slight trend to greater stabilization at 12 ($P = 0.18$), 18 ($P = 0.16$), and 24 months ($P = 0.11$), but toxicity proved to be a major obstacle for these patients. Severe nausea and vomiting were experienced by all patients given maintenance boosters despite efforts to reduce this with parenteral antiemetics. Only half of the patients who stabilized would have considered continuing treatment after the 2-year study period. Preliminary data from a much larger recently reported study[30] indicate that bimonthly boosters as administered by Goodkin et al were associated with a significant improvement in stabilization rates for as long as 30 months in chronic progressive MS patients ($P = 0.032$).

Monthly intravenous CTX was administered to relapsing or remitting patients by Killian et al.[31] The CTX group (n=6) had fewer exacerbations than did the pacebo group (n=8) after 12 months of therapy (P=0.03). When each group served as their own controls, the CTX group had a significant decrease in exacerbations, but the placebo group did not. The results of this study should be interpreted cautiously, since exacerbations during the study were clearly defined but prestudy exacerbations were not.

The first single-blind, randomized, placebo-controlled trial of CTX was reported by Likosky et al in 1988.[32] In this study, 44 patients with clinical MS were randomized to treatment with intravenous CTX (400 to 500 mg, five times a week, until a WBC count of less than 4,000 cells/mL was achieved) or intravenous folic acid placebo (1 mg, five times a week) and assessed by blinded examining physicians at baseline, 12, 18, and 24 months. In contrast to prior unblinded studies, no concomitant ACTH or steroid was administered. At 12 months, changes in disability ratings (EDSS and AI) in the CTX and folic acid groups were very similar. Preliminary data from a double-blind CTX treatment trial reported by Noseworthy[33] failed to demonstrate any significant benefit for chronic progressive MS patients after 1 year of treatment with high-dose intravenous CTX and prednisone, daily oral CTX and alternate day oral prednisone, or placebo medication and sham plasmapheresis. The final results of this study are awaited with considerable interest.

In summary, recent data obtained from properly controlled, randomized, blinded studies of CTX in chronic progressive MS do not support earlier claims of efficacy, and enthusiasm for this therapy in MS has considerably lessened in the last year. In addition, con-

siderable patient discomfort due to nausea, vomiting, frequent hematological toxicity, and alopecia, and the threat of future infertility or malignancy are factors which must be taken into consideration when administering this medication.[34,35]

CYCLOSPORINE TRIALS

Background

Cyclosporine is a cyclic undecapeptide which was initially isolated from two soil fungi and recognized as an antifungal metabolite. This drug has proven effective in preventing host vs graft and graft vs host responses when used alone or in combination with other more conventional agents, and it has been reported to be useful in treating a variety of putative autoimmune diseases in man.[36] Interest in using this drug for human neurologic diseases quickly followed these initial reports.

Much work has been done in the attempt to clarify cyclosporine's mechanism of action. Many of its in vitro effects can be explained by the observed inhibition of the production of a number of lymphokines, including interleukin-2 (Il-2), interleukin-3, migration inhibitory factor, and gamma interferon.[37] Reduced levels of Il-2 messenger RNA inhibit Il-2 production,[38] and the same mechanism appears to inhibit other lymphokines.[39] However, cyclosporine appears to spare T lymphocytes that secrete a soluble factor which is critical for the expansion of nonspecific suppressor T cells. It appears possible that this T cell subpopulation belongs to the CD4+CD45R+ subset of T cells known as suppressor inducers.[40]

Clinical experience

Three major studies have assessed the efficacy of cyclosporine in MS. In the first of these, Kappos et al[41] reported the results of a double-blind, controlled trial of 194 patients with clinically definite active relapsing MS: 98 were randomized to treatment with cyclosporine (5 mg/kg/day), and 96 underwent treatment with AZA (2.5 mg/kg/day). Eighty-five patients in the cyclosporine group and 82 in the AZA group completed a treatment period of 24 to 32 months, in accordance with the study protocol. No significant changes were detected in EDSS, frequency of relapse, or overall treatment efficacy as assessed by patients and investigators at the end of the trial. Overall, only minor deterioration occurred in both groups during the trial. The incidence of side effects in the cyclosporine group was more than two times that in the AZA group (particularly hypertrichosis, gingival hyperplasia, paresthesias, elevated serum creatinine, and elevated blood pressure). The authors concluded that cyclosporine as a single agent could not be the drug of final choice in the long-term immunosuppressive treatment of relapsing MS.

The second study was a double-blind, placebo-controlled trial with patients in centers at London (N = 44) and Amsterdam (N = 38).[42] Participants in this study had either relapsing or chronic progressive clinically definite MS. The patients were begun on cyclosporine 10 mg/kg/day for 2 months, which was thereafter adjusted to minimize toxicity for the final 22 months of observation. The mean maintenance dose differed at the two sites (London, 7.2 mg/kg/day, and Amsterdam, 5.0 mg/kg/day). A variety of outcome measures including the EDSS were used. Investigators in Amsterdam concluded that no beneficial effects were seen and that side effects from cyclosporine presented a major problem.[43] However, the investigators at the London site separately reported a statistically significant early benefit for the patients treated with cyclosporine at that site. These patients had fewer relapses and a longer interval to first relapse on treatment over the 2-year study and better overall functional assessments for the first 6 months of treatment.[42]

The most recent study was a multicenter effort undertaken in the United States.[44] In this study, clinically definite moderately disabled (EDSS 3.0 to 7.0) MS patients were randomized to receive cyclosporine (N = 273) or placebo (N = 274) for at least 2 years. The dosage was adjusted for toxicity, resulting in trough whole-blood levels from 310 to 430 mg/mL. The mean worsening in EDSS score for cyclosporine-treated patients (0.39±1.07 points) was significantly less (P = 0.002) than for placebo-treated patients (0.65±1.08). Three primary efficacy criteria were used in this study: time to becoming wheelchair-bound, time to "sustained progression of disability", and a composite score of "activities of daily living." Cyclosporine treatment delayed patients' ultimate confinement to wheelchair (P = 0.038), but statistically significant effects were not observed for the other criteria.

Active treatment did have a favorable effect on several secondary measures of disease outcome. A large and differential withdrawal rate (cyclosporine = 44%, placebo = 32%) complicated the analysis but did not appear to explain the observed effect of cyclosporine in delaying time to wheelchair confinement. Nephrotoxicity and hypertension were common toxicities which accounted for most of the excess loss of patients

in the cyclosporine arm of the study. The authors concluded that cyclosporine was associated with a modest benefit in the chronic progressive MS patients in the study, but these benefits were not evident until 18 to 24 months after initiating therapy. This delay in measurable benefit and the high incidence of toxicity create practical limitations for using this drug, particularly for patients who are experiencing rapid functional deterioration. It is difficult to directly compare this study with the German multicenter study, since whole-blood trough levels and types of MS patients treated were significantly different.

INTERFERON TRIALS

Background

In 1957, Isaacs and Lindenmann[45] described a substance secreted by virus-exposed cells which "interfered with" viral replication and was accordingly termed "interferon." Since the original description, three classes of interferon (alfa, beta, and gamma) have been defined, initially by their antigenicity and subsequently by molecular cloning.

Though all three interferon classes possess antiviral activity, their other biologic activities have important differences. Interferon alfa and interferon beta are designated type I interferons. Human interferon alfa and beta genes are structurally similar, and the amino acid sequences of the various type I interferons are highly homologous. The human interferon alfa gene family, comprised of at least 24 genes and pseudogenes, and the single human interferon beta gene are both located on chromosome 9. The type I interferons utilize a common receptor. Reflecting their structural similarity, biological effects of the type I interferons are virtually indistinguishable in most assay systems. By contrast, interferon gamma, which is designated as a type II interferon, is encoded on a different chromosome, is structurally dissimilar to type I, acts through a different receptor, and has dramatically different biological activities in several assay systems.[46]

The immunologic activities of the interferons are expressed most prominently through macrophage activation, immunoglobulin synthesis, delayed-type hypersensitivity reactions, natural killer cell function, and major histocompatibility complex (MHC) antigen display. The regulation of these effects in vivo and in vitro by interferons depends critically upon the concentration of agent and timing of administration.[46] In some instances, type I and II interferons may exert opposing actions, as in the regulation of class II MHC expression. In particular, interferon gamma has been demonstrated to increase class II MHC expression by a wide variety of cell types, including astrocytes, endothelial cells, and monocyte/macrophages.[47] Interferon-gamma-induced class II MHC expression can be efficiently antagonized by interferon beta or alfa in some but not all of these cell types.[48-50]

The antiviral and immunomodulatory properties of interferon,[51] its limited toxicity with clinical use,[52] and reports of deficient production of interferon by cells isolated from MS patients[53] constitute the rationale for using interferons as experimental therapeutic agents in MS. Both recombinant (synthesized in cell cultures programmed with interferon genes) and natural (purified from cell cultures stimulated to produce interferon) reagents have been administered. The greater purity and unlimited availability of recombinant interferons have made them agents of choice for these applications.

Clinical experience

The first study of interferon alfa in MS was reported in 1984 by Knobler et al.[54] In this randomized, double-blind, placebo-controlled crossover study, 24 patients with frequent exacerbations were treated with either 5×10^6 IU of natural human interferon alfa or placebo daily for 6-month periods. A 6-month washout period followed each treatment. Exacerbation rates were reduced during interferon and placebo phases compared with pre-study rates.

A formal assessment of patient and physician blinding was not performed. However, patients who received interferon after placebo improved significantly more than those who received interferon before placebo, and the investigators concluded that the patients were able to deduce which course of treatment they had been given. This suggested that the blind had not been effective and that a "learning effect" could account for the benefits of interferon in this study. This underscores the critical importance of ensuring masking of treatments in interferon clinical trials.

Camenga et al[55] investigated the role of systemic recombinant human interferon alfa-2 in relapsing MS. Ninety-eight clinically definite, actively relapsing MS patients were admitted to this randomized, double-blind, placebo-controlled trial. Patients injected themselves with 2×10^8 IU of interferon alfa-2 or placebo three times each week for up to 52 weeks. No significant toxicity was noted. During the trial, exacerbation rates were reduced in both groups. In the 3-month period after stopping treatment, more interferon

recipients became worse neurologically than did those on placebo. More patients who had received interferon changed from exacerbating to progressive MS during the trial than did those on placebo. It is reassuring that this observation has not been confirmed by other investigators.

A more recent 3-year multicenter, prospective, double-blind, placebo-controlled trial of natural interferon alfa combined with transfer factor failed to demonstrate either benefit or deleterious effect in 182 relapsing and progressive MS patients.[55] Similarly, no benefit or worsening was observed with systemic lymphoblastoid interferon in chronic progressive MS.[56]

In theory, the effect of interferon beta treatment on the course of MS should not differ from that of other type I interferons. However, as noted above, the timing, dose, and route of administration may affect interferon action, and results of interferon beta clinical trials have been somewhat more encouraging than those obtained with interferon alfa. In particular, natural human interferon beta administered intrathecally appeared to reduce exacerbation rates MS in two therapeutic trials. The rationale for the intrathecal route of administration was concern that interferons might fail to cross the blood-brain barrier, since systemically administered interferon could not be detected in the cerebrospinal fluid.

An initial trial of intrathecal interferon beta utilized a randomized, controlled but unblinded study design in 20 patients.[57] Fewer exacerbations occurred in interferon recipients than controls, and exacerbation rates of recipients during the study were lower than prestudy rates. Subsequently, a randomized, double-blind trial of intrathecal interferon beta in MS using sham lumbar-puncture controls demonstrated a significant difference in exacerbation rates between treated and control patients, and the efficacy of the blinding technique was formally documented.[58,59] Interferon beta exhibits two properties that could help to explain this beneficial effect: it down-regulates interferon-gamma-induced class II MHC expression, and reverses the T lymphocyte suppressor cell defect observed consistently in MS.[47]

The aggregate results of these trials supported the notion that type I interferons could favorably modify the course of MS. However, the most encouraging results were obtained with a natural interferon beta preparation of limited availability administered via the cumbersome intrathecal route. Therefore, two multicenter trials of systemic recombinant human interferon beta have been undertaken. The peripheral route of administration could be justified, since sensitive assays for interferon-regulated gene expression has shown that systemic interferon clearly exerted biologic effects within the central nervous system compartment.[60,61] Although there are some differences in study design and dosage, it is anticipated that results of the two ongoing multicenter trials of interferon for relapsing-remitting MS will provide a definitive characterization of the efficacy of type I interferons for this indication.

A phase I trial of interferon gamma for MS produced a drastically disparate result from trials of type I interferons.[62] During one month in which intravenous recombinant human interferon gamma (dosage range 1.5×10^4 IU to 1.5×10^7 IU) was administered twice weekly to 18 patients with relapsing-remitting MS, exacerbations occurred in 7 patients in low-, intermediate-, and high-dosage groups. The study was discontinued, and it was proposed that interferon gamma be contraindicated for MS.[63] Administration of interferon gamma to these MS patients was associated with laboratory evidence of immune activation: increases in numbers of class II-MHC-positive circulating monocytes, augmented lymphocyte natural killer activity and enhanced lectin-driven proliferative responses.[64] These provocative observations strongly implicated the immunologic effects of interferon gamma in the pathogenesis of MS disease activity.

PLASMAPHERESIS TRIALS

Background

Plasmapheresis is effective in managing autoantibody-mediated diseases, including Guillain-Barré syndrome and myasthenia gravis, and it has been considered a potential therapy for MS because of these beneficial responses and the demonstration of increased amounts of immunoglobulin gamma in the brain tissue and cerebrospinal fluid of MS patients.

The rationale for plasmapheresis is based on assumptions that the pathogenic antibody is present in the vascular compartment, that it is capable of migrating from the vascular compartment to the central nervous system target site, and that removing it from the blood will therefore result in clinical improvement. Antibodies specific for myelin basic protein have been demonstrated in the cerebrospinal fluid of MS patients, but they have not been demonstrated in the blood. However, complement-fixing antibodies binding to saline extracts of brain tissue, as well as antimyelin and anti-oligodendroglial antibodies, have been found in the sera of MS patients.[65]

Clinical experience

The initial reports of plasmapheresis for MS appeared in 1980. In Dau's study,[66] seven of eight patients with progressive MS who were receiving long-term plasmapheresis together with azathioprine and pulsed prednisone demonstrated modest improvement of neurologic function. IgG levels in the cerebrospinal fluid decreased in six of seven of these patients. Whether this was attributable to plasmapheresis or to steroid administration was uncertain. Three additional patients who were in the midst of acute severe exacerbations refractory to prednisone made substantial recoveries. The improvement began within 3 or 4 exchanges, but seemed to plateau by the 10th exchange. One patient showed decreased latency of an abnormal somatosensory-evoked response within hours after completing plasmapheresis.

Weiner[67] reported improvement in either timed ambulation or upper extremity coordination in six of eight patients who underwent plasmapheresis. This improvement lasted 2 to 3 months in four patients and 6 months in two patients. Non-standardized treatment within this study limits the ability to draw conclusions from these data.

The first double-blind, controlled trial of plasmapheresis was reported by Gordon et al in 1985.[68] Twenty patients receiving prednisone 30 mg qod alternating with 5 mg qod and AZA 150 mg/day were randomized to undergo plasmapheresis or sham plasmapheresis three times weekly for 2 weeks, then twice weekly for the third week. Blinding was satisfactory, and patients in both treatment arms were well matched. Modest improvement was found on clinical examination at the conclusion of plasmapheresis in 7 of 10 plasmapheresis patients and in 3 of 10 control patients, but DSS score change was seen in only 1 plasmapheresis patient, and was no longer evident by 3 months.

Khatri et al[69] treated 54 chronic progressive randomized MS patients with plasmapheresis or sham plasmapheresis. In addition each patient received prednisone 1 mg/kg qod and oral CTX 1.5 mg/kg/day for 21 weeks, and pooled human serum immune globulin 40 mL over 2 days following treatment. Blinding was satisfactory and patient groups were well matched. Immunosuppressive therapy alone (control group, n = 29) resulted in DSS improvement in 8 patients at 5 months, which was sustained in 5 patients at 11 months. Three patients in this treatment arm were worse at 5 months and 6 were worse at 11 months. The plasmapheresis group (n = 26) demonstrated improvement in 14 patients at 5 months and 11 patients at 11 months. One patient was worse at 5 months and 3 were worse at 11 months. The difference between the treatment arms significantly favored plasmapheresis ($P = 0.007$). This study has been widely criticized because several of the patients who improved in the plasmapheresis treatment group did so by several DSS points, a finding which is unusual in chronic progressive MS patients. It has been speculated that some of the patients in this study who improved may have been recovering from exacerbations experienced just prior to starting therapy.

Weiner et al[70] recently completed a multicenter, randomized, double-blind, controlled trial to determine whether plasmapheresis is effective therapy for acute attacks. An 8-week course of 11 plasma exchange treatments was given to 116 patients who were experiencing exacerbations. Both plasmapheresis and control groups received identical treatment with intramuscular ACTH (tapered from 40 u IM bid over 14 days) and oral CTX (2 mg/kg/day for 12 weeks). Serum IgG decreased more in the plasmapheresis group at completion of the procedure than in the control group. Plasmapheresis-treated patients demonstrated greater improvement in DSS than the control group at 4 weeks and at 12 months after completing plasmapheresis, but this benefit was not seen after 4 weeks in the relapsing-remitting patients. The median time to recover to the baseline disability level was shorter in plasmapheresis patients than in controls. Numerous additional outcome measures were used, but the statistical analysis did not adjust for multiple outcome measures. Realizing this limitation, the authors suggested that the results be interpreted with considerable caution.

The role of plasmapheresis combined with immunosuppression for chronic progressive and relapsing MS patients remains controversial, and plasmapheresis as monotherapy has not yet been investigated. Given the limited benefits and the cost of this procedure, it seems unlikely that plasmapheresis will become a widely used therapy for MS.

TOTAL LYMPHOID IRRADIATION

Background

Total lymphoid irradiation (TLI) was initially developed to treat Hodgkin's disease and shortly thereafter was used in the treatment of rheumatoid arthritis.

The rationale for its use included its ability to induce a long-lasting suppression of T-cell immune responses in animals receiving skin grafts and bone

marrow transplants[71] and the absence of long-term sequelae, such as hematologic malignancies.[72] The beneficial effects seen with TLI in rheumatoid arthritis patients[73] resulted in its application to MS patients.

Clinical experience

Hafstein et al[74] administered TLI 2000 rad (200 Gy) or sham TLI in double-blind fashion to 19 randomized MS patients who had subjectively experienced chronic deterioration for at least 1 year prior to study entry. Patients were significantly disabled (DSS score 4 to 8) and had not received prior immunosuppressive therapy, but the randomization process did not yield demographically equal treatment groups. All patients underwent monthly evaluations for 6 months by the same neurologist. Trends favoring the irradiated group were seen using the DSS and muscle testing scale, but the differences were not statistically significant. An assessment of the blinding demonstrated that patients were able to correctly guess which treatment they received.

This study was later extended to include a total of 40 patients with an entry DSS score of 4 to 8 and disease duration of 3 or more years who were evaluated monthly for 6 months and thereafter at 3- month intervals for a total of 36 months.[75] The irradiated patients demonstrated a significantly better clinical course over the 2-year treatment period ($P < 0.05$) as measured by mean time to first sustained progression of disability at 6 and 24 months using a functional scale created by the investigators. Subgroup analysis showed that patients with total lymphocyte counts less than 900 for the first 3 months after treatment experienced better outcomes at all subsequent 3-month intervals up to 30 months following therapy.[76] Data related to progression of disability (eg, DSS, EDSS, or AI scores) were not provided in these reports. The blinding procedure was assessed as adequate "shortly" after treatment, but a reassessment of the blinding at 24 months demonstrated that patients were able to accurately guess which treatment they had received.

Toxicity from this treatment was initially reported as mild. Subsequently, however, three patients receiving TLI died of sepsis (Stuart Cook, personal communication). Three other instances of death due to sepsis in TLI recipients have appeared in the literature, all involving patients with rheumatoid arthritis.[72] Persistent lymphopenia and suppression of in vitro lymphocyte functions was evident in all patients and theoretically could have contributed to some of the deaths. The future of TLI therapeutic trials is uncertain at present.

ADDITIONAL CONSIDERATIONS

The problems of evaluating the effectiveness of treatment methods in MS result in large part from the variable and unpredictable course of the disease, and by its multiplicity of clinical manifestations. The design of future clinical trials and the interpretation of trial outcomes must take these factors into account.

Measures of disability or functional impairment may not accurately reflect underlying disease activity. Recent improvements in trial design, statistical methodology, diagnostic criteria, standardized neurological examinations, and other outcome measures have enabled clinical investigators to report more meaningful data. For example, serial magnetic resonance imaging and standardized neuropsychological testing can supplement traditional methodologies of monitoring disease activity.

The immunomodulatory therapies reviewed here show only minimal or modest benefits and are at times associated with troublesome or severe toxicity. Though these studies increase our understanding of how the immune system functions in MS, the future of global immunosuppression as a therapeutic strategy depends upon developing more effective and less toxic applications. Towards this end, we are currently investigating the role of less toxic agents, such as weekly administration of low-dose oral methotrexate in chronic progressive MS, and high-dose bimonthly intravenous pulses of methylprednisolone in relapsing MS.

The potential for developing specific, rather than global, immunomodulatory strategies increases with growing understanding of immunoregulatory mechanisms (including the characterization of the trimolecular complex, delineation of the roles of immunoactive cytokines, and genetic control of immune competence and disease susceptibility).

While we await the development of more successful interventional pharmacotherapy, we should not underestimate the importance of treatments which minimize functional impairment, such as physical and occupational therapy, supportive personal and family counselling, patient education, and use of medication which is effective in managing symptomatic complaints (bowel and bladder dysfunction, spasticity, pain, tremor, impotence, and fatigue).

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