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Murine and human studies on the immunology of Copaxone™ (copolymer 1)

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Copolymer 1 or Cop1 is the active ingredient of Copaxone™, a drug approved worldwide for the treatment of relapsing remitting multiple sclerosis (MS). Cop1 consists of the acetate salt of a synthetic copolymer of four naturally occurring amino acids, L-glutamic acid, L-lysine, L-alanine and L-tyrosine, with an excess of positive charge. Cop1 has been used in our laboratory since 1967 in the investigation of the MS animal model disease - experimental autoimmune encephalomyelitis - EAE. Cop1 was shown to suppress EAE induced by various encephalitogens [e.g. myelin basic protein (MBP), proteolipid protein (PLP) and myelin oligodendrocyte glycoprotein (MOG)] in several species including primates. The studies in experimental animals led to clinical studies culminating in the approval by the FDA in 1996 and since then in many other countries.

Extensive studies conducted during the last decade have characterized the immunological properties of Cop1.

a. Cop1 was found to be cross reactive with MBP both at the humoral level using mainly monoclonal antibodies, and at the cellular levels in both in vivo and in vitro assays.

b. Cop 1 exhibits a very rapid, high and efficient binding to different MHC class II haplotypes of murine and human origin. As a result Cop1 is capable of competing for binding with myelin antigens (MBP, PLP and MOG) and can efficiently displace them from the MHC binding site.

c. It has been demonstrated that Cop1 can competitively inhibit the immune response to myelin antigens of diverse antigen specific T cell lines of murine and human origin.

d. In vivo studies have demonstrated that Cop1 treated animals (either by subcutaneous injections or oral administration) develop Cop1 specific T suppressor (Ts) cells in the peripheral immune system. These cells can adoptively transfer protection to EAE and were characterized as Th2/3 type cells.

Recent studies have indicated that these cells can cross the blood-brain barrier and accumulate in the CNS, where the pathological processes of EAE and MS occur. This was demonstrated by their isolation from the CNS of actively sensitized mice, as well as, by their localization in the brain, after their passive transfer to the periphery (Fig. 1). The Cop1 specific T-cells, isolated from either spleens or brains, exhibited a confined Th2/Th3 response to both Cop1 and MBP, when stimulated on peripheral as well as on CNS originated antigen presenting cells. These results lend support to the notion that induction of Th2/3 Ts cells is the major important mechanism operating in vivo.

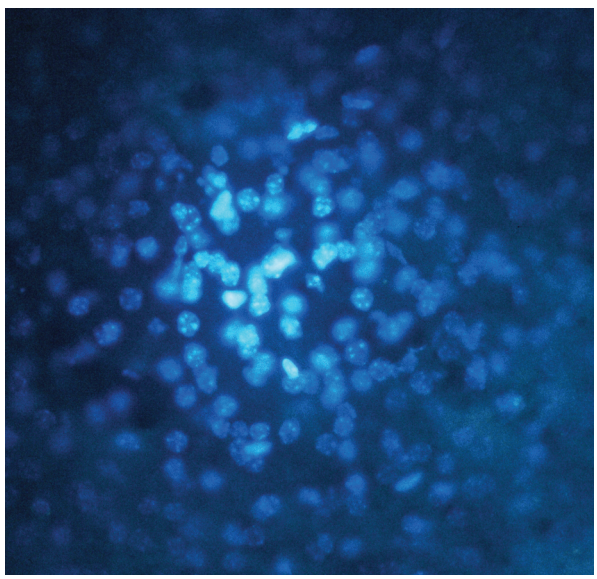

Fig. 1 *Brain section of mouse adoptively transferred with Cop 1-specific T cell. Activated Cop 1-specific cells were labeled with Hoechst dye and injected intaperitoneally to normal mice. After 7 days, brains were removed, fixed and sectioned.*

The clinical trials with Cop1 have enabled a more rigorous study on the in vivo immunological effects of Cop1 in MS patients. We followed the humoral and cellular immune responses in MS patients treated with Cop1, who participated in three different trials involving a total of 217 patients.

Antibody response

Evaluation of the immunological responses to Cop1 in MS patients revealed that all patients treated with Copaxone™ developed anti Cop1 antibodies, whereas placebo treated

patients were negative. The antibody level peaked at 3 months after initiation of treatment and reached a level of 8-20 fold above baseline. It decreased at 6 months and remained low. IgG1 levels were 2-3 fold higher than those of IgG2 at all time points examined. The anti Cop1 antibodies are non-neutralizing and they do not interfere with the therapeutic effect of Cop1 nor do they correlate with the reported side effects of Cop1.

T cell response to Cop1 in naïve MS patients

Several studies have demonstrated the presence of Cop1 reactive T cells in peripheral blood mononuclear cells (PBMC) of both untreated MS patients and normal individuals. The proliferative response to Cop1 in naïve MS and normal individuals could be inhibited by anti DR but not anti DQ antibodies, indicating that the proliferation induced by Cop1 is mediated by the T cell receptor (TCR) and is MHC class II restricted. Thus, there is compelling evidence that Cop1 is recognized as a conventional antigen and not as a mitogen or superantigen, and the response is a recall response to a commonly occurring antigenic determinant.

T cell response to Cop1 in treated MS patients

The proliferative responses to Cop1, MBP and PPD were followed up for two years in 86 patients participating in the phase III open-label study in Israel. Following an initial, slight increase, the response to Cop1 was markedly and gradually reduced as a function of time in trial. The proliferative response to MBP which was low at baseline, showed also a trend toward reduction with time of borderline significance. On the other hand, the response to the non-relevant antigen - PPD, which was high at baseline, did not change during the trial. The decline in the proliferative response to Cop1 may reflect an antigen induced cell death due to the repetitive stimulations, anergy or a shift to a Th2 type of response.

Thus, Cop1 elicited immunological reactions in MS patients that were modulated upon long-range treatment. The response to Cop1 does not interfere with its therapeutic activity nor can it induce any adverse reactions. These results are consistent with the findings that the therapeutic effects of Cop1 are maintained and enhanced upon long-term treatment.

Selected Publications

Aharoni, R, Teitelbaum, D, Sela, M. and Arnon R. (1997) Copolymer 1 induces T cells of the T helper type 2 that crossreact with myelin basic protein and suppress experimental autoimmune encephalomyelitis. Proc. Natl. Acad. Sci. USA. 94, 10821-10826.

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Aharoni, R, Teitelbaum, D, Arnon R. and Sela, M. (1999) Copolymer 1 acts against the immunodominant epitope 82-100 of myelin basic protein by T cell receptor antagonism in addition to major histocompatibility complex blocking. Proc. Natl. Acad. Sci. USA. 96, 634-639.

Teitelbaum, D., Arnon, R. and Sela, M. (1999) Immunomodulation of autoimmune encephalomyelitis by oral administration of Copolymer 1. Proc. Natl. Aad. Sci. USA 96, 3842-3847.

Aharoni, R., Teitelbaum, D., Leitner, O., Meshorer A., Sela, M. and Arnon, R. (2000) Specific Th2 cells are present in the central nervous system of mice protected against EAE by Copolymer 1. Proc. Natl. Acad. Sci. USA. 97, 11472-11477.

Brenner, T., Arnon, R., Sela, M. et al. (2001) Humoral and cellular immune responses to Copolymer 1 in multiple sclerosis patients treated with Copaxone. J. Neuroimmunol. 115, 152-160.