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Activities and characteristics of transfer factors

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Abstract

This report summarizes three components of our transfer factor research program. Several clinical studies have used oral administration of transfer factor containing materials. Sceptics have rejected these findings by assuming that the acidic and enzymatic environment of the gastrointestinal tract would destroy the factors. To further examine this issue, we have conducted dose-response studies of the delayed-type hypersensitivity reaction in mice that were given transfer factor either by gavage or subcutaneously. There were no difference in the responses that were related to the route of administration. We conclude that oral route of administration is efficacious and should be used when possible.

We have also studied the effects of transfer factors on immune responses by recipients. The details of this research are presented in the paper by Dr. Alvarez-Thull. Briefly, the study showed that recipients of a specific transfer factor responded to the antigen for which the factor was specific by secreting gamma-IFN, but no other cytokines.

The structures of transfer factor molecules are unknown. We have developed a process for isolating transfer factors in pure form and we have obtained preliminary data concerning amino acid sequences. Our goal is to obtain the complete primary structure of several transfer factor molecules.

Abbreviations: CMV: cytomegalovirus; HBSS: Hanks' balanced salt solution; IFN-g: gamma interferon; KLH: keyhole limpet hemocyanin.

There are many transfer factors

Several years ago our group [1,2] and Borkowsky and Lawrence [3] described affinity purification methods that enabled one to separate specific transfer factors from more complex mixtures such as whole leucocyte dialysates. Subsequently, this technique in association with HPLC and other chromatographic procedures has allowed purification of specific transfer factors with preservation of antigen specificity [4]. These observations provide conclusive evidence for the specificity of transfer factors and should put to rest alternative models for transfer factors such as those that involve adjuvant activity or non-specific effects on activation of cell-mediated immunity. It would seem appropriate to refer to transfer factors in publications.

Oral versus parental administration of transfer factors in clinical studies.

Most of the clinical trials with transfer factors have used parental administration [5–8].

An interesting feature of other studies has been oral administration of the transfer factors. Jeter and associates [9,10] described successful transfers of delayed-type hypersensitivity to tuberculin, coccidioidin and the contact sensitizer, 1-chloro–2,4-dinitrobenzene (CDNB), in humans who were fed the dialysable material from 1.5 x 10^9 lymphocytes from sensitized bovine donors. Burger and co-workers [11] transferred delayed-type hypersensitivity to keyhole limpet hemocyanin (KLH) to humans with orally administered transfer factor-containing dialysates from KLH-immune calves. The Herpes simplex patients treated by Viza and associates [7,12] were treated intramuscularly at entry into the study, but they subsequently received oral treatment during the maintenance stage.

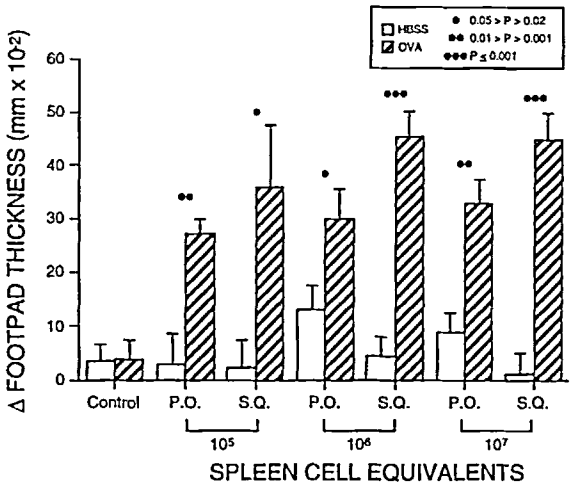

Figure 1. Delayed hypersensitivity responses in transfer factor recipients. Both the gavage and subcutaneous routes produced significant hypersensitivity in the recipients.

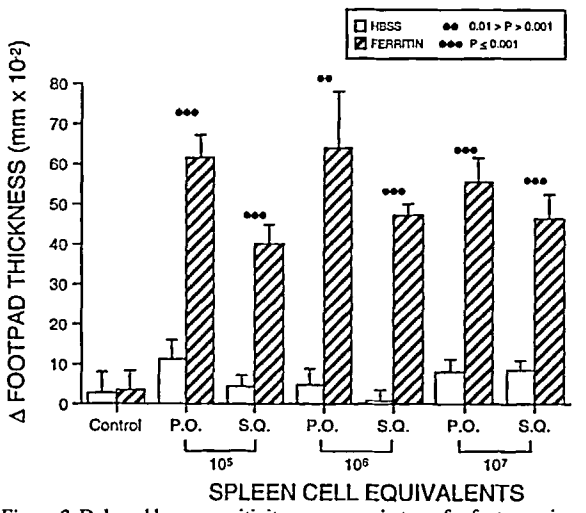

Figure 2. Delayed hypersensitivity responses in transfer factor recipients. Both the oral and subcutaneous routes of treatment produced equivalent sensitivity in the recipients.

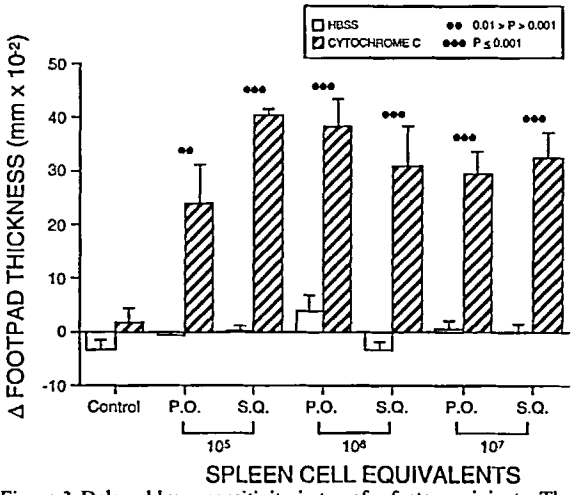

Figure 3. Delayed hypersensitivity in transfer factor recipients. The oral and subcutaneous routes of administration produced comparable degrees of delayed hypersensitivity in the recipients.

In a study of treatment of intestinal cryptosporiosis in patients with AIDS, only oral therapy was used [13].

As part of development of a procedure for isolating and purifying individual transfer factors, we devised calculations for units of activity and for specific activity [4]. Subsequently, we have used dose-response studies to compare cell-mediated immune responses by recipients of transfer factors that were administered either subcutaneously or by gavage. The results showed that there is little, if any, loss of transfer factor activity during oral administration.

Materials and methods

Transfer factors that were specific for ovalbumin (OVA), horse spleen ferritin or cytochrome c were prepared by sensitizing BALB/c mice with the antigen emulsified in Freund's complete adjuvant. Spleen cells were collected after three weeks. Single cell suspensions were prepared, the cells were lysed and dialysed and the dialysates were subjected to affinity extraction of the specific transfer factors [4]. The transfer factors were given to BALB/c mice either by gavage or by subcutaneous injection. Twenty-four hours later delayed hypersensitivity to the antigen was measured with the footpad swelling test [14]. Dose-response relationships were analysed by the analysis of variance.

Results and discussion

The results of these experiments are summarized in Figures 1–3. Briefly, the oral route of administration was effective in sensitizing recipients for expression of delayed-type hypersensitivity. There were no significant differences — assayed by ANOVA — in the footpad responses between recipients of subcutaneous or oral transfer factors, but in each experiment, the responses to the appropriate test antigen were significantly greater than the responses to the Hanks' balanced salt solution (HBSS).

These results virtually refute the arguments that the acidic or enzymatic environment of the gastrointestinal tract would prevent oral therapy with proteinaceous materials such as transfer factors.

As described above, orally-administered transfer factor preparations have been efficacious in clinical studies. In addition, Jones and co-workers [15] have reported a four-year-old male with persistent infections with both Epstein-Barr virus and cytomegalovirus (CMV). Oral administration of transfer factor from a calf that had delayed hypersensitivity to bovine rhinotracheitis virus transferred immunity to CMV as demonstrated by a positive lymphocyte transformation test. The patient was treated monthly for six months during which there was clinical improvement and chronic CMV viruria ceased. He remained well during the year after cessation of treatment.

Subsequently, Jones and associates [16] described beneficial effects of long-term oral treatment with bovine transfer factor in a patient with the hyper-IgE syndrome who had repeated staphylococcal infections and chronic oral candidiasis.

Recently, oral protein therapy has been evaluated in autoimmune diseases. Beneficial results have been described in some multiple sclerosis patients who received oral myelin basic protein [17] and rheumatoid arthritis patients who were fed type II collagen [18]. Taken together, the laboratory and clinical results justify the oral route of administration in clinical trials with transfer factors.

Effects of transfer factors on immune responses

The results of these experiments are presented in detail by Dr. Alvarez-Thull elsewhere in these Proceedings [19]. Briefly, the objective was to compare cytokine responses by spleen cells of mice that were sensitized to human Herpes simplex virus by infection or by administration of a Herpes simplex-specific transfer factor. It was found that sensitization by infection resulted in spleen cells that secreted IL–2 and IFN-g, but transfer factor therapy allowed recipients to produce IFN-g, but not IL– 2. No Th–2 cytokine production was detected.

Structure of transfer factors

For several years, our laboratory has been studying the chemical nature and structure of molecules with transfer factor activity. Thus far, our data indicate a proteinaceous structure. We have not been able to confirm the nucleoprotein or nucleopeptide structures that have been proposed by others [20–22]. We also propose that the primary structure of each transfer factor determines its specificity.

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