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PROSPECTS FOR A NONLIVING VACCINE AGAINST SCHISTOSOMIASIS BASED ON CELL-MEDIATED IMMUNE RESISTANCE MECHANISMS

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We have designed a vaccine model based on induction of cell-mediated immunity and shown that it protects mice against Schistosoma mansoni *infection. Mice are immunized by intradermal injection with schistosome antigens plus BCG. Resistance is dependent on the route of antigen presentation and the adjuvant chosen. The pattern of resistance correlates with sensitization of T lymphocytes for production of gamma interferon, a macrophage activating lymphokine that stimulates the cellular effector mechanism of protection. Purified schistosome paramyosin, a muscle cell component present in soluble parasite antigenic preparations, is immunogenic for T lymphocytes and induces resistance when given intradermally with BCG. It is likely that this protein, and possibly other soluble molecules that are released by the parasites of a challenge infection, induce a cellular inflamatory response resulting in larval trapping and/or killing by activated macrophages. These results verify the feasibility of a vaccine against schistosomiasis based on induction of cell-mediated immune resistance mechanisms.*

Because of recent advances in identification of chemically defined parasite antigens with protective potential in experimental animals, it is anticipated that a safe and effective vaccine may consist of a mixture of two or more defined antigens administered in a way that stimulates both humoral and cell-mediated immunity against subsequent infection.

It is only relatively recently that the possibility of a vaccine against schistosomiasis based on cell-mediated immune responses has been considered. The basis of protection under these conditions would be vaccine-induced sensitization of T lymphocytes to parasite antigens. Upon challenge infection, antigen-specific memory T cells would proliferate and produce lymphokines that stimulate the differentiation of other immune effector cells and their localization at the site of antigen presentation. Furthermore, certain lymphokines also activate these effector cells for more efficient killing of the invading parasites. For example, gamma interferon activates macrophages to kill newly transformed schistosomula as well as older (17 day) worms *in vitro* (James, 1986a; Pearce & James, 1985). Susceptibility of the parasites does not depend on recognition of a surface antigenic structure by the macrophages, as these cells can also kill unrelated targets such as tumor cells (James, 1986a). Schistosomes killed by lymphokine-activated macrophages show complete disorganization of internal structure, while retaining an intact-appearing tegumental membrane (McLaren & James, 1985).

There are several theoretical advantages to a vaccine against schistosomiasis based on cell-mediated immunity. Among these are: a) the fact that macrophage-mediated killing is not an antigen-specific event, and thus is not deterred by the parasite's immune evasion strategies of antigenic masking or shedding, b) the localization of immune response at the site of antigen stimulation due to the short half-life of most lymphokines, thus minimizing systemic pathology, c) the fact that any parasite molecule to which the correct subset of T cells is sensitized and which is expressed by the challenge parasites at a time when they are vulnerable to activated macrophages can theoretically function as a protective antigen, so that soluble as well as surface antigens may be protective, and finally d) the tendency of T cells to recognize protein rather than carbo-

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hydrate antigens, and primary or secondary structure rather than conformational determinants due to the requirement for antigen processing and presentation in the context of molecules of the major histocompatibility complex, suggesting that T cell epitopes would easily synthesized.

RESULTS

We have designed a vaccine model based on induction of cell-mediated immunity and shown that it protects mice against *Schistosoma mansoni* infection (James, 1985; James, 1986b). In this model, immunization with crude schistosome antigens plus *Mycobacterium bovis*, strain BCG, by the intradermal (i.d.) or subcutaneous route results in approximately 50% resistance to challenge infection. Intravenous or intramuscular injection with the same antigens is ineffective. Pertussis vaccine and saponin are also functional adjuvants in this system, but *C. parvum* and alum do not enhance protective immunity. This pattern of response correlates with sensitization of T lymphocytes for production of gamma interferon (James, 1986b). Intradermal immunization can protect B cell-deficient μ-suppressed mice and strains with defects in aspects of immediate hypersensitivity, but not T cell deficient nude mice or strains with defects in macrophage activation, verifying the T cell-mediated basis of resistance (James & DeBlois, 1986). Soluble supernatant fluids from homogenized schistosomula or adult worms protect as well as do whole disrupted schistosomula (James et al., 1985). Paramyosin, a myofibrillar protein that is a component of invertebrate muscle structure and is present in these soluble antigenic preparations, has been identified as a protective antigen in this model (Pearce et al., 1986; Lanar et al., 1986). This molecule is uniquely recognized by antibodies from mice immunized by i. d. injection of crude antigen preparations plus BCG, and stimulates T cells from these animals to proliferate and produce gamma interferon. Moreover, paramyosin purified to homogeneity by either affinity chromatography or salt fractionation and administered i. d. with BCG can protect mice against *S. mansoni* infection (Pearce et al., 1988). It is envisioned that parasites of the challenge infection release paramyosin as an excretory/secretory factor, eliciting a localized cell-mediated inflammatory reaction resulting in parasite trapping and/or killing by activated macrophages. This hypothesis is substantiated by the observation that culture supernatants from living schistosomula contain paramyosin epitopes in a quantity and form capable of stimulating T cells from mice immunized with the native molecule to produce gamma interferon. The possibility remains that other parasite antigens are also capable of contributing to a protective cell-mediated immune response, since soluble adult worm antigen preparations depleted of paramyosin by affinity chromatography continue to induce resistance when administered i.d. with BCG. The nature of other protective soluble antigens is currently under investigation.

DISCUSSION

These studies show that cell-mediated immune mechanisms can be protective against schistosome infection *in vivo*, and therefore that induction of cell-mediated immunity could be a useful facet of any eventual vaccine against human schistosomiasis. Moreover, they indicate that soluble molecules, such as parasite excretory/secretory factors, can function as protective antigens by stimulating cell-mediated immune response. Finally, the results described here emphasize the contribution of the appropriate delivery system to the protective efficacy of a given antigen, since the route of administration and adjuvant chosen can profoundly effect the type of immune response that is invoked. It is likely that different antigens can be protective in the context of different immune mechanisms, and that a successful vaccine will require a combined approach.

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