

Zeitschrift: Acta Tropica
Herausgeber: Schweizerisches Tropeninstitut (Basel)
Band: 44 (1987)
Heft: (12): Prospects for immunological intervention in human schistosomiasis

Artikel: Immunization with schistosome membrane antigens
Autor: Harn, A.
DOI: <https://doi.org/10.5169/seals-313840>

Nutzungsbedingungen

Die ETH-Bibliothek ist die Anbieterin der digitalisierten Zeitschriften. Sie besitzt keine Urheberrechte an den Zeitschriften und ist nicht verantwortlich für deren Inhalte. Die Rechte liegen in der Regel bei den Herausgebern beziehungsweise den externen Rechteinhabern. [Siehe Rechtliche Hinweise.](#)

Conditions d'utilisation

L'ETH Library est le fournisseur des revues numérisées. Elle ne détient aucun droit d'auteur sur les revues et n'est pas responsable de leur contenu. En règle générale, les droits sont détenus par les éditeurs ou les détenteurs de droits externes. [Voir Informations légales.](#)

Terms of use

The ETH Library is the provider of the digitised journals. It does not own any copyrights to the journals and is not responsible for their content. The rights usually lie with the publishers or the external rights holders. [See Legal notice.](#)

Download PDF: 08.01.2025

ETH-Bibliothek Zürich, E-Periodica, <https://www.e-periodica.ch>

Department of Medicine, Robert B. Brigham Hospital, Boston, Massachusetts, USA

Immunization with schistosome membrane antigens

A. HARN

In schistosomiasis, animals harboring experimental, natural infections or those immunized with irradiated cercariae, develop significant levels of resistance to challenge infection [1–4]. The surface membranes of cercariae and early schistosomula have been considered to be the major targets of protective host mechanisms responsible for resistance to challenge infection. This view has been based on many *in vitro* observations which demonstrated that unlike lung or adult worms, young schistosomula are susceptible to a wide variety of immune effector mechanisms [5, 6].

Therefore, studies on the detection and characterization of surface membrane antigens present on cercariae and developing schistosomula were initiated by many different groups. To characterize these antigens, our laboratory and several others, have produced and selected monoclonal antibodies which bind to the surface membranes of cercariae and/or developing schistosomula, and which also are able to passively transfer resistance to cercarial challenge in naive animals [7–15].

Protective monoclonal antibodies have been produced with both rat and mouse lymphocytes. The level of protection in passive transfer experiments has ranged from 30–45% in mice and as high as 60% in rats. These monoclonal antibodies have been produced from animals harboring natural infections [7–9, 13], immunized with membrane enriched extracts of cercariae or schistosomula [11, 12, 14, 15], or immunized with eggs or attenuated larvae [10, 15].

To date, monoclonal antibodies which protect *in vivo* have described surface membrane antigens at the following molecular weights: 38 kD [8]; 160 and 130 kD [9, 10]; 155 kD [16]; 28 kD [11]; 22 kD [12]; 30, 45 kD [13]; 32 kD and 17–18 kD [15].

Additionally, it should be noted that a number of surface membrane antigens have been detected by monoclonal antibodies which may be effective *in vitro* but do not passively transfer immunity *in vivo* [17–22].

The purification of putatively protective antigens has been accomplished

Correspondence: Dr. Donald A. Harn, Department of Medicine, Robert B. Brigham Hospital, The Seeley G. Mudd Building, 250 Longwood Avenue, Boston, MA 02115, USA

using the monoclonal antibodies in an affinity column, or by excising the antigen from a polyacrylamide gel after SDS-PAGE [23] separation of a parasite extract. In the latter method, it is then possible to immunize with the antigen in the polyacrylamide, or it may be eluted from the gel. The purity of the antigens and the relative yield may be estimated by SDS-PAGE.

Using an immunoaffinity column with protective monoclonal antibodies as ligands, Smith and Clegg [16] first reported on the isolation of membrane antigens from both adult worms and schistosomulae. The antigens had molecular weights of 155 kD and 53 kD as determined by Western blot analysis [24]. Trial vaccination studies with the purified antigens were administered intramuscularly in alhydrogel, and the animals were challenged with cercariae two weeks after the boost. Immunized mice had a statistically significant reduction in adult worms, 26%, when compared to sham immunized controls. The levels of protection ranged from 0.0% to 64%. In the only experiment with monkeys, the three which were immunized with the 155 kD antigen had adult worm burdens reduced by 30% when compared to three sham immunized controls.

In the only other published report which I am aware of, Hazdai et al. [13] immunized mice with purified antigen from cercariae. The antigen was initially administered with complete Freund's adjuvant, however no adjuvant was used in the boost. In this experiment mice were challenged with cercariae and the immunized group showed a significant level of protection which was apparently dose dependent. Animals which received 5 μ g of antigen per injection had a 34% reduction in adult worm burden while mice which received only 1.5 μ g of antigen had a 17% reduction in adult worms.

Our laboratory has been involved with the purification of three distinct membrane antigens from schistosomula. The molecular weights as determined by Western blots and immunoprecipitations with Bolton-Hunter [25] radiolabeled samples are: 22 kD, 28 kD, and 160 kD. Both the 22 kD and the 28 kD antigens can be purified from a membrane enriched deoxycholate extract of schistosomula [10] by immunoaffinity chromatography. However, the relative yields of antigen obtained in this manner are low. We estimate, based on silver staining, that we are able to obtain 5 μ g of pure 22 kD and 8 μ g of pure 28 kD per 100,000 schistosomula extracted. Thus we initiated trial vaccinations with the 22 kD antigen using small quantities of antigen.

We immunized naive mice intradermally with approximately 50 ng of pure antigen mixed with BCG as described by James et al. [26]. Additionally, we used maalox at a 1:1 volume for volume ratio. Mice were boosted 4 times at three week intervals. Four weeks after the last boost, mice were challenged with cercariae by belly penetration. The immunized mice had adult worm burdens reduce 27% $p < 0.01$ compared to sham immunized controls. In subsequent experiments where fewer boosts were given, the protection level dropped to 10–17%. However, the antigen did appear to invoke a memory response as the animals were challenged 4 weeks after the last boost.

We next immunized mice with purified 28 kD antigen. For these experiments we immunized mice with immune complexes. The complexes consisted of protein A-sepharose beads to which we bound our anti-28 kD antibody. Antigen was next bound to the beads, and the beads were injected subcutaneously. The amount of antigen injected was estimated at 5 μ g per mouse. The mice were boosted 4 weeks after the prime. We waited 4 weeks after the boost before we challenged the vaccinated mice with cercariae. Six weeks post-infection, adult worms were harvested. The mice vaccinated with the 28 kD antigen had adult worm burdens reduced 38% compared to sham immunized controls.

The last antigen which we are working with is the antigen detected by our protective anti-egg monoclonal antibody [10]. The amount of antigen in the schistosomula extracts is small compared to what we can get from soluble egg antigen, so we have performed these experiments with cross-reactive egg antigen. We primed and boosted naive mice with approximately 50–100 ng of pure antigen. The antigen was mixed with BCG and maalox as described earlier. Mice were immunized intradermally and intramuscularly. Four weeks after the boost the mice were challenged with cercariae. The vaccinated mice had adult worm burdens reduced 37% when compared to sham immunized controls.

Although these are preliminary results, they are encouraging. When taken together with the two published studies, they clearly demonstrate that defined antigens can be used to vaccinate naive mice. Our studies also indicate that it is possible to induce a memory response.

Because these preliminary studies have been successful, we are optimistic that future research concerning optimal antigen doses, the different types of adjuvants, and even combinations of defined antigens will give rise to far superior vaccines. When considering studies where multiple antigens may be employed, it should be noted that all of the studies other than with the 22 kD antigen which I have mentioned are early stage antigens. Thus it is likely a vaccine which would combine epitopes from early and late stages may achieve protection levels as high as 80–90%.

- 1 Stirewalt M. A.: The influence of previous infection of mice with *Schistosoma mansoni* on a challenging infection with the homologous parasite. *Amer. J. trop. Med. Hyg.* 2, 867 (1953).
- 2 Hunter G. W., Crandall R. B., Zickafoose D. E., Purvis Q. B.: Studies on schistosomiasis. XVIII. Some factors affecting resistance to *Schistosoma mansoni* infection in albino mice. *Amer. J. trop. Med. Hyg.* 11, 17 (1962).
- 3 Minard P., Dean D. A., Jacobsen R. H., Vannier W. E., Murrell K. D.: Immunization of mice with cobalt-60 irradiated *Schistosoma mansoni* cercariae. *Amer. J. trop. Med. Hyg.* 27, 76 (1978).
- 4 Eveland L. K., Morse S. E.: *Schistosoma mansoni*: infectivity and immunising effects of in vitro derived schistosomula attenuated by X irradiation. *Exp. Parasit.* 45, 19 (1978).
- 5 Butterworth A. E., Taylor D. W., Veith m. C., Vadas M. A., Dessein A., Sturrock R. F., Wells E.: Studies on the mechanisms of immunity in human schistosomiasis. *Immunol. Rev.* 61, 5 (1982).
- 6 Capron A., Dessaint J. P., Capron M., Joseph M., Torpier G.: *Immunol. Rev.* 61, 41 (1982).
- 7 Smith M. A., Clegg J. A., Snary D., Trzidowicz A. J.: Passive immunization of mice against *S. mansoni* with an IgM monoclonal antibody. *Parasitology* 84, 83 (1982).

- 8 Dissous C., Grzych J.-M., Capron A.: *Schistosoma mansoni* surface antigen defined by a rat monoclonal IgG2_a. J. Immunol. 129 (5), 2232 (1982).
- 9 Zodda D., Phillips S. M.: Monoclonal antibody-mediated protection against *Schistosoma mansoni* infection in mice. J. Immunol. 129, 2326 (1983).
- 10 Harn D. A., Mitsuyama M., David J. R.: *Schistosoma mansoni*: Anti-egg monoclonal antibodies protect against cercarial challenge in vivo. J. exp. Med. 159, 1371 (1984).
- 11 Harn D. A., Mitsuyama M., Huguenel E. D., Oligino L., David J. R.: Identification by monoclonal antibody of a major (28 kDa) surface membrane antigen of *Schistosoma mansoni*. Mol. Biochem. Parasit. 16, 345 (1985).
- 12 Harn D. A., Mitsuyama M., Huguenel E. D., David J. R.: *Schistosoma mansoni*: detection by monoclonal antibody of a 22,000-dalton surface membrane antigen which may be blocked by host molecules on lung stage parasites. J. Immunol. 135, 2115 (1985).
- 13 Hazdai R. T., Levi-Schaffer F., Brenner V., Horowitz S., Eshhar Z., Arnon R.: Protective monoclonal antibody against *Schistosoma mansoni*: antigen isolation, characterization, and suitability for active immunization. J. Immunol. 135, 2772 (1985).
- 14 Horowitz S., Brenner V., Arnon R.: In vivo protection against *S. mansoni* infection by monoclonal antibodies. Ummunol. Lett. 9, 69 (1985).
- 15 Bickle Q. D., Andrews B. J., Raylor M. G.: *Schistosoma mansoni*: characterization of two protective monoclonals. Parasite Immunol. 8, 95 (1986).
- 16 Smith M. A., Clegg J. A.: Vaccination against *Schistosoma mansoni* with purified surface antigens. Science 227, 535 (1985).
- 17 Taylor D. W., Butterworth A. E.: Monoclonal antibodies against surface antigens of schistosomula of *Schistosoma mansoni*. Parasitology 84, 65 (1982).
- 18 Strand M., McMillan A., Pan X. Q.: *Schistosoma mansoni*: reactivity with infected human serum and monoclonal antibody characterization of glycoprotein in different developmental stages. Exp. Parasit. 54, 145 (1982).
- 19 Aronstein W. S., Norden A. P., Strand M.: Tegumental expression of a major schistosome structural glycoprotein. Amer. J. trop. Med. Hyg. 32, 334 (1983).
- 20 Aronstein W. S., Strand M.: Lung stage expression of a major schistosome surface antigen. J. Parasit. 69, 1027 (1983).
- 21 Norden A. P., Aronstein W. S., Strand M.: *Schistosoma mansoni*: identification, characterization and purification of the spine glycoprotein by monoclonal antibody. Exp. Parasit. 54, 432 (1982).
- 22 Tavares C. A. P., DeRossi R., Payares G., Simpson A. J. G., McLaren D. J., Smithers S. R.: A monoclonal antibody raised against adult *Schistosoma mansoni* which recognizes a surface antigen on schistosomula. Z. Parasitenk. 70, 189 (1984).
- 23 Laemmli U. K.: Cleavage of structural proteins during the assembly of the head of bacteriophage T4. Nature (Lond.) 227, 680 (1971).
- 24 Towbin H. T., Staehlin T., Gordon J.: Electrophoretic transfer of proteins from polyacrylamide gels to nitrocellulose sheets: procedure and some applications. Proc. Nat. Acad. Sci. 76, 4350 (1979).
- 25 Bolton A. G., Hunter W. J.: The labeling of proteins to high specific radioactivities by conjugation to a 125I-containing acylating agent. Biochem. J. 133, 529 (1973).
- 26 James S. L., Pearce E. J., Sher A.: Induction of protective immunity against *Schistosoma mansoni* by a non-living vaccine. I. Partial characterization of antigens recognized by antibodies from mice immunized with soluble schistosome extracts. J. Immunol. 134, 3432 (1985).