on the molecular biology of the *E. histolytica* cell. Egbert Tannich and his colleagues (Bernard Nocht Institute for Tropical Medicine, Hamburg, Germany) and Juan Engel (University of California, San Francisco, USA) had observed significant variations in the pattern of pulsed-field gel electrophoresis (PFGE) of *Entamoeba* DNA during manipulation of the organisms in culture. Engel had observed differences in both the position and copy number of protease genes when clones of HM1 strain *E. histolytica* were analysed by field inversion gel electrophoresis. Tannich observed that some bands seen on PFGE were Exol-S1 nuclease-resistant, and his group isolated DNA from these individual bands for rotary shadowing and electron microscopy. To his surprise, several of these bands contained DNA molecules that were clearly circular! Aside from these variations in 'chromosome' linearity or size, how much true genetic rearrangement occurs in *E. histolytica*? The isolation by several laboratories (John Samuelson, Harvard School of Public Health, Boston, MA, USA; William Petri, University of Virginia Medical Center, Charlottesville, VA, USA; Jonathan Ravdin, VA Medical Center, Cleveland, OH, USA; Bruce Torian, Idaho State University, Pocatello, ID, USA; Samuel Stanley, Washington University School of Medicine, St. Louis, MO, USA, and others) of individual *E. histolytica* genes may now allow some genetic linkage groups to be established and this key question addressed. Several speakers (Orozco, Tannich, McKerrow) also confirmed that gene amplification is common in *E. histolytica*.

Subunit Vaccines

As is the case with several other major parasitic diseases, interest has grown in the potential for development of a subunit vaccine for amebiasis. Several candidate antigens were discussed, including the galactose-inhibitable adherence protein (William Petri and Barbara Mann, University of Virginia, USA; and Jonathan Ravdin), and a serine-rich surface molecule (Samuel Stanley), to name just two. While a promising avenue of investigation, work on immunoprophylaxis is just beginning, and the surprises and problems encountered in vaccine development for malaria and schistosomiasis may still be on the horizon.

Final Comment

While all the work presented at this symposium cannot be summarized in this short review, a final comment on the growth of cell biology studies on *Entamoeba histolytica* is worth making. The past five years have seen an explosion in our knowledge of the molecular biology of the eukaryotic cell, and few organisms are so fascinating and yet so mysterious in their cell biology as *Entamoeba histolytica*. Martha Espinosa Cantellano presented work from Adolfo Martinez-Palomo's group at CINVESTAV-IPN, Mexico City. New cryofixation techniques are allowing unparalleled visualization of the fine structure of *Entamoeba*, and ultrastructural analysis suggests many promising avenues for research, including the molecular mechanisms of endocytosis and motility. Isaura Meza and her group, also at CINVESTAV-IPN, have begun to unravel the events that lead to movement of trophozoites on an extracellular matrix. An elegantly orchestrated series of changes in the content and organization of polymerized actin, changes in cell shape and the release of proteases is envisioned as contributing to the 'footprint' of trophozoites as they adhere, release and move on a tissue matrix.

Acknowledgements
The Fogarty International Center Symposium on Host–Parasite Relationships in Amebiasis was held in Bethesda, MD, USA, 8–10 September 1993. This meeting should certainly be viewed as a call to young (and not so young) investigators to look anew at *Entamoeba histolytica* as an exciting focus of research in parasitology.

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A Perspective on Schistosomiasis Research

P. Hagan and B. Gryseels

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Ten years ago, the Commission of the European Communities established the Life Sciences and Technologies for Developing Countries (STD) Programme, addressing health problems relevant to developing countries. The aim was to co-ordinate the research efforts of European scientists and their counterparts in developing countries, with a view to establishing genuine partnerships that would tackle the complex problems presented by, among others, the major endemic parasitic diseases. For schistosomiasis, this vision of an integrated research programme has now become a reality. STD-supported scientists from 20 countries gathered in Noordwijk to report on the latest progress on their different projects, to reinforce existing links, to establish new links and to decide on future directions.

Epidemic Schistosomiasis

There have been few opportunities that have allowed the study of schistosomiasis as an epidemic disease. The outbreak of *Schistosoma mansoni* infection in Richard Toll, Senegal has provided an opportunity for the study of the relationships between some of the major factors that are thought to influence the acquisition of infection (B. Gryseels*, University of Leiden, The Netherlands). In particular, the importance of age, exposure and immunity can be investigated, since individuals across the age spectrum are having their first exposures to infection. The first case of schistosomiasis mansoni was reported in Richard Toll in 1988, with the prevalence of infection rising to 76% in 1990. Near by, in the smaller

*Only project co-ordinators are listed, throughout.

community of Ndombo, prevalence of infection in those above five years of age is 100% and such high prevalences are maintained across the adult age range. Infection intensities as high as 26000 eggs per gram of faeces have been recorded. In addition, 41% of the population excrete more than 1000 eggs per gram of faeces. One surprising finding was that the characteristic decline in egg counts and worm loads (as confirmed by antigen levels) seen in older age groups within established endemic foci was also reported within the study cohorts in Ndombo. Four staggered study cohorts, each a random sample of the community, are now being followed up parasitologically, immunologically and for exposure, before and after treatment. There was some evidence of the early and rapid development of immune responses but the results have to be interpreted in the light of information on exposure to infection and seasonal transmission. The relative inefficiency of treatment with praziquantel in people participating in this study (with a cure rate of only 18% at 12 weeks after treatment) had been a matter of some concern. Obviously, the susceptibility of the parasite to praziquantel has to be considered. A more-likely explanation was very rapid re-infection (including maturation of prepatent infections) and/or a reduced synergy between praziquantel and the immune response in this new focus. The poor cure rate with praziquantel was confirmed through the application of the circulating anodic and circulating cathodic antigens. These are being used for the first time as a routine epidemiological tool to monitor the dynamics of infection in the community. Ultrasonography showed there was, as yet, no evidence of hepatosplenic involvement, which may be due to the recent history of infection. Intestinal morbidity was highly prevalent but could not be unequivocally attributed to schistosome infection. Transmission is intense and seasonal; the peculiar features of this focus which have permitted this epidemic of infection are probably related to recent man-made ecological changes. The changing situation in Richard Toll provides a grim reminder of the impact environmental modification can have on disease distribution.

Transmission and Focality of Infection

Studies in Mali relevant to the National Schistosomiasis Control Programme have concentrated on the factors that contribute to the endemicity and focality of schistosome infections of the *S. haematobium* group (J. Vercruysse, University of Gent, Belgium). For the focality studies, circulating antigen detection was evaluated with results that proved to be comparable to or better than those obtained through the detection of eggs in faeces or urine, particularly in low endemicity foci.

Changes in schistosome distribution are obviously linked to the distribution of the intermediate snail hosts and their compatibility with different parasite strains. Besides conventional isoenzyme studies to identify snails and parasites, random amplification of polymorphic DNA (RAPD) has opened up hitherto unavailable options for parasite and snail strain identification, relevant to epidemiological and immunological studies as well as to surveillance during schistosomiasis control programmes. These new methods are being applied to studies of the schistosomes of the *S. haematobium* group and their intermediate hosts in Mali, Senegal and Zambia. Already the assumption that all schistosome parasites of one species are the same is no longer tenable.

Schistosoma intercalatum transmission was investigated in the Dogon Country, where *S. haematobium* is the major species; European tourists had apparently contracted *S. intercalatum* in this area, whereas it has never been found in the local populations. No evidence could be found to support the hypothesis that *S. intercalatum* is transmitted locally, even though polymorphic eggs resembling *S. intercalatum* were found in 33 of 1398 stool specimens. In nine cases, eggs were identified which were possibly hybrids between *S. haematobium* and *S. intercalatum*.

In human populations, there is little opportunity to design and implement experimental studies, apart from those created naturally (as in Richard Toll in Senegal, or Masongaleni in Kenya). The situation is quite the opposite for animal schistosomes, where experimental studies are possible and the perfusion of animals permits accurate counting of adult worm burdens. This situation has been exploited in Zambia where, in slaughterhouse surveys, the prevalence of bovine schistosomiasis is 51%, 90% of which are *S. mattheei*. However, infection intensities were generally low, with less than 100 adult worm pairs in the intestinal veins of animals. Studies on a dairy farm near Lusaka under conditions of natural challenge, including studies of imported tracer calves, provided evidence that some degree of acquired resistance to infection developed in animals as early as ten months after exposure to infection. This resistance was manifest in two ways: (1) a decline in faecal egg counts which were high in young animals and virtually zero in adult cows; and (2) the limitation of adult worm burdens in a situation akin to the 'concomitant immunity' seen in other systems. *Schistosoma mattheei* faecal egg counts appeared not to be a good indication of worm burdens.

Studies of *S. haematobium* and *S. mansoni* in the human population are also under way in Zambia where comparisons of disease transmission are being made between urban, farming and lake-side communities.

Diagnosis

One of the most exciting aspects of this meeting was the number of reports of the use of circulating anodic antigen (CAA) and circulating cathodic antigen (CCA) detection assays in various epidemiological situations, and the reports on the latest progress in development of field-applicable assay systems (A.M. Deelder, University of Leiden, The Netherlands). A range of assays has now been produced and these have been tailored to the needs of particular situations.

A main thrust of the research programme has been to produce a simple 'dipstick' assay. Significant progress has been made with a new dipstick assay based on the anti-CCA sandwich enzyme-linked immunosorbent assay (ELISA), which is able to detect CCA in urine of patients infected with *S. mansoni*. Using nitrocellulose as the carrier, with development using metal (nickel)-enhanced diamino-benzidine, sensitivity was 92.3%. The assay could be completed in 75 min, a significant improvement on ELISA. The results were qualitatively as good as those obtained by ELISA or by faecal examination by the Kato–Katz method. The simplicity and speed of the antigen dipstick may ultimately lead to its more-extensive application in screening programmes. Comparisons of the antigen detection assay with the Kato–Katz method from Brazil and the Philippines revealed that the ELISAs were sensitive enough to pick up apparently egg-negative cases, but did miss some egg-positives. Complementarity of gut- and egg-associated antigens in the diagnosis of low levels of infection was demonstrated

from studies in China, where chronic infections persist despite many years of intervention. In studies on *S. haematobium* in Cameroun, their effectiveness in monitoring post-chemotherapy re-infection was impressive. Quantitative antigen detection assays are also of use in measuring infection intensity and associated disease severity. A much more sophisticated time-resolved immunofluorometric assay (TR-IFMA) can be used to process specimens for quality control from different geographical locations. CAA and CCA detection on specimens from the previously described *S. mansoni* focus of Maniema in Eastern Zaire showed that the age profiles of these antigens closely mirrored those of faecal egg counts. A comparison of CAA and CCA data from Maniema, Zaire and Ndombo, Senegal was interpreted in favour of the hypothesis that the same amount of eggs was produced by fewer worms in Senegal than in Zaire. The implication is that worm fecundity in the chronic endemic focus in Maniema might be more affected by host factors. Anti-fecundity effects have also been the centre of a study of *S. haematobium* infection in laboratory animals and in field studies in humans in Kenya. The evidence from these studies strongly supported the view that anti-fecundity effects are important, and may be more significant in *S. haematobium* infections than in *S. mansoni* infections. Circulating antigen assays have moved beyond their straightforward diagnostic use and are now seen as being valuable tools for epidemiological investigations. Furthermore, they will be of value in monitoring large-scale interventions such as chemotherapy and vaccination programmes.

Immunity and Morbidity

One of the most important long-term studies of immunity and morbidity in human *S. mansoni* infection has been under way in Kenya for the past 13 years (A.E. Butterworth, University of Cambridge, UK). Chemotherapy directed at schoolchildren has proved an effective intervention in reducing the morbidity due to *S. mansoni* infection. The role of specific IgE antibodies in protective immunity was confirmed, as was the additional blocking functions of IgG4 antibodies. Recently the recognition of a 22 kDa antigen by antibodies in serum of exposed individuals has been shown to be associated with low levels of re-infection. Other antigens have been identified and responses to them are now being analysed in different mouse strains.

Field studies have now been extended to examine the interactions between schistosomiasis and nutritional status and other infections. Combined infections with *S. mansoni* and *Trichuris muris* in mice have shown that the presence of *S. mansoni* infection can cause a switch of the response from a T-helper (Th1) cell type, normally associated with the responses to *T. muris*, to that of a Th2 cell type. This results in interleukin production (IL-4 and IL-5) and the early expulsion of *T. muris* from the gut. Human T-cell responses to schistosome infection need further investigation. The experience in the Kenyan studies was that proliferative responses to adult worm and schistosomula antigens were inversely correlated with re-infection in those between 14 and 35 years of age, but not in those in the 9–13 year range. This result was confirmed even after allowing for the effects of age, sex and exposure to infection in multiple regression analysis. There was also an inverse relationship between the production of interferon-γ (IFN-γ) and IL-5, suggesting crossregulation of their production. A negative association was recorded between re-infection and IL-5 production to egg antigen stimulation, but this effect could not be dissociated from the confounding effects of age and exposure to infection.

Studies on the cytokine regulation of the human cellular immune response to *S. mansoni* antigens were also reported from Brazil (R.A. Wilson, University of York, UK). The role of cytokines on the peripheral blood mononuclear cell proliferation responses of *S. mansoni* patients was examined by adding anti-cytokine monoclonal antibodies or recombinant cytokines to PBMCs, cultured with soluble egg (SEA), adult worm (SWAP) and seven days cultured, lung-stage-like schistosomula antigens (SSP). The results varied between different clinical cases and provided evidence that different immunoregulatory mechanisms might function when the immune system is stimulated by different developmental stages of the parasite.

An important factor in any work on schistosomiasis is the role played by exposure to infection in determining levels of re-infection. Studies of snail, infected snail and cercarial densities are not easy to perform, but even these difficulties are dwarfed by the problems in measuring water contact. Normally, investigators attribute weightings (reflecting the risk of exposure) to particular water-contact behaviours. Given the difficulties in measuring accurately the duration of contact and the influence that weightings can have on the interpretation of the data, and consequently on the conclusions regarding resistance or susceptibility to infection, a degree of caution should be exercised whenever water-contact studies are interpreted.

New projects on the epidemiology, immunology and morbidity of *Schistosoma haematobium* infections have been approved (P. Hagan, University of Glasgow, UK; B. Vennervald, Danish Bilharziasis Laboratory, Copenhagen, Denmark). One of these is designed to investigate the assessment of morbidity using simple, non-invasive methods and in particular the detection of eosinophil cationic protein in urine (B. Vennervald). Initial results show great promise and extensive evaluation is now being done.

Vaccine Strategies

Considerable progress has been made in the development of the *S. mansoni* 28 kDa glutathione-S-transferase molecule (Sm28GST) as a vaccine candidate (A. Capron, Institut Pasteur, Lille, France). The distribution of Sm28GST in the different life cycle stages has now been mapped. The molecule is present in all stages, but is most abundant on the surface tubercles of adult worms and in the eggs of the parasite. It is now thought that the miracidium is responsible for most of the Sm28GST production and this has some implications for the effects of the immune response targeted against this molecule. Sm28GST has been crystallized and the three-dimensional structure of the molecule has been elucidated and the natural dimeric form of the molecule confirmed. Sm28GST is a parasite enzyme, and its enzymatic activity resides in the N-terminal (residues 10–43) and C-terminal (residues 190–211) domains, which are brought close to one another as a result of the folding of the molecule. The core of the molecule (residues 115–131) is highly exposed and forms the major dominant antigen. This core domain is also the main region of diversity of the molecule and has been shown to differ between the different schistosome species. This probably means that a vaccine which will protect against more than one schistosome species would need to be based on the N-terminal and C-terminal portions of the GST molecule even though experimental studies have shown that the core 115–131 peptide can, by

itself, stimulate a protective immune response. The chromosomal gene for Sm28GST has been identified. The promoter region for the molecule has binding sites for transcription factors, so regulation of expression of the molecule is likely to occur. In addition, nuclear components in schistosomes have been demonstrated to bind transcription factors of mammalian genes, raising the possibility that schistosomes may interfere with the regulation of the production of components of the immune response, for example the regulation of cytokine production. The *S. haematobium* and *S. bovis* GSTs have also been cloned, sequenced and expressed. Sm26GST is a relatively minor component in the 'African' schistosomes but is a major constituent of *S. japonicum*. The possibility that Sm28GST can stimulate IgA responses that may influence worm fecundity, suggested by observations on human sera from Kenya and Senegal, is being investigated with a view to finding oral vaccination regimes. Work is under way on the use of liposomes, immunostimulatory complexes (ISCOMs) and the live vectors, such as *Salmonella typhimurium* and *Bordetella pertussis*. The ability of IgA to participate in antibody-dependent cellular cytotoxicity reactions has been established definitively during the course of these studies. The evidence that vaccination with Sm28GST affects worm numbers as well as worm fecundity and the hatchability of eggs, bodes well for its potential as an anti-morbidity vaccine. Sm28GST is now also being investigated as a diagnostic reagent for use in antigen detection.

Extension of the vaccination studies to *Schistosoma haematobium* and *S. bovis* in primates and ruminants has, so far, yielded promising results. One important point emerging from these studies is that the immunization protocol and vaccine formulation are critical to outcome. Besides the work on Sm28GST, other molecules with a potential role in vaccination are being explored. Glutathione peroxidase of *S. mansoni* has been cloned but its expression in bacterial systems has proven problematic. A 45 kDa molecule identified as *S. mansoni* calreticulin has high- and low-affinity calcium binding sites and an endoplasmic reticulum retention sequence. It crossreacts with the Ro/SS-A antigen, the target of autoimmune responses in people suffering from systemic lupus erythrematosis (SLE); serum from *S. mansoni* patients recognize the Ro/SS-A antigen. No protection data are available for calreticulin or glutathione peroxidase.

There has also been progress on the development and use of defined antigen vaccines against *S. bovis* and *S. japonicum* in cattle (M.G. Taylor, London School of Hygiene and Tropical Medicine, UK). Irradiated vaccines have been highly successful in protecting animals against *S. bovis* and *S. japonicum* in experimental and natural field challenges, but are impracticable for continued large-scale field applications. Crude antigen vaccines have not been successful, but defined antigen vaccines have shown much promise. Four defined parasite antigens were studied in vaccination experiments: glutathione-S-transferase, keyhole limpet haemocyanin (KLH), paramyosin and a 23 kDa antigen of *S. japonicum*, Sj23. Antifecundity effects have been recorded following immunization with GST and KLH. While vaccination with GST can give a persistent IgG response, the IgG response to KLH wanes rapidly. The effect of immunization with GST and KLH against *S. bovis* in cattle in Sudan was to reduce faecal egg output, but tissue egg loads were not significantly reduced. The interpretation of this result requires some caution as the experiment was hampered by the loss of some control animals. In studies in China, it was found that the dose of antigen given in the vaccination procedure is critical, with higher doses invariably giving higher levels of protection when GST and KLH (both with complete Freund's adjuvant) were used to immunize sheep against *S. japonicum*. Immunized animals had high antibody responses against the molecules, and there was some indication that granuloma sizes were reduced in immunized animals. Whilst they may have restricted field application, irradiation attenuated parasites help to define antigens which act as important targets of protective immune responses. When irradiated parasites are used to immunize animals, antibody responses are stimulated against antigens of the parasite which are not stimulated when the animals are given a normal infection. Using this approach, antigens of 13, 23, 40 and 90 kDa have been identified as potential vaccine candidates. Reactivity to these molecules has been investigated using passive serum transfer experiments in mice and compared with the effectiveness of transferring serum taken from water buffalo. Additional studies are required before firm conclusions can be drawn. The molecular approach to work on *S. japonicum* is now at a relatively advanced stage. The GST molecule used by this group has been found to be identical to that produced by the Philippine strain of *S. japonicum*, allowing the pGEX vector to be used as a source of recombinant material. The 28 kDa GST has a four-nucleotide change in sequence resulting in a conservative amino acid substitution and continued enzymatic activities. Part of the paramyosin molecule has been expressed as a 33 kDa fusion protein with the pTricHis vector which can be readily purified on a nickel chelated sepharose column. Sj23 was initially identified as a molecule with immunodiagnostic potential. The molecule has two extracellular hydrophilic domains and protective monoclonal antibodies have been shown to bind to the larger of these. Following disappointing results in preliminary vaccination experiments with Sj23 in mice, alternative constructs and routes of presentation are now being investigated and the experiments will be extended to vaccination of cattle.

Conclusions and Recommendations

The immunological, genetic and neuroendocrinological factors that determine resistance and susceptibility to infection are not yet fully understood. As with studies of exposure to infection, some standardization of the methods and protocols used by different research teams is desirable, and would ultimately strengthen the confidence in the conclusions which can be drawn from such studies. Mathematical modelling of the epidemiology and control of infection transmission and morbidity could play an important role in these evaluations and also in allowing those responsible for schistosomiasis control programmes to investigate the likely outcome of alternative intervention strategies.

In the field of diagnostics, ultrasound techniques and classification systems should be standardized and geographical variations in morbidity should be investigated. The search must be continued for alternative markers of infection and morbidity that might permit rapid screening of populations. Other areas in which more information is required are in defining 'nonspecific' morbidity, and in assessing the impact of other infections on schistosome morbidity and vice versa.

Significant progress has been made in the development of the Sm28GST vaccine candidate. Work on this molecule, as well as on the other vaccine candidate antigens for the major schistosome species, will continue. Additional

studies of vaccine efficacy in animals (including primates) are needed.

There is an increasing awareness of the importance of strain variations in snails and in parasites, with implications for schistosome transmission and disease morbidity. Recent progress has been significant, in particular regarding the application of RAPD analyses. Establishing a reference source for parasite strains would also allow changes in drug susceptibility of parasites to be monitored.

Studies on host–parasite interactions in animals, and their epidemiology and immunology in natural systems, should be continued. In particular, more work should be done on *S. japonicum*; as this is the parasite about which we know least.

Opportunities for interactions with other networks within the CEC/STD programme, and with control and development programmes supported by the CEC and by individual member states, holds potential benefits for all of those involved. Mechanisms to encourage and facilitate such interactions should be established. In recent months, there has also been progress towards establishing a dialogue between the CEC and other international scientific bodies, in particular, with WHO/TDR and NIH. Further contacts and co-ordination between funding agencies were considered to be crucial to the scientific community involved in tropical disease research.

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Comment

Trans-sialidases in the Insect-vector Stages of African and American Trypanosomes

A.C.C. Frasch

Trans-sialidase is an enzyme that seemed to be exclusive to the American parasite *Trypanosoma cruzi*. However, recent reports have shown that *T. cruzi* shares this enzyme with the insect vector stages of African trypanosomes[1–4]. African and American trypanosomes use different mechanisms to survive inside their hosts, namely antigenic variation (for *T. brucei* trypomastigotes) and penetration into mammalian cells (for *T. cruzi*). In the case of *T. brucei*, after ingestion by the tsetse, bloodstream trypomastigotes differentiate into procyclic trypomastigotes in the midgut and then migrate to the salivary gland where they attach to the epithelial cells and transform into epimastigotes. Finally, epimastigotes differentiate into metacyclic trypomastigotes, which detach from the epithelial cells, invading the mammalian host through the saliva (reviewed in Ref. 5). In the case of *T. cruzi*, bloodstream trypomastigotes are ingested by the triatomine with the bloodmeal and differentiate into epimastigotes, which proliferate in the gut. After attachment to the epithelial cells at the rectal ampoule, epimastigotes transform into metacyclic trypomastigotes able to infect the mammalian hosts (reviewed in Ref. 6).

In 1985, a trans-sialidase activity was suggested to be present in *T. cruzi*[7] to explain the fact that the parasite contains sialic acid, but is unable to synthesize it[8]. This enzyme differs from neuraminidases/sialidases in that it preferentially transfers sialic acid to an acceptor molecule other than water; from sialyltransferases in that it uses donors of sialic acid other than cytidine monophosphate-sialic acid; and from both enzymes in eukaryotes in that it is located on the surface membrane of trypomastigotes and shed into the medium (reviewed in Refs 9,10). Thus, trans-sialidase can act on the outer side of the plasma membrane. This enzyme has been implicated in the process of invasion of mammalian cells through several possible mechanisms (reviewed in Ref. 10).

Reviews dealing with the biochemical properties[9,11,12] and gene structure and gene family organization[9,12,13] of the trans-sialidase have recently been published.

Trypanosoma cruzi trans-sialidase, previously considered to be expressed exclusively in the bloodstream trypomastigote stage, has now been shown to be present in stationary-phase cultured epimastigotes[14]. The enzyme is developmentally regulated, however, since trypomastigote and epimastigote trans-sialidases are expressed from different genes. The enzyme in epimastigote forms lacks the shed acute phase antigen (SAPA) repeats, a highly immunogenic domain encoded in the genes expressed in the bloodstream and metacyclic trypomastigote stages[14] (reviewed in Ref. 9). Trans-sialidase is also present in the African trypanosomes *T. b. brucei*, *T. b. gambiense*, *T. b. rhodesiense* and *T. congolense*[1–4,15] (M. Engstler, R. Schauer and R. Brun, unpublished). In all cases, the enzyme was found to be developmentally regulated, being expressed only in procyclic trypomastigotesis. Parasites (such as *T. evansi* and *T. equiperdum*) that are mechanically transmitted and lack the insect-vector stages did not present trans-sialidase activity in the bloodstream trypomastigote stage[15] (M. Engstler, R. Schauer and R. Brun, unpublished). Trans-sialidase from African trypanosomes has enzymatic properties similar to the trans-sialidase of *T. cruzi*[2].

Trypanosoma brucei and *T. cruzi* have few things in common in their life cycles. Thus, it seems likely that trans-sialidase has an important function in the insect-vector stages of both parasites. Acceptor/donor molecules of sialic acid were recently identified[2,3,17]. In African trypanosomes, the procyclic acidic repetitive protein (PARP, also