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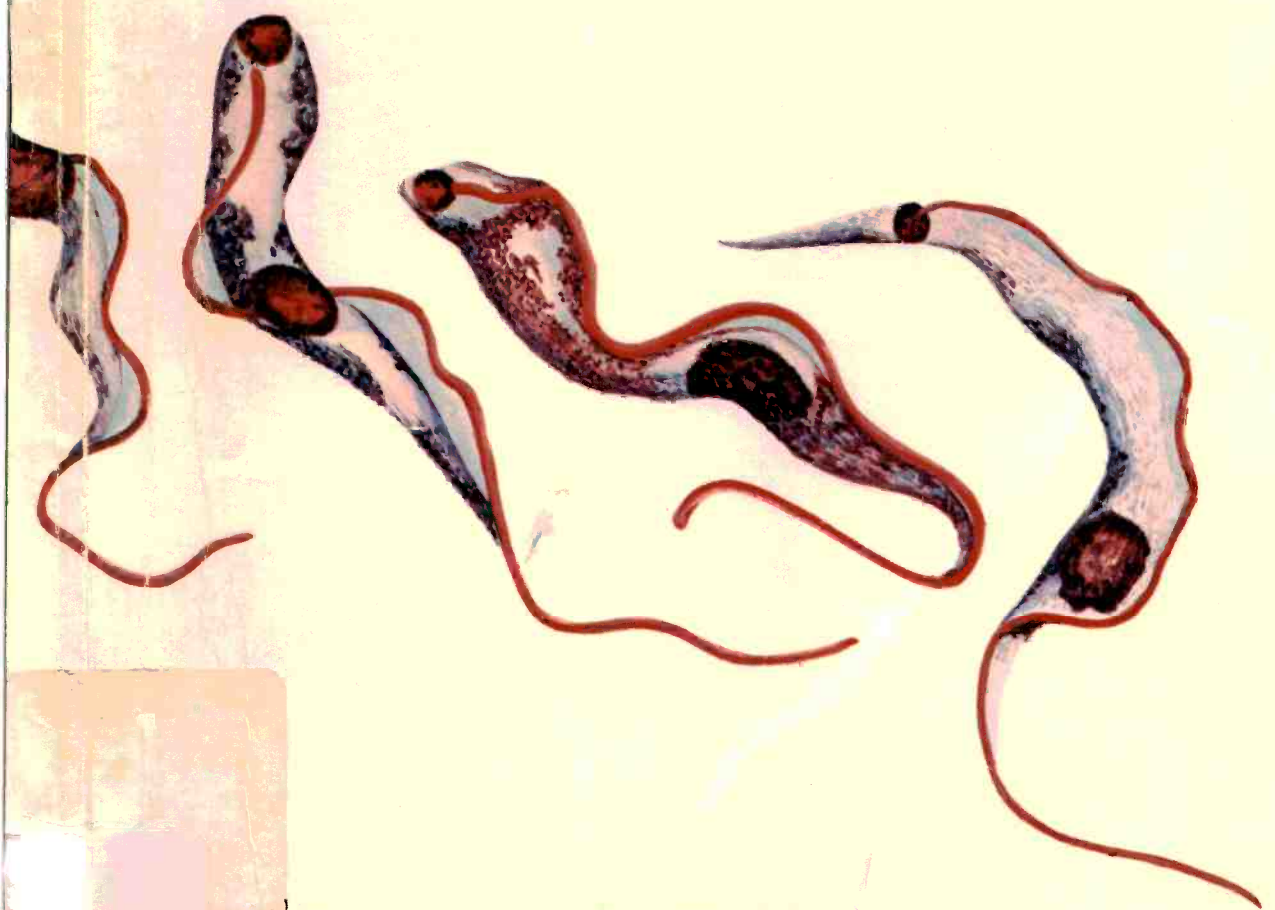
Pathogenicity of Trypanosomes

Proceedings of a workshop held at Nairobi, Kenya, 20-23 November 1978



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Editors: **George Losos[1] and Amy Chouinard[2]**

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Rodent trypanosomiases

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Abstract. Although the agents of the rodent trypanosomiases are generally considered benign, they can produce severe, and at times fatal, infections in young rodents, in pregnant hosts, and, with concomitant infections, in mature animals. The severity of such infections results from an immature or inadequate immune response. However, even in fully immunocompetent hosts, infections can be associated with splenomegaly, hepatomegaly, anemia, hypocomplementemia, glomerulonephritis, and immunodepression. Although these changes are similar to those produced by the pathogenic species of trypanosomes, they are much less severe and there is a relatively rapid return to normal. This results from the host's immune response, and especially the production of a reproduction-inhibiting antibody, ablastin, which controls the parasitemia until trypanocidal responses eliminate the parasites. In the absence of, or with an inadequate, ablastic response, the rodent trypanosomes may produce fulminating infections strikingly similar to those of the pathogenic species. Like the pathogenic African species, they are capable of antigenic variation, but they are ultimately blocked by the host's development of reproduction-inhibiting immunity, a distinguishing feature of the rodent diseases. An understanding of this unique response, which renders potentially lethal parasites, benign, may be valuable for the control of the pathogenic species.

The rodent trypanosomiases are caused by a large group of parasites that are distributed worldwide and include more than 40 named species of *Trypanosoma* of the subgenus *Herpetosoma* (Hoare 1972). These trypanosomes are generally considered benign or nonpathogenic (D'Alesandro 1970; Hoare 1972; Molyneux 1976; Mansfield 1977). Moreover, with few exceptions, they show a high degree of host specificity and are uninfective for rodents other than the ones in which they occur naturally. These features are in contrast to the African trypanosomes and *T. cruzi*, the agent of American trypanosomiasis, which are considered pathogenic and show broader host ranges (Hoare 1972; Mansfield 1977). Although the appellations "pathogenic" and "nonpathogenic" are convenient designations for these groups of trypanosomes, they are not strictly accurate, for the rodent trypanosomes do harm their hosts and under certain conditions can cause death. Conversely, although the African trypanosomes and *T. cruzi* are usually pathogenic and frequently lethal in man and certain domestic animals, they appear to be well tolerated in many species of wild animals that may also serve as reservoir hosts. Therefore, with both groups of trypanosomes, apparent nonpathogenic infections occur; in the case of the rodent trypanosomiases, this favourable balance has an immunologic basis, but the factors involved in comparable infections with the pathogenic trypanosomes have not yet been identified (D'Alesandro 1970).

In spite of the fairly large number of species of rodent trypanosomes, only a few of them have been studied in detail, and the studies made, though numerous, have usually been immunologic. Studies of the abnormalities produced by these trypanosomes are relatively few and have been confined to two species: *T. lewisi* of the rat (*Rattus* spp.) and *T. musculi* of the house mouse (*Mus musculus*). This limitation has probably resulted from practical considerations, i.e., ease of maintaining laboratory strains of rats and mice. Nevertheless, the information available on these two species will probably be applicable to other members of the group. Moreover, the studies have the advantage that they are based on natural host–parasite relationships.

The pathologic changes in rodent trypanosomiasis have not been studied as

thoroughly as those caused by the pathogenic trypanosomes. It is clear nonetheless, that although they are not usually as severe and extensive as the latter, there are some common features.

Age of Host

Most species of rodent trypanosomes are well tolerated by their hosts, but naturally pathogenic strains of *T. lewisi* that produce lethal infections in rats of all ages have been described by Brown (1914) and have been experimentally developed by rapid passage through rats (Roudsky 1910a, b; 1911a, b). The latter are also infective for mice and other rodents. All strains, however, are more pathogenic for young rats as first reported by Jürgens (1902) and confirmed by others. Severe, and at times fatal, infections with marked anemia were noted in young hosts by Marmorston-Gottesman, Perla, and Vorzimer (1930). In more detailed studies, death rates of 80–90% in rats less than 30 days old were recorded by Herrick and Cross (1936); rats more than 40 days old always survived with few apparent ill effects. Similar results were obtained by Duca (1939), who found that infections killed more than 70% of rats 25 days old or younger in contrast to 6% of animals more than 25 days old. In rats of all ages, however, secondary anemias were observed. Similarly, young mice have been reported to be more susceptible than older animals to infection with *T. musculi* (Culbertson 1941).

The basis of the age resistance of rats to infection with *T. lewisi* appears to be immunologic. Following immunization with formalin-killed blood trypomastigotes, rats less than 25 days old produced low titred agglutinins and showed low resistance to challenge infections, at times succumbing as readily as unvaccinated controls (Culbertson and Kessler 1939). In contrast, older animals readily produced agglutinins and had solid immunities. Further studies (Culbertson and Wotton 1939) showed that young rats were unable to control the reproduction of the trypanosomes, indicating inadequate production of the reproduction-inhibiting antibody, ablastin (Taliaferro 1932). These observations indicate that the generally severe infections of young rodents result from their immunologic immaturity.

Intercurrent Infections

The potential pathogenicity of rodent trypanosomes can be expressed in older, immunocompetent hosts that experience intercurrent infections with other agents. The additional stress imposed on the host by the dual infection can be sufficient to upset the host–parasite balance so that severe or fatal infections occur. *Haemobartonella muris*, which is frequently latent in otherwise normal rats and mice (Marmorston-Gottesman and Perla 1930; Baker, Cassell, and Lindsey 1971; Bartlett and Pease 1975; Cox and Calaf-Iturri 1976; Sogandares-Bernal and Chandler 1978), is most commonly found associated with *T. lewisi*. Such dual infections are especially severe in young animals (Marmorston-Gottesman, Perla, and Vorzimer 1930), but even in mature rats, death rates of up to 50% can occur within the 1st week of the trypanosome infection (D'Alesandro unpublished results). Similarly, *Plasmodium berghei*, which seldom kills older rats, has caused death rates of 50% (Hughes and Tatum 1956) to 68% (Jackson 1959) when injected simultaneously with *T. lewisi*. In contrast, none of the mice simultaneously inoculated with avirulent *P. yoelii* and *T. musculi* died. When trypanosomes were inoculated into mice that had been injected with *P. yoelii* 7 days earlier, however, mortality was more than 60% (Cox 1975). The deaths were attributed primarily to the immunodepressive effects of the malaria on the ability of the host to respond to the trypanosomes. In similar dual infections of mice with virulent *P. berghei* and *T. musculi* (Bungener 1975), all the animals died regardless of the timing of the inoculations, but it was noted that with simultaneously initiated dual infections, the trypanosome parasitemia was stabilized before death.

It is clear from the studies of age resistance and intercurrent infections that an effective immune response is necessary to suppress the potential virulence of the rodent trypanosomes. Nevertheless, even in fully immunocompetent hosts that are able to terminate their infections with little apparent difficulty, some pathogenic changes occur — striking evidence that even under the most favourable circumstances, the rodent trypanosomes are not truly benign.

Splenomegaly and Hepatomegaly

Probably the most obvious abnormality resulting from rodent trypanosomiasis is splenomegaly. In one of the earliest studies, rats infected with *T. lewisi* and *Haemobartonella* had spleens that were six to seven times as large as those in uninfected animals (Marmorston-Gottesman, Perla, and Vorzimer 1930), although it is difficult to determine how much of this increase was due to the trypanosomiasis. In similar studies with

Haemobartonella-free rats Taliaferro, Cannon, and Goodloe (1931) found that *T. lewisi* infections caused only a doubling of spleen size, whereas concomitant infections with *Haemobartonella* resulted in spleens three times the normal; comparable degrees of splenomegaly were reported by Becker, Manresa, and Johnson (1943) and Thoongsuwan and Cox (1978). In one of the most recent studies, presumably in *Haemobartonella*-free rats, about a ninefold increase in spleen size was observed (Ferrante, Jenkin, and Reade 1978). The variations aside, *T. lewisi* clearly induces splenomegaly. Similarly, mice infected with *T. musculi* can have spleens 10 times larger than normal (Albright, Albright, and Dusanic 1977).

Studies of the kinetics of splenomegaly indicate there is a general, positive correlation with the parasitemia. Most of the increase in spleen size occurs within the first 7–10 days of infection with *T. lewisi* and *T. musculi*, and this corresponds to the time of peak parasitemia (Marmorston-Gottesman, Perla, and Vorzimer 1930; Taliaferro, Cannon, and Goodloe 1931; Albright, Albright, and Dusanic 1977), although Ferrante, Jenkin, and Reade (1978) observed maximum spleen enlargement on the 20th day of infection. Parasitemia is terminated shortly after maximum splenomegaly is attained, and the spleen gradually returns to near normal size. Histologically, there is a marked hyperplasia of the white pulp (lymphoid elements) of the spleen so that as the lymphoid follicles increase in size and number, the division between white and red pulp becomes less distinct (Marmorston-Gottesman, Perla, and Vorzimer 1930; Taliaferro, Cannon, and Goodloe 1931; Albright, Albright, and Dusanic 1977; Ferrante, Jenkin, and Reade 1978). There is also hyperplasia of the red pulp (Taliaferro, Cannon, and Goodloe 1931) apparently resulting from erythropoiesis and an increase in lymphoid elements (Ferrante, Jenkin, and Reade 1978). As the spleen returns to normal size, it also regains its normal structure.

Infections with *T. lewisi* and *T. musculi* also cause hepatomegaly, but the liver size does not exceed twice normal (Albright, Albright, and Dusanic 1977; Ferrante, Jenkin, and Reade 1978). In rats, maximum hepatomegaly occurs several days later than the peak of splenomegaly, and the liver returns to normal size more slowly than does the spleen (Ferrante, Jenkin, and Reade 1978); in mice, the liver returns to normal size before the spleen (Albright, Albright, and Dusanic 1977). Histological studies in the rat have attributed liver enlargement to degenerative changes characterized by cloudy swelling of parenchymal cells (Ferrante, Jenkin, and Reade 1978); in addition, there is a marked round-cell infiltration. Ultrastructural studies of liver from *T. lewisi*-infected rats have revealed parenchymal cells with dilated cisternae of the endoplasmic reticulum and swollen and more numerous mitochondria (Simaren 1973; Lee and Barnabas 1974). Hepatocytes with focal areas of lipid and fatty infiltrations were also observed, but normal structural appearance was regained about 5 weeks after infection. Coincident with the general decline and termination of the parasitemia, between the 14th and 21st day of infection, residual lysosomes were found in Kupffer cells, which also contained phagocytized trypanosomes (Lee and Barnabas 1974).

The marked lymphoid hyperplasia and consequent splenomegaly probably result from the intense antigenic stimulation that occurs during infections with rodent trypanosomes. Mitogens of parasite origin may also be involved, as suggested by a recent study (Hazlett and Tizard 1978). Another factor very likely involved is the anemia associated with infection, because erythrophagocytosis has been reported in the spleens of infected rats (Marmorston-Gottesman, Perla, and Vorzimer 1930; Greenblatt 1973; Thoongsuwan and Cox 1978) as well as erythropoiesis (Greenblatt 1973; Ferrante, Jenkin, and Reade 1978).

Anemia

The anemia associated with *T. lewisi* infections of rats has been known for a long time (Marmorston-Gottesman, Perla, and Vorzimer 1930; Duca 1939; Saul and Becker 1949; Barnes 1951; Sherman and Ruble 1967; Tawil and Dusanic 1971; Shaw and Dusanic 1973; Greenblatt 1973; Thoongsuwan and Cox 1978). In some of the earlier studies, concomitant *Haemobartonella* infections complicated the results (Marmorston-Gottesman, Perla, and Vorzimer 1930), but in later work with *Haemobartonella*-free rats, it was clearly demonstrated that *T. lewisi* alone can induce anemia. More recently, anemia has been reported in mice infected with *T. musculi* (Jarvinen and Dalmasso 1977a). In all these studies, the anemia can generally be correlated with changes in the number of parasites and is usually most severe near the time of peak parasitemia. In rats, erythrocyte numbers can decrease from about 7 to $4 \times 10^6/mm^3$ (Duca 1939; Thoongsuwan and Cox 1978; D'Alesandro unpublished results) and hematocrit levels from about 45 to 35% (Sherman and Ruble 1967; Tawil and Dusanic 1971; Shaw and Dusanic 1973); similar changes in hematocrit values occur in mice (Jarvinen and Dalmasso 1977a).

The mechanism of anemia in rodent trypanosomiasis is not fully understood. There does

not appear to be a suppression of hematopoiesis because normoblasts and compensatory reticulocytosis occur during the infection (Duca 1939; Greenblatt 1973; Jarvinen and Dalmasso 1977a). Hemolysins of parasite origin have been suggested as the cause of these secondary anemias (Duca 1939), but there is no evidence of intravascular lysis: no hemoglobinuria or hemoglobinemia has been found. Erythrophagocytosis does occur, however (Marmorston-Gottesman, Perla, and Vorzimer 1930: Greenblatt 1973; Thoongsuwan and Cox 1978), and could account for the loss of erythrocytes. Recently Jarvinen and Dalmasso (1977a) have found evidence for an immunologic mechanism of the anemia in *T. musculi*-infected mice. Direct antiglobin tests of erythrocytes were positive: 50% of the mice had only IgG1 on their erythrocytes; the remaining animals had different combinations of IgG2, IgM, IgA, and C3 in addition to IgG1. Because similar degrees of anemia occurred in C5-deficient and normocomplementemic strains of mice and because the presence of C3 in addition to the immunoglobulins on the erythrocytes did not appear to aggravate the anemia, the investigators concluded that intravascular hemolysis does not occur and that C3 is not essential for erythrophagocytosis. Apparently they did not attempt to determine whether parasite antigens were also present on the surface of the erythrocytes, but it is likely that the bound immunoglobulins were part of an antigen–antibody complex that would make the cells susceptible to phagocytosis via one of the mechanisms postulated by Jennings (1976).

The anemia associated with *T. lewisi* infections in rats has not yet been similarly studied, but it may also involve immunologic factors. Exoantigens are produced by *T. lewisi* and remain in the blood throughout the infection (D'Alesandro 1972, 1975; Bawden and Stauber 1974). Also it was noted many years ago by Marmorston-Gottesman, Perla, and Vorzimer (1930) that the serum of *T. lewisi*-infected rats is anticomplementary (i.e., fixes complement), and this finding was interpreted as evidence of circulating antigen–antibody complexes. Possibly, the complexes adsorb temporarily to the surfaces of the erythrocytes and activate complement, thereby promoting opsonization by a mechanism similar to that proposed by Assoku (1975) to explain the anemia in rats infected with *T. evansi*; alternatively, antibody may combine with adsorbed parasite antigens on the erythrocyte surface with subsequent release of the complex. Consistent with these possibilities is the marked reduction in total complement that occurs during *T. lewisi* infections in rats (Jarvinen and Dalmasso 1976; Nielsen, Sheppard, Holmes et al. 1978). In a recent study (Thoongsuwan and Cox 1978), cold-active hemagglutinins for trypsinized rat erythrocytes were detected in plasma from *T. lewisi*-infected rats. However, the role of these antibodies in the anemia is unclear because the induction of similar autoantibodies in uninfected rats did not result in anemia. Nevertheless, other observations argue against an immunologic mechanism. It has been found by Balber (1974), for example, that immunosuppression of mice by treatment with corticosteroids attenuates the anemia associated with *T. brucei* infections. However, treatment with cortisone (Sherman and Ruble 1967) or antilymphocyte serum (Tawil and Dusanic 1971) of *T. lewisi*-infected rats exacerbates the anemias. Possibly, such treatments also affect erythropoiesis, as antilymphocyte treatment alone causes a mild, transient anemia, and cortisone treatment alone, although it causes no anemia, reduces the number of reticulocytes. Furthermore, Greenblatt (1973, 1975) has found evidence of a developmental link between plasma cells and blood cells in his studies of infected rats' spleens and has suggested that because of the intense antigenic stimulus provided by *T. lewisi*, anemia occurs because of competition between lymphopoiesis and hematopoiesis for a common stem cell.

Complement Depletion

Recent studies by Jarvinen and Dalmasso (1976) have shown that a massive activation of complement occurs via the classical pathway in *T. lewisi*-infected rats. Total complement and C4 levels are reduced to less than 10% of preinfection values, irrespective of parasite numbers; C3 levels are inversely proportional to parasitemia, dropping to 35% of normal values with heavy infections; and C6 levels are unaffected. In genetically C4-deficient rats, parasitemias and C3 levels were found to be similar to those of normocomplementemic controls, and the use of cobra venom factor at various times during infection to deplete C3 and late-acting components in C4-deficient and normal rats had no effect on the course of infection. These results indicate that complement is not essential to, or at least does not play a major role in, the control and elimination of the trypanosomes. In contrast, Nielsen, Sheppard, Holmes et al. (1978) found that decomplemented rats developed significantly higher parasitemias than did controls. Their findings may be related to their methods: the cobra venom factor was administered 24 hours before infection with the trypanosomes and in larger doses than in the studies by Jarvinen and Dalmasso (1976), who

suggest that immune complexes are responsible for the complement depletion. It is likely that complement plays a role in the anemia because the level of C3 and the severity of the anemia are both proportional to the parasitemia. Is C3 bound to the surface of erythrocytes of *T. lewisi*-infected rats, and does C3 depletion attenuate the anemia? Unfortunately, these questions have not yet been answered.

In contrast to *T. lewisi* infections, *T. musculi*-infected mice show unchanged or slightly increased levels of C1 and C3 (Jarvinen and Dalmasso 1977b); only in genetically C5-deficient mice are occasional, moderate reductions of these components observed. In normocomplementemic and C5-deficient mice, however, the course of infection is similar (Dusanic 1975b; Jarvinen and Dalmasso 1977b). In addition, treating mice with cobra venom factor late in the infection reduces the rate of parasite elimination and prolongs the infection (Jarvinen and Dalmasso 1977b). Therefore, two closely related species, *T. lewisi* and *T. musculi*, evoke surprisingly different responses in their hosts. The results suggest that elimination of the parasites depends upon complement-mediated opsonization, although, if this is true, a high synthetic rate would be necessary to maintain the generally unchanged levels of complement components observed throughout infection. These essentially unchanged levels, however, are consistent with the conclusion that C3 is not involved in the anemia caused by *T. musculi* (Jarvinen and Dalmasso 1977a).

It has recently been reported that after several hours of incubation at 20 °C, suspensions of *T. lewisi* blood trypomastigotes release anticomplementary factors that can activate bovine, human, and guinea pig complement in vitro (Nielsen and Sheppard 1977); subsequent work has shown that one of the major, active components that can be prepared from cellular homogenates of the parasite is a carbohydrate-rich compound (Nielsen et al. 1977). How these findings are related to the complement depletion that occurs in the natural host is not clear. Rat serum was apparently not tested, and anticomplementary activity was not detected until suspensions of cells had been incubated in buffer for 3.5 h, at which time very few parasites were still viable (Nielsen et al. 1978b). Jarvinen and Dalmasso (1976) found that *T. lewisi* cells can immediately activate rat complement in vitro but only when incubated in serum from immune, not normal, rats. Therefore, although other species of trypanosomes may activate complement directly (See Diffley 1978a, b; Diffley and Honigberg 1978), the available evidence indicates that, during *T. lewisi* infections, complement depletion occurs through the participation of antibody.

The host is especially vulnerable to the rodent trypanosomes during pregnancy when embryonal and maternal death can be common consequences of infection. In the studies of Shaw and Dusanic (1973), it was found that if infections with *T. lewisi* in rats were initiated early in the 1st week of pregnancy, fetal resorption readily occurred and parasitemias were similar to those of nonpregnant controls; in rats infected late in the 1st week of pregnancy, however, resorption was more difficult, and half the females died shortly before parturition, even though parasitemias were not elevated. The midterm of pregnancy was found to be the most vulnerable period, for most of the rats infected at this time had extremely high parasitemias and died at parturition (80%) without giving birth. Conversely, during the last week of pregnancy, resistance appeared to be enhanced, for rats infected at this time had significantly lower parasitemias than did controls and produced normal litters. The anemia associated with the infection was exacerbated in all the infected, pregnant rats except in those infected during the last week of pregnancy. It was also found that the placentas of rats infected at midpregnancy harboured an unusually large number of multinucleated parasites that contained 8–16 nuclei and kinetoplasts, but parasites were not found in fetuses. The mechanism of fetal death is unknown but does not appear to be hormone depletion (Shaw and Quadogno 1975).

Similar observations have been made by Krampitz (1975) in his studies of pregnant mice infected with *T. musculi*. Here, too, the host was most vulnerable during the midterm of pregnancy (between the 4th to 14th days) when fetal resorption, abortion, and maternal death frequently occurred and parasitemias were 10-fold greater than normal; when infections were initiated during the last week of pregnancy, however, parturition and parasitemias followed a regular course. The focus of the enhanced parasitemias appears to be the placenta where immense masses of reproducing trypanosomes are found in unusually large rosette formations. Krampitz (1975) has observed that parasitemias show an immediate decline following normal delivery, abortion, or the surgical removal of the pregnant uterus; extirpation of the embryos alone with preservation of the placenta in situ has no marked effect. Krampitz (1975) suggested that an immune response develops during the susceptible period but is somehow held in abeyance by hormonal or other factors until the uterus is emptied. It is also possible that there is simply an excess of antigen relative to circulating antibody at this time, and the balance is shifted to antibody

excess, with consequent removal of the parasites, when the focus of intense antigen production is removed. That such severe infections also occur naturally is indicated by the intense parasitemias found in wild, pregnant house mice by Krampitz (1975).

Immunodepression

Albright, Albright, and Dusanic (1977, 1978) have convincingly demonstrated that immunodepression can occur in rodent trypanosomiasis. They found that in mice infected with *T. musculi* there is a marked correlation between the kinetics of parasitemia and splenomegaly and the depression of humoral immune responses (see also Hazlett and Tizard 1978). At the time of maximum splenomegaly (day 14), the in vivo response to sheep erythrocytes was 10% of normal, and the in vitro response of infected spleen cell cultures was completely suppressed; following the termination of the parasitemia and the return of the spleen toward normal size, immune responsiveness was regained. Analysis of cell types in infected spleens showed that although the normal ratio of T cells to B cells was doubled at maximum splenomegaly, their absolute numbers were actually increased 10-fold and 5-fold, respectively, because of the hyperplasia. Paralleling the depressed humoral antibody response was a virtually complete suppression of the response of infected-spleen cell cultures to T-cell and B-cell mitogens. Preliminary experiments indicate that humoral substances directly mediate the immunodepression: it was observed that serum from infected mice, saline extracts of blood trypomastigotes, and living blood trypomastigotes ($>10^3$ cell/ml) strongly inhibited the humoral antibody response of normal spleen cell cultures; marked inhibition also occurred when such cultures were separated from the living parasites by membranes with a pore size of 0.22 μm. Whether *T. lewisi* or other rodent trypanosomes have similar immunodepressive effects is at present unknown.

Glomerulonephritis

Viable, infective stages of *T. musculi* can be found in the kidneys of immune mice almost 1 year after termination of parasitemia (Viens et al. 1972; Targett and Viens 1975). How this prolonged presence relates to pathologic changes is not clear because the kidneys, although they become enlarged (up to twice normal), return to normal size about 1 month after infection (Albright, Albright, and Dusanic 1977). Ultrastructural studies have shown, however, that at the peak of parasitemia, when glomerular diameters increased two or three times and trypanosomes were present in glomerular capillaries, there was an infiltration of the glomeruli by eosinophils, neutrophils, and other leukocytes (Molyneux, Kaddu, and Suzuki 1973; Molyneux 1976). The changes associated with glomerulonephritis were not found until 21 days after infection when electron-dense material and a thickening of the basement membrane appeared in glomerular capillaries.

T. lewisi, too, has been found in the kidneys of infected rats (Ormerod 1963, 1975) but, in contrast to *T. musculi*, only during the period of patent parasitemia and not after recovery (Wilson et al. 1973; Targett and Viens 1975). Ultrastructural studies by Simaren (1974) of rat kidneys 14 days after infection have also demonstrated cytopathologic changes, most notably, irregular thickening and splitting of capillary and tubular basement membranes where parasites were localized. In the most recent study, in addition to thickening of Bowman's membrane and tubular basement membrane, hypercellularity of the glomerular tuft, swelling of vascular endothelium and tubular epithelium, and abnormal numbers of hyaline casts were observed (Thoongsuwan and Cox 1978). The etiology of glomerulonephritis in rodent trypanosomiasis is unknown, but very likely immune complexes are involved.

Discussion and Conclusions

Available evidence strongly indicates that the relatively benign nature of the rodent trypanosomiases has an immunologic basis. As pointed out by Taliaferro (1929), the unique aspect of these rodent diseases is the ability of the host to produce a reproduction-inhibiting antibody, ablastin, that controls the parasitemia until trypanocidal responses destroy the parasites. The resulting discontinuous reproductive activity of the rodent trypanosomes contrasts sharply, for example, with the African trypanosomes, which show continuous reproductive activity throughout the infection. The periodic crises observed in the latter, however, are only temporarily effective because of the marked antigenic lability of the pathogenic species. The rodent trypanosomes also undergo antigenic variation, although to a more limited degree (D'Alesdandro 1970, 1976), but the key element in their control is the host's ablastic response. Thus, if the host is adequately immunosuppressed by a variety

of standard methods, an ablastic response does not occur, and the rodent trypanosomes produce fulminating, fatal infections that are strikingly similar to those of the pathogenic species (see D'Alesandro 1970; Dusanic 1975b). Therefore, although both groups of trypanosomes elicit trypanocidal responses, it is apparently the failure of the host to produce an ablastic response against the pathogenic species that allows full expression of their pathogenic potential. Even despite a well developed ablastic response, the rodent trypanosomes cause some pathologic changes, which ironically, are apparently the by-products of the vigorous, effective immune response.

Although the abnormalities associated with rodent trypanosomiases are not generally as severe as those associated with the pathogenic trypanosomes, there are common features. Splenomegaly and hepatomegaly, for example, are clinical features of African trypanosomiasis in humans, domestic animals, and experimental hosts (Apted 1970; Ormerod 1970; Losos and Ikede 1972; Murray, P.K. et al. 1974a; Mansfield and Bagasra 1978); these conditions generally develop quickly and persist throughout the infection. In rodent trypanosomiasis there is a relatively rapid return to normal that parallels the decline and termination of infection. Moreover, the degree of organ enlargement can be much greater with the pathogenic species, at least under experimental conditions. Thus, *T. musculi*-infected mice show a 10-fold increase in spleen size (Albright, Albright, and Dusanic 1977), whereas mice infected with *T. rhodesiense* may have spleens 20 times as large as normal (Mansfield and Bagasra 1978). In both cases, organ growth can be attributed primarily to marked hyperplasia of lymphoid elements, but the observed erythrophagocytosis (Greenblatt 1973; Goodwin 1974; Murray, P.K. et al. 1974a) is probably also a contributing factor.

Anemia has long been recognized as a common feature of the various pathogenic forms of trypanosomiasis as well as other parasitic diseases (Mulligan 1970; Woodruff et al. 1973; Goodwin 1970, 1974; Jennings 1976), and accumulating evidence strongly suggests that immunologic mechanisms promoting erythrophagocytosis are involved (see Jennings 1976; Kobayashi, Tizard, and Woo 1976; Jarvinen and Dalmasso 1977). Similarly, the anemia in rodent trypanosomiasis appears to be immunologically mediated, but because studies have been so limited, it is not clear which of the several possible mechanisms described by Jennings (1976) is operative. Most likely, immune complexes that promote erythrophagocytosis rather than intravascular hemolysis play a major role.

Human, animal, and experimental infections with the pathogenic trypanosomes have been reported to cause complement depletion (Ormerod 1970; Nagle et al. 1974; Greenwood and Whitttle 1976b; Kobayashi and Tizard 1976; Diffley 1978b), and there is evidence to suggest that immune complexes are also involved in this process (Nagle et al. 1974; Houba 1976; Neilsen and Sheppard 1977). Thus far, in studies of rodent trypanosomiasis, significant complement depletion has been reported only in *T. lewisi*-infected rats (Jarvinen and Dalmasso 1976), but here, too, immune complexes appear to play a major role. The thickened basement membranes reported in the glomerulonephritis of infected rodents, for example, most likely result from deposits of complement-containing immune complexes, which have been reported in experimental infections with pathogenic trypanosomes (Nagle et al. 1974; Houba 1976). It has been suggested, however, that other mechanisms of complement consumption, such as the release of complement-activating factors by the parasite, may also occur in trypanosomiasis (Nielsen and Sheppard 1977; Nielsen, Sheppard, Tizard et al. 1978). In this regard, Diffley (1978a, b) and Diffley and Honigberg (1978) have convincingly demonstrated that complement consumption can occur in the absence of an immune response. They found that in immunosuppressed rats infected with *T. congolense* or *T. rhodesiense*, complement levels were depressed and to a degree generally proportional to the number of circulating parasites. Moreover, the addition of trypanosomes from immunosuppressed hosts to fresh normal rat, human, and bovine sera caused immediate complement consumption in vitro, and C3 was detected on the surface of such parasites by immunofluorescence techniques. Similar direct activation of complement in vitro by suspensions of living *T. brucei* cells derived from lethally irradiated hosts as well as by isolated variant-specific surface antigen was recently reported by Musoke and Barbet (1977). In rodent trypanosomiasis, however, as mentioned earlier, there is as yet no evidence for direct activation of complement.

The significance of hypocomplementemia in trypanosomiasis is not fully understood, but its possible role in immunodepression, polyclonal stimulation, susceptibility to secondary infections, and evasion by the trypanosomes of the host's immune response has been discussed by others (Losos and Ikede 1972; Nielsen and Sheppard 1977; Nielsen, Sheppard, Holmes, et al. 1978; Diffley 1978; Cross 1978a). In rodent trypanosomiasis, there is evidence to suggest that depressed complement levels are directly related to

lowered resistance to secondary bacterial infection. In a study by Nielsen, Sheppard, Holmes, et al. (1978), it was found that if rats were inoculated with *Salmonella typhimurium* 1 week after infection with *T. lewisi*, a time when the hypocomplementemia is most severe, all the animals died within 5–12 hours of enteritis; in contrast, among control animals infected with bacteria alone, the death rate was only 10%.

Immunodepression is now a well recognized and studied feature of pathogenic forms of trypanosomiasis as well as other parasitic and infectious diseases (Goodwin 1970; Goodwin et al. 1972; Murray, P.K. et al. 1974a; Houba 1976; Clinton et al. 1975; Rowland and Kuhn 1978a, b; Ramos et al. 1978). Thus far, however, immunodepression in rodent trypanosomiasis has been described only in *T. musculi*-infected mice (Albright, Albright, and Dusanic 1977, 1978; Hazlett and Tizard 1978). On the basis of the limited data available, some interesting differences are apparent. In *T. musculi* infections, although there is an absolute increase in splenic T and B cells, the proportion of B to T cells declines to about half the normal value. In *T. rhodesiense*-infected mice there is also an absolute increase in B and T cells of the spleen, but the ratio of B to T cells increases to more than 6 times the normal (Mansfield and Bagasra 1978). The marked hyperplasia of plasma cells in experimental African trypanosomiasis has been noted by others (see Murray, P.K. et al. 1974a) and is no doubt the basis of the elevated IgM levels found in this disease, although the initiating factors are not completely understood (Clarkson 1976). Moreover, recent studies have provided evidence implicating suppressor T. cells and macrophages in the immunodepression of African trypanosomes (Corsini et al. 1977; Eardley and Jayawardena 1977); the trypanosomes do not appear to play a direct role (Eardley and Jayawardena 1977). In contrast, there is evidence that humoral factors of parasite origin have a direct immunodepressive effect in *T. musculi* infections (Albright, Albright, and Dusanic 1977, 1978). Nevertheless, these apparent differences in mechanisms do not alter the potentially important consequences of immunodepression, especially those related to secondary infections (see Murray, P.K. et al. 1974a).

It is apparent from comparisons of the abnormalities produced by the pathogenic and the rodent trypanosomes that there are many similarities. In rodent trypanosomiasis, however, even when pathologic changes approach in severity those produced by the pathogenic species, the duration of the changes is relatively short and there is a return to the normal state with few, if any, lasting effects. There is no doubt that the rapid immune response of the host attenuates the pathogenicity of the rodent trypanosomes and is the basis of the favourable host–parasite relationship.

Moreover, the central element of this response is ablastin, the unique reproduction-inhibiting antibody whose mode of action is still not completely understood (see D'Alesandro 1970, 1975; Giannini and D'Alesandro 1978). Present knowledge of rodent trypanosomiases has had no practical applications, but further studies leading to identification of the factors responsible for the comparatively benign nature of these diseases may well provide the key to control of their pathogenic counterparts.