section B — abstracts
1. general (including land use)
   [See also 18: no. 8868.]
   Hospital for Tropical Diseases, St Pancras Way, London NW1 0PE, UK.
   This editorial recounts the steps taken by Dr David Bruce to determine the cause of nagana in Zululand 100 years ago. By a series of elegant parasitological and entomological experiments, he established beyond doubt that nagana is caused by a ‘Haematozoa’ (later named Trypanosoma brucei) and that this is conveyed by an infected tsetse fly, which is itself infected by feeding on game animals which form the major reservoir of infection. He was thus the first investigator to demonstrate transmission of a protozoan parasite by insect bite and the first to demonstrate the developmental cycle within the tsetse fly. He also satisfied himself that dosing with arsenic had an inhibitory effect on trypanosomes in the blood of animals.

   Itty: Department of Agricultural Economics, Swiss Federal Institute of Technology, ETH Zentrum SOL, 8092 Zurich, Switzerland.
   Trypanosomiasis is a major constraint to livestock production and mixed farming in Africa. Due to the prevalence of trypanosomiasis and skin diseases in much of Zaire, it was concluded that cattle production was only feasible through the introduction of trypanotolerant cattle. This study examines the economics of village cattle production using N’Dama, a trypanotolerant breed, in a tsetse affected area which was until recently devoid of cattle. A social-level economic analysis and a private-level financial analysis were performed using a herd model. The study reveals that cattle production was profitable socially and privately, and that the cattle lease scheme provided substantially higher returns than if farmers had to purchase all their stock. The introductory scheme was successful as cattle are now part of the farming system and their numbers have been increasing. Results suggest that introducing N’Dama cattle into village farming systems of areas with no tradition in cattle husbandry is feasible and profitable.

   Itty: Department of Agricultural Economics, Swiss Federal Institute of Technology, ETH Zentrum SOL, 8092 Zurich, Switzerland.
   This study examines the economics of the village production of trypano-tolerant cattle recently introduced in a trypanosomiasis affected area of Togo that was previously virtually devoid of cattle. Social-level economic and private-level financial analyses are performed using a herd model. The results show that cattle production was profitable to society and to cattle owners but that private returns were especially vulnerable to alterations in costs of inputs, e.g. cattle purchase,
veterinary care. Private returns were only greater than the opportunity cost of capital because such inputs were highly subsidised. Foreign financed subsidies did not enhance farmers' participation in the development process and distorted the incentive structure. In countries such as Togo where trypanotolerant breeds are available, importation of N'Dama cattle would need careful appraisal as the costs incurred depress social returns.


The historical occurrence and current situation with regard to human trypanosomiasis in the Bouenza region, south-west Congo, is outlined. Although written records of the disease are rare, at least three outbreaks were recorded in the seventeenth century in the neighbouring regions of Niari and Louozi, and in the southern and western provinces of the ancient kingdom of Kongo where sleeping sickness is said to have ravaged the population for 50 years. Caravans are known to have skirted round much of the Bouenza region to escape the epidemic in the eighteenth century. At the beginning of the twentieth century, discovery of the pathogenic agent led to the setting up of a research laboratory in Brazzaville. However, a flare-up of the disease occurred in Bouenza and the surrounding areas in 1907. Mobile health teams were set up by the colonial administration to screen the rural population and arrange treatment, but since this service has disappeared there has been a dangerous increase in a whole range of endemic diseases. A heartfelt plea for re-establishment of mobile teams is made. Other suggestions to help the present situation include distancing the banana trees from the villages to avoid the tsetse flies which infest them in the wet season, weeding the areas around dwellings, penning the pigs, using tsetse traps, and installing wells in villages.


Barrett: NRI, Central Avenue, Chatham Maritime, Chatham, Kent, ME4 4TB, UK.

The potential contribution of social scientists to planning and appraising operations to control the transmission of animal trypanosomiasis by tsetse flies is illustrated by three examples. The first considers aspects of social feasibility in the design of tsetse control programmes involving community participation. The second concerns the scope for improving the linkage between tsetse control operations and planning sustainable land use in tsetse-freed areas, with emphasis on the social aspects of dealing with potential environmental degradation associated with overstocking with cattle. The final example concerns the potential problem that, while farmers may benefit substantially from tsetse control, some of the poorest farmers may not only not benefit but may also be disadvantaged by the schemes. From an understanding of social structures and processes, the social scientist can provide a perspective on these problems which should be useful, relevant and cost-effective in the planning and appraisal of tsetse control and related development projects. Increased attention to social
aspects of animal trypanosomiasis control appears justified.


ILRAD, P.O. Box 30709, Nairobi, Kenya.
The biotechnology work carried out at ILCA and ILRAD is briefly described under the headings: animal breeding (embryo transfer technology, animals of defined genotypes, identical twin cattle, trypanotolerant cattle, bovine chimeras); animal genetics (resource cattle populations); genetic markers of trypanotolerance (marker-assisted selection, from markers to genes, transgenesis, DNA fingerprinting and conservation); diagnosis (new diagnostic tests, parasite antigen detection tests, parasite DNA detection tests); vaccination (current vaccination against theileriosis, new vaccines for theileriosis, vaccination against trypanosomiasis).

### 2. TSETSE BIOLOGY

(a) REARING OF TSETSE FLIES
(b) TAXONOMY, ANATOMY, PHYSIOLOGY, BIOCHEMISTRY


Department of Biochemistry and Applied Molecular Biology, University of Manchester Institute of Science and Technology, P.O. Box 88, Manchester M60 1QD, UK.
The ultrastructural and biochemical features of the death of the thoracic eclosion muscles in *Glossina morsitans* are described. Sequential dismantling of the myofilaments occurs within an intact membrane. Trypan Blue is excluded from muscle fibres for the duration of the degeneration process. There is a great deal of RNA production that increases during muscle destruction, nuclei and mitochondria remain active and rough endoplasmic reticulum proliferates. No sign of DNA breakdown to smears or ladders is apparent. These results suggest apoptosis rather than necrosis in the eclosion muscles.


Elsen: Laboratory of Entomology, Prince Leopold Institute of Tropical Medicine, 155 Nationalestraat, B-2000 Antwerp, Belgium.
The genetics of two laboratory colonies of *G. p. gambiensis* were characterised by C-banding and isoenzyme studies. The colonies, derived from flies collected in the same locality, had different histories in the laboratory and different susceptibilities to trypanosome infection. Although the two lines were also found to differ in the frequencies of chromosome and isozyme variants, the variation was not enough to put their specific status in doubt; it was probably the result of genetic drift since the foundation of the colonies.


Grubhoffer: Institute of Parasitology, Czech Academy of Sciences, Branisovská 31, 37005 České Budejovice, Czech Republic.
Lectin activities were studied in five different species of tsetse flies (*G. tachinoides*, *G. palpalis*, *G. austeni*, *G. pallidipes*, *G. brevipalpis*). Different native or enzymatically treated human or animal red blood cells were used to detect haemagglutination activity in midgut extracts. Two inducible lectin systems in the midgut of *G. tachinoides* were distinguished.


ODA Tsetse Research Project, Box CY52, Causeway, Zimbabwe.

Tsetse flies *Glossina morsitans morsitans* and *G. pallidipes* were marked and released within 12 h of emergence at Rekomitjie Research Station, Zambezi Valley, Zimbabwe, and on Redcliff Island, Lake Kariba. Ovarian dissections were performed on recaptured flies and on wild collected samples. At Rekomitjie > 90% of female *G. m. morsitans* were inseminated by age 4 days and *G. pallidipes* by 7 days. For both species at both sites the largest oocyte, for flies in ovarian category zero, increased in length approximately linearly for about the first 6 days and was ovulated at c. 6-8 days. The largest oocyte grew significantly more slowly in later cycles. For *G. m. morsitans*, but not for *G. pallidipes*, the rate increased with temperature; the rates were always higher than observed in the laboratory. At Rekomitjie, for both species and at a mean screen temperature of 22°C, the first larva was produced at c. 18 days and subsequent larvae at 11-day intervals; the intervals decreased with increasing temperature by c. 0.5 days/°C. On Redcliff Island the intervals for both species were 2 days shorter than at Rekomitjie at any given screen temperature and were sometimes as short as 7 days. The length of the larva *in utero* increased exponentially during pregnancy.


Ochanda: Department of Biochemistry, University of Nairobi, P.O. Box 30197, Nairobi, Kenya.

The efficacy of bloodmeal digestion in teneral *G. m. centralis* fed on rabbits immunised with tsetse fly midgut extracts was progressively monitored over a period of 96 h. Flies fed on immunised rabbits showed a reduced rate of bloodmeal digestion as compared to the controls. Although there was insignificant difference in the rate of bloodmeal digestion up to 24 h post-feeding, in later stages of digestion there was quite a significant difference. Polyacrylamide gel electrophoretic patterns of bloodmeal drawn from the posterior sections of the midgut demonstrated that the bloodmeal is completely degraded in the midgut after 96 h in the control flies, while a substantial amount is still undigested in the experimental flies. However, not much difference in the rates of digestion was observed with bloodmeal drawn from the anterior section of the midgut. These results suggest that, when flies are fed on rabbits immunised with tsetse fly midgut extract, there is an impairment in the efficiency of digestion. The anti-midgut antibodies could be interfering with either the induction or the proteolytic activity of the midgut enzymes.

(c) DISTRIBUTION, ECOLOGY, BEHAVIOUR, POPULATION STUDIES

[See also 18: no. 8860.]

8855 **Amsler, A., Filledier, J. and Millogo, R., 1994.** Efficacité comparée de
The efficiency of five traps, unbaited with olfactory attractants, for catching G. tachinoides was compared along the Comoé river, Burkina Faso, during both the cool and hot periods of the dry season. The biconical trap (Challier-Laveissière) gave the best results, while the monoconical trap (Mérot) and the F3 trap (Flint) showed very little efficiency. The Vavoua trap (Laveissière) and the screen trap (Gouteux and Noireau) gave intermediate results. Quantitatively as well as qualitatively, the catches varied over the dry season: the number of flies caught increased during the hot dry season, and more females than males were caught throughout the experiment, especially during the second period.


Mboungou-Mouanda: Centre de Recherches Vétérinaires et Zoo-techniques, Brazzaville, Congo.

A month-long entomological survey was carried out in 1990 in five villages, Inoko, Mimbelli, Mindzoukou, Issombé and Mimputu, situated along the Ibenga river in Enyellé, northern Congo. Two Lancien monoconical traps were used for 16 h per day for 6 days in each village. The results obtained confirmed the presence of Glossina fuscipes fuscipes and G. tabaniformis at a relative density varying between 0.83 and 1.66 flies/trap/day.


Tsetse and Trypanosomiasis Control Branch, Department of Veterinary Services, P.O. Box 8283, Causeway, Harare, Zimbabwe.

Three new trapping devices (M1, M2 and M3 traps) were constructed by modifying F3 and Epsilon traps by the addition of extra external entrances. These were tested and compared with unmodified F3 and Epsilon traps for tsetse flies in Zimbabwe. The most effective was the M3 trap, a modified Epsilon trap with three entrances, which caught 80% and 73% more male and female Glossina morsitans and 110% and 39% more male and female G. pallidipes respectively than the standard Epsilon trap. The mean daily catch for the standard Epsilon trap, which was the least effective of the traps tested, was 6 male and 24 female G. morsitans and 76 male and 199 female G. pallidipes.


Glogauer Weg 12, D-84130 Dingolfing, Germany.

The behaviour of G. tachinoides at odour-baited and unbaited biconical traps was studied by visual observation and the use of electrocuting nets. The phenolic fraction of cow urine in combination with 1-octen-3-ol served as olfactory attractant.
attractant. Both baited and unbaited traps attract significantly more females than males in their vicinity, but thereof only 18% of males and 9.3% of females enter the trap. These percentages are termed trap efficiencies; odour baiting increases them significantly, by 66% for males and 94% for females. Long-range attractiveness and efficiency of baited and unbaited biconical traps are less for G. tachinoides compared to G. morsitans group flies observed elsewhere. As 94% of landing G. tachinoides alight on the blue cone, it is recommended for tsetse campaigns that insecticide impregnation be restricted to the blue cone of the biconical trap. Up to 74% of landing tsetse alight on the lower half of the blue cone, which forms only 12% of the whole external trap surface. 500 individuals of G. tachinoides were examined for age, nutritional status and trypanosome infection. Of those tsetse flies approaching biconical traps, there is a significant tendency for the younger and those with lower body weight and lipid content to enter the traps. These three physiological groups of tsetse tend also to be more strongly attracted by odours. No significant difference could be shown between trypanosome-infected and uninfected G. tachinoides relative to trap-orientated behaviour and nutritional status.

3. TSETSE CONTROL (INCLUDING ENVIRONMENTAL SIDE-EFFECTS)

[See also 18: nos. 8848, 8855, 8857, 8858, 8890.]


Department of Civil Engineering, University of Strathclyde, Glasgow G4 0NG, UK.

The paper describes a methodology for specifying the deposition of spray droplets on flying insects and insects at rest on a quantitative basis. Processes of deposition by sedimentation and inertial impaction are viewed in terms of a deposition velocity defined by \( v = v_a + E u a \), in which \( v \) is the droplet settling velocity, \( u \) the air speed relative to the insect, \( E \) a collection efficiency associated with inertial impaction and \( a \), \( a \) signify horizontal and vertical projections of a representative trapping area. Published data on the deposition of small droplets (diameter < 20 \( \mu \)m) on mosquitoes show that the trapping efficiency is dominated by inertial impaction and it is assumed that the collection efficiency is specified by \( E = St^2/(St + 0.2)^2 \) in which St is the Stokes number. Analysis of a second data set regarding the deposition of larger drops on flying locusts shows that the dependence of the observed collection efficiency on droplet size can be explained satisfactorily by this simplified approach. Further, it demonstrates that a characteristic length which forms an essential component of the Stokes number is matched to the general size of the insect. The paper concludes with an analysis of a further data set concerning the deposition of small drops onto resting tsetse flies: this provides insight into the effective air speed controlling the deposition process.


Baylis: Department of Arbovirology, Institute for Animal Health, Ash Road, Pirbright, Surrey GU24 0NF, UK.
An experiment was conducted at Galana Ranch, Kenya, which examined, under natural conditions, whether treatment of oxen with insecticidal pour-ons affects the success with which tsetse flies, *G. pallidipes*, feed on them. An incomplete ring of electric nets was used to sample *G. pallidipes* approaching and departing from oxen that were either untreated, or treated 6-12 days previously with pour-ons containing deltamethrin or cypermethrin. Eight animals of each treatment were used. There was no evidence suggesting that pour-on application affected the number of *G. pallidipes* attracted to oxen. A positive relationship was observed between the number of *G. pallidipes* that approached an ox and the frequency with which it made anti-fly movements. There was also a significant, negative relationship between the rate of anti-fly movements and the proportion of *G. pallidipes* that fed on the oxen. However, there was no effect of pour-on application on either the rate of anti-fly movement or on the proportion of tsetse that fed. It is concluded that even recent application of deltamethrin or cypermethrin pour-ons to an ox does not affect the ability of *G. pallidipes* to feed; and that the feeding success of *G. pallidipes* is density-dependent because when more tsetse approach an ox its rate of anti-fly movements increases and the proportion of tsetse that feed decreases.


2% Cypermethrin high cis (Ectopor) as a pour-on was tested for the control of tsetse flies at Buhuri Cattle Farm near Tanga Town. The farm harbours three species of *Glossina*, namely *G. pallidipes*, *G. morsitans* and *G. brevipalpis*, which transmit *Trypanosoma congolense*, *T. vivax* and *T. brucei* amongst the farm cattle. Ectopor was applied on a few selected animals in the farm at an interval of 2 weeks. The flies were collected and their apparent densities recorded; these decreased as the application of Ectopor continued. Moreover there was a marked decline in trypanosomiasis cases among the experimental animals for a period of about 6 months after first application.

**4. EPIDEMIOLOGY: VECTOR-HOST AND VECTOR-PARASITE INTERACTIONS**

[See also 18: nos. 8847, 8852, 8860, 8880, 8947, 8948.]


An index of epidemiological risk was developed for the foci of human African trypanosomiasis (HAT) in the forest zone of Côte d'Ivoire, based on the following characteristics of *Glossina palpalis palpalis* populations: daily survival rate, apparent density of teneral males and females, and frequency of human-fly contact. The index agreed well with HAT prevalence. It varied according to ethnic groups and with seasonal changes in agricultural activities and fell rapidly to zero following the start of an anti-vector control campaign. Further studies in different biogeographical zones are desirable in order to substantiate the validity
of the index.


Maudlin: Tsetse Research Group, Department of Veterinary Medicine, University of Bristol, Bristol BS18 7DU, UK.

The routes of establishment and maturation of *Trypanosoma brucei* and *T. congolense* in tsetse flies remain contentious. This review is concerned rather with the mechanisms involved in the differentiation of trypanosomes within the vector. To date, work has concentrated on factors controlling refractoriness/susceptibility to establishment of midgut infections in tsetse (role of lectin, endochitinase activity of midgut bacterium). The mechanism of maturation appears to be more complex and is profoundly influenced by fly species, fly sex and trypanosome genotype. The possible biochemical mechanisms involved are discussed.


Mehlhorn: Department of Zoology and Parasitology, Ruhr-University Bochum, Universitätstrasse 150, D-44780 Bochum, Germany.

The presence and survival of pathogens inside the gut of leeches bought from German pharmacies were studied by means of light and electron microscopy. Ingested red and white blood cells survived for long periods at both high (22°C) and low (3°C) temperatures and pathogens persisted in large numbers. Protozoan parasites such as *Trypanosoma brucei brucei* were even capable of reproducing inside the gut of the leech. No evidence was seen of the penetration of pathogens into the salivary glands. Nevertheless, transmission is considered possible, especially when the leeches are squeezed or manipulated during attachment to the host.


Institut für Parasitologie und Tropenveterinärmedizin, Freie Universität Berlin, Berlin, Germany.

Tsetse collected in an endemic area of Côte d'Ivoire were examined for intestinal procyclic trypanosomes by three methods: 10% were found infected by the dissection method, 28.5% by microscopic examination of homogenised gut and 47.1% by *in vitro* culture of gut tissues. To test for metacyclic forms, tsetse salivary glands were inoculated into *Mastomys coucha*; this animal test detected two cases of salivary gland infection not found on dissection and subsequent microscopic examination. These results suggest that the dissection method used routinely in the field underestimates *Trypanosoma congoense* and *T. brucei* infections. The possibility of mechanical transmission of trypanosomes in the field was not confirmed. Furthermore, transmission of *T. brucei* from an infected to an uninfected pig using teneral *Glossina palpalis palpalis* did not succeed. *In
vitro cultured trypanosomes were characterised by thin-layer starch-gel electrophoresis. The proportion of ALAT-II to ALAT-III in procyclic forms of *T. brucei* from *Glossina* agreed with that of trypanosomes previously recovered from domestic animals. An ALAT II/ASAT II combination (recently included in the *T. b. gambiense* isoenzyme pattern) is reported for the first time for trypanosomes isolated from *Glossina*. This rare aminotransferase combination was found in 1983 in a *Trypanosoma* isolate from a local pig; trypanosomes with this enzyme pattern are thus still circulating in this area.

5. HUMAN TRYPANOSOMIASIS

(a) SURVEILLANCE  
[See also 18: no. 8847.]


Kloos: 2307 N. Backer Avenue, Fresno, CA 93703, USA.

Epidemiological studies were carried out among 180 randomly chosen settler and 180 non-settler households in the three resettlement schemes of Kishe, Gera and Didessa located in river valleys and highland areas of Illubabor Administrative Region in western Ethiopia. All 628 slides of blood taken by the finger prick method were negative for trypanosomes and no tsetse flies were noted on study team members. However, household questionnaire surveys revealed high livestock mortality from trypanosomiasis in the lowland schemes of Kishe and Didessa. More epidemiological, including entomological, studies are needed to determine transmission patterns and to evaluate the feasibility of different control strategies.


National Sleeping Sickness Control Programme, Ministry of Health, P.O. Box 1241, Jinja, Uganda.

Diagnosis of sleeping sickness is difficult since the clinical manifestations are highly variable and unspecific. The available methods of parasitological and serological diagnosis are described. Parasitological diagnosis is difficult because of the frequently low and fluctuating parasitaemia, and most of the available serological tests can only detect the presence of antibodies to trypanosomes and therefore only indicate exposure to trypanosomes rather than active infection. Treatment remains largely dependent on the use of suramin and pentamidine for early stage cases and Mel B (melarsoprol) for late stage cases with CNS involvement. Recent trials have shown eflornithine (DFMO) to be effective against *gambiense* sleeping sickness.

(b) PATHOLOGY AND IMMUNOLOGY


Bentivoglio: Istituto di Anatomia Umana, Facoltà di Medicina, Strada Le Grazie, 37134 Verona, Italy.

In 1903 Aldo Castellani first observed trypanosomes in the cerebrospinal fluid of
sleeping sickness patients. A brief biographical sketch of Castellani is given, and the etiology, clinical features and neuropathological picture of sleeping sickness are described. The molecular and cellular mechanisms of the interplay between the parasite and the host are reviewed with particular reference to recent findings in the experimental rat model of African trypanosomiasis.


Kristensson: Division of Neurodegenerative Disease Research, Department of Neuroscience, Karolinska Institute, S-17177 Stockholm, Sweden.

In African trypanosomiasis the occurrence of antibodies to interferon-\(\gamma\) (IFN-\(\gamma\)) was studied in both humans and experimental rats. Sera from patients infected with \textit{Trypanosoma brucei gambiense} showed increased levels of antibodies to IFN-\(\gamma\) as compared with controls from the same regions in Africa. In rats infected with \textit{T. b. brucei} an early appearance of IFN-\(\gamma\)-producing spleen cells was observed, followed by an increase in levels of antibodies against IFN-\(\gamma\) in the sera. Previously, IFN-\(\gamma\) has been found to play a crucial role in trypanosome infections in rats by promoting proliferation of \textit{T. b. brucei}. The appearance of antibodies to IFN-\(\gamma\) in humans, as in rats, indicates that this cytokine is produced also in the human infection. Its parasitic growth-stimulating and pathophysiological effects on the organism may be reduced by the antibodies.


Brandenberger: LPPE, Strasbourg, France.

Plasma prolactin and renin were measured at 10 min intervals, and sleep was recorded polygraphically throughout the 24 h period, in six sleeping sickness patients (Congo) and in five healthy controls (Côte d'Ivoire). Prolactin profiles of the sleeping sickness patients did not show any systematic variations throughout the 24 h, reflecting the disappearance of the circadian alternation of sleeping and waking in these patients. However, the onset of REM sleep occurred in the descending phases of prolactin pulses, as in normal subjects. Renin release was increased during sleep episodes and exactly reflected the sleep stage distribution, with NREM sleep occurring during the ascending phases and REM sleep during the descending phases of the oscillations as in normal subjects. During some waking periods, renin showed sharp increases, which reflected slow wave activity observed in some patients upon awakening from slow-wave sleep.


Buguet: Physiology Department, CRSSA, B.P. 87, F-38702 La Tronche Cedex, France.

Eight sleeping sickness patients in Côte d'Ivoire at an early stage of meningoencephalitis were observed during two 24 h periods. In the most severely
sick patient, the structure of sleep and wakefulness was altered due to numerous bursts of slow waves invading the EEG trace, and distinction between stages 1 and 2 and between stages 3 and 4 was not possible. In the other patients, sleep patterns were distinct. Disturbances of the circadian sleep-wake cycle organisation were observed in all eight patients and were proportional to the severity of the disease. Polygraphic traces of another ten patients from Congo also showed disorganisation of the sleep-wake circadian alternation proportional to the degree of severity of the clinical symptoms. Hypersomnia was not seen, total sleep time being equal to or less than 8 h. Unlike the Côte d'Ivoire patients, those from Congo had only small amounts of slow-wave sleep. In the most severely sick, REM sleep represented almost the only sleep state.


Buguet: Physiology Department, CRSSA, B.P. 87, F-38702 La Tronche Cedex, France.

The plasma melatonin profile in nine sleeping sickness patients (Brazzaville, Congo) at an early stage of meningoencephalitis and six controls (Abidjan, Côte d'Ivoire) was determined from blood sampled hourly for 24 h through an indwelling catheter. In patients, the circadian periodicity of the sleep-wake cycle was disturbed proportionally to the degree of severity of the disease. Patients' plasma melatonin profiles showed a marked rhythm with undetectable levels during the daytime, and night-time concentrations similar to those of controls. The disappearance of the circadian sleep-wake cycle despite the normal secretion of the endogenous synchroniser melatonin suggests a dysregulation at the level of the main pacemaker of the circadian timing system, the supra-chiasmatic nuclei.


Hamon: Laboratoire de Psychologie Expérimentale et Comparée, Université de Nice Sophia-Antipolis, B.P. 209, Nice, France.

The late components (N2 and P3 waves) of ERP (which are related to stimulus evaluation and decision making processes) and the contingent negative variation (which is linked both to attention and motivation) elicited by simple and paired auditory stimuli during a sensorimotor task were recorded in 16 patients at the meningoencephalitic stage of *gambiense* sleeping sickness. The latency of N2 and P3 auditory ERP components and the reaction time were significantly increased, while the amplitude of the contingent negative variation and N2 wave were severely decreased in patients compared to control subjects. These findings are consistent with those obtained from totally sleep-deprived subjects.


Radomski: Defence and Civil Institute of Environmental Medicine, Toronto,
Blood samples were taken hourly over a 24 h period via an indwelling catheter from eight sleeping sickness patients at an early stage of meningoencephalitis and from six healthy controls and analysed for plasma cortisol and prolactin levels. Disruptions were seen in the circadian rhythms of cortisol and prolactin secretion in all patients, with more severe disruption in the three most severely sick patients who had been shown previously to have the most disturbed sleep-wake cycles.

(c) TREATMENT


Plasma concentrations of pentamidine were measured up to 1-8 months after a single 2 h i.v. infusion of 3.0 to 4.8 mg/kg pentamidine isethionate in 11 patients with late stage T. b. gambiense sleeping sickness. Maximum plasma drug concentrations varied between 713 and 2461 nmol/l. After termination of infusion, a rapid distribution phase over 10 min was followed by a slower distribution phase and an elimination phase prolonged over weeks to months. The 'terminal' elimination rate constant could be determined in six patients and subsequent kinetic calculations showed a three- to fourfold variation in plasma clearance and 'terminal' half-life (median 1126 (range 553-2036) ml/min and 265 (107-446) h, respectively). The median apparent volume of distribution (Vss) was 11,850 l. Renal clearance accounted for a median of 11% of total plasma clearance, indicating that metabolism is a major route of pentamidine elimination in man. Side effects were few and mild and a slight or moderate decrease in blood pressure was the most common registered adverse reaction observed in four subjects. The prolonged elimination of pentamidine seems inconsistent with the present recommended dosage regimen of pentamidine for treatment of trypanosomiasis of 7 to 10 parenteral doses given once daily or every second day.


A novel method for the control of Trypanosoma brucei gambiense trypanosomiasis was evaluated in an endemic focus of Zaire where a high incidence has persisted despite massive participation in active case-finding surveys based on lymph node puncture. All inhabitants of three villages were examined with a card agglutination serological test (CATT), and parasitological examinations were performed on those who were CATT+. Individuals in whom
we detected trypanosomes were treated as usual. A lumbar puncture was carried out on CATT+/parasitology− subjects; those whose cerebrospinal fluid showed more than 3 white blood cells (WBC) per mm³ were treated with a full course of melarsoprol while those with a CSF WBC count between 1 and 3 per mm³ were given a single injection of diminazene (7 mg/kg). Three such surveys were performed, with a 6-month interval, during which 282 `serological suspects' received diminazene, 39 `clinical cases' were given melarsoprol and 82 `parasitological cases' were treated according to standard protocols. The annual incidence of trypanosomiasis decreased rapidly from 10.4-41.1/1000 inhabitants (mean: 17.6/1000) during the 10 years before the intervention to 1.1-2.6/1000 (mean: 1.7/1000) in the 3 years following the intervention. No major adverse effect was seen with diminazene. Among the 282 serological suspects, an elevated CSF WBC count was later documented in 12 individuals, who were all cured with melarsoprol. The incidence increased 5 years after the intervention (7.1/1000 in 1992), which might have been avoided had we carried out similar interventions in adjacent foci.


Institute of Tropical Medicine, 155 Nationalestraat, B-2000 Antwerp, Belgium.

From the first decade of this century arsenicals have been the most universal and most effective drugs for all cases of sleeping sickness. Melarsoprol, introduced in the 1940s, remains the most universal of these compounds. However, resistance of trypanosomes and toxicity that may be fatal for the patient are two major shortcomings. Pentamidine, suramin and Berenil are active only in the first stage of the disease, when the parasites are confined to blood and lymph. Nifurtimox taken orally for 1–2 months and alpha-difluoromethylornithine (α-DFMO) with an administration scheme spread over 5 weeks, including 14 days of i.v. injections, provide interesting alternatives for all cases, since they reach the central nervous system. However, DFMO is known to be less active against Trypanosoma brucei rhodesiense.

Imidazoles, new arsenical derivatives and antimitotics have been successfully tested in experimental models. Combinations of drugs with additive or potentiating effects mainly based on inhibition of decarboxylase enzymes or exposure to oxidative stress appear promising.

6. animal trypanosomiasis

(a) SURVEY AND DISTRIBUTION


Daniel: Veterinary and Livestock Studies Division, NITR, P.M.B. 03, Vom-Jos, Plateau State, Nigeria.
The prevalence of trypanosomosis was studied during April-June 1991 in sheep and goats kept peridomestically in Alkaleri and Gombe local Government areas of Bauchi State in Northern Nigeria. A total of 615 animals, consisting of 258 sheep and 357 goats, were examined for trypanosome infection. Of this total, 19 (7.4%) sheep and 18 (5.0%) goats were positive, giving a total infection rate of 37 (6.0%), 22 being positive with *Trypanosoma vivax*, 9 with *T. congolense* and 6 with *T. brucei*. In order to elucidate the most appropriate tool for surveying trypanosomosis in small ruminants under Nigerian field conditions, the sensitivity of four techniques currently in use for the parasitological diagnosis of trypanosomosis was investigated. The concentration methods (haematocrit centrifugation and buffy coat method) were more accurate than the standard trypanosome detection methods (wet film and thin film). Due to the prevalence of the disease, sheep and goats must be treated as well as cattle in the region.


CRTA-CIRDES, 01 B.P. 454, Bobo-Dioulasso 01, Burkina Faso.
This report summarises the work carried out with financial support from UREF on the introduction of new, more sensitive techniques for the diagnosis of trypanosomiasis at CRTA, in cooperation with ILRAD and other laboratories. A new protocol for the ELISA technique, giving improved reproducibility of results, has been introduced, and an antigen ELISA test has been evaluated. An important development has been the introduction of DNA probes for the diagnosis and characterisation of trypanosomes, particularly in the tsetse vector. Work on nine stocks of *Trypanosoma congolense* from CRTA has been carried out at the University of Bordeaux II to compare radioactive and non-radioactive marking of probes, and at ORSTOM-Montpellier to gain familiarity with chemoluminescent marking. This latter technique has been adopted by CRTA and will be used in future epidemiological studies.


Komoin-Oka: Laboratoire de Pathologie Animale, LANADA, B.P. 206, Bingerville, Côte d'Ivoire.
Compared with numerous studies of trypanosomosis in domestic animals, few such studies have been carried out on wild animals in West Africa. Preliminary results on the comparison of three detection methods (thin smears, antigen ELISA and kit for in vitro isolation of trypanosomes (KIVI)) in wild animals (elephant, buffalo, roan antelope, hartebeest, waterbuck and warthog) in the Comoé Game Reserve in Côte d'Ivoire confirm the presence of trypanosomes; however, no
accurate identification of species has been possible, but work is in progress to clarify the taxonomic status of stocks isolated by KIVI.


Masake: ILRAD, P.O. Box 30709, Nairobi, Kenya.

Four Boran cattle were infected with *T. brucei* using *Glossina morsitans centralis* and were left untreated throughout the experimental period of 18 months. During this period, sequential blood samples were collected and examined for the presence of anti-trypanosome antibodies and their antigens. Using the buffy coat technique (BCT), trypanosomes were detected in 38 (16.3%) of the 233 blood samples. Unlike the BCT, antigen-detection enzyme-linked immunosorbent assay (Ag-ELISA) diagnosed infections in 189 (81.1%) of the blood samples. These results were supported by the presence of anti-trypanosome antibodies in the same samples. Thus Ag-ELISA was 5.5 times more sensitive than the BCT. Towards the end of the observation period, *G. m. centralis* were fed on the aparasitaemic cattle to determine whether they still harboured the infection as the persistent antigenaemia seemed to suggest. Bloodmeals from the four cattle were infective to tsetse, thus emphasising the importance of Ag-ELISA in diagnosis of sub-patent infections.


Department of Parasitology, Animal Health Research Institute, Dokki, Cairo, Egypt.

Blood samples were collected from 100 dromedary camels aged 3-4 years slaughtered at El Monieb abattoir and from 25 mice infected 2 days earlier with *T. evansi* isolated from a naturally infected camel. Each sample was tested for trypanosomes by microscopic examination of wet smears as well as by the silicone centrifugation technique (SCT), m-AECT and the HCT. The results indicated that the SCT is as sensitive as the other concentration methods for detection of low parasitaemia. The SCT, however, is advantageous over the other concentration techniques in that it is simple, rapid and can be performed within 15 min, enabling early diagnosis of the disease. Moreover, it can be reused several times, stored at room temperature and no special buffer is needed to run the assay.

(b) PATHOLOGY AND IMMUNOLOGY

[See also 18: no. 8906.]


Agag: Animal Health Research Institute, Dokki, Cairo, Egypt.

Five 7-12 year old dromedaries in Qena Governorate, Egypt, naturally infected with *T. evansi* showed weakness, emaciation, lachrymation, pale to icteric mucous membranes, dry skin and intermittent fever; two cases also suffered
subcutaneous oedema of the limbs and abdomen. The animals were treated i.v. with 10 ml Trypamidium (isometamidium chloride) and were cured within 1 month.


Akinbamijo: Department of Animal Production Systems, P.O. Box 338, 6700 AH Wageningen, Netherlands.

The effects of infection with *T. vivax* in mid- or late pregnancy on food intake and utilisation, liveweight changes, abortion rate and lamb growth rate were investigated in West African Dwarf ewes at Ibadan, Nigeria, in 1990. Rate of liveweight gain by ewes infected during mid-pregnancy (IMH) was 16 g/day compared with 33 and 37 g/day for the uninfected ewes offered medium (CM) or high (CH) plane diets. Although digestibility coefficients were not affected, intake of digestible organic matter was higher in CH ewes than in IMH and CM ewes. Nitrogen retention at mid-pregnancy on a metabolic size basis was higher in CH ewes than in CM and IMH ewes. Lamb birth weight and survival rate were lower in infected ewes. Lambs from ewes infected in mid-pregnancy (IMH) and in late pregnancy (ILH) had mean birth weights of 1.4 and 1.0 kg compared with those from CM and CH ewes, which had mean birth weights of 1.9 and 2.0 kg respectively. Observed survival rates were 63, 15, 75 and 80% for lambs nursed by IMH, ILH, CM and CH ewes respectively. During the first 6 weeks postpartum, lamb growth rate in all groups did not differ. However, during weeks 7-12 postpartum, lambs nursed by IMH ewes had significantly lower growth rates. Weaning weight was also lower in lambs from IMH dams (5.0 kg) than in lambs from CM and CH dams (7.1 kg). Infection during late pregnancy was more severe and all infected ewes lost weight due to reduced feed intake and fever. *T. vivax* infection in sheep is responsible for reproductive wastage, abortion, poor lamb growth and ewe mortality.


Akinbamijo: Department of Animal Production Systems, P.O. Box 338, 6700 AH Wageningen, Netherlands.

The effects of trypanosomiasis on digestible organic matter intake, milk yield and composition, dam liveweight changes during lactation and lamb growth rates were investigated at Ibadan, Nigeria, in 1991/92, using 20 West African Dwarf sheep nursing single lambs. Although digestibility coefficients were affected neither by infection nor by level of feed intake, organic matter intake during early and late lactation was significantly lower in infected dams. Nitrogen retained in late lactation was lower in infected animals due to reduced feed intake. Mean daily milk yields were not affected by the infection during early lactation; however, during the second half of lactation, average daily milk yields were significantly lower in infected animals than in uninfected controls. Variations in milk component concentrations between experimental groups did not attain statistical significance throughout lactation. While control ewes on a high plane
of nutrition (CH) gained 12.1 g/day, infected ewes (IH) and uninfected control ewes on a medium plane of nutrition (CM) lost 45 and 5.4 g/day respectively during lactation. Liveweight gain in the lambs was not affected by infection in the dams. This study demonstrated reduction in feed intake, late lactation milk yield and dam liveweight gain with no adverse effect on digestibility coefficients, milk composition, early lactation milk yield and lamb weight gain during *T. vivax* infection of lactating ewes.


Elhassan: Pathology Division, NITR, Kaduna, Nigeria.

Ten West African Dwarf ewes were inoculated with *T. vivax* and, at varying intervals, treated subcuratively with diminazene aceturate to maintain the infection. Soon after infection all ewes had anoestrus for 40 to 96 days and five died by day 110 p.i. Compared to control animals, infected ewes had prolonged low levels of plasma progesterone until recovery or death. However, no gross or histological lesions were detected in the endocrine or reproductive organs. Of the survivors, the five that were aparasitaemic subsequently became pregnant and had normal gestations.


Gaidulis: ILRAD, P.O. Box 30709, Nairobi, Kenya.

A method was developed to detect expression of macrophage-derived cytokines at the mRNA level using a quantitative PCR. Expression of TNF-α was monitored throughout experimental infection of cattle with either *Trypanosoma congolense* IL 1180, which causes a gradual drop in PCV, or *T. vivax* IL 2337, which causes a severe acute drop in PCV. TNF-α expression was induced by both infections but during the early stages of infection was far more pronounced in the *T. vivax* infection.


Harrus: School of Veterinary Medicine, Hebrew University of Jerusalem, P.O. Box 12, Rehovot 76-100, Israel.

Trypanosomiasis, caused by *T. congolense*, was diagnosed for the first time in Israel in two boxer dogs imported from Kenya. The dogs developed clinical signs two days after arrival and succumbed to the disease within 4 days. The major clinical and clinicopathological findings included anaemia, haemorrhages, lymphadenomegaly, hepatosplenomegaly and neurological signs. Histopathology showed lymphocytic-plasmacytic infiltration in the skin, brain, meninges, kidney and liver. The dogs were suspected of having been in the chronic stage of the disease when they left Kenya, and the acute flare-up and severity of the cases presented may be explained by the stress initiated by air travel and environmental changes to which the dogs were exposed.

Department of Veterinary Medicine, Veterinary College, Gujarat Agricultural University Campus, Anand 388 001, Gujarat, India.

Eight donkeys infected with *T. evansi* showed listlessness, staggering gait and subcutaneous oedema of the head and forelegs, together with a rise in temperature and in respiration and pulse rates. Blood samples showed significantly decreased haemoglobin concentration, PCV and total erythrocyte count, and little improvement was seen 2 weeks after treatment with diminazene aceturate (3.5 mg/kg). Hypoglycaemia and elevated blood urea nitrogen were also observed, the latter returning to normal after treatment.

Hugh-Jones: Department of Epidemiology and Community Health, Louisiana State University, Baton Rouge, LA 70803, USA.

A total of 854 cattle, equivalent to 1099 animal-years, were monitored over a period of 2 calendar years (June 1990-June 1992) in four villages in the Didessa Valley (western Ethiopia) to assess possible associations between the tsetse control programme and the health and productivity of local Zebu cattle kept under traditional management. The four villages had different levels of trypanosomosis prevalence. The initial and final cattle population compositions showed that male adult cattle accounted for the highest proportion in all villages as they were kept primarily for draught power. Standardised mortality rates differed among villages. The risks of cattle dying in the tsetse-unprotected villages ranged from 4 to 9 times higher than in the tsetse-protected village (Meti). Recorded calving rates were 81% for the single protected village and 64% for the highest of the unprotected villages. The estimated offtake rates were in opposite directions: a positive 16% in Meti (tsetse-protected village) versus a negative 18% in the nearest tsetse-unprotected village to Meti. Thus, there are strong suggestions that the tsetse control has affected the health and productivity of cattle in the valley.

Kamboj: Department of Veterinary Medicine, Punjab Agricultural University, Ludhiana 141 004, Punjab, India.

Blood examination of two dogs, aged 2 and 4.5 years, with anaemia, raised temperature and corneal opacity, revealed *Trypanosoma evansi* infection. They were treated s.c. with quinapyramine prosalt at 0.025 ml/kg bodyweight.

Karram: Department of Animal Medicine, Faculty of Veterinary Medicine, Assiut University, Assiut, Egypt.

Five hundred camels aged 10-13 years from Assiut Governorate, Egypt, were examined for infection by *T. evansi* and microfilariae. *T. evansi* was detected in 15 camels and microfilariae in 12. Clinical signs of trypanosomiasis included emaciation, pale mucous membranes, dry coat, intermittent fever, weakness and lachrymation. Blood samples showed a significant decrease in haemoglobin and PCV, and normocytic hypochromic anaemia was associated with leucocytosis, eosinophilia and monocytosis. The infections were successfully treated with an
i.v. dose of 50 ml of 10% solution of Naganol (suramin) followed by a further injection 30 days later.


Kumi-Diaka: Department of Population Medicine, Ontario Veterinary College, University of Guelph, Guelph, Ontario, Canada.

Over a period of 6 years (1976-81), 4730 bulls between 2.5 and 11 years of age were examined in various abattoirs in Northern Nigeria. Gross lesions were seen in the genitalia of 270 (5.7%). Haemoparasites were found in 1150 (46%) of 2500 bulls from which blood samples were taken: *Babesia bigemina* in 539 (21.6%), *Anaplasma marginale* in 720 (28.8%), *Trypanosoma congolense* in 89 (3.5%) of which 24 had testicular degeneration, *T. vivax* in 132 (5.3%) of which 28 had testicular degeneration. Twenty-one bulls with mixed infections of *T. congolense* and *T. vivax* were dehydrated, cachectic and in poor body condition; 11 with *T. congolense* and *B. bigemina* all showed testicular degeneration. Morphological abnormalities were more common in the semen of bulls with testicular degeneration.


Fifteen dogs of Liberian origin were experimentally infected with trypanosomes by the bite of infected tsetse flies. Those infected with a *T. congolense* stock originally from Côte d’Ivoire developed chronic, asymptomatic infection; trypanosomes disappeared from peripheral blood after 218 days. Those infected with a *T. congolense* stock originating from cattle in Togo developed acute infection which needed trypanocidal treatment. Subsequent infection with *T. brucei* of East African origin resulted in acute or subacute disease, fatal after 15-30 days, in all dogs.


Luckins: CTVM, Easter Bush, Roslin, Midlothian, EH25 9RG, UK.

*T. congolense* develops in the skin of sheep at the site of inoculation of metacyclic trypanosomes, forming a chancre containing large numbers of parasites. By cannulating the afferent and efferent lymphatic ducts draining the skin and regional lymph node, the progressive development and migration of trypanosomes from the chancre was monitored and the expression of metacyclic antigen types (M-VATs) was determined. The kinetics of development of parasitosis in the afferent and efferent lymph was similar. Trypanosomes were detected in lymph 5-6 days after the inoculation of cultured metacyclic trypanosomes, at the same time as the chancre first appeared in the skin. The numbers of trypanosomes in the lymph reached their peak levels 8-10 days p.i. and thereafter numbers fell, although there were still many parasites in the lymph after the chancre had regressed. Trypanosomes in the afferent lymph expressed mainly M-VATs and the absolute numbers of four M-VATs which were
monitored increased up to 9 days p.i. There was a fall in numbers by day 10, but 92% of the trypanosomes in the afferent lymph continued to express M-VATs. In contrast, trypanosomes from the efferent lymph were found not to express M-VATs, suggesting that a major switch in VAT expression occurs in the lymph node. Specific antibody responses, measured by neutralisation tests, were evident 16-20 days p.i. in afferent lymph but only low levels of antibodies were found in efferent lymph.


Powell: Skirball Institute of Biomolecular Medicine, New York University Center, 540 First Avenue, New York, NY 10016, USA.

Cattle that were inoculated with an antigen derived from the flagellar pocket of *T. b. rhodesiense* and then infected with *T. congolense* and *T. vivax* were compared with unvaccinated cattle when both groups of cattle were placed in regions of Kenya endemic for tsetse flies known to harbour *T. congolense* and *T. vivax*. In one trial, 90 cattle were employed, 40 untreated controls, 30 cattle given prior treatment with samorin, and 20 inoculated with a flagellar pocket (Fp) antigen derived from *T. b. rhodesiense*, with bovine serum albumin as the carrier and alum as the adjuvant. The animals were monitored for parasitaemia, by buffy coat analysis, during one rainy season. The untreated controls had 58% infection, the samorin-treated cattle had 43% infection, and the immunised cattle had 26% infection. Simultaneously, a second trial was conducted using 250 cattle, 100 untreated controls and 150 inoculated with the above antigen, carrier and adjuvant. At the end of the same rainy season, the untreated controls had 22% infection while the immunised animals had 9% infection. In a third experiment, on the same ranch as the latter experiment, ovalbumin was employed as the carrier. After 15 months, or over three rainy seasons, 13% of the untreated controls became infected while of the 177 immunised animals 0.9% became infected. These results are the first report of heterologous immunoprotection against trypanosomiasis in cattle.


Mutayoba: Department of Veterinary Physiology, Biochemistry, Pharmacology and Toxicology, Sokoine University of Agriculture, P.O. Box 3017, Morogoro, Tanzania.

The effect of trypanosomiasis on adrenal function was studied in 10 pubertal Scottish Blackface rams infected with *T. congolense* and nine uninfected controls. Plasma cortisol concentration was measured by radioimmunoassay in samples obtained twice a week for 3 weeks before infection and three times a week for 79 days p.i. There was a significant (*P* < 0.001) increase in cortisol concentration in all the infected rams after the onset of parasitaemia 9-16 days p.i. This was followed by a transient non-significant decrease in cortisol levels between 19 and 41 days and a variable and parasitaemia-dependent increase in cortisol levels between 44 and 79 days p.i. Marked hypertrophy of the zona fasciculata-reticularis, infiltration of mononuclear cells into the cortical and medullary zones,
hyperaemia and focal coagulative necrosis were evident in the adrenal glands of infected rams killed at the end of the study. Trypanosome infection induced a low grade persistent pyrexia, marked anaemia, reduced growth rates and general loss of body condition. These results demonstrate that *T. congolense* infection in sheep causes marked pathological changes in the adrenal cortex and changes in the secretion of cortisol.


Eckersall: Department of Clinical Veterinary Biochemistry, University of Glasgow Veterinary School, Bearsden, Glasgow G61 1QH, UK.

Changes in pulsatile secretion of luteinising hormone (LH) and testosterone and responses to exogenous GnRH were assessed at different stages of *T. congolense* infection in Scottish Blackface rams. Jugular blood samples were collected every 15 min for 6 h followed by immediate injection of GnRH (20 µg i.v.) and further sample collection after 10, 20, 40, 60, 80, 100 and 120 min. This sampling and injection regimen was performed 5 days before infection (day −5) and 23 and 52 days after infection. *T. congolense* infection increased (*P* < 0.05) the mean plasma LH concentration over 6 h on day 23 (3.2 ± 0.2 ng/ml) and decreased (*P* < 0.05) the mean LH concentration on day 52 (1.2 ± 0.2 ng/ml) compared with day −5 values (2.0 ± 0.2 ng/ml). Trypanosome infection induced a rapid decline in plasma testosterone concentration from a mean of 7.5 ± 1.4 nmol/l on day −5 over 6 h to 3.6 ± 0.4 nmol/l (*P* < 0.05) on day 23 and 1.7 ± 0.3 nmol/l (*P* < 0.001) on day 52. The observed decline in plasma LH concentration in infected rams was not associated with reduced sensitivity of the pituitary to GnRH or its ability to release LH, as the LH response to exogenous GnRH was not impaired throughout the period of infection. However, the testosterone response to GnRH-induced LH stimulation was depressed on both days 23 and 52 after infection. It was concluded that the decline in plasma LH concentration in infected rams was caused by reduced GnRH stimulation of the pituitary, whereas the decline in plasma testosterone was partly caused by reduced sensitivity of the Leydig cells to circulating LH.


Mutayoba: Department of Veterinary Physiology, Biochemistry, Pharmacology and Toxicology, Sokoine University of Agriculture, P.O. Box 3017, Morogoro, Tanzania.

To investigate whether the aberrations in adrenocortical and gonadal activity observed in trypanosomiasis may be induced by the refractoriness of the pituitary to hypotalamic liberins, the responses of the pituitary and adrenal glands and the testes to stimulation with ovine corticotrophin-releasing hormone (CRH) were studied in Scottish Blackface rams 23 days (acute phase) and 65 days (chronic phase) after they were infected with *T. congolense*. On both occasions a peak of plasma ACTH was observed within 20 min of the injection of CRH but the rate of
increase in ACTH and the mean peak values in the infected rams were significantly lower ($P < 0.001$) on day 23 but higher ($P < 0.05$) on day 65 than in the uninfected control rams. Plasma cortisol concentration increased in all the rams after the injection of CRH. The rate of increase in plasma cortisol and the mean peak values were not significantly different between the control and infected rams on day 23 but were significantly ($P < 0.001$) higher in the infected rams on day 65. However, the post peak concentrations of ACTH declined more rapidly in the infected rams than in the controls on both days 23 and 65. The plasma concentration of luteinising hormone did not change after the injection of CRH, whereas the testosterone levels showed a delayed response and its concentration increased when plasma ACTH and cortisol concentrations declined in both groups. On day 23, there was a greater increase in testosterone in the infected than in the control rams. These results demonstrate that the responsiveness of the pituitary corticotrophs to CRH is depressed during the acute phase and enhanced during the chronic phase of *T. congolense* infection in rams, whereas the adrenal cortisol response is less affected. The results are also consistent with the hypothesis that the modulation of the pituitary-adrenal axis by infective trypanosomes may exacerbate the changes in testicular steroidogenesis frequently observed in trypanosomiasis.


Department of Veterinary Pathology, University of Ibadan, Nigeria.

Serum and plasma biochemical values were determined in female West African Dwarf sheep experimentally infected with *T. brucei*. The results showed an increase in the values of serum iron, chloride, bicarbonate, inorganic phosphate, creatinine, urea, total protein, globulin and plasma fibrinogen. The serum albumin, albumin/globulin ratio, potassium, copper and magnesium values were depressed. These findings suggest defective re-utilisation of iron in erythropoiesis and probable parathyroid gland, hepatic and/or renal malfunction.


Omotainse: Veterinary and Livestock Studies Division, NITR, Vom, Plateau State, Nigeria.

Four dogs, 7 months to 2 years of age, were inoculated with $2 \times 10^6$ *T. brucei*, while two others were kept as uninfected controls. Parasitaemia was first observed 5-10 days p.i. and was associated with increased temperature, anaemia and oedema. Corneal opacity and partial blindness occurred in two dogs. The plasma levels of ASAT, ALAT, bilirubin, blood urea nitrogen and creatinine increased in the infected dogs, while there was no significant change in alkaline phosphatase or in total plasma protein level. However, a slight increase in fibrinogen level was observed. Dogs with a higher PCV tolerated the infection better than those with lower PCV.

8902 Osaer, S., Goossens, B., Clifford, D.J., Kora, S. and Kassama, M., 1994. A comparison of the susceptibility of Djallonké sheep and West African Dwarf goats to experimental infection with two different strains of *Trypanosoma*
Two cloned strains of *T. congolense*, of West and East African origin, were used to infect by intradermal inoculation two groups of young adult female Djallonké sheep and West African Dwarf goats. For a 3 month period p.i., PCV, parasitaemia, body weight and clinical parameters were followed to evaluate their trypanotolerant nature and to compare the pathogenicity of the two strains of *T. congolense*. Although the West African strain of *T. congolense* was more pathogenic than the East African strain, it seemed that the Djallonké sheep and the West African Dwarf goats, despite high levels of parasitaemia and a concomitant drop in PCV, showed a high degree of trypanotolerance, as reflected by zero mortality and an increase in body weight during 12 weeks of observation.


Department of Medicine, Nagpur Veterinary College, Nagpur 440 006, India.

Twelve dogs were inoculated s.c. with *Trypanosoma evansi* and six were given 4 mg dexamethasone (a corticosteroid widely used in canine practice) daily i.m. for 7 days. Clinical symptoms were similar in the two groups but dexamethasone reduced the prepatent period, increased the severity of the symptoms and resulted in death in a shorter time.


Rowlands: ILCA, P.O. Box 46847, Nairobi, Kenya.

Approximately 320 East African Zebu cows over 36 months of age were monitored monthly from 1986 to 1992 in nine village herds in an area of high trypanosomiasis risk in south-west Ethiopia where there was resistance to all available trypanocidal drugs. Cows were individually treated with diminazene aceturate when their PCV either decreased below 26% and were detected parasitaemic, or when they showed clinical signs of trypanosomiasis. The average annual monthly trypanosome prevalence was 25% (range 18-39%). Average cow body weight was 196 kg but was 16 kg lower on average during 1988, a year when early rains failed. The median calving interval was 463 days, ranging from 379 to 620 days for different years and seasons. Cows detected parasitaemic in more than half of the monthly samples taken over the period 1-150 days post partum had an average calving interval 39 ± 18 (SE) days longer than those not detected parasitaemic. Calving interval was also inversely related to both post partum body weight and to the change in body weight between 1 and 150 days post partum. The median age at first calving was 40.5 months. Heifers detected parasitaemic at least once between 19 and 30 months of age calved at an average age 2.9 ± 1.2 (SE) months older than those not detected parasitaemic. Over 8% of calvings resulted in abortions or still births and there was a significant increase from 7 to 10% in the rate of abortion associated with cases of parasitaemia detected during the last 3 months of pregnancy. Annual abortion rate and annual trypanosome prevalence also appeared to be correlated. Except for the high incidence of abortions, the effects of trypanosomiasis on reproduction
Trypanosoma evansi infection was diagnosed in 30 of 100 Egyptian dromedary camels using blood smears and serochemical tests (formol gel, mercuric chloride) in addition to clinical examination. Infected animals showed emaciation, atrophy of the hump, general weakness, oedema of the ventral parts of the abdomen and occasionally fever. The gross lesions consisted of accumulation of a large amount of serous exudate in the pericardial and peritoneal cavities and pulmonary oedema. Histopathological examination showed degenerative changes, especially fatty change, necrosis, congestion of the central vein and hepatic sinusoids, focal leukocytic aggregation and granuloma formation in the liver. In the kidneys, mild glomerulo- and interstitial nephritis, tubular degeneration and necrosis were seen, while in the lung there was thickening of most of the alveoli with some areas of emphysema.

(c) TRYPANOTOLERANCE
[See also 18: nos. 8845, 8846, 8849.]

Data on five transhumant and four sedentary trypanotolerant Borgou herds in north-east Benin were collected over a period of 5 years. The calving rate was 65.4%, the average age at first calving was 43 months ± 16 days and the average calving interval was 16 months ± 3 days. The total mortality rate was 7.5%, with the mortality rate of adult animals being 3.1% and that of calves 23.1%. The offtake was 11.8%, while the increase in herd size was 3.9%. Comparison of the two livestock production systems revealed significantly lower calf mortality and higher offtake in the sedentary herds. Borgou cattle
are well adapted to their environment and their performance is similar to that of other trypanotolerant breeds living under the same traditional conditions. The productivity index was calculated as being 27.4 kg.


By amplification of pools of DNA representative of different bovine populations with single short oligonucleotide primers of random sequence, we were able rapidly to identify markers which distinguish the two major subspecies of domestic cattle, Bos taurus and B. indicus. One of the marker polymorphisms was found to be in a novel, dispersed DNA sequence which occurs in several ruminant species. The marker will assist in the detection of crossbreeding between Zebu and B. taurus types where this threatens a potentially valuable trypanosomiasis-resistant B. taurus genetic resource in West Africa. In addition, the marker will be useful for exploration of the evolutionary relationships of the major subspecies of domestic cattle. The general approach used to identify population-specific DNA polymorphisms has potentially broad application in definition of species, breeds and populations and will be of generic value in studies of genome evolution.


Trypanotolerance of N’Dama cattle is reviewed and also their resistance to gastrointestinal helminths, ticks and some tick-transmitted diseases.


The main force driving gene mapping in domestic animals is the potential for using gene maps as a means of identifying and mapping the genetic loci responsible for genetic variation in traits of economic importance. The ultimate objective is utilising this information for marker-assisted selection (MAS) using DNA level polymorphisms. This overview considers: (i) the need for MAS, (ii) the relationship of gene mapping to MAS, and (iii) implementing MAS. Examples illustrating the potential contribution of MAS to animal genetic improvement are presented, including introgression of trypanotolerance from N’Dama to other cattle breeds.

(d) TREATMENT


The drugs used for the treatment and prophylaxis of animal trypanosomiasis are tabulated, with details of dilution, injection route, dose, volume of solution to
inject, and an indication of which drugs should be used against which trypanosome species and for which animals. In order to avoid the appearance of resistant strains it is imperative to respect the rules of cleanliness while injecting the drugs, to assess correctly the weight of the animal to be treated and respect the prescribed dosages and dilutions, to treat the animal while it still has enough reserves of energy to help fight the parasite, and to inject by the deep i.m. route for products requiring this method (do not exceed 15 ml per injection site, and massage the site).


**Peregrine:** ILRAD, P.O. Box 30709, Nairobi, Kenya.

A PCR technique for distinguishing different types of *T. congolense* was developed and its sensitivity and specificity evaluated. The technique was used to screen for the diminazene-sensitive clone in trypanosome populations collected from infected goats, both before and after treatment with diminazene acetate. Three groups of five goats each were infected with IL 1180 (diminazene-sensitive, group A), IL 3274 (diminazene-resistant, group B) or both clones simultaneously (group C), and treated with diminazene acetate at a dose of 7.0 mg/kg body weight following detection of trypanosomes. Three other groups of three goats each were similarly infected and kept as untreated controls. All group A animals were cured, while all in group B and four animals in group C relapsed. Trypanosomes were harvested from all animals at regular intervals up to 60 days post treatment. Using the PCR technique, IL 1180 DNA could not be detected in any post-treatment trypanosome DNA sample. It therefore appeared, on the basis of the sensitivity of the DNA detection systems used, that IL 1180 is unable to survive treatment with diminazene acetate when mixed with IL 3274 in goats.


**ILRAD,** P.O. Box 30709, Nairobi, Kenya.

Experiments were carried out in goats to determine the frequency with which diminazene-resistant trypanosomes occur in parasite populations before and after the i.m. treatment of the goats with diminazene acetate. *T. congolense* IL 3274, a diminazene-resistant clone, was used to initiate infections in three groups of five goats. The goats in the first group were treated with diminazene acetate at a dose of 7.0 mg/kg bodyweight within 10 s of infection; one of the goats was cured. All of the second group, which received no treatment, became parasitaemic. The third group of goats received the same dose of drug as the first group but 3 days after all of them were first detected parasitaemic; trypanosomes reappeared in all the five goats. When this third group was treated, the frequency of trypanosomes resistant to the drug dosage was estimated to be less than 1 in 10⁷. The parasites which reappeared after the treatment of these animals were used to infect two additional groups of five goats i.v. The goats in one group
were treated with the same dose of drug as before, within 10 s of infection, and were all cured. In contrast, the five goats in the second, untreated group became parasitaemic. Finally, when the goats in which the infections had relapsed were retreated with diminazene aceturate at the same dose rate, the level of parasitaemia temporarily decreased by at least 10^3 trypanosomes/ml. These findings suggest that diminazene-resistant *T. congolense* occur at low levels in trypanosome populations despite attempts to select for a population resistant to the dose of drug used.


Radioisotope Laboratory, KETRI, P.O. Box 362, Kikuyu, Kenya.

A survey on veterinary drug residues in meat was carried out in the central region of Kenya. A total of 240 samples from different animals were collected from various slaughter houses and meat retail markets in Athi River, Dagoretti and Ngong/Kiserian areas. The samples were tested for antibiotics and trypanocides. From all the samples collected, 144 samples had drug residues. The 90 lean meat samples from Athi River area had significant amounts of isometamidium and homidium ranging from 3.85 µg/g to 14.25 µg/g and from 22.21 ng/g to 302 ng/g of tissue respectively. 20% of kidney samples from this area contained antibiotics, while 40% of kidney samples from Ngong/Kiserian and Dagoretti areas both contained antibiotics. This study reveals that there is a problem of violative drug residues in animal products in Kenya due to misuse of these drugs by the farmers and those concerned with animal production. There is need to determine the extent of the problem and assess the overall effect of the residues to the consumer.


Department of Medicine, Nagpur Veterinary College, Nagpur 440 006, India.

Twelve dogs, aged 6 months to 2 years, were infected by s.c. inoculation of *T. evansi*. Six of the dogs were left untreated and all died within 14-23 days of infection. The other six dogs were given a single i.m. dose of diminazene aceturate at 10 mg/kg body weight on day 10 p.i., supported by i.v. 5% dextrose saline at 20 ml/kg for 2 days. Treatment was well tolerated and the dogs all recovered.


Peregrine: ILRAD, P.O. Box 30709, Nairobi, Kenya.

Electrophoretic karyotyping and the arbitrary primer PCR (AP-PCR) were used to infer the existence of drug-resistant *T. evansi* in field situations, and to describe the epidemiology of such parasites in two herds of camels in northern Kenya. One herd at South Horr (c. 150 animals) was refractory to treatment with quinapyramine prosalt and also melarsomine and isometamidium chloride; the other at Olturot (80 animals) had a long history of treatment with quinapyramine prosalt. The *T. evansi* populations were found to be apparently homogeneous in each herd, having a single karyotype pattern and exposing high levels of
resistance to quinapyramine. This is in contrast to a high level of heterogeneity in molecular karyotypes that was observed amongst multiple drug resistant isolates of *T. congolense* in Ethiopia in a previous study. The similarity in karyotype patterns of the drug-resistant isolates from South Horr and Olturot indicates that the parasites are derived from common progenitors at the two sites; the introduction of other drug-resistant populations does not appear to have occurred. Similar isolates to those at South Horr were seen 3 years earlier in a camel herd at Ngurunit and it is possible that parasites with this karyotype pattern were introduced when some camels were transferred between herds. These results suggest that intensive use of trypanocides may lead to karyotype homogeneity and thus that karyotype homogeneity in such a situation may be used to infer drug resistance.

7. **experimental trypanosomiasis**

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[See also 18: nos. 8868, 8869.]


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8921 **El-Sawak, A.A. and Abd-Tabo, T.M.A., 1993.** Pathological studies on laboratory animals experimentally infected with *Trypanosoma evansi*. [Mice,
Department of Pathology, Faculty of Veterinary Medicine, Kafr-El-Sheikh, Egypt.

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8927 **Magez, S., Lucas, R., Pays, E. and Baetselier, P. de, 1994.** mTNF plays a protective role during the initial phase of experimental infection with *Trypanosoma brucei brucei*. [Mice.] (Meeting abstract.) *European Cytokine Network*, 5 (2): 204.
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8. TRYPANOSOME RESEARCH
(a) CULTIVATION OF TRYPANOSOMES
(b) TAXONOMY, CHARACTERISATION OF ISOLATES

[See also 18: no. 8865.]


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We provide evidence from isoenzyme analysis, hybridisation with repetitive DNA probes, behavioural studies and morphometrics that four trypanosome isolates from Glossina morsitans submorsitans in The Gambia constitute a new species now named Trypanosoma (Nannomonas) godfreyi. The bloodstream trypomastigotes of T. (N.) godfreyi are relatively small with a mean length of 13.7 µm (range: 9.1-21.8 µm) and a mean width of 1.65 µm (range 0.65-2.69 µm). There is no free flagellum and the marginal kinetoplast is subterminal to a rounded posterior end; the undulating membrane is usually conspicuous. As with other Nannomonas, T. godfreyi developed in the midgut and proboscis of Glossina and infections matured in 21-28 days in laboratory G. m. morsitans. In The Gambia the normal vertebrate host appears to be the warthog, Phacochoerus aethiopicus, although elsewhere other wild and domestic suids may also be implicated in the life cycle. T. godfreyi was identified unequivocally using a 380 bp DNA probe specific for a major genomic repeat sequence; its isoenzyme profile distinguished it clearly from T. simiae and three strain groups of T. congolense: savanna, riverine forest and kilifi.

Stevens: School of Biological Sciences, University of Bristol, Woodland Road, Bristol BS8 1UG, UK.

In many previous characterisation studies of Trypanozoon, isolates have been subpassaged numerous times in laboratory rodents until a quantity of trypanosomes sufficient for analysis has been obtained. In addition to the numerous biochemical effects of such a process on the parasite, it appears probable that adaptation to an unnatural host may also serve to filter out less virulent populations from mixed infections, leading to an underestimate of the true level of genetic diversity. By the early cloning of trypanosomes from susceptible captive flies infected from the primary isolate – the midgut of a wild tsetse – the present study provides evidence of the range of genetically different T. brucei populations which may coexist within the midgut of individual tsetse flies in nature. The three primary isolates from tsetse yielded one, five and nine genetically distinct populations. Cloned populations were confirmed as T. brucei using the polymerase chain reaction, and were characterised by karyotype.
analysis and multilocus isoenzyme electrophoresis. These data allowed a limited assessment of the level of genetic variability in natural populations of *T. brucei*.


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