

TSETSE AND TRYPANOSOMIASIS INFORMATION QUARTERLY

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section b - abstracts

1. GENERAL (INCLUDING LAND USE)

10013 **Food and Agriculture Organization of the United Nations, 1994.** *Residues of some veterinary drugs in animals and foods.*

(Monographs prepared by the 42nd meeting of the Joint FAO/WHO Expert Committee on Food Additives, Rome, 1-10 February 1994.) Rome; FAO (FAO Food and Nutrition Paper, no. 41/6). 87 pp.

FAO, Viale delle Terme di Caracalla, 00100 Rome, Italy.

This report on the safety of certain veterinary drug residues includes data on diminazene. See **20**: nos.

10023 and 10024 for parallel WHO documents and recommendations.

10014 **Gouteux, J.P. and Artzrouni, M., 1996.** Faut-il ou non un contrôle des vecteurs dans la lutte contre la maladie du sommeil? Une approche bio-mathématique du problème. [Is vector control needed in the fight against sleeping sickness? A biomathematical approach.] *Bulletin de la Société de Pathologie exotique*, **89** (4): 299-305.

Gouteux: ORSTOM-URA CNRS 1204, Laboratoire de Mathématiques Appliquées, Université de Pau et des Pays de l'Adour, 64000 Pau, France.

Vector control and the detection (followed by treatment) of infected individuals are the two methods currently available for the control of sleeping sickness. The basic reproduction rate of a compartmental model (Kermack and McKendrick) is used to analyse and compare the two strategies. The model shows that when there is a long first stage, characteristic of an endemic situation, the detection of sick individuals is more efficient than vector control. This higher efficiency of detection decreases in an epidemic situation. In this case vector control in the form of a decrease in vector density and/or an increase in vector mortality is relatively more efficient than detection. Because it is squared in the basic reproduction rate, the probability of a tsetse blood meal on humans is an important and sensitive parameter in the study of control strategies. This sensitivity has been observed previously and empirically by field workers. When the probability of a tsetse blood meal on humans is above a certain value, vector control becomes warranted or even necessary.

10015 **Kadohira, M., McDermott, J.J., Shoukri, M.M. and Thorburn, M.A., 1997.** Assessing infections at multiple levels of aggregation. *Preventive Veterinary Medicine*, **29** (3): 161-177.

McDermott: Department of Population Medicine,

University of Guelph, Guelph, Ontario N1G 2W1, Canada. The patterns of seroprevalence of antibodies to four infectious diseases (brucellosis, contagious bovine pleuropneumonia, infectious bovine rhino-tracheitis, trypanosomiasis) were investigated at three levels of organisation (farm, area and district). Three contrasting districts in Kenya were compared: an arid and pastoral area (Samburu), a tropical highland area (Kiambu), and a tropical coastal area (Kilifi). Cattle were selected by two-stage cluster sampling between August 1991 and 1992. Schall's algorithm, a generalised linear mixed model suitable for multi-level analysis, was compared to ordinary logistic regression (OLR), which ignores clustering of responses; generalised estimating equations (GEE) or Jackknife, to account for clustering at the farm level; and SAS VARCOMP, which provides normal-theory random-effects models. Schall's algorithm provided similar estimates to GEE (regression effects) and Jackknife (standard errors) for farm-level clustered data. Extending Schall's procedure for additional district and area-within-district random effects usually provided additional information. In general, models that included only a farm-level random effect consistently provided larger estimates of farms' variance components than did models with additional district and area random effects. The four diseases exhibited various amounts of clustering. Brucellosis had moderate farm clustering plus some area and district clustering. Contagious bovine pleuro-pneumonia had only a small amount of clustering, mostly by area. Infectious bovine rhinotracheitis exhibited a large amount of clustering, primarily at the farm level. Trypanosomiasis antibody prevalence varied by district, area and farm. It is suggested that patterns of disease clustering identified by multi-level analysis could be used to better target high-risk units for disease control and guide research to understand disease transmission factors.

10016 **Mhlanga, J.D.M., 1996.** Sleeping sickness: perspectives in African trypanosomiasis. *Science Progress*, **79** (3): 183-214.

Karolinska Institute, Doktorsringen 17, S-17177 Stockholm, Sweden.

This review covers: historical background to African trypanosomiasis; its impact on public health and economic and social development in sub-saharan Africa; the trypanosome; immunopathology; geographical ranges

and host specificities; African trypanosomiasis in humans and in livestock; control of African trypanosomiasis – chemotherapeutic, chemoprophylactic and vector control; mechanisms of action of chemical agents used against African trypanosomiasis (suramin, pentamidine, melarsoprol, tryparsamide, eflornithine, homidium bromide, diminazene aceturate, isometamidium chloride); vector control (small section only); conclusions. Current molecular and biochemical studies on the African trypanosome suggest a need for reappraisal of strategies for the diagnosis and treatment of both the chronic and acute forms of sleeping sickness. These studies have also highlighted the complexity of animal trypano-somiasis. There is an urgent need to understand first, fundamental elements of protection by the immune system, especially in the light of recent findings on the interaction(s), at the outset, between T-cell subsets, B cells, cytokines and parasites and/or parasite derived components (trypanokines), and second, the mechanisms of action of the drugs currently used.

10017 **Olliaro, P., 1997.** Will the fight against tropical diseases benefit from orphan drug status? (Editorial.) *Tropical Medicine and International Health*, **2** (2): 113-115.

UNDP/World Bank/WHO Special Programme (TDR), 1211 Geneva 27, Switzerland.

Despite progress in the identification of chemotherapeutic targets and lead compounds, the drug development pipeline is drying up, and the new avenues in research appear to have little impact on the ultimate availability of new drugs for tropical diseases. Apart perhaps from malarial drugs, the global market for tropical diseases is too small to warrant private-sector investment. The EU is discussing a regulation for orphan medicinal products analogous to the legislation in place in the USA and Japan. Such legislation would allow tax exemption and exclusivity, which might or might not be an incentive. Other elements, such as provision for accelerated authorisation procedures and for a private/public sector partnership in research and development and distribution of products, might be of greater appeal to industry.

10018 **Petney, T.N., 1997.** Ecological implications of control strategies: arthropods of domestic and production animals. *International Journal for Parasitology*, **27** (2): 155-165.

Department of Parasitology, Hygiene Institute, University of Heidelberg, D-69120 Heidelberg, Germany. The control of arthropods of veterinary importance represents a disturbance for the ecosystem and its animal community. This disturbance can influence the densities of target and non-target organisms and their associated indirect interactions in the food web, leading to reductions in the species richness and diversities in the communities involved. The effects of control of arthropod pests of domestic and production stock must be seen against a background of more general effects caused by rural development, which acts to modify the environment continuously over a long time span. This review includes a discussion of the tsetse eradication versus land degradation debate: it concludes that the latter is caused by uncontrolled immigration of people and livestock into the tsetse-cleared area rather than by planned land-use. There is obviously a conflict of interest between human development and maintenance of natural ecosystems. The effect of tsetse control on non-target organisms is reviewed: there has been a decline in species richness of some vertebrates and invertebrates, and the adverse effects on birds, fish, mammals, reptiles and amphibians are noted.

10019 **Ssennyonga, J.W., Goodell, G., Lako, G.T. and Tedla, S., 1994 [1996].** Social and economic aspects of integrated pest management in the tropics. *Insect Science and its Application*, **15** (6): 633-648.

Ssennyonga: ICIPE, P.O. Box 30772, Nairobi, Kenya. This paper reviews major concepts and achievements made in the tropics in tackling five major social and economic issues associated with research, development and adoption of integrated pest management technologies during the past two decades. Major focus has been placed on (i) integrating IPM into farming and health systems, (ii) monitoring pest populations, (iii) information and management intensity inherent in IPM, (iv) economic viability of IPM, and (v) institutional factors affecting IPM. Emphasis is placed on pinpointing ways in which success can be achieved and how constraints can be overcome in order to make IPM work better in future. Three case studies of community participation are given: these include the community-based management of tsetse and trypanosomiasis in Lambwe Valley, western Kenya, using the NGU trap. ICIPE researchers trained a catalytic group of 42 farmers who in turn trained and mobilised their

community in organisation management, mobilisation of resources, trap deployment and servicing, and impact assessment.

10020 **Tedla, S., 1994 [1996].** Social and economic aspects of integrated vector management as it relates to disease control. *Insect Science and its Application*, **15** (6): 649-676.

P.O. Box 5998, Addis Ababa, Ethiopia.

This paper examines social and economic aspects of integrated vector management in relation to the control of various vector-borne diseases. The role of the human environment and activities in transmitting and in controlling human diseases is described. Common vector-borne human and domestic animal diseases in the tropics are listed (including trypanosomiasis) and described in relation to their carriers (vectors), geographical regions of prevalence, approximate numbers of humans affected and likely remedial strategies. The benefits of integrated vector control as opposed to chemical control are discussed and supported with examples from various parts of the world. The paper ends by listing World Health Organisation recommendations on how to control human vector-borne diseases.

10021 **Wilson, C.J., Reid, R.S., Stanton, N.L. and Perry, B.D., 1997.** Effects of land-use and tsetse fly control on bird species richness in southwestern Ethiopia. *Conservation Biology*, **11** (2): 435-447.

Wilson: ILRI, P.O. Box 30709, Nairobi, Kenya.

Successful control of tsetse (*Glossina* spp.)-transmitted trypanosomiasis in the Ghibe Valley, Ethiopia, appears to have accelerated conversion of wooded grassland into cropland. Land conversion, in turn, may have fragmented wildlife habitat. Our objective was to assess the influence of the expansion of agricultural land-use, brought about by tsetse control, on ecological properties by using bird species richness and composition as indicators of environmental impacts. We sampled bird species richness and composition (using Timed-Species counts) and habitat structure (using field sampling and remote sensing) in four land cover/land-use types in areas subjected to tsetse fly control and adjacent areas without control. At the height of the growing season bird species numbers and vegetative complexity were greater in the small-holder, oxen-ploughed fields and riparian woodlands than in wooded grasslands or in large-holder, tractor-ploughed fields. Species composition was highly dissimilar (40-

70% dissimilarity) comparing among land-use types, with many species found only in a single type. This implies that trypanosomiasis control that results in land conversion from wooded grasslands to small-holder farming in this region may have no adverse impacts on bird species numbers but will alter composition. These results also suggest that moderate land-use by humans (e.g. small-holder field mosaics) increases habitat heterogeneity and bird species richness relative to high levels of use (e.g. tractor-ploughed fields). Tsetse control may be indirectly maintaining species richness in the valley by encouraging the differential spread of these small-scale, heterogeneous farms in place of large-scale, homogeneous farms. Nevertheless, if the extent of small-holder farms significantly exceeds that of present levels, negative impacts on bird species richness and large shifts in species composition may occur.

10022 **World Health Organization, 1993.** *Toxicological evaluation of certain veterinary drug residues in food.* (Monographs prepared by the 40th meeting of the Joint FAO/WHO Expert Committee on Food Additives, Geneva, 9-18 June 1992.) Geneva; WHO (WHO Food Additives Series, no. 31). 213 pp. WHO, 1211 Geneva 27, Switzerland.

This report summarises the safety data on selected veterinary drug residues reviewed by the Committee. The drugs include the trypanocide isometamidium. See *TTIQ*, **17**: nos. 8178 and 8188 for parallel FAO and WHO publications; see no. 8188 for recommendations.

10023 **World Health Organization, 1994.** *Toxicological evaluation of certain veterinary drug residues in food.* (Monographs prepared by the 42nd meeting of the Joint FAO/WHO Expert Committee on Food Additives, Rome, 1-10 February 1994.) Geneva; WHO (WHO Food Additives Series, no. 33). 152 pp. WHO, 1211 Geneva 27, Switzerland.

This report summarises the safety data on selected veterinary drug residues reviewed by the Committee, including the trypanocide diminazene. See **20**: nos. 10013 and 10024 for parallel FAO and WHO publications and recommendations.

10024 **World Health Organization, 1995.** *Evaluation of certain veterinary drug residues in food.* (Forty-second report of the Joint FAO/WHO Expert Committee on Food Additives, Rome, 1-10 February 1994.) Geneva; WHO (WHO Technical Report Series, no. 851). 41 pp. WHO, 1211 Geneva 27, Switzerland.

The first part of this report outlines the principles for evaluating the safety of residues in edible animal

products and for establishing acceptable daily intakes (ADIs) and recommended maximum residue limits (MRLs). A summary is then given of the Committee's evaluations and toxicological and residue data on selected veterinary drugs, including the trypanocide diminazene. The ADI for diminazene is established at 0-100 µg/kg body weight. MRLs in cattle are: muscle 500 µg/kg; liver 12,000 µg/kg; kidney 6000 µg/kg; fat - no MRL allocated; milk 150 µg/l. (See also **20**: nos 10013 and 10023.)

2. tsetse biology

(a) REARING OF TSETSE FLIES

(b) TAXONOMY, ANATOMY, PHYSIOLOGY, BIOCHEMISTRY

10025 **Challoner, C.M. and Gooding, R.H., 1997.** A white eye color mutant in the tsetse fly *Glossina morsitans submorsitans* Newstead (Diptera: Glossinidae). *Genome*, **40** (1): 165-169.

Gooding: Department of Biological Sciences, University of Alberta, Edmonton, Alberta T6G 2E9, Canada.

A spontaneous mutation in *G. m. submorsitans* is described. The mutant, designated *wht*, has white compound eyes but the ocelli and testes have normal coloration. Mutants have lower than normal amounts of xanthommatin and pteridines in their heads. The lesion occurs late in the tryptophan to xanthommatin pathway, in the storage of xanthommatin in the compound eyes, or, most likely, in the transport of precursors into the compound eyes. The locus *wht* is on the X chromosome.

10026 **Dagnogo, M., Lohuirignon, K. and Traore, G., 1997.**

Determination of the insemination time of the *Glossina palpalis* Robineau-Desvoidy, 1830 in the forest belt of Daloa, Côte d'Ivoire. *Acta Tropica*, **63** (2-3): 111-115.
Dagnogo: CEMV, 01 B.P. 2597, Bouaké 01, Côte d'Ivoire.
Observations on the insemination of *G. palpalis* were made in the forest belt of Daloa in Côte d'Ivoire by studying the spermathecae of females caught in different biotopes. Of 8342 females tested, only 0.88% had empty spermathecae. Most of the flies caught were teneralis. No virgin female flies were found among the young parous *Glossina* (ovarian categories I-III) above the age of 9 days. Of 3841 old parous *Glossina* (ovarian categories IV or more), 0.08% were found to have empty spermathecae. The results suggest that insemination of *G. palpalis* takes place early in the forest belt of Côte d'Ivoire, between 1 and 3 days after emergence.

10027 **Krafsur, E.S., Griffiths, N., Brockhouse, C.L. and Brady, J., 1997.** Breeding structure of *Glossina pallidipes* (Diptera:

Glossinidae) populations in East and southern Africa. *Bulletin of Entomological Research*, **87** (1): 67-73.

Krafsur: Department of Entomology, Iowa State University, Ames, IA 50011-3222, USA.

Gene diversity and gene flow in *G. pallidipes* were studied by using allozyme electrophoresis on samples collected from southern and East Africa in 1994 and 1995.

Recorded were 30 alleles segregating at eight loci.

Gene diversity was 0.212 ± 0.085 S.E. in four southern African populations (three from Zimbabwe and one from Mozambique) and 0.163 ± 0.076 in seven Kenyan populations. All loci were in Hardy-Weinberg equilibrium.

Pgm and *6pgd* were sex linked. Spatial components of gene diversity were measured by using F statistics. Mating was random within each population.

The 'fixation index' F_{ST} was 0.133 ± 0.062 among southern African populations. Among the Kenyan populations, F_{ST} was 0.159 ± 0.069 . F_{ST} was 0.238 ± 0.051 among all F_{ST} populations.

Analysis of variance of gene frequencies showed that 65.8% of the genetic variance lay within populations and 34.2% of the genetic variance lay between Kenyan and southern African populations. These data suggest a strong measure of genetic drift among tsetse populations even in a region where it is thought they are continuously distributed. The causes of this drift require investigation.

10028 **Solano, P., Duvallet, G., Dumas, V., Cuisance, D. and Cuny, G., 1997.** Microsatellite markers for genetic population studies in *Glossina palpalis* (Diptera: Glossinidae). *Acta Tropica*, **65** (3): 175-180.

Cuny: Centre ORSTOM, Département Santé, LEMV, B.P. 5045, 34032 Montpellier Cedex, France.

Little is known about tsetse intraspecific variability and its consequences on vectorial capacity. Since isoenzyme analyses revealed little polymorphism, microsatellite markers have been developed for *G. p. gambiensis*. Three loci have been identified and showed size polymorphisms for insectarium samples. More-over, amplifications were observed in different species belonging to the *palpalis* group. These molecular markers will be useful to estimate gene flow within *G. p. gambiensis* populations, and analyses could be extended to related species.

10029 **Stark, K.R. and James, A.A., 1996.** Anticoagulants in vector arthropods. *Parasitology Today*, **12** (11): 430-437.

Department of Molecular Biology and Biochemistry, University of California, Irvine, CA 92697-3900, USA.

In this review, the diversity of arthropod anticoagulants and their role in haematophagy and potential implications for parasite transmission are examined. Following a discussion of the significance of coagulation in bloodfeeding, there are sections on anticoagulants in the different arthropod groups (including a short section on tsetse anticoagulants), convergent evolution, and implications for parasite transmission.

(c) DISTRIBUTION, ECOLOGY, BEHAVIOUR, POPULATION STUDIES

[See also **20**: nos. 10026, 10041, 10052.]

10030 **Dagnogo, M., Lohuirignon, K. and Traore, G., 1997.**

Diversity of *Glossina* in the forest belt of Côte d'Ivoire. *Acta Tropica*, **65** (3): 149-153.

Dagnogo: CEMV, 01 B.P. 2597, Bouaké 01, Côte d'Ivoire. Collections of *Glossina* with a biconical trap have been made for 6 years in the forest area of Daloa in Côte d'Ivoire. Three species of *Glossina* have been identified in the village of Batéguédéa II as well as in the plantations and gallery forests. They are: *G. palpalis*, *G. pallicera* and *G. nigrofuscus*. *G. palpalis* was predominant in the village in 1984, representing 74% of the collection, while *G. pallicera* and *G. nigrofuscus* were more common in plantations and gallery forest (*G. pallicera* 42% and 51% respectively; *G. nigrofuscus* 40% and 35% respectively). In 1987 *G. palpalis* was the most common species in each of the three biotopes (village 96%, plantation 88% and forest 57%). *G. palpalis* was even more common in 1990, following a decline in the population of *G. pallicera* and *G. nigrofuscus* in the village (0.2% for *G. pallicera*; 0.8% for *G. nigrofuscus*) and the plantations (0.7% for *G. pallicera*; 1.3% for *G. nigrofuscus*). However, these two species were still found in small numbers in the gallery forest (4% for *G. pallicera*; 10% for *G. nigrofuscus*).

10031 **Groenendijk, C.A., Griffiths, N.T. and Takken, W., 1996.**

The initial flight direction of tsetse (Diptera: Glossinidae) exposed to natural and synthetic ox odour. *Proceedings of the Section Experimental and Applied Entomology of the Netherlands Entomological Society*, **7**: 241-246.

Groenendijk: Department of Entomology, Wageningen Agricultural University, P.O. Box 8031, EH Wageningen, Netherlands.

The behaviour of *Glossina pallidipes* released 10 m downwind of an odour source was studied in the Zambezi Valley, Zimbabwe, using a video camera and electric nets. Video studies showed that in the absence of odour, 46%

of the released tsetse flies turned downwind and 32% turned upwind. Tsetse left the tsetse release box (TRB) at a constant rate. When an artificial odour mixture containing carbon dioxide, acetone, octenol and phenols was used, significantly fewer tsetse (35%) turned downwind and more tsetse (37%) turned upwind. In the presence of odour, tsetse left the TRB later and not at a constant rate. When the TRB was placed in a complete ring of electric nets, the release of natural ox odour changed the distribution of tsetse to the downwind electric nets compared to the no odour treatment. Artificial odour, with and without carbon dioxide, had no effect on the distribution of tsetse over the electric nets. The difference between the video study and the electric net study is attributed to the 50% efficiency of electric nets. It is inferred that 10% of the tsetse departing from the TRB react to the presence of odour immediately.

10032 **Omoogun, G.A., 1994 [1996].** The design and construction of the Nitse trap. *Insect Science and its Application*, **15** (4-5): 535-539.

NITR, P.M.B. 03, Vom, Plateau State, Nigeria.

The design and construction of the Nitse trap are described. The materials required are: royal blue cotton, black cotton, mosquito netting, brass pipe, metal rods and galvanised wire. The cutting and joining of the various parts to produce the trap and the method of placement are also described. Finally a rough estimate of the cost of the trap is given.

10033 **Omoogun, G.A., Onyiah, J.A. and Shaida, S.S., 1994 [1996].** An improved tsetse trap for *Glossina tachinoides* in Nigeria, the Nitse trap. *Insect Science and its Application*, **15** (4-5): 529-534.

Omoogun: NITR, P.M.B. 03, Vom, Plateau State, Nigeria. The Nitse trap is a new trap designed to resemble the ungulate hosts of tsetse flies. It is horizontally cylindrical, consists mainly of royal blue cotton, with a top of white synthetic mosquito netting. It has four large rectangular openings, one at each corner of the base, in locations corresponding to the flanks of the tsetse host. The Nitse II has additional white stripes on the black interior screens near the openings. Field tests were carried out to compare the performance of the Nitse (I and II) with the biconical, F₃ and NGU traps at the Yankari Game Reserve, Bauchi State, Nigeria, which harbours *G. palpalis*, *G. tachinoides* and *G. morsitans submorsitans*. The biconical trap was used as the standard. Two areas were used for the study: A, an

area with high tsetse density, and B, an area with low-medium tsetse density. Five sites were selected in each area and cleared of vegetation; they were 5-10 m away from vehicle paths and about 100 m apart. Each trap was moved rotatingly until all five traps were exposed to each of the five sites. Traps were emptied each morning after 24 h and the tsetse sorted according to species and sex. The data on the different trap catches were analysed using the one-way ANOVA and the multiple range test via the SPSS statistical package. No significant difference in performance between the Nitse I and II was seen. In area A (high tsetse density), the Nitse performed significantly better than all the other three traps for *G. tachinoides*; the Nitse and the biconical trap performed significantly better than the F₃ for *G. m. submorsitans*. In area B (low tsetse density), only the biconical trap performed significantly better than the F₃ trap for both species; there was no significant difference between the Nitse and the other traps. The new Nitse trap was more effective for *G. tachinoides* than for *G. m. submorsitans*, but there was a significant difference between the catches of *G. tachinoides* and *G. m. submorsitans* only in area A.

10034 **Rogers, D.J., 1995.** Remote sensing and the changing distribution of tsetse flies in Africa. *In*: Harrington, R. and Stork, N.E. (eds), *Insects in a changing environment* (17th Symposium of the Royal Entomological Society, Harpenden, UK, 7-10 September 1993) (London, UK; Academic Press), pp. 177-193.

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This paper examines the four categories of evidence used by researchers, and the activities they undertake, in order to understand the distribution of animals in space and time: presence, precedence, paradigms and predictions. Examples are given of each activity, drawn from experience with tsetse flies (*Glossina* spp.) in Africa. Present-day distribution maps are often compendia of historical surveys, but provide the best-guesses available to us at the present time.

Historical maps, if made at precise moments in time and especially if they precede or follow important ecological events such as disease panzootics, provide an insight into a species' range under either adverse or particularly favourable conditions. The ebb and flow of a species' distribution may be analysed to reveal the abiotic constraints operating on the species, and provide a stimulus for field work to test

the predictions arising from such 'hind-casting' exercises. Models of species' distributions are of two main sorts, biological and statistical. The advantages of each are briefly mentioned and it is concluded that a synthesis of the two approaches will lead to the most rapid progress in understanding the major determinants of each species' range. Arising from the statistical analysis of the distribution of *G. morsitans* and *G. pallidipes* in Kenya and Tanzania, several predictions are made of the impact of global warming of 1 and 3°C on these species' distributions. The predictions depend to a large extent upon the variables within the data set; the omission of only one of these can result in diametrically opposite conclusions. It is concluded that statistical analyses need to be reinforced with biological observations for progress to be made.

3. TSETSE CONTROL (INCLUDING ENVIRONMENTAL SIDE-EFFECTS)

[See also **20**: nos. 10014, 10018, 10021, 10032, 10033.]
10035

Maniania, N.K., 1994 [1996]. A laboratory technique for infecting adult tsetse with a fungal pathogen. *Insect Science and its Application*, **15** (4-5): 421-426.

ICIPE, P.O. Box 30772, Nairobi, Kenya.

A technique for infecting adult tsetse with an entomopathogenic fungus was developed and tested in the laboratory. A nitrocellulose filter membrane (Millipore) was used as substrate for retaining conidia after filtration, and a cylindrical plastic tube served as a chamber for the flies. The technique was designed to allow accurate estimation of inoculum on the substrate, thereby permitting comparison of virulence during the screening of fungal pathogens against tsetse flies. *Glossina morsitans centralis* adults were susceptible to *Metarhizium anisopliae* at all doses tested, and mortalities were dose dependent. No significant differences in mortalities were observed when flies were exposed for different durations to the pathogen. The number of conidia picked up by flies varied considerably with exposure time and concentration.

10036 **Nagel, P., 1995.** Ecological side effects of tsetse control. *In: Abstracts of VIII International Conference of Institutions of Tropical Veterinary Medicine, 25-29 September 1995, Berlin, Germany*, p. 90.

Institute of Environmental Sciences - Biogeography,
University of Basel, St Johannis-Vorstadt 10, CH-4056
Basel, Switzerland.

In the past 15 years, field data on the ecological side

effects of tsetse control operations have been collected in different parts of West and southern Africa. SIT is the only technique with no or negligible side effects but it can only be applied together with other techniques. Artificial attractive devices, such as insecticide-impregnated, odour-baited cloth targets, attract also a small range of non-target insects. Effects on these are usually negligible but may be more severe in small, isolated, sensitive habitats. Large-scale aerosol applications of insecticides affect a wide range of terrestrial and aquatic non-target animals. Generally these effects are judged tolerable, although more severe effects have occurred, for example on fish and certain predaceous diving beetles after endosulfan applications and on certain spiders and predaceous diving beetles after deltamethrin application. Residual applications of insecticides are rarely used today but remain an option (especially the use of deltamethrin and other pyrethroids). Certain arthropod taxa may be severely affected especially in small-scale operations in dense vegetation; the side effects of large-scale residual applications in dense woodland or riverine vegetation are not tolerable. Indirect effects of tsetse control were investigated in a case study in Côte d'Ivoire. Biomonitoring and remote sensing showed that overgrazing was normally limited to the immediate vicinity of certain villages, certain sections of transhumance routes and some fenced pastures. Severe grazing damage was observed occasionally in protected woodlands. The decline in dense riverine vegetation and woodland and the parallel increase in agricultural areas pre-dated tsetse control operations, and must be attributed to other factors. The necessity of continued monitoring of future tsetse control operations is stressed since conditions vary and results from one area cannot be extrapolated to other areas.

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

[See also **20**: nos. 10015, 10029, 10031, 10099.]
10037 **Baylis, M., 1997.** The daily feeding rate of tsetse (Diptera: Glossinidae) on cattle at Galana Ranch, Kenya and comparison with trypano-somiasis incidence. *Acta Tropica*, **65** (2): 81-96.
Department of Arbovirology, Institute for Animal Health, Ash Road, Pirbright, Surrey GU24 0NF, UK.

At Galana Ranch, south-eastern Kenya, for 2 days each month from January to May 1993, *Glossina pallidipes* and *G. longipennis* were sampled around a heifer for 30 min every hour from 06:00 to 19:00. There was a seasonal decline in tsetse abundance; estimates of the total number attracted to the heifer in 1 day ranged from 556 *G. pallidipes* in January to 0 in May and 122 *G. longipennis* in February to 27 in May. The number of tsetse estimated to have fed on the heifer in 1 day during peak months was 260 *G. pallidipes* and 15 *G. longipennis*. Trypanosome infection rates of tsetse, obtained from trapped flies, suggest that the heifer received 1.1 *Trypanosoma congolense*- and 2.2 *T. vivax*-infected bites per day in January but only 0.007 *T. congolense*- and 0.047 *T. vivax*-infected bites per day in May. These predictions were compared with the observed incidence of trypanosomiasis in a nearby herd of cattle. There was a linear relationship between the estimated daily rate of infected flies feeding on the heifer and the incidence of trypanosomiasis the following month. The slope of this relationship suggests that the transmission efficiencies of *T. congolense* and *T. vivax* from tsetse to cattle are 0.84 and 2.36%, respectively, considerably lower than has been reported elsewhere. Possible reasons for this are discussed and it is suggested that previous estimates may be too high.

10038 **Bossche, P. van den and Staak, C., 1997.** The importance of cattle as a food source for *Glossina morsitans morsitans* in Katete District, Eastern Province, Zambia. *Acta Tropica*, **65** (2): 105-109.

Bossche: RTTCP, P.O. Box A560, Harare, Zimbabwe. The feeding habits of *G. m. morsitans* in the Eastern Province of Zambia were studied. A total of 687 meals were identified. Results show that 75.1% of the meals were taken on cattle, even when other domestic animals (mainly goats, pigs and dogs) were present, in contrast to other results in Zambia and elsewhere which showed that suids (particularly warthogs) were the main host. It is suggested that, with extending cultivation, as livestock have replaced its wild hosts, *G. m. morsitans* has adapted to cattle because domestic pigs are only seasonally available. The implications for the control of tsetse and trypano-somosis in the study area are discussed.

10039 **Diallo, B.P., Truc, P. and Laveissière, C., 1997.** A new method for identifying blood meals of human origin in tsetse flies. *Acta Tropica*, **63** (1): 61-64.

Diallo: IPR, OCCGE, B.P. 1500, 01 Bouaké, Côte

d'Ivoire.

A new sensitive technique using the electrophoresis of superoxide dismutase to distinguish between tsetse blood meals of human and non-human origin is described. In Côte d'Ivoire, 602 blood meals collected from *Glossina palpalis palpalis* in the Sinfra area were analysed: 170 were from man (28.3%), 377 from animals (62.6%) and 55 were unidentified (9.1%) since no pattern was observed. The index of epidemiological risk, calculated from these results, was found to be strongly correlated with the incidence of sleeping sickness cases, a large number of human blood meals in an area indicating a high risk of disease transmission.

10040

Jura, W.G.Z.O. and Otieno, L.H., 1994 [1996].

Infectivity and virulence of *Trypanosoma brucei* metacyclics from *Glossina morsitans morsitans* salivary glands infected with tsetse DNA virus. *Insect Science and its Application*, **15** (4-5): 551-556.

Jura: ICIPE, P.O. Box 30772, Nairobi, Kenya.

The pathogenicity of cyclically transmitted C16 clone 1 of *T. brucei* metacyclics which developed within normal and hypertrophied DNA virus-infected salivary glands of *G. m. morsitans* was studied in inbred, BALB/c mice.

Microscopic examination of salivary glands obtained from *G. m. morsitans* with combined virus and *T. brucei* infections (i.e. the flies which transmitted the trypanosome infection to the test group of mice, group A) revealed that the glands comprised hyperplastic epithelial cells, some of which were fragmented, and numerous metacyclic trypanosomes. All the mice which contracted trypanosomiasis from *G. m. morsitans* with only *T. brucei*, i.e. the control mice (Group B: $n = 4$), and the mice in the test group ($n = 3$) developed high parasitaemia and died. Repeated-measures analysis of variance using adjusted mean rate revealed that the mean prepatent periods and the mean times to death in both the control and the test groups of the BALB/c mice were not significantly different ($P > 0.05$). Analysis of log-transformed data fitted to a logistic growth model revealed that the rate of multiplication of *T. brucei* parasites in the blood of the test group BALB/c mice ($r = 2.373$) was greater than in the control mice ($r = 0.808$) and that the maximum carrying capacity was also attained earlier in the former group. These observations imply that the development of *T. brucei* metacyclics within hypertrophied salivary glands and their co-existence with the DNA virus particles might have enhanced their infectivity and virulence in the

mice.

10041 **Mohamed-Ahmed, M.M. and Odulaja, A., 1997.** Diel activity patterns and host preferences of *Glossina fuscipes fuscipes* (Diptera: Glossinidae) along the shores of Lake Victoria, Kenya. *Bulletin of Entomological Research*, **87** (2): 179-186.

Mohamed-Ahmed: ICIPE, P.O. Box 30772, Nairobi, Kenya. Diurnal activity patterns and host preference of *G.f. fuscipes* were studied in forest and linear habitats along Lake Victoria shore, Kenya. The objective was to identify the preferred host of *G.f. fuscipes*, the emanations of which may be attractive to this species. Hourly catches of flies in biconical traps were related to changes in the weather and the prevalence of hosts in the vicinity of traps. Flies were mainly active between 0800 and 1600 h, with males peaking around 1100 h and females around 1300 h. Activity of flies correlated directly with light intensity and temperature, but indirectly with relative humidity. Humans, livestock and the monitor lizard, *Varanus niloticus*, were the predominant hosts, although a significant positive correlation with fly catches could only be established with the prevalence of lizards. Blood meal identification by microscopic and serological methods showed that 73-98% of *G.f. fuscipes* fed on monitor lizards irrespective of host prevalence, season or location. The significance and possible epidemiological importance of the relationship between *G.f. fuscipes* and monitor lizards are discussed.

10042 **Mwanje, J.I. and Mwanje, M.T., 1995.** Evolutionary, regulatory and mediation aspects of *T. b. rhodesiense* and its endemicity in Lambwe Valley, Kenya. *African Journal of Health Sciences*, **2** (4): 364-371.

J.I. Mwanje: University of Nairobi, P.O. Box 30197, Nairobi, Kenya.

The transmission of human African trypanosomiasis depends on environmental factors, such as habitat, temperature, humidity, the distribution and types of vegetation, vector density, etc., operating at the mega-, macro- and micro-scale levels. At the micro-scale level (ecological biotopes), the metacyclic development processes of *Trypanosoma brucei rhodesiense* are controlled by evolutionary, regulatory and mediation factors. These include selective pressures acting on host-parasite interactions and phenotypic variation. This paper considers the underlying dynamics which account for the endemicity of *T. b. rhodesiense* infection in the Lambwe Valley ecosystem, with special reference to

the transmission of the parasite and parasitaemia in reservoir agents and humans.

5. human trypanosomiasis

(a) SURVEILLANCE

[See also **20**: no. 10014.]

10043 **Hove, D. van, 1997.** Sleeping sickness in Zaire. (Letter.) *Lancet*, **349** (9049): 438.

Belgian Development Cooperation, P.O. Box 81, Kigali, Rwanda.

This letter refers to an article in a previous issue of *Lancet* (see *TTIQ*, **20**: no. 9785), outlining the history of sleeping sickness in Zaire (Congo) from 1926 to 1994, and recommending mass screening and treatment of serological suspects as well as parasitologically confirmed cases to break the chain of transmission. It is pointed out that this strategy, although useful, is unlikely to compensate for a very low (50%) participation rate of exposed populations in case-finding surveys, due mainly to fear of drug-induced mortality. Closely supervised effective treatment is therefore needed, as well as a widespread and judicious use of serological and more refined parasitological techniques.

10044 **Koko, J., Duffillot, D., Gahouma, D., Amblard, J. and Kani, F., 1997.** Trypanosomose humaine africaine chez l'enfant.

Expérience d'un service de pédiatrie à Libreville, Gabon. [Human African trypanosomiasis in children. Experience in a pediatrics department in Libreville, Gabon.] *Bulletin de la Société de Pathologie exotique*, **90** (1): 14-18.

Koko: Service de Pédiatrie Générale, Hôpital Pédiatrique d'Owendo, B.P. 1208, Libreville, Gabon. During a period of six years from 1 January 1989 to 31 December 1994, seven children with trypanosomiasis were admitted to the Department of Pediatrics of Owendo Pediatric Hospital. They were five boys and two girls, aged 4-17 years, five of them under 15 years. Delay in diagnosis ranged from 2 to 9 months. The main reasons for hospitalisation were somnolence (4 cases), psychical disorders (5 cases), neurological disorders (4 cases), asthenia (3 cases), loss of weight (3 cases) and fever (3 cases). Increased sedimentation rate (5 cases) and hypergammaglobulinaemia (6 cases) were the most important biological disturbances. Serodiagnosis (CATT, indirect immunofluorescence test) was positive in all cases. The parasite was detected four times in

both blood and cerebrospinal fluid and three times in blood alone. According to CSF status, six children were classified as being in the second stage of the disease. Six patients were treated with melarsoprol, and one with eflornithine. Tolerance and response to treatment were good in six cases. Three children presented sequelae when leaving hospital. No patient was seen again after the study.

(b) PATHOLOGY AND IMMUNOLOGY

[See also 20: no. 10016.]

10045 **Pentreath, V.W., Alafiatayo, R.A., Barclay, G.R., Crawley, B., Doua, F. and Oppenheim, B.A., 1997.** Endotoxin antibodies in African sleeping sickness. *Parasitology*, **114** (4): 361-365.

Pentreath: Department of Biological Sciences, University of Salford, Salford M5 4WT, UK.

Antibodies to the core region of endotoxin (endotoxin core antibodies, EndoCAB), which cross-react with endotoxin from a range of Gram-negative bacteria, are maintained in relative homeostasis in health, but undergo marked changes in a number of different diseases associated directly or indirectly with endotoxaemic or septicaemic states. The levels of EndoCAB IgG in the blood and CSF of 35 late-stage sleeping sickness (*Trypanosoma brucei gambiense*) patients and 9 healthy control individuals were measured by ELISA. EndoCAB levels were significantly elevated in the patient blood (mean EndoCAB value 290 MU/ml, control 182 MU/ml, $P < 0.001$) and CSF (mean EndoCAB value 254 MU/ml, control 150 MU/ml, $P < 0.001$). EndoCAB IgG levels correlated with endotoxin levels in patient blood ($r = 0.78$, $P < 0.001$) but not in the CSF, and were not reduced 6 weeks following chemotherapy, unlike the endotoxin levels. It is concluded that late-stage sleeping sickness is associated with chronic exposure to endotoxins from Gram-negative bacteria.

10046 **Rhind, S.G., Sabiston, B.H., Shek, P.N., Buguet, A., Muanga, G., Stanghellini, A., Dumas, M. and Radomski, M.W., 1997.** Effect of melarsoprol treatment on circulating IL-10 and TNF- α levels in human African trypanosomiasis. *Clinical Immunology and Immunopathology*, **83** (2): 185-189.

Rhind: Defence and Civil Institute of Environmental Medicine, Toronto, Ontario M3M 3B9, Canada.

The pathogenesis of human African trypanosomiasis has been the object of considerable research interest but has remained incompletely understood. The importance of cytokines in the pathophysiology of this protozoan

infection is now widely recognised, but the full spectrum of cytokines involved has yet to be determined. In the present investigation we compared the plasma concentrations of TNF- α and IL-10 in normal African controls and patients suffering from advanced meningoencephalitic (late-stage) *Trypanosoma brucei gambiense* infections, before and after treatment with the arsenical trypanocide melarsoprol. We found that patients with late-stage *T. b. gambiense* infection exhibit chronically elevated circulating levels of both of these cytokines, and that these levels quickly decline following melarsoprol treatment. These findings confirm that TNF- α is involved in the immunopathogenesis of late-stage African trypanosomiasis and suggest that IL-10 may also play an important regulatory role in this disease.

(c) TREATMENT

[See also 20: nos. 10016, 10017.]

10047 **Ericsson, O., Schweda, E.K.H., Bronner, U., Rombo, L., Fridén, M. and Gustafsson, L.L., 1997.** Determination of melarsoprol in biological fluids by high-performance liquid chromatography and characterisation of two stereoisomers by nuclear magnetic resonance spectroscopy. *Journal of Chromatography (B)*, **690** (1-2): 243-251.

Ericsson: Hospital Pharmacy and Unit of Tropical Pharmacology, Department of Clinical Laboratory Medicine and Technology, Huddinge University Hospital, S-14186 Huddinge, Sweden.

The analysis of melarsoprol in whole blood, plasma, urine and cerebro-spinal fluid is described. Extraction was made with a mixture of chloroform and acetonitrile followed by back-extraction into phosphoric acid. A reversed-phase liquid chromatography system with ultraviolet detection was used. The relative standard deviation was 1% at concentrations around 10 $\mu\text{mol/l}$ and 3-6% at the lower limit of determination (9 nmol/l in plasma, 93 nmol/l in whole blood, 45 nmol/l in urine and 10 nmol/l in CSF). Melarsoprol is not a stable compound and samples to be stored for longer periods of time should be kept at -70°C . Plasma samples can be stored at -20°C for up to 2 months. Chromatography showed that melarsoprol contains two components. Using nuclear magnetic resonance spectroscopy the two components were shown to be diastereomers which slowly equilibrate by inversion of the configuration at the As atom.

- 10048 **Frayha, G.J., Smyth, J.D., Gobert, J.G. and Savel, J., 1997.**
The mechanisms of action of antiprotozoal and anthelmintic drugs in man. *General Pharmacology*, **28** (2): 273-299.
Frayha: Rue de Javel Tour Espace 2000, 75015 Paris, France.
This review includes sections on the mode of action of pentamidine, diminazene, melarsoprol and tryparsamide.
- 10049 **Huebert, N.D., Schwartz, J.-J. and Haegele, K.D., 1997.**
Analysis of 2-difluoromethyl-DL-ornithine in human plasma, cerebrospinal fluid and urine by cation-exchange high-performance liquid chromatography. *Journal of Chromatography (A)*, **762** (1-2): 293-298.
Huebert: Marion Merrell Research Institute, 16 rue d'Ankara, 67080 Strasbourg Cedex, France.
An analytical method has been developed based on cation-exchange liquid chromatography for the measurement of 2-difluoromethyl-DL-ornithine (DFMO) in human plasma, cerebrospinal fluid (CSF) and urine. Fluorescence detection at excitation/emission wavelengths of 340/440 nm is followed by postcolumn derivatisation with *o*-phthalaldehyde-2-mercaptoethanol. All calibration ranges yielded linear relationships with correlation coefficients better than 0.999. In each case the limit of quantitation was equal to the lowest value of the standard curve. The variability of the assay, expressed as relative standard deviations, was less than 7.1%, 15.3% and 7.1% for plasma, CSF and urine, respectively. The accuracy of the assay (expressed as relative errors) ranged between -4.3% and 2.0% for plasma analysis, between -0.1% and 14.0% for CSF analysis and between -8.0% and 2.0% for urine analysis. Plasma, CSF and urinary DFMO concentrations were measured in samples obtained from patients undergoing treatment for trypanosomiasis. The method was found to be applicable for the measurement of DFMO levels in human body fluids for the determination of pharmacokinetic parameters in clinical studies.
- 10050 **Watts, R.G., Conte, J.E., Zurlinden, E. and Waldo, F.B., 1997.**
Effect of charcoal hemoperfusion on clearance of pentamidine isethionate after accidental overdose. *Journal of Toxicology, Clinical Toxicology*, **35** (1): 89-92.
Watts: Division of Hematology-Oncology, University of Alabama, Birmingham, AL 35233, USA.
Pentamidine isethionate is an antimicrobial agent effective in the treatment of *Pneumocystis carinii* pneumonia, trypanosomiasis and leishmaniasis. Severe and fatal toxicity is reported with pentamidine use. A

patient (a 17-month-old child) received an accidental overdose (40 times the prescribed dose) of i.v. pentamidine due to a pharmacy mixing error. Charcoal haemoperfusion was successfully utilised to lower the serum concentration of pentamidine and lessen toxicity. This technique may represent a useful modality in the management of pentamidine isethionate overdosage.

6. animal trypanosomiasis

(a) SURVEY AND DISTRIBUTION

[See also **20**: nos. 10015, 10059, 10073.]

10051 **Desquesnes, M., 1997.** Evaluation of a simple PCR technique for the diagnosis of *Trypanosoma vivax* infection in the serum of cattle in comparison to parasitological techniques and antigen-enzyme-linked immunosorbent assay. *Acta Tropica*, **65** (3): 139-148.

CIRAD-EMVT-GUYANE, Institut Pasteur, B.P. 6010, 97306 Cayenne, French Guiana.

Polymerase chain reaction (PCR) with specific oligonucleotides for the amplification of *T. vivax* DNA has been developed by Masiga *et al.* (1992) to detect the presence of *T. vivax* DNA in biting flies. The aim of this experiment was to evaluate the efficacy of this technique when applied directly on cattle serum, without DNA purification, to detect infection. The sensitivity of this PCR technique was compared with parasitological techniques, namely haematocrit centrifuge technique (HCT) and buffy coat method (BCM), and with the antigen-enzyme-linked immunosorbent assay (Ag-ELISA) for *T. vivax* developed by Nantulya and Lindqvist (1989). Blood and serum samples were collected from four calves experimentally infected with a stock of *T. vivax* from French Guiana (IL 4007). During the first 51 days of infection, a total of 164 samples were collected and processed using the four tests. Mean percentages of positive results were 68% with HCT, 59% with BCM, 4% with Ag-ELISA and 64% with PCR. Parasitological and PCR techniques yielded approximately the same sensitivities. PCR was able to detect active infection in serum samples when parasitaemia was over 10^3 trypanosomes/ml. With this isolate of *T. vivax* the Ag-ELISA was not found to be sensitive enough to be used as a diagnostic tool. The sensitivity of this PCR technique is not greater than parasitological techniques but it allows delayed processing of the samples and gives a highly species-specific diagnosis. This simple PCR technique should be evaluated for field diagnosis because it makes

retrospective epidemiological survey using serum banks possible. Moreover, it can be substituted for parasitological techniques when immediate examination is not feasible.

10052 **Kalu, A.U. and Uzoigwe, N.R., 1996.** Tsetsefly and trypanosomosis on the Jos Plateau: observations on outbreaks in Barkin-Ladi Local Government Area. *Tropical Veterinarian*, **14** (3-4): 117-126.

Kalu: Department of Veterinary Public Health and Preventive Medicine, University of Maiduguri, P.M.B. 1069, Maiduguri, Nigeria.

Epidemiological investigations involving two outbreaks of trypanosomosis in ruminant herds in Barkin-Ladi LGA of Jos Plateau are reported. The first outbreak was characterised by high prevalence in all bovine herds ranging from 37.6% (confidence interval CI: 0.34, 0.38) to 50% (CI: 0.40, 0.60) with a mean of 43.0% (CI: 0.38, 0.48). All infections in animal hosts and 12.5% in the vector (*Glossina palpalis palpalis*) were caused by *Trypanosoma vivax*. Lower infection rates in the second outbreak (mean 6.93%, CI: 0.05, 0.09) were due to a low tsetse population and high incidence of biting flies (*Stomoxys*, tabanids) and were caused by *T. vivax* and *T. congolense*. Several factors favourable to the breeding of riverine tsetse were observed in Sholong and Bachit districts. Findings from tsetse ecology in the area are presented and a review of the tsetse status of the Jos Plateau is suggested.

10053 **Katakura, K., Lubinga, C., Chitambo, H. and Tada, Y., 1997.** Detection of *Trypanosoma congolense* and *T. brucei* subspecies in cattle in Zambia by polymerase chain reaction from blood collected on a filter paper. *Parasitology Research*, **83** (3): 241-245.

Katakura: Department of Tropical Medicine, Jikei University School of Medicine, 3-25-8 Nishi-shinbashi, Minato-ku, Tokyo 105, Japan.

To facilitate epidemiological studies of African trypanosomiasis in cattle in Zambia, we adapted a PCR method using blood spotted on filter papers. For easy preparation of template DNA from the dried blood, we adapted a simple DNA extraction method using Chelex-100, an anion-exchange resin. Using primers directed for repetitive nuclear DNA sequences, species-specific DNA amplifications were detected from the blood of rats infected with Zambian isolates of *T. congolense* and *T. brucei* subspecies. The method was sensitive enough to detect a single trypanosome for both species. In the Eastern Province of Zambia, 240 cattle were examined for motile

flagellates in the buffy coat by the microhaematocrit method, and 100 of them tested positive. These 100 animals were further examined by thin blood smears and PCR for species identification. The thin blood smear revealed 62 and 14 animals with *T. congolense* and *T. brucei* subspecies infection, respectively, whereas the PCR detected 73 of the former and 38 of the latter species. These results indicate that dried blood spots on filter papers are a useful source of DNA for detection of African trypanosomes by PCR.

10054 **Nawathe, D.R., Srivastava, G.C., Basu, A.K. and Kollere, M.A., 1995.** Trypanosomiasis in small ruminants in the arid zone, Nigeria. *Bulletin of Animal Health and Production in Africa*, **43** (4): 293-294.

Nawathe: University of Maiduguri, Maiduguri, Nigeria. A survey of trypanosomiasis prevalence in sheep and goats in the arid zone of Borno State was carried out using parasitological methods. Blood samples were collected from animals slaughtered at Maiduguri abattoir (catchment area c. 100 km radius) twice a week from November 1991 to April 1992 (dry season). PCV was also determined but did not reveal the presence of severe anaemia. Only a few samples were positive for *Trypanosoma vivax*: 2 females out of 147 Uda sheep; 2 females out of 77 Red Sokoto goats; 2 males out of 273 Borno White goats. No positive cases were seen among 167 Yankasa sheep. These low prevalence rates of 0.6% in 314 sheep and 1.1% in 350 goats were expected in an area without tsetse where *T. vivax* is transmitted by biting flies. Buffy coat examination was the most satisfactory parasitological method used.

(b) PATHOLOGY AND IMMUNOLOGY

[See also **20**: nos. 10016, 10068.]

10055 **Agur, Z. and Mehr, R., 1997.** Modelling *Trypanosoma congolense* parasitaemia patterns during the chronic phase of infection in N'Dama cattle. *Parasite Immunology*, **19** (4): 171-182.

Agur: Department of Cell Research and Immunology, Life Science Faculty, Tel-Aviv University, Ramat-Aviv, 69978 Tel-Aviv, Israel.

We re-analysed parasitaemia profiles of the trypanotolerant N'Dama cattle (*Bos taurus*), consecutively infected with the same four clones of *T. congolense*. Our analysis shows that each individual parasitaemia is characterised by progressively longer intervals between

parasite waves. This pattern is most visible during the chronic phase of infection. In addition, the last of the four infections had a significantly larger overall duration of inter-wave intervals. We retrieved these patterns by numerical simulations of a mathematical model, which incorporates assumptions about the molecular basis of antigenic variation and about the anti-parasitic major immune processes. Six potential factors that may determine parasitaemia pattern were studied: carrying capacity of the host environment, intrinsic growth rate of the parasite, affinity maturation of the immune response, immune cell birth and death rate, levels of antibodies to variant surface glycoprotein and levels of antibodies to invariant antigens. Our simulations suggest that the first five factors are not likely to determine the chronic phase parasitaemia pattern whereas the sixth one, namely antibody response to invariant antigens, yielded profiles consistent with the experimental data. Being cumulative, the immune response to invariant antigens may be increasingly effective as infection proceeds and in successive infections. Comparisons between N'Dama and Zebu and between chronic and acute phases will be needed to make a statement on the role of this phenomenon in trypanotolerance.

10056

Anosa, V.O., Logan-Henfrey, L.L. and Wells, C.W., 1997.

The haematology of *Trypanosoma congolense* infection in cattle I. Sequential cytomorphological changes in the blood and bone marrow of Boran cattle. *Comparative Haematology International*, **7** (1): 14-22.

Logan-Henfrey: USDA, ARS, National Program Staff, Animal Health, Room 203, Building 005, BARC-West, Beltsville, MD 20705, USA.

Five adult Boran cattle (*Bos indicus*), infected with a clone of *T. congolense* IL13-E3 three years earlier and treated, were re-challenged with the same clone. Changes in the peripheral blood were monitored twice weekly, and events in the bone marrow were assessed by weekly biopsies of the sternal bone marrow, until day 98 p.i. when the three surviving animals were treated with diminazene aceturate. One animal died on day 57 p.i. whereas another was treated on day 63 p.i. when the PCV was 15%. The infected animals developed anaemia, leucopenia and thrombocytopenia during the first peak of parasitaemia which persisted until the experiment was terminated. Three phases of bone marrow response were demonstrated on light microscopic examination of bone marrow smears. The first, the

preparasitaemic phase represented by samples taken on day 15 p.i., was an immunological response with slight but significant increases in lymphoblasts, lymphocytes, plasma cells and macrophages whereas erythroid and granulocytic cells were unchanged. The second, the early parasitaemic or acute phase (21-57 days p.i.) associated with the development of anaemia, leucopenia and thrombocytopenia, was characterised by intensification of the immunological response, and an early but transient granulocytic hyperplasia. The third, the late parasitaemic or chronic phase (63-98 days p.i.) associated with persisting pancytopenia, was characterised by erythroid, megakaryocytic and macrophage hyperplasia, dyserythropoiesis, granulocyte hypoplasia and return of lymphoid cell counts to preinfection numbers. Transmission electron microscopy confirmed these findings and showed that intact trypanosomes were not observed in the sinusoids and haemopoietic compartment of the bone marrow. This study demonstrates that *T. congolense* infection affects haemopoiesis, downregulating or upregulating the various blood cell lineages depending on the stage of infection. This suggests a fine control mechanism, presumably cytokine-mediated. Erythropoiesis, thrombopoiesis and monocytopoiesis were generally upregulated, whereas granulopoiesis was downregulated. However, haemopoiesis was generally ineffective as numbers of circulating blood cells remained below preinfection levels throughout the period of the study.

10057 **Anosa, V.O., Logan-Henfrey, L.L. and Wells, C.W., 1997.**

The haematology of *Trypanosoma congolense* infection in cattle II. Macrophage structure and function in the bone marrow of Boran cattle. *Comparative Haematology International*, **7** (1): 23-29.

Logan-Henfrey: USDA, ARS, National Program Staff, Animal Health, Room 203, Building 005, BARC-West, Beltsville, MD 20705, USA.

Macrophages in smears and sections of sternal bone marrow derived by weekly sequential biopsies from five adult Boran cattle re-challenged with *T. congolense* were studied by light and transmission electron microscopy (TEM). Cells of the mononuclear phagocyte system including monoblasts, promonocytes, monocytes and macrophages increased several-fold in the sinusoids and haemopoietic compartment of the bone marrow during infection. Macrophage activation occurred with significant increases ($P < 0.001$) in macrophage size and numbers of organelles including mitochondria,

lysosomes and rough endoplasmic reticulum. Light microscopic examination of the bone marrow smears showed that 25.8% of 1200 macrophages examined phagocytosed many non-mitotic haemopoietic cells of the erythroid and granulocytic series as well as mature erythrocytes and thrombocytes but seldom lymphocytes from day 29 p.i., when the first peak of parasitaemia occurred, until termination of the experiment on day 98 p.i.. Some of the macrophages with phagocytosed cells (10.4%) had cells from more than one lineage. TEM confirmed cytophagia and showed that the process begins with cell to macrophage attraction characterised by development of microvilli at the surface of contact by the target cell and of enveloping pseudopodia by the macrophage. This was followed by target cell to macrophage adhesion and finally phagocytosis. The cells being phagocytosed and those freshly engulfed appeared morphologically normal. Many macrophages were heavily laden with haemosiderin in the chronic phase of the infection (78 and 98 days p.i.). TEM showed that the activated macrophage in the bone marrow developed extensive contacts through reciprocal blunt microvilli with the haemopoietic cells. Macrophages were absent from the sinusoids of the bone marrow prior to infection but became numerous during infection, and were adhered to sinusoidal endothelial cells by reciprocal blunt microvilli. These macrophages phagocytosed blood cells (erythrocytes, neutrophils, thrombocytes) and free trypanosomes which, though present in the arterioles of the bone marrow, were never seen in the sinusoids and haemopoietic compartment of the bone marrow. This study indicates that the macrophage plays very vital roles in regulating and executing the events in the bone marrow during *T. congolense* infection of cattle.

10058 **Dam, J.T.P. van, Schrama, J.W., Vreden, A., Versteegen, M.W.A., Wensing, T., Heide, D. van der and Zwart, D., 1997.** The effect of previous growth retardation on energy and nitrogen metabolism of goats infected with *Trypanosoma vivax*. *British Journal of Nutrition*, **77** (3): 427-441.

Dam: Department of Animal Husbandry, Wageningen Institute of Animal Sciences, P.O. Box 338, 6700 AH Wageningen, Netherlands.

The effect of growth retardation, resulting from feed restriction for a prolonged period, on the course of infection with *T. vivax* was studied. Twelve male castrated West African Dwarf goats were subjected to a restricted feeding regimen of 55 g pelleted lucerne

(*Medicago sativa*) /kg body weight^{0.75}/day for on average 17 weeks. Twelve other animals were fed on pelleted lucerne *ad libitum*, resulting in a normal growth pattern. After this period, all animals were fed on pelleted lucerne *ad libitum*, and six animals of each previous feeding regimen treatment were infected with *T. vivax*. The other animals served as controls. In weeks 2 and 4 p.i., energy and N balances were measured. In the week before infection and during infection blood biochemical and clinical variables were measured. At 2 weeks before, and 4 weeks after infection, a liver biopsy was taken for measurement of triacylglycerol. Infection caused intermittent fever and anaemia. The first peak of fever persisted longer in infected animals with normal growth than in infected animals with retarded growth. Gross energy and metabolisable energy intake, and energy retention, were reduced in infected animals. Metabolisable energy requirements for maintenance were increased by infection. Plasma non-esterified fatty acids (NEFA) and glucose concentrations were increased in infected animals, whereas serum triiodothyronine and thyroxine concentrations were decreased. Plasma urea concentration and liver triacyl-glycerol were unaffected. No interaction of growth retardation with infection with respect to blood biochemical variables was found, apart from plasma NEFA in week 2 p.i. N retention was not significantly affected by treatments. In conclusion, minor indications were found for an interaction between growth retardation, as applied in the present study, and trypanosomiasis infection in West African Dwarf goats with respect to energy and N metabolism.

10059 **Dehoux, J.-P., Diaw, M. and Buldgen, A., 1996.**

Observation d'une flambée de trypanosomose équine due à *Trypanosoma vivax* en zone urbaine au Sénégal.

[Observation of an outbreak of equine trypano-somiasis due to *T. vivax* in an urban environment in Senegal.]

Tropicultura, **14** (1): 35-36.

Dehoux: Département des Productions Animales, Ecole Nationale Supérieure d'Agriculture (ENSA), B.P. 296, Thiès, Senegal.

An outbreak of *T. vivax* trypanosomiasis in imported and local horses and ponies occurred in September 1994 in an equestrian centre near Dakar, resulting in five mortalities (including a local pony) among 20 infected animals. The clinical signs were fever, depression, emaciation, anaemia and oedema. Following diagnosis of the disease, curative treatment with deep i.m.

diminazene aceturate (3.5 mg/kg) was given, followed by prophylactic i.v. isometamidium (0.5 mg/kg) in October 1994 and again in July 1995. *Glossina palpalis gambiensis* was isolated near the farm.

10060

Katunguka-Rwakishaya, E., Murray, M. and Holmes, P.H.,

1997. Susceptibility of three breeds of Ugandan goats to experimental infection with *Trypanosoma congolense*.

Tropical Animal Health and Production, **29** (1): 7-14.

Katunguka-Rwakishaya: University of Glasgow Veterinary School, Bearsden Road, Glasgow G61 1QH, UK.

The susceptibility of Kigezi, Mubende and Small East African (SEA) goats to *T. congolense* infection was studied at the Faculty of Veterinary Medicine, Kampala, Uganda. Five goats of each breed were infected i.v. with 10^5 *T. congolense* while another five were kept as uninfected controls. All infected goats showed detectable parasitaemia 5-7 days p.i. Over a period of 7 weeks p.i., infected Kigezi goats lost 0.8 kg, infected Mubende goats maintained their weight and infected SEA goats gained 0.9 kg, while the controls gained 2.0-2.5 kg. PCV had decreased from 25 \square 3% to 13 \square 2% 77 days p.i. in Kigezi goats, from 31 \square 4% to 16 \square 2% 42 days p.i. in Mubende goats and from 30 \square 3% to 18.0 \square 2% 84 days p.i. in SEA goats (control goats 22-31%). Two of the Kigezi goats died 77 and 80 days p.i. Four weeks after treatment with diminazene aceturate (at 84 days p.i.), the PCV values of SEA goats had returned to normal while those of Kigezi and Mubende goats were still lower than control values. SEA goats thus appear to be least susceptible to *T. congolense* infection and Kigezi most susceptible.

10061

Zia-ur-Rahman, Butt, A.A., Asif, M.M., Haq, I.U., Ahmad, A.,

Asghab, M. and Shaukat, S.A.J., 1996. Concentration of serum micro and macro-elements in the sera of control and trypanosome-infected camel. *Tropical Veterinarian*, **14** (3-4): 133-136.

Zia-ur-Rahman: Department of Veterinary Physiology and Pharmacology, University of Agriculture, Faisalabad 38040, Pakistan.

Serum was obtained from nine trypanosome-infected and three uninfected control camels for the determination of micro- and macro-elements by atomic absorption and flame photometry. No significant differences were observed in copper (Cu^{2+}), iron (Fe^{2+}), magnesium (Mg^{2+}), sodium (Na^+) and potassium (K^+) concentrations of infected camels compared to controls. Significant differences were observed in serum zinc (Zn^{2+}) and calcium (Ca^{2+}) concentrations. The concentration of

Zn²⁺ was 6.31 \pm 0.80 mg/l in the infected group and 3.96 \pm 0.81 mg/l in the control group. The concentration of Ca²⁺ was 7.73 \pm 0.67 mEq/l in the controls and 5.13 \pm 1.16 mEq/l in infected camels.

(c) TRYPANOTOLERANCE

[See also **20**: nos. 10055, 10060, 10074, 10081, 10083.]
10062

Awolaja, O.A., Antia, R.E. and Oyejide, A., 1997. Trace element levels in plasma/serum and erythrocytes of Keteku and White Fulani cattle. *Tropical Animal Health and Production*, **29** (1): 2-6.

Awolaja: Faculty of Veterinary Medicine, University of Ibadan, Ibadan, Nigeria.

In order to determine the probable role of trace elements in the maintenance of trypanotolerance in tropical cattle, the levels of zinc, copper, manganese and iron in plasma/serum and erythrocytes of trypanotolerant Keteku and trypano-susceptible White Fulani cattle were measured by atomic absorption spectro-photometry and compared by breed or sex. Trypanotolerance was associated with significantly higher ($P < 0.05$) erythrocyte levels of zinc and manganese. The Keteku breed showed higher erythrocyte levels of zinc and manganese than the White Fulani. The serum manganese level was also significantly higher ($P < 0.01$) in Keteku than in White Fulani cattle.

However, PCV and total protein concentration were not significantly different by breed or sex.

10063 **Gelhaus, A., Hanotte, O., Agaba, M., Horstmann, R.D. and Teale, A.J., 1996.** Genetic mapping of trypanosusceptibility in the F₂ generation of two full-sibling N'Dama \times Boran families². (Meeting abstract no. B006.) *Animal Genetics*, **27** (Suppl. 2): 44.

Gelhaus: Department of Molecular Genetics, Bernhard Nocht Institute for Tropical Medicine, 20359 Hamburg, Germany.

To study the genetic background of trypanotolerance, trypanotolerant N'Dama bulls and trypanosusceptible Boran cows were mated at ILRI to produce three-generation full-sibling families in which trypanotolerance genes were expected to segregate. *BoLA-DRB3* in the *MHC* class II region on chromosome 23 was chosen as a candidate gene. In two ILRI families *DRB3* polymorphisms were found to be fully informative and the respective F₂ generations were typed using PCR-RFLP. Anaemia was taken as a phenotypic marker for quantifying trypanotolerance, and the PCV was measured on day 150 after challenge (PCVd150). Analysis of 57

F₂ animals revealed linkage between the *DRB3* locus and the PCVd150 indicating the presence of a gene on bovine chromosome 23 which might confer trypanotolerance or trypanosusceptibility. Further analysis using *t*-test statistics indicated that the allele *DRB3**1301 might be associated with trypanosusceptibility. The PCVd150 of animals carrying this particular allele were significantly lower than the PCVd150 of animals not carrying *DRB3**1301 ($P < 0.005$).

10064 **Hanotte, O., Okomo, M., Verjee, Y., Ochieng, J., Teale, A.J. and Rege, E., 1996.** Assessment of genetic diversity and breed relationships of sub-Saharan African cattle by microsatellite analysis. (Meeting abstract no. A017.) *Animal Genetics*, **27** (Suppl. 2): 21.

Hanotte: ILRI, P.O. Box 30709, Nairobi, Kenya. As part of a large-scale study of African cattle biodiversity, two sets of autosomal microsatellite loci (StockMark™ and ILRI kit) and a Y-specific microsatellite locus were used for an analysis of the genetic diversity and relationship of 10 cattle breeds including pure *Bos taurus* breeds (Friesian and N'Dama), supposedly pure *B. indicus* breeds (Highland zebu) and presumed hybrid breeds between the two species/subspecies (sanga breeds). Population heterozygosity ranged from 0.260 ± 0.041 (Muturu) to 0.690 ± 0.033 (Drakensberger). Nei's standard genetic distances ranged from 0.081 ± 0.069 between the Fogera and the Highland zebu to 0.7523 ± 0.1700 between the Muturu and the Highland zebu. A dendrogram constructed by the neighbour-joining method suggests a gradient of *B. taurus* introgression amongst the sanga breeds.

10065 **Hendrickx, G., Meghen, C., Napala, A., MacHugh, D.E., Bradley, D.G. and Dao, B., 1996.** Phenotypic designations of Togolese cattle correlate strongly with microsatellite allele frequency data. (Meeting abstract no. A018.) *Animal Genetics*, **27** (Suppl. 2): 21.

Hendrickx: GCP-TOG-013-BEL, B.P. 114, Sokodé, Togo. A phenotype map of the sedentary cattle herds of Togo was constructed using morphological criteria. Four classes of cattle, based on the degree of zebu influence, were recorded. The proportion of animals classified as taurine within 1/8th degree grid squares and within individual herds was found to be an excellent predictor of trypanotolerance. Microsatellite loci which display zebu-specific alleles were used to calculate the extent of zebu influence in a representative sample of 722 Togolese cattle. Independent phenotypic designations were made for the

same cattle. Strong correlation was observed between the proportion of zebu alleles detected in each herd and the average phenotype. This correlation between phenotype and microsatellite frequency data suggests a novel means of investigating breed composition and breed relationships.

10066 **Rink, A., Urban, B., Hess, M., Förster, B., Mehlitz, D. and Horstmann, R.D., 1996.** Bovine analogues of human trypanolytic factors. (Meeting abstract no. C030.) *Animal Genetics*, **27** (Suppl. 2): 63.

Rink: Department of Molecular Genetics, Bernhard Nocht Institute for Tropical Medicine, 20359 Hamburg, Germany.

Recently haptoglobin-related protein (HRP) and paraoxonase arylesterase (PONA) have been claimed to be trypanolytic factors in human serum. To investigate the mechanism of trypanotolerance expressed by certain cattle breeds, bovine analogues of human HRP and PONA were sought. Two full-length cDNA clones were isolated. The coding sequence of one of them showed 84% similarity with the human *PONA* gene for both the nucleotide and the deduced amino acid sequence. The bovine cDNA clone identified with an HRP probe resembles human HRP and human haptoglobin (HPP). Comparison of the nucleotide and deduced amino acid sequence with HRP and HPP revealed similarities of 78% and 74%, respectively.

10067 **Uza, D.V., 1997.** The productivity of Muturu cattle (*Bos brachyceros*) under ranching conditions in the southern Guinea Savanna of Benue State, Nigeria. *Outlook on Agriculture*, **26** (1): 19-23.

Department of Animal Production, University of Agriculture, P.M.B. 2373, Makurdi, Nigeria.

An analysis of the productivity of Muturu cattle, a native breed with tolerance to trypanosomiasis, under ranching conditions was carried out at Raav, Benue State, during 1981 to 1991. Six calving seasons occurred during this period. When the performance of the Muturu under ranching and village management conditions was compared, age at first service, age at first calving and calving interval appeared to be similar. However, calf mortality rate in the ranch herd (5%) was lower than that in the village herd (10.8%). When the productivity of the Muturu in the ranch and village herds were compared to the Bunaji, the predominant indigenous zebu cattle breed in Nigeria, the Muturu cattle were superior in age at first service, age at first calving, calving interval

and calf mortality rate. These results suggest that the Muturu should be studied further with a view to enhancing their productivity.

(d) TREATMENT

[See also **20**: nos. 10013, 10016, 10022-10024, 10059.]

10068 **Akingbemi, B.T., Madekurozwa, M.N. and Joshua, R.A., 1995.** Effect of chemotherapy on some haematological and serum biochemical parameters in Mashona goats experimentally infected with *Trypanosoma congolense*. *Bulletin of Animal Health and Production in Africa*, **43** (4): 269-275.

Akingbemi: Department of Preclinical Veterinary Studies, University of Zimbabwe, P.O. Box MP 167, Harare, Zimbabwe.

Haematological and serum biochemical parameters were studied in indigenous Mashona goats experimentally infected with *T. congolense*. Four weeks after infection, there were significant changes ($P < 0.05$) in these parameters. The erythrocyte count, haematocrit and haemoglobin concentration with initial values of 13.40 ± 1.10 ($\times 10^{12}/l$), $32.00 \pm 1.77(\%)$ and 13.57 ± 0.90 (g/dl) had decreased ($P < 0.05$) to 3.55 ± 0.62 , 12.24 ± 2.98 and 5.07 ± 0.81 , respectively, 4 weeks p.i. Similarly, there were significant decreases ($P < 0.05$) in serum alkaline phosphatase activity and concentration of albumin, cholesterol and creatinine, along with increases in the concentration of globulins, total bilirubin and urea. There were no changes in these values in the control animals over time. The values obtained from the infected animals, however, returned to normal levels after a single administration of diminazene aceturate at 7.0 mg/kg body weight. The infected animals, however, appeared to have been able to curtail parasite growth as parasitaemia was latent and therefore could not be quantified microscopically. These findings further suggest that diminazene may be of good therapeutic value in controlling trypanosomiasis in these animals.

10069 **Eisler, M.C., Gault, E.A., Molloo, S.K., Holmes, P.H. and Peregrine, A.S., 1997.** Concentrations of isometamidium in the sera of cattle challenged with drug-resistant *Trypanosoma congolense*. *Acta Tropica*, **63** (2-3): 89-100.

Eisler: University of Glasgow Veterinary School, Bearsden Road, Glasgow G61 1QH, UK.

The relationship between serum concentrations of the prophylactic trypanocidal drug isometamidium chloride and protection against tsetse challenge with two populations of *T. congolense* was investigated in Boran (*Bos*

indicus) cattle, using an isometamidium-ELISA. Isometamidium chloride (Samorin) was administered to cattle at a dose rate of 1.0 mg/kg body weight by deep i.m. injection. Thereafter, the animals were challenged at monthly intervals with either a drug-sensitive clone (*T. congolense* IL 1180) or a clone expressing a moderate level of resistance to isometamidium (*T. congolense* IL 3343). Untreated control cattle were used to confirm the infectivity of each challenge. Of ten drug-treated cattle that were challenged with *T. congolense* IL 3343, all were refractory to infection at the first challenge, 1 month after drug administration. However, all ten animals succumbed to infection at either the second (seven cattle) or third (three cattle) monthly challenges. By contrast, all five drug-treated cattle challenged with *T. congolense* IL 1180 resisted four monthly challenges. The mean isometamidium concentration at the time of the first, 1 month, challenge was 5.6 ± 2.8 ng/ml. At the time of the second monthly challenge the mean concentration was 2.0 ± 0.86 ng/ml: at this time, concentrations were not significantly different between those cattle refractory to challenge with *T. congolense* IL 3343 and those cattle that were not. Thus, differences in susceptibility to challenge at this time would appear to be due to differences in the drug sensitivity of the parasite challenge. Finally, the mean isometamidium concentration in uninfected cattle at the time of the fourth monthly challenge was 0.4 ± 0.18 ng/ml. These results indicate that when *T. congolense* infection occurs in cattle under isometamidium prophylaxis, the parasites may be considered at least moderately drug resistant if the concentration of isometamidium in serum is 2.0 ng/ml. At concentrations between 0.4 and 2.0 ng/ml a low level of drug resistance may be inferred. Below 0.4 ng/ml, however, no inference regarding drug resistance should be made.

10070 **Geerts, S., Kageruka, P., Deken, R. de, Brandt, J.R.A., Kazadi, J.M., Diarra, B., Eisler, M.C., Lemmouchi, Y., Schacht, E. and Holmes, P.H., 1997.** Prophylactic effects of isometamidium- and ethidium-sustained release devices against *Trypanosoma congolense* in cattle. *Acta Tropica*, **65** (1): 23-31.

Geerts: Institute of Tropical Medicine, Nationalestraat 155, 2000 Antwerp, Belgium.

Two successive experiments were carried out in which three cows were treated by i.m. injection of either 0.5 mg/kg isometamidium or 1 mg/kg ethidium and compared with another group of three cows which received a

subcutaneously implanted sustained release device (SRD) containing the same dose of drug. The prophylactic effect of both drug formulations was evaluated by exposing the animals at monthly intervals to *Glossina morsitans morsitans* infected with *T. congolense*. The average protection period using the isometamidium- and the ethidium-SRD was extended by a factor of 3.2 and 2.8, respectively, in comparison with the i.m. injection of the drugs. In the analysis of isometamidium concentrations in the serum of the animals using a competitive drug-ELISA the drugs remained present for much longer periods in the sera of the implanted animals than in those of the i.m. treated cattle. The animals were still protected, however, a long time after the disappearance of detectable drug levels in the serum. No difference in drug sensitivity could be observed when breakthrough isolates were compared from animals which received the ethidium-SRD and those treated i.m., although a slight loss of sensitivity occurred in the breakthrough isolates as compared to the parent trypanosome population.

10071 **Mulugeta, W., Wilkes, J., Woudyalew Mulatu, Majiwa, P.A.O., Masake, R. and Peregrine, A.S., 1997.** Long-term occurrence of *Trypanosoma congolense* resistant to diminazene, isometamidium and homidium in cattle at Ghibe, Ethiopia. *Acta Tropica*, **64** (3-4): 205-217.
Peregrine: ILRI, P.O. Box 30709, Nairobi, Kenya.
Ten trypanosome isolates were collected at random from cattle at Ghibe, Ethiopia, in February 1993 and all shown to be savanna-type *T. congolense*. When inoculated into naive Boran (*Bos indicus*) calves, all 10 isolates were resistant to diminazene aceturate (Berenil), isometamidium chloride (Samorin) and homidium chloride (Novidium) at doses of 7.0 mg/kg, 0.5 mg/kg and 1.0 mg/kg body weight, respectively. In order to determine whether this multiple drug resistance was expressed by individual trypanosomes, clones were derived from two of the isolates and characterised in mice for their sensitivity to the three compounds: by comparison to drug-sensitive populations, the two clones expressed high levels of resistance to all three trypanocides. In experiments to characterise the uptake kinetics of [¹⁴C]-Samorin, the maximal rates of uptake (V_{max}) for four Ghibe isolates ranged from 9.2 to 15.0 ng/10⁸ trypanosomes/min. In contrast, V_{max} for the isometamidium-sensitive clone *T. congolense* IL 1180 was 86.7 ± 8.6 ng/10⁸ trypanosomes/min. Lastly, molecular

karyotypes were determined for eight isolates: seven different chromosome profiles were observed. These data indicate that in February 1993 there was a high prevalence of drug-resistant trypanosome populations with different chromosome profiles in cattle at Ghibe. Since a similar situation existed at the same site in July 1989, this suggests that the drug-resistance phenotype of trypanosomes at Ghibe had not altered over a 4-year period.

7. experimental trypanosomiasis

(a) DIAGNOSTICS

10072 **Diallo, P.B., Truc, P., Meda, H.A. and Kamenan, A., 1996.** Diagnostic sérologique de la trypanosomose humaine africaine à *Trypanosoma brucei gambiense*. 1 - Obtention et utilisation d'antigènes bruts dans les tests ELISA et d'agglutination au latex. [Serological diagnosis of human African trypanosomiasis due to *T. b. gambiense*. 1 - Production and use of crude antigens for ELISA and latex agglutination tests.] *Bulletin de la Société de Pathologie exotique*, **89** (4): 262-268.

Diallo: IPR/OCCGE, 01 B.P. 1500, Bouaké 01, Côte d'Ivoire.

A major difficulty with the extraction of crude antigens from trypanosomes for immunodiagnosis has been obtaining a sufficiently large number of parasites. Various techniques used by different authors are described, some of them sophisticated and time-consuming. In this study, the rat *Cricetomys gambianus* was experimentally infected with *T. b. gambiense* to produce a high level of parasitaemia (10^8 - 10^9 trypanosomes/ml). Extraction and filtration of blood from infected rats produced extracts of 50-60 mg of crude antigen. In order to establish a specific and sensitive immunological test for Gambian trypanosomiasis, crude antigen from different strains of *T. b. gambiense* was compared using ELISA and latex agglutination tests. Confirming previous work, the results showed a high sensitivity but questionable specificity for both techniques.

10073 **Masake, R.A., Majiwa, P.A.O., Molloo, S.K., Makau, J.M., Njuguna, J.T., Maina, M., Kabata, J., ole-MoiYoi, O.K. and Nantulya, V.M., 1997.** Sensitive and specific detection of *Trypanosoma vivax* using the polymerase chain reaction. *Experimental Parasitology*, **85** (2): 193-205.

Masake: ILRI, P.O. Box 30709, Nairobi, Kenya. The nucleic acid probes that are currently in use detect and distinguish *T. vivax* parasites according to their geographic origin. To eliminate the need for using multiple DNA probes, a study was conducted to evaluate the suitability of a tandemly reiterated sequence which encodes a *T. vivax* diagnostic antigen as a single probe for detection of this parasite. The antigen is recognised by monoclonal antibody Tv27 currently employed in antigen detection ELISA (Ag-ELISA). A genomic clone which contained a tetramer of the 832-bp cDNA sequence was isolated and shown to be more sensitive than the monomer. Oligonucleotide primers were designed based on the nucleotide sequence of the 832-bp cDNA insert and used in amplifying DNA sequences from the blood of cattle infected with *T. vivax* isolates from West Africa, Kenya and South America. The polymerase chain reaction (PCR) product of approximately 400 bp was obtained by amplification of DNA from all the isolates studied. The oligonucleotide primers also amplified DNA sequences in *T. vivax*-infected tsetse flies. Subsequently, PCR was evaluated for its capacity to detect *T. vivax* DNA in the blood of three animals experimentally infected with the parasite. *T. vivax* DNA was detectable in the blood of infected animals as early as 5 days p.i. Blood and serum samples from the three cattle and from six other infected animals were also examined for the presence of trypanosomes and *T. vivax*-specific diagnostic antigen. Trypanosomes appeared in the blood 7-12 days post-challenge, while the antigenaemia was evident on days 5-20 of infection. Analysis of the data obtained in the three animals during the course of infection showed that the buffy coat technique, Ag-ELISA and PCR revealed infection in 42, 55 and 75% of the blood samples, respectively. PCR amplification of genomic DNA of *T. vivax* is thus superior to the Ag-ELISA in the detection of *T. vivax*. More importantly, both the *T. vivax* diagnostic antigen and the gene encoding it are detectable in all the *T. vivax* isolates examined from diverse areas of Africa and South America.

(b) PATHOLOGY AND IMMUNOLOGY

[See also 20: nos. 10118, 10121, 10130.]

10074 **Clapcott, S.J., Kemp, S.J., Gill, J.J.B. and Teale, A.J., 1996.**

Identification of imprinted genes controlling

resistance to trypano-somiasis in mice. (Meeting abstract no. B037.) *Animal Genetics*, **27** (Suppl. 2): 51.
Clapcott: Department of Genetics and Microbiology, University of Liverpool, Liverpool, UK.

10075 **Ekanem, J.T., Akanji, M.A. and Odutuga, A.A., 1996.** Elevated erythrocyte calcium and Ca²⁺-ATPase activity in *Trypanosoma brucei*-infected rats. *Biomedical Letters*, **54** (213): 7-11.

Ekanem: Department of Medical Biochemistry and Biophysics, Umeå University, S-90187 Umeå, Sweden.

10076 **Gouteux, S. and Buguet, A., 1996.** Analyse actimétrique des perturbations du rythme circadien du cycle veille-sommeil chez le rat infesté par *Trypanosoma brucei brucei*. [Actimetric analysis of circadian disturbances of the sleep-wake cycle in rats infected with *T. b. brucei*.] *Travaux scientifiques des Chercheurs du Service de Santé des Armées*, no. 17 (1995): 193-194.

Gouteux: CRSSA, B.P. 87, 38702 La Tronche Cedex, France.

10077 **Hiepe, F., Jungnitz, S. and Hiepe, T., 1996.** Production of auto-antibodies against DNA and collagen after inoculation of rabbits with *Trypanosoma equiperdum*. *Applied Parasitology*, **37** (4): 266-274.

F. Hiepe: Medizinische Klinik und Poliklinik III des Universitäts-klinikums Charité, Schumannstrasse 20/21, D-10117 Berlin, Germany.

10078 **Keita, M., Bouteille, B., Enanga, B., Vallat, J.-M. and Dumas, M., 1997.** *Trypanosoma brucei brucei*: a long-term model of human African trypanosomiasis in mice, meningo-encephalitis, astrocytosis, and neurological disorders. *Experimental Parasitology*, **85** (2): 183-192.

Keita: Institut d'Epidémiologie Neurologique et de Neurologie Tropicale, 2 rue du Docteur Raymond Marcland, F-87025 Limoges, France.

10079 **Kennedy, P.G.E., Rodgers, J., Jennings, F.W., Murray, M., Leeman, S.E. and Burke, J.M., 1997.** A substance P antagonist, RP-67,580, ameliorates a mouse meningoencephalitic response to *Trypanosoma brucei brucei*. *Proceedings of the National Academy of Sciences of the United States of America*, **94** (8): 4167-4170.

Kennedy: Department of Neurology, Glasgow University, Institute of Neurological Sciences, Southern General Hospital, Glasgow G51 4TF, UK.

10080 **Ojok, L. and Weiss, E., 1995.** Light and electron

microscopical studies of *in vivo* phagocytosis of *Trypanosoma congolense* by circulating blood leukocytes in *Mastomys natalensis* (multimammate rats). *Bulletin of Animal Health and Production in Africa*, **43** (2): 95-103.

Ojok: Department of Veterinary Pathology, Faculty of Veterinary Medicine, Makerere University, P.O. Box 7062, Kampala, Uganda.

10081 **Teale, A., Iraqi, F., Darvasi, A., Hanotte, O., Gathuo, H., Sileghem, M., Womack, J.E., Soller, M. and Kemp, S., 1996.** Genetics of resistance to trypanosomiasis in mice and livestock. [*T. congolense*, *T. vivax*.] *Animal Genetics*, **27** (Suppl. 2): 5-6.

Teale: ILRI, P.O. Box 30709, Nairobi, Kenya.

10082 **Tomlinson, S., Muranjan, M., Nussenzweig, V. and Raper, J., 1997.** Haptoglobin-related protein and apolipoprotein AI are components of the two trypanolytic factors in human serum. *Molecular and Biochemical Parasitology*, **86** (1): 117-120.

Tomlinson: Department of Pathology, New York University Medical Center, 550 First Avenue, New York, NY 10016, USA.

10083 **Uimari, P., Kemp, S.J., Dekkers, J.C.M., Teale, A.J. and Kennedy, B.W., 1997.** Sensitivity of segregation analysis to data structure and transformation: a case study of trypanotolerance in mice. [*T. congo-lense*.] *Heredity*, **78** (4): 424-432.

Uimari: Centre for Genetic Improvement, Department of Animal and Poultry Science, University of Guelph, Guelph, Ontario N1G 2W1, Canada.

10084 **Veiga Fernandes, J.H., Atouguia, J.M., Peleteiro, M.C., Jennings, F.W. and Rosário, V.E., 1997.** Post-treatment hind-leg paralysis in mice infected with *Trypanosoma brucei brucei*: a light microscopic study. [Diminazene.] *Acta Tropica*, **63** (2-3): 179-184.

Rosário: Centro de Malária e outras Doenças Tropicais (CMDT), Rua da Junqueira 96, 1300 Lisbon, Portugal.

(c) CHEMOTHERAPEUTICS

[See also **20**: nos. 10079, 10084, 10100.]

10085 **Anene, B.M., Udechukwu, A.C. and Anika, S.M., 1995.** Effects of DFMO alone and in combination with levamisole in the treatment of experimental *Trypanosoma congolense* infection of rats. *Bulletin of Animal Health and Production in Africa*, **43** (2): 143-144.

Anene: Faculty of Veterinary Medicine, University of Nigeria, Nsukka, Nigeria.

10086 **Bellevue, F.H., Boahbedason, M., Wu, R., Woster, P.M., Casero, R.A., Rattendi, D., Lane, S. and Bacchi, C.J., 1996.** Structural comparison of alkylpolyamine analogues with potent *in*

- vitro* antitumor or anti-parasitic activity. [*T. b. brucei*, *T. b. rhodesiense*.] *Bioorganic and Medicinal Chemistry Letters*, **6** (22): 2765-2770.
Woster: Department of Pharmaceutical Sciences, Wayne State University, Detroit, MI 48202, USA.
- 10087 **Kaminsky, R., Schmid, C. and Brun, R., 1996.** An 'in vitro selectivity index' for evaluation of cytotoxicity of antitrypanosomal compounds. *In Vitro Toxicology*, **9** (3): 315-324.
Kaminsky: Swiss Tropical Institute, Socinstrasse 57, 4002 Basel, Switzerland.
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8. trypanosome research

(a) CULTIVATION OF TRYPANOSOMES

10097 **Hirumi, H., Martin, S., Hirumi, K., Inoue, N., Kanbara, H., Saito, A. and Suzuki, N., 1997.** Cultivation of bloodstream forms of *Trypanosoma brucei* and *T. evansi* in a serum-free medium. *Tropical Medicine and International Health*, **2** (3): 240-244.

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(b) TAXONOMY, CHARACTERISATION OF ISOLATES

10098 **Lukes, J., Jirku, M., Dolezel, D., Kral'ová, I., Hollar, L. and Maslov, D.A., 1997.** Analysis of ribosomal RNA genes suggests that trypanosomes are monophyletic. [Incl.

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Maslov: Department of Biology, University of California, 3401 Watkins Drive, Riverside, CA 92521, USA.

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Fifty-two *T. brucei* stocks isolated in Côte d'Ivoire from sympatric locations were analysed by cellulose acetate electrophoresis of isoenzymes. Of 13 genetic loci surveyed, 5 appeared as variable, which made it possible to delimit 12 different zymodemes. The most abundant zymodeme involved stocks isolated from both humans and pigs, which is consistent with the hypothesis that pig is a reservoir of human African trypanosomiasis in Côte d'Ivoire, as already proposed by other authors. Population genetic analysis of the isozyme data indicated a strong linkage disequilibrium, which suggests that genetic recombination is severely restricted in this sample and favours the hypothesis that the trypanosome populations surveyed are basically clonal. Nevertheless, additional studies are required to better estimate the long-term stability of these clones and the possible interference of gene exchange at an evolutionary scale. The results corroborate the hypothesis that a majority of human *T. brucei* stocks from West Africa correspond to a fairly homogeneous cluster of genotypes (*T. brucei gambiense* 'Group I', Gibson 1986).

(c) LIFE CYCLE, MORPHOLOGY, BIOCHEMICAL AND MOLECULAR STUDIES

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