

### NETHERLANDS SOCIETY FOR PARASITOLOGY

## AUTUMN MEETING OF THE NETHERLANDS SOCIETY FOR PARASITOLOGY

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Dr. E. J. RUITENBERG in the chair

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#### Communications

1. E. SCHOLTYSECK (Abteilung für Protozoologie, Zoologisches Institut der Universität, Bonn, Federal Republic of Germany)

Host-parasite relationships in the Sporozoa (Supported by the "Deutsche Forschungsgemeinschaft")

The nature of the relationship between two partners in a host-parasite system is dependent on numerous factors. Among these are: species of the parasite and host involved, the kind of parasitism (i.e. intra- or extracellular), virulence of the parasite, resistence and immunity of the host, developmental stages of the parasite, site and kind of tissue or cells in the host affected, age of the host, etc.

The sporozoan group of protozoans offers an especially desirable and interesting model to study the reactions which occur between a host-parasite association. This is true because these organisms have more than one route of invasion (i.e. ingestion, insect transmission), they infect a variety of organs and tissues, they undergo sexual and asexual reproduction, they are both intra- and extracellular and many stages are morphologically distinct.

The following examples will be used for illustration of host-parasite relationships: the invasion of host-cells by infective stages of *Toxoplasma*, *Eimeria*, and *Plasmodium*. The intake of nutrients in *Toxoplasma*, *Eimeria*, and *Plasmodium*. Special structures occurring in the interface between intracellular parasites and their host cells (i.e. parasitophorous vacuole, intravacuolar folds and tubules, crescent bodies, etc.).

It may be possible that the host-parasite relationship influences the development of the parasite by determining the number of schizogonic generations, the initiation of sexual development and even differentiation of micro- and macrogamonts as shown by Grell in *Eucoccidium* (1953).

Some aspects of host-parasite relationships can be studied more conveniently by use of cultured cells as compared to *in vivo* infections. *In vitro* studies are practical for demonstrating the activities of enzymes in the parasite as well as the host cell because it is likely that this relationship is determined mostly by physiological factors.

2. W. TADROS (Laboratory of Parasitology, University of Amsterdam)

Recent advances in research on toxoplasmid coccidia

For many years Toxoplasma, Sarcocystis, Besnoitia and Frenkelia were collectively referred to as the Toxoplasma — a group of uncertain toxonomic affinities. The discovery of the coccidian life-cycle of Toxoplasma (1969) by Hutchison, revolutionized our understanding of the epidemiology of toxoplasmosis and paved the way for the elucidation of the life-cycles of the other three members of the Toxoplasmea. In 1972, the coccidian life-cycle of bovine, ovine and porcine Sarcocystis were demonstrated by Rommel et al. in Germany. Ingestion of Sarcocystis by the appropriate final host, man, dog or cat initiating gametogony in the lamina propria of the intestine, which results in the excretion of fully sporulated I. hominis-type sporocysts.

The adaptation of the indirect immunofluorescence technique (I.F.A.) to detect circulating

Trop. geogr. Med., 28 (1976) 362-364

antibodies against I.hominis is described and the results of a serological survey of selected human and cattle sera in the Netherlands is briefly outlined. Preliminary results of the cross reaction of Sarcocystis, Toxoplasma, Frenkelia and Besnoitia in the I.F.A. are presented. The development of a convenient rodent-mustelid model for the investigation of obscure aspects of Sarcocystis-induced isosporiasis is described. Sarcocystis of the vole, Microtus arvalis was shown to undergo sexual development in the intestine of the weasel Mustela nivalis. The I.hominis-type sporocysts (12  $\times$  9 $\mu$ m) excreted seven days post infection (patency ± 30 days), when fed to laboratory-bred voles, induced schizogony in lymph nodes and visceral organs, followed by sarcocyst formation in the muscle.

The recent elucidation of the life-cycle of Besnoitia as a coccidian of cats (Wallace and Frenkel, 1975; Peteshev et al., 1974) and of Frenkelia as a stage in the life-cycle of Isospora buteonis of the

common buzzard is diagrammatically illustrated.

Finally, speculation on the evolutionary gradation from monoxenous, through facultative heteroxenous to obligatory heteroxenous life-cycle pattern within the eimerid coccidia is presented and discussed.

The original work presented was carried out in collaboration with Prof. J. J. Laarman. The serological investigation was performed in Prof. C. F. A. Bruyning's Laboratory, University of Leiden, with the skilful assistance of Miss A. van den Eijk.

3. J. P. OVERDULVE (Institute of Tropical and Protozoan Diseases, Faculty of Veterinary Medicine, University of Utrecht)

Toxoplasma in the cat (Sustained by the Netherlands Organization for the Advancement of Pure Research (Z.W.O.), grant nr. 93-29, and by the Deutscher Akademischer Austauschdienst, DAAD)

Light microscopical and partly also electronmicroscopical investigations of the small intestines of cats, orally infected with Toxoplasma cysts, have revealed at least four different types of merozoites, provisionally called M1 to M4 (M1: orange segment shaped, 3.5. to 4.5 by 2.5  $\mu m$ ; nucleus about  $^2$   $\mu$ m; M2: banana shaped, 5 to 6 by 2 to 3  $\mu$ m, nucleus about 2  $\mu$ m; M3: slender, sickle shaped, 8 to 9 by 1 to 1.5  $\mu$ m, nucleus small, 1 to 1.5 by 2  $\mu$ m; M4: large, bean shaped or ellipsoid, up to 12 by  $6 \mu m$ , nucleus large, up to  $4 \mu m$ ) and at least three different types of schizonts, provisionally called S1 to S3 (S1: small, 4 to 8 M1-merozoites arranged fan-wise around a residual body, merozoite formation often not synchronous; S2: large, developing from M4-merozoites, up to 36 M2-merozoites arranged in a rosette, variable amount of residual cytoplasm; S3: small, compact, 8-16 M3-merozoites, no residual body), as well as macrogametes and microgamonts and -gametes.

Evidence was obtained that:

(1) there is no direct intra-intestinal development from cystozoites (bradyzoites) to gamonts but that in between there exists an extra-intestinal pregametogonic stage in the intestinal organs from which parasites re-invade the intestinal epithelium to form gamonts;

(2) some of the intra-epithelial merozoites and schizonts, described above, are not infective for mice; (3) sexual differentiation starts already before the actual formation of gamonts, i.e. gamontforming merozoites and the schizonts from which these merozoites are derived, are already sexually differentiated, M2 merozoites probably developing to macrogamonts and M3 merozoites to microgamonts.

# 4. A. HILL-KROOS (Laboratory of Zoology, University of Nijmegen)

The life-cycle of hypopi living "phoretical" upon small mammals

The hypopus, a heteromorphic deutonymph, has neither a mouth-opening nor mouth parts. Their occurrence on insects, mammals and birds is considered phoretical.

106 species were mentioned in the review of hypopi living on mammals (FAIN, 1969), from those

only three species had known life-cycles.

In Nijmegen a cultivation-method was established that allowed the study of the life-cycle of seven species.

As an example the life-cycle of a parasite living on the native wood mouse is discussed. The free-living stages in the nest of the host have very short developing times, since the parasite and non-phoretic hypopus stage lasts about eleven months. The parasitic- and host-reproduction are closely related; changing of the hormonetiter of the host may be the stimulus for the development of the

Trop. geogr. Med., 28 (1976) 362-364

5. J. J. MELLINK (Laboratory of Medical Parasitology, Free University, Amsterdam)
On the potential relevance of vector-host interactions for the infection process.

Transmission of infectious agents by haematophagous arthropods is, in general, directly linked to the act of blood feeding of the vector. Course and consequences of the latter could therefore probably affect the initial phases of ensuing arthropod-borne infections.

In this respect very little is known so far, concerning the possible relevance of qualities of the blood feeding process. The same applies to the responses of the host to the feeding activities of the vector. Especially in the latter case conjectures appear inciting. Host responses may reciprocally act on the vector, including its biting performance. A whole complex of vector — host interactions is emerging, with all sorts of potential repercussions on a concurrent or subsequent infection process.

The present situation in this field of research was surveyed, in connection with the presentation of an experimental approach. Some preliminary results of the project refered to above, were used to indicate its eventual perspectives.

### 6. J. H. E. Th. MEUWISSEN (Laboratory of Parasitology, University of Nijmegen)

The malaria parasite inside and outside the erythrocyt

A short review is given of recent studies (Miller, Aikawa, Dvorak etc.). Attention is paid to the E.M. studies on the Caveola vesicle complex in erythrocytes infected with vivaxtype Plasmodia, to the studies on the receptor site for merozoites on host red blood cells and finally to the mechanism of the host cell penetration by merozoites, as an introduction to the NIH film "Invasion of erythrocytes by malaria parasites", produced by Dvorak, Miller and Whitehouse.

# 7. W. J. TERPSTRA, H. P. T. VAN HELDEN, B. M. OKOT-KOTBER AND V. M. EYAKUZE (East African Institute for Medical Research, Mwanza, Tanzania)

Immunofluorescence patterns in schistosomiasis

During serological studies in schistosomiasis with the Indirect Fluorescent Antibody Technique using sections of adult male *Schistosoma mansoni* worms for antigen, distinct immunofluorescence patterns were observed. Essentially two patterns, "focal" and "diffuse" fluorescence which may occur alone or in combination, could be distinguished. In the "focal" pattern fluorescence is observed localized in the worm's intestine and/or a few adjacent areas. In the "diffuse" pattern, fluorescence is observed evenly distributed throughout the worm tissues. An intermediate state demonstrates both patterns. In a longitudinal study in experimental animals infected with *S.haematobium* and people naturally infected with *S.haematobium* or *S.mansoni*, the phenomenon of "focal" fluorescence was found to be the first serological sign of an infection. In a later stage of the infection the development of "diffuse" fluorescence was observed.

# 8. H. P. T. VAN HELDEN, W. J. TERPSTRA, G. J. J. M. VAN EYS AND V. M. EYAKUZE (East African Institute for Medical Research, Mwanza, Tanzania)

The epidemiological significance of "focal" immunofluorescence in schistosomiasis

While applying the Indirect Fluorescent Antibody Technique for serodiagnostic purposes in East Africa, using worm sections of *S.mansoni* as antigen, essentially two patterns of fluorescence, "focal" and "diffuse" could be distinguished. The "focal" immunofluorescence pattern was found to be characteristic of a young infection.

A serologic prevalence survey was carried out in an area highly endemic for *S. mansoni* and an area highly endemic for *S. haematobium* infections. In both areas "focal" fluorescence, alone or in combination with "diffuse" fluorescence was found to decrease in prevalence with age. Pure "focal" fluorescence was observed mainly in the youngest groups, which confirms the view that in highly endemic areas people usually become infected at an early age.

Trop. geogr. Med., 28 (1976) 362-364