

**Bulletin
of the
SCANDINAVIAN SOCIETY
FOR PARASITOLOGY**



**PROCEEDINGS OF THE XIX SYMPOSIUM OF THE SCANDINAVIAN
SOCIETY FOR PARASITOLOGY, ICELAND, 8-11 MAY, 1999**

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BULLETIN OF THE SCANDINAVIAN SOCIETY FOR PARASITOLOGY

The Bulletin is a membership journal of the Scandinavian Society for Parasitology. Besides membership information, it also presents articles on all aspects of parasitology, with priority given to contributors from the Nordic countries and other members of the Society. It will include review articles, short articles/communications. Comments on any topic within the field of parasitology may be presented as Letters to the Editor. The Bulletin is also open for a short presentation of new projects. All contributions should be written in English. Review articles are commissioned by the editor, however, suggestions for reviews are welcomed.

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Cover: In Norse mythology, the giant ash tree - Yggdrasill - spreads its limbs over the entire mankind. The ash has three roots, each of them sucking water from its own spring.

The first spring- Hvergelmir - is found in the ice cold North; next to the spring, the serpent Níðhoggr is ceaselessly gnawing at the roots of the ash. The second spring - Mímisbrunnr - is the source of wisdom and is guarded by Mímir. The third spring - Urðarbrunnr - is guarded by three women, the Norns, which mete out man's thread of life.

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WELCOME BY THE PRESIDENT OF THE SCANDINAVIAN SOCIETY FOR PARASITOLOGY

By TELLERVO VALTONEN

Department of Biological and Environmental Science, University of Jyväskylä, Finland

The XIXth meeting of the Scandinavian Society for Parasitology in Iceland is a historical occasion. The meeting of the Society, a bi-annual occurrence, is here for the first time. Although this Nordic country has a population of 275,000, only five of these are members of this society! Consequently, the organisation of this meeting has involved a tremendous amount of work for those involved. The local organisers have demonstrated their activity and industriousness, which became apparent at two previous successful meetings held in Iceland under the auspices of this Society. These meetings were the symposium on "Parasites and ecology of marine and coastal birds" at Stykkisholmur in 1996 and one on "Parasites of biological and economic significance in the aquatic environment" at Heimaey in 1994. I would like to thank our Icelandic friends and colleagues for their hard work also for the present meeting, which has resulted in such large numbers of us visiting this lovely and very special country: a land of volcanoes, hot springs, glaciers, fishes, sagas, trolls and other ancient traditions which have unfortunately been lost in many other parts of the western world.

Today, on the behalf of the Scandinavian Society for Parasitology, I have the pleasure to welcome you all to our 19th Symposium. A successful meeting is not only the result of good organisation, but also the product of contributions from all those attending. Parasitological meetings are of great interest, not only because we are parasitologists, but because we represent so many different kinds of background: we are biologists of various kinds, microbiologists, veterinarians and medics. In our research, we use techniques and methods which are widely utilised in science generally, from those of molecular microbiology to those involved in testing theories of evolutionary ecology. Indeed, parasites, the subjects of our research are phenomenally adaptable in the way in which they utilise their hosts and survive in virtually all environments. A parasite takes the meal and asks the host to pay the bill, while protecting itself in order to survive in this evolutionary arms race.

At this meeting we have many very interesting papers; our invited speakers will enlighten us on topics, such as: "how to study ecological laws", "parasite mediated competition", "parasitological problems in different production systems" and "cell-to-cell interactions in malaria". A special welcome and thanks are directed towards our invited speakers, and in fact to all participants, for their valuable contributions. Finally I shall thank the local organising committee, Drs S.H. Richter, K. Skirnisson and M. Eydal, and the scientific committee headed by K. Skirnisson, who have performed this formidable task of preparation. We are also indebted to various agencies and sponsors who have contributed the success of this meeting.

INVITED PAPERS

HOW TO STUDY ECOLOGICAL LAWS

By P. ARNEBERG

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Ecological laws are typically proposed to hold for a large number of species over large geographic areas and long periods of time. Most ecological research, on the other hand, is carried out on a scale of few species over small areas and short time periods. Because small scale studies cannot be used to generalise about large scale laws without serious problems, most ecological research does not focus directly on general laws (e.g. Pimm, 1991). Rather, assessing the validity of ecological laws empirically requires an approach that focuses directly on a scale as large as the laws are proposed to apply at. One way to do so is to use the comparative method. The comparative method uses data from large number of species collected over large areas and long time periods to test general hypotheses by examining patterns among species (Harvey & Pagel, 1991). An example of a central law within parasite population ecology, that has been studied using this approach, is the assumption that parasite transmission rate is a positive function of host population density. By using data from 19 mammalian species, representing examination of more than 6,500 host individuals, a positive relationship was found between host population density and abundance of gastrointestinal strongylid nematode parasites, suggesting that the proposed law is indeed generally valid for this group of parasites and hosts (Arneberg *et al.*, 1998).

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ECOLOGY AND EPIDEMIOLOGY OF HANTAVIRUSES: A EUROPEAN PERSPECTIVE

By H. HENTTONEN¹, O. VAPALAHTI², A. PLUYSNIN², Å. LUNDKVIST³, J. NIEMIMAA¹ & A. VAHERI²

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Hantaviruses are enveloped RNA viruses carried by different rodent species. When transmitted to man, some Eurasian hantaviruses, carried by murine or arvicoline rodents, cause different forms of haemorrhagic fever with renal syndrome (HFRS). Hantavirus pulmonary syndrome (HPS) is caused by hantaviruses found in New World rats and mice (Sigmodontinae) in the Americas. Two hantaviruses, Puumala (PUU), carried by the bank vole *Clethrionomys glareolus*, and Dobrava (DOB), carried by yellow-necked mouse *Apodemus flavicollis* and striped field mouse *A. agrarius*, are known to cause HFRS in Europe. PUU virus generally causes a milder disease with fever, headache, gastrointestinal symptoms and impaired renal function, while DOB infections have more often also haemorrhagic complications. Tula virus (TUL) from common voles *Microtus arvalis* and *M. rossiaemeridionalis* is not known to cause HFRS. PUU and its host *C. glareolus* are found in most of Europe excluding the Mediterranean region, while DOB is found mainly in the Balkans and Eastern and Central Europe. It seems that the DOB strain found in *A. flavicollis* causes a more severe HFRS in the Balkans than the one in *A. agrarius* in Eastern Europe.

The population dynamics of rodents in Europe vary geographically within and between the species, and these differences are reflected in the seasonal and multianual patterns of human epidemiology. In the north, pronounced regular 3-4 year vole cycles characterize dynamics, while in Central Europe occasional mast years increase rodent densities. In the north, human epidemics peak in late autumn when bank voles invade human settlements while in southern Europe epidemics peak in summer and are obviously related to crop harvesting.

The serological relationships of hantaviruses mirror their genetic distances, which, in turn, follow the phylogeny of the carrier rodents. Closely related hantaviruses (e.g. within murines, or arvicoline) cross-react, and this has caused confusion in many earlier reports. E.g. there is no reliable documentation of the real Hantaan (HTN) virus either in rodents or humans in Europe, and obviously many earlier reports mentioning HTN in fact deal with DOB. Neither is there reliable documentation of HFRS by rat-borne Seoul virus (SEO) in Europe. The reliable documentation of specific hantaviruses should be based on RT-PCR and subsequent sequencing or neutralization assays comparing titers to all hantaviruses. Only convalescent-phase sera should be used in the latter assays because of unspecific cross-reactivity of acute-phase antibodies. On the other hand, the cross-reactivity between PUU and DOB (as

well as HTN) is very weak, particularly in the acute phase. Therefore, for human diagnostics in Europe, both PUU and DOB (or HTN that cross-reacts strongly with DOB) antigens are needed to reveal the hantavirus infections in Europe, also infections by those viruses yet to be found in murine and arvicoline rodents. The diagnosis of acute human hantavirus infection is based on detection of specific IgM or low avidity of specific IgG antibodies.

PARASITE-MEDIATED COMPETITION

By P.J. HUDSON

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Parasites are believed to have an important role in influencing biodiversity in species by generating genetic diversity on the one hand, and also by reducing the competitive ability of individuals and influencing abundance and distribution on the other. This talk will specifically address the question, "Do shared parasites influence the distribution of their host species?" The talk will review aspects of parasite-mediated competition and the theory of multiple host systems. This will be illustrated with detailed experimental work on *Heterakis gallinarum* shared between pheasants and grey partridges, providing evidence that pheasants can eliminate grey partridge locally.

ECHINOCOCCOSIS IN ICELAND - A HISTORICAL REVIEW

By P.A. PÁLSSON

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Hydatid disease of humans, sheep and cattle has been known in Iceland for centuries. In the old Icelandic literature from the 12th and 13th century, cases of the disease are described. Until the beginning of this century, most people in Iceland lived on farms where sheep, cattle, dogs and humans lived in close contact, often even under the same roof. Under such conditions; hydatid disease flourished, as long as the nature of the disease was not understood. It has been estimated by physicians working in Iceland in the middle of the 19th century that one out of every six Icelanders was infected with hydatid cysts.

Following H. Krabbe's studies on echinococcosis in Iceland in the year 1863, a campaign against this disease was initiated and supported by a decree in the year 1869. It was based primarily on enlightening the public, explaining the life-cycle of *Echinococcus granulosus*, emphasising immediate destruction of all cysts and infected offal, reduction of the dog population by taxation. Some years later, yearly anthelmintic treatment of all dogs at the end of the slaughter season was introduced. The

health authorities impressed upon the public the need to observe strict caution when dealing with dogs and never feed them raw offal or meat.

Reports from necropsies performed during the years 1932-1992 strongly indicate that infection with *E. granulosus* was practically eradicated in the population by the beginning of the 20th century. Surveys by meat inspectors indicate that hydatidosis in sheep and cattle was first brought under control 30-40 years later.

Although all evidence indicate that echinococcosis has now been reduced to the point of extinction, both in humans and animals, all control measures against the disease are still enforced.

REVIEW ON PARASITOLOGICAL PROBLEMS IN DIFFERENT PRODUCTION SYSTEMS

By A. PERMIN

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Poultry are kept in backyard or commercial production systems in most areas of the world and have within the last 40 - 50 years become one of the most important protein sources for man throughout the world. The total number of poultry in the world has been estimated by the Food and Agriculture Organization of the United Nations (1997) to be nearly 15,000 million. The most commonly kept poultry are domestic chickens (*Gallus* spp.), ducks (*Carina* spp.), geese (*Anser* spp.) and turkeys (*Meleagris* spp.). Among these, domestic chickens (*Gallus domesticus*) are the most important. Of the total meat production (beef, pork and poultry) poultry meat has increased from 20% to 30% in the last 10 years. In Africa, poultry meat is estimated to represent almost 25% of all meat and in some areas it even accounts for 100% of the animal protein available. In Asia and Europe poultry meat accounts for more than 20% of all meat produced, whereas in North and Central America more than 40% of all meat produced is poultry meat. This expansion in poultry production is in part due to easy industrialisation, e.g. short turnover, low establishment costs and efficient disease prophylaxis, compared with other livestock productions. The continuing growth of the poultry industry is supported by efficient disease prophylaxis, efficiency of poultry to convert vegetable protein into animal protein, its competitive cost and the implementation of new technologies.

Basically, there are two production systems. The modern commercialized intensive system and the traditional extensive or rural scavenging system. Approximately 75% of all chickens are kept in traditional, rural scavenging systems.

The mortality in commercial systems is in the range of 10% or less per year, whereas in the traditional systems the mortality has been estimated to be in the range of 80 - 90% within the first year after hatching! Chicks under 3 months of age have especially high mortality rates.

Among diseases in rural chickens, viral infections such as Newcastle Disease have attracted most attention, but recent studies have shown that a wide range of parasitic diseases are present in these systems with prevalences close to 100%. Knowledge related to parasitic infections is scarce and research in this area is almost non-existent. Also in the commercial systems, especially in deep-litter and free-range systems, parasites are widely distributed and contribute significantly to the low productivity, where the most commonly mentioned parasites are *Eimeria* spp., *Ascaridia galli* and *Heterakis gallinarum*.

Although it is known that parasites constitute a health problem in poultry, there are only a few reports on the prevalence and significance of endo-, ecto- and haemoparasites in the different poultry production systems. The following tables present the most important parasites of poultry:

Table 1 Ectoparasites of importance in poultry production.

Parasite	Hosts	Predilection site
The fowl tick: <i>Argas persicus</i>	chickens, turkeys, pigeons, ducks, geese	skin
Mites: <i>Dermanyssus gallinae</i> , <i>Ornithonyssus sylviarum</i> , <i>O.bursa</i>	chickens, turkeys, ducks, wild birds	skin
Mite: <i>Cnemidocoptes mutans</i>	chickens, turkeys	under the skin on legs, occasionally on comb and wattles
Flea: <i>Echidnophaga gallinacea</i>	chickens and other birds	head

Table 2 Haemoparasites of importance in the poultry production.

Parasite	Hosts	Predilection site
<i>Leucocytozoon</i> spp.	chickens, ducks, geese, turkeys	leucocytes, erythrocytes
<i>Plasmodium</i> spp.	chickens, turkeys	erythrocytes
<i>Haemoproteus</i> spp.	ducks, geese, chickens	erythrocytes
<i>Aegyptinella</i> spp.	chickens, turkeys, ducks, geese	erythrocytes

Table 3 Endoparasites of importance in poultry production.

Parasite	Hosts	Predilection site
Nematodes		
<i>Oxyuris mansoni</i>	chickens, turkeys, guineafowls, peafowl	eye, lacrimal duct
<i>Syngamus trachea</i>	pheasants, chickens, turkeys, geese, guineafowls, quails, peafowls	trachea, lungs
<i>Gongylonema ingluvicola</i>	chickens, turkeys, partridges, pheasants, quails	oesophagus, crop
<i>Tetrameres</i> spp.	chickens, turkeys, ducks, grouse, pigeons, quails, guineafowls, geese,	proventriculus
<i>Dispharynx nasuta</i>	chickens, turkeys, grouse, guineafowls, partridges, pheasants, pigeons, quails	oesophagus, proventriculus
<i>Acuaria hamulosa</i>	chickens, turkeys, grouse, guineafowls, pheasants, quails	gizzard
<i>Amidostomum anseris</i>	ducks, geese, pigeons	gizzard
Nematodes		
<i>Capillaria</i> spp.	chickens, turkeys, geese, grouse, quails, guineafowls, partridges, pheasants, pigeons	entire intestinal tract
<i>Ascaridia galli</i>	chickens, turkeys, doves, ducks, geese	small intestine, occasionally oviduct
<i>Ascaridia dissimilis</i>	turkeys	small intestine
<i>Heterakis</i> spp.	chickens, turkeys, ducks, geese, grouse, guineafowls, partridges, pheasants, quails	caeca
<i>Allodapa suctoria</i>	chickens, turkeys, doves, ducks, grouse, guineafowls, partridges, pheasants, quails	caeca
Cestodes		
<i>Raillietina</i> spp.	chickens, turkeys, guineafowls, pigeons	small intestine
<i>Davainea proglottina</i>	fowls, pigeons	small intestine
<i>Choanotaenia infundibulum</i>	fowls, turkeys	small intestine
<i>Hymenolepis</i> spp.	fowls, ducks, geese	small intestine
Trematodes:		
<i>Echinostoma revolutum</i>	ducks, geese	rectum, caeca
<i>Prosthogonimus</i> spp.	fowls, ducks, geese	bursa Fabricius, oviduct, cloaca, rectum
Protozoa:		
<i>Eimeria</i> spp.	Chickens, turkeys, ducks	small intestine
<i>Histomonas meleagridis</i>	turkeys, chickens,	caeca, liver

CELL-TO-CELL INTERACTIONS OF IMPORTANCE FOR THE DEVELOPMENT OF SEVERE *Plasmodium falciparum* MALARIA

By M. WAHLGREN

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It has previously been found that the capacity of *P. falciparum*-infected erythrocytes to bind uninfected erythrocytes (form rosettes) is associated with the virulence of the parasite. Importantly, rosetting infected erythrocytes also bind to endothelial cells. For example, *P. falciparum*-infected RBC (PRBC) selected for rosetting are also spontaneously upregulated for adhesion to platelet/endothelial cell adhesion molecule-1 (PECAM-1)/CD31 on the vascular endothelium. Soluble PECAM-1/CD31 and monoclonal antibodies (mAbs) specific for the amino terminal segment of PECAM-1/CD31 (domains 1-4) blocked the binding. IFN- γ , essential for the development of cerebral malaria in the mouse, was found to augment adhesion of human PRBC to PECAM-1/CD31 on endothelial cells. This may explain the curious age-dependent increase of cerebral malaria with the acquisition of immunity. Our results suggest that PECAM-1/CD31 is a virulence associated endothelial receptor of *P. falciparum* infected RBC. Rosetting parasites carry adhesive electron-dense fibrillar strands at the "knobs" on the infected erythrocyte surface.

Immunoglobulins are the major components of the strand. Similar fibrillar strands are also found in the cerebral microvasculature at autopsy, thus suggesting that rosetting parasites are equivalent to virulent parasites.

PfEMP1 (*Plasmodium falciparum* erythrocyte membrane protein 1, Mr 200,000-350,000) has been suggested as the important parasite-derived adhesion ligand expressed on the surface of the malaria-infected erythrocyte. By single-cell RT-PCR and cDNA cloning we have found that the adhesive rosetting PfEMP1 contains clusters of glycosaminoglycan-binding motifs. A recombinant fusion protein (DBL-1-GST) was found to adhere directly to normal erythrocytes, disrupt naturally formed rosettes, block rosette-reformation and bind to a heparin-sepharose matrix. The adhesive interactions could be inhibited with heparan sulfate or enzymes that remove heparan sulfate from the cell surface, while other enzymes or similar glycosaminoglycans of a like negative charge did not affect the binding. PfEMP1 is suggested to be the rosetting ligand and heparan sulfate, or a heparan sulfate-like molecule, the receptor both for PfEMP1 binding, and naturally formed erythrocyte rosettes.

SUBMITTED PAPERS - ORAL PRESENTATIONS

PREDICTABILITY OF METAZOAN PARASITE COMMUNITIES OF SOME ISOLATED CRUCIAN CARP (*Carassius carassius*) POPULATIONS

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Objective: The predictability of the parasite fauna and factors determining it have been subject of studies and under discussion during the last two decades. So far there is, however, no general agreement on main factors determining the composition of parasite faunas. In particular, the question of predictability of parasite communities has not been resolved to date. The aim of this study is to evaluate the predictability of one of the most isolated freshwater fish populations; crucian carp (*Carassius carassius*), which lives naturally in small, easily outlined, very isolated ponds.

Materials and methods: During the summer of 1998 on average 20 crucian carps of size about 10 cm in length were studied from 8 separate ponds in three different geographical location in Finland. All adult metazoan parasites were identified to species and larvae stages were also counted from fresh slides.

Results: Three dactylogyrids (*Dactylogyrus formosus*, *D. intermedius* and *D. wegneri*) were found from gills, and *Gyrodactylus carassii* from gills and fins. No other metazoan parasites were found. *D. formosus* and *D. wegneri* were present in all ponds. *D. intermedius* was present in 5 ponds and *G. carassii* in 5 ponds, making 2 to 4 species present in each pond. The prevalence of infection of different species varied between 10 to 100%. The abundance was highest for *D. intermedius*, whose numbers rose to over 3,000 individuals on one fish in one of the ponds.

Conclusion: As expected, the parasite communities within each pond were very homogenous due to overcrowded populations. Also the number of parasite species were low. All species found were specialists and also had a direct life-cycle, an indication of the extreme conditions where crucian carps live. A surprising observation was the similarity between these isolated populations, the same four monogenean species being found in all geographical locations studied. The benefit of specialization as well as the factors determining predictability are discussed.

ANALYSIS OF CYTOKINE EXPRESSION IN PIGS INFECTED WITH *Schistosoma japonicum*

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Objective: We have studied local Th2 (Il-10 and Il-4) and Th1 (IFN- γ) cytokine expression in pigs infected with the zoonotic blood fluke *Schistosoma japonicum*.

Materials and methods: Twelve 10-12 week-old, specific pathogen-free Danish Landrace/Yorkshire/Duroc crossbred pigs were infected by intramuscular injection (n=6) or per oral infection (n=6) of 1,000 *S. japonicum* cercariae. Three other pigs served as non-infected controls. The pigs were killed ten weeks after infection. Tissue samples of liver, small intestine, caecum, colon and rectum were taken from lesion sites as well as nearby areas without obvious lesions. These samples were kept in a RNase inhibitor solution at -80°C until a reverse transcriptase polymerase chain reaction (RT-PCR) was performed to obtain cDNA and subsequently amplify genes encoding the Il-10, Il-4 and IFN- γ pig cytokines.

Results: The *S. japonicum* infection induced Th2 cytokine responses in the liver, caecum and colon. This response was especially pronounced in the caecum and colon of the orally infected pigs. We then compared the cytokine expression levels in the lesion sites against the non-lesion sites of the liver, rectum, colon and caecum. Subsequent to an oral infection the Th2 type cytokine production was higher in the lesion sites of the liver and rectum. Interestingly, an apparent lower level of Il-4 and Il-10 was detected in lesion sites of the colon and caecum when compared to non-lesion sites.

Conclusion: We conclude that the RT-PCR is an appropriate technique to analyse cytokine expression in pigs which are used as an animal model for human *S. japonicum* infection.

THE MALE NEMATODE TAIL

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In the nematode male, the size and shape of its tail, cuticular ornamentation, number, size and position of caudal sensory papillae, as well as size, and shape of the spicules are taxonomically important. However, understanding copulation in nematodes is an exercise in interpreting functional morphology. The nematode body, being turgid and almost circular in cross section, poses problems for the male in finding and lock-

ing himself to the female's vulvar region. In many nematode groups, the male coils his posterior tightly round the female, but he should be able to flatten his ventral body to make intimate contact. In many nematode taxa, including the ascarids, so-called copulatory muscles between ventral mid-line and lateral body in front of cloaca, pull the ventral side in. The male tail thus becomes more or less spoon-shaped, the caudal papillae being in contact with female body. In cucullanids and heterakids some latero-ventral muscles are arranged as a rosette, forming a sucker.

In the strongylids the male tail forms a copulatory bursa, the papillae forming, so to speak, the rays in an umbrella, which may grip and hold on to the female. The male spirurids have a "cork-screw" tail, usually with one long, and one short spicule. Can the male coil either dextrally or sinistrally around the female?

Assuming that in most nematodes the male coils himself more or less transversely around the female, how is the female vagina arranged to receive the spicules? And why do some nematodes have exceedingly long spicules?

DIGENEANS IN COASTAL ECOSYSTEMS OF SW-ICELAND - FAUNA COMPOSITION AND TRANSMISSION PATTERNS

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Objective: Investigate the species composition of digenean parthenites and larvae in intertidal molluscs of SW-Iceland with emphasis on their transmission pattern in the coastal ecosystem.

Material and methods: Molluscs (Gastropoda, Prosobranchia) were collected at 11 intertidal sampling stations in Skerjafjörður and Grindavík (SW-Iceland) in August-September 1998. Totally, 899 *Littorina saxatilis*, 622 *L. obtusata*, 136 *L. mariae*, 325 *Onoba aculeus*, 349 *Nucella lapillus* and 80 *Ephera vineta* were dissected under a stereo microscope. Species determination of parthenites, cercariae and metacercariae was carried out in vivo using a light microscope.

Results: Totally, 19 digenean species were recorded: *Microphallus pygmaeus*, *M. piriformes*, *Microphallus* sp. 1 (*M. pseudopygmaeus*), *M. triangulatus*, *M. similis*, *Cercaria islandica* I, *Cercaria littorina saxatilis* VII, *Maritrema linguilla* (Microphallidae), *Renicola thaidus*, *Cercaria parvicaudata* (Renicolidae), *Cryptocotyle lingua* (Heterophyidae), *Parvatrema homoeotecnium* (Gymnophallidae), *Himasthla elongata*, *Cercaria littorinae obtusata* (Echinostomatidae), *Paramonostomum chabaudi*, *Notocotilidae* gen.sp. (Notocotilidae), *Parapronocephalum symmetricum* (Pronocephalidae), *Parorchis acanthus* (Philophthalmidae) and *Podocotyle atomon* (Opecoelidae). Fourteen of the 19 species are new for Iceland, and *Cercaria islandica* I from *Littorina obtusata* is new for the science. With the exception of *Podocotyle atomon*, which adults develop in fishes, marine and coastal birds are final

hosts of these digeneans. The representatives of microphallids of the "pygmaeus" group (*M. pygmaeus*, *M. piriformes*, *M. pseudopygmaeus* and *M. triangulatus*) have a so-called autonomic life cycle without second intermediate host and free-swimming larval stages (miracidium and cercaria). With the exception of *P. symmetricum*, however, all the remaining species have one or two free-swimming larvae in their complicated life-cycles. In *L. saxatilis*, *L. obtusata* and *O. aculeus* the prevalence (P) of microphallids of the "pygmaeus" group was 1-7%. Pooled P of the other species varied from 2 to 22%. We did not find significant P differences in microphallids of the "pygmaeus" group in the snails examined from Skerjafjörður and Grindavík. The same result was obtained for *R. thaidus* and *P. acanthus* in *N. lapillus* while the P of *M. similis*, *C. lingua*, *C. parvicaudata* and *C. littorinae obtusata* in periwinkles in Grindavík was significantly higher than in Skerjafjörður.

Conclusion: Species composition of digenean parthenites and larvae in intertidal molluscs of Iceland is richer than on the coasts of the Barents Sea and NE-Norway (Galaktionov & Bustnes, 1996). This is defined by the increase of number of species with complicated life-cycle. These species predominate in the total prevalence of digeneans in Icelandic molluscs while microphallids of the "pygmaeus" group play this role in arctic regions. Seemingly, the relatively mild climate, abundance and diversity of potential final hosts (birds), and second intermediate hosts (invertebrates and fishes), promote the successful completion of complicated life-cycles in the intertidal zone of SW-Iceland. Peculiarities of molluscan infection in Skerjafjörður and Grindavík are presumably determined by specific features in the distribution of the final hosts. A stable population of common eiders (*Somateria mollissima*) in both areas is regarded to be responsible for comparable infection levels of microphallids belonging to the "pygmaeus" group. However, relatively higher prevalences of gull digeneans (*M. similis*, *C. lingua*, *C. littorinae obtusata*) in the molluscs in Grindavík are regarded to be a result of the relatively high concentrations of herring gulls (*Larus argentatus*) and great black-backed gulls (*L. marinus*) which are attracted to the study area in all seasons by the fishing-port of Grindavík and a nearby aqua culture farm.

THE POSSIBLE INTERSPECIFIC COMPETITION BETWEEN THE NEMATODE-
TRAPPING FUNGUS *Duddingtonia flagrans* AND SELECTED FUNGI AND
BACTERIA ISOLATED FROM COW PATS AND SOIL

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The soil fungus, *Duddingtonia flagrans*, has the capacity to be able to trap and kill nematodes, e.g. larvae of the cattle parasite *Cooperia oncophora*, when placed in cattle faeces. Interspecific competition between this fungus and other microorganisms has been investigated.

Faeces from two stabled cows was collected, and by a selection program on TSA-'agar Petri dishes', 20 coprophilic bacteria were isolated from each portion of faeces. Five bacteria, isolated from faeces from one of the cows, were antagonistic (showed interspecific competition) to the nematode-trapping fungus *D. flagrans* (strain CI3 and Trol A), by depressing its growth on PDA-'agar Petri dishes'. Bacteria isolated from faeces from the other cow did not show any antagonistic properties. The 5 antagonistic bacteria (2 *Bacillus subtilis*, 2 *Pseudomonas* spp. and 1 *Streptomyces* sp.) will be tested in cattle faecal cultures containing *D. flagrans* chlamydospores and eggs of *C. oncophora*, to investigate if these bacteria are able to depress the nematode-trapping capacity of *D. flagrans* in cattle faeces.

In another experiment, the nematode-trapping capacity of *D. flagrans* (Trol A) was challenged, in a cattle faecal culture, by the coprophilic fungus *Pilobolus kleinii*. But even very high inoculations of faeces by *P. kleinii* spores did not suppress the ability of *D. flagrans* to trap and kill developing larvae of *C. oncophora*. In future experiments on agar, *D. flagrans* will be challenged by other fungi isolated from faeces and soil.

ECTOMYCORRHIZAE REDUCE DAMAGE OF RUSSIAN LARCH BY *Otiorhyncus* LARVAE

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The effect of ectomycorrhiza on damage caused by *Otiorhyncus* larvae and on plant growth was evaluated in a three-year field experiment. Russian larch seedlings, inoculated with ectomycorrhizal fungi, were compared with uninoculated plants. Assessment of larval damage at the end of the first growing season showed a significant effect of treatment on damage intensity, but not on damage incidence; 11.2% of uninoculated plants were severely damaged, but only 3.5% of inoculated plants. There was a significant effect of treatment on plant mortality. The accumulated mortality in the end of the third growing season was 34.5% for uninoculated plants, but 17.6% for inoculated plants. The height increment of inoculated plants was significantly greater than that of uninoculated plants in the first growing season; 4.5 cm for inoculated plants, but 2.7 cm for uninoculated plants. No difference in height increment between treatments was observed during the second and third growing seasons.

THE DISTRIBUTION OF A NEMATODE INFECTION IN A DANISH ROE DEER (*Capreolus capreolus*) POPULATION

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Objective: The size of a host and the numbers of parasites that they harbour are often expected to be negatively correlated because of the pathological effect related to worm burden. This should be the case for food transmitted parasites, with a direct life-cycle. But if a parasite has an indirect transmission, using an intermediate host which is associated with the host's preferred food, this correlation should be positive, if access to this food is linked to dominance. These relationships were examined in a roe deer population infected with lungworms and abomasal nematodes.

Materials and methods: Seventy-eight roe deer were shot during the hunting season of 1997 and 1998 at The Frijsenborg Estate, Denmark. The abomasum of all were examined for adult nematodes with direct life-cycle. First-stage protostrongylid larvae, with indirect transmission, were recovered from faeces. The intensity of the two

groups of nematode species were correlated to body weight and age. The hosts were grouped by gender.

Results: The prevalence of abomasal nematodes was 100%. For the protostrongylids it was 48.6% for the male, and 69.6% for the female roe deer. Generally, young hosts (1-2 yr.) harboured most parasites of both groups. There was no correlation between the intensities of protostrongylids and abomasal nematodes. As expected, we found the highest intensities of abomasal nematodes in the lightest roe deer. Unexpectedly, we found the same relationship between intensities of protostrongylids and body weight of the host. The heaviest roe deers tended to have small, or no infection with protostrongylids.

UNTANGLING THE WEB FROM INSIDE - PARASITES IN BIO-DIVERSITY STUDIES.

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Ecosystems are made up of populations of interacting species. Recent studies indicate that ecosystems have properties that can only be studied at the ecosystem level. An example of this is food web structure. It is normally a daunting task to set out to study food webs. However, we argue that macroparasites can contribute much ecological information in such studies.

A field study of freshwater fish in Costa Rica was carried out to contribute information to an ATBI (all-taxa-biodiversity-inventory) in one of the conservation areas in the country. We examined 21 species of fish representing 9 families. We have quantitative data on macroparasites from 10 fish species representing four families.

The number of parasite species among fish species varied between one and six. The highest number of parasite species was found in fish species which had plants, invertebrates and fish included in their diet. There was some exchange of parasite species within each fish family, but little indication of such exchange between families.

The information about parasite species, their intermediate hosts and the exchange of parasites between fish species will be used to discuss interactions among fish and their biotic environment.

THE EFFECT OF A PRIMARY *Toxocara canis* INFECTION ON A SUPERIMPOSED *Ascaris suum* INFECTION IN PIGS

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Objective: At present, it is not known whether concurrent infections with *Ascaris suum* and *Toxocara canis* results in interaction in the pig. Therefore, the development of cross-resistance against *A. suum* following a *T. canis* infection was studied with respect to population kinetics of *A. suum* and host immune response.

Materials and methods: Group A was primary inoculated with 60,000 *T. canis* eggs and four weeks later challenge-infected with 1,000 *A. suum* eggs; group B was inoculated with 1,000 *A. suum* eggs at the time of challenge, only. Subgroups from group A and B were slaughtered 7, 14 and 21 days after challenge infection (p.c.i.). Individuals from an uninfected group C were used as negative controls for blood parameters.

Results: Total recovery of *A. suum* was significantly reduced by 85% 7 days after challenge in group A, compared with group B. Fourteen days after challenge, the reduction was 20%. However, by day 21 the larval recovery was higher in group A compared to group B. The liver pathology was more severe in group A compared with group B on day 7 and 14 p.c.i. No cross-reaction was observed between sera from *A. suum* infected pigs and *T. canis* antigen. In contrast, sera from the *T. canis* infected pigs cross-reacted with *A. suum* antigen.

Conclusion: The *A. suum* larvae was temporarily arrested during the migration probably due to the strong pathological reactions in the liver. However, this did not affect the further migration and survival of the larvae. Serological cross-reaction was observed and should be taken into account when using serological tests for diagnosing.

APPLICATION OF AN *in ovo* CULTURE TECHNIQUE TO HELP SOLVE TAXONOMIC CONFUSION IN MICROPHALLID TREMATODES

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Objective: Accurate identification of larval trematodes often proves very difficult because these organisms are frequently devoid of anatomical features which can be used to assist in separation of closely related species. The *in ovo* culture technique

provides a method by which microphallid metacercariae can be cultured to adult flukes which, having well developed reproductive systems, can be more accurately identified.

Materials and methods: Encysted metacercariae are induced to excyst by incubation at 41°C in a saline solution containing digestive enzymes and bile salts. Unencysted metacercariae are treated in the same manner to ensure that they are activated. The resulting organisms are inserted into the chorio-allantois of incubating chicken embryos and, after approximately five days, mature microphallid flukes are recovered.

Results: Adult specimens of a number of species have been cultured. Comparison of these organisms with descriptions in the scientific literature have shown that they are morphologically identical to specimens obtained from their natural hosts.

Conclusion: Application of this technique has helped solve confusion arising from larvae having been described and named in isolation from their adult forms. It has also provided adult worms from which life cycles have been completed in the laboratory. This has facilitated the formulation of accurate descriptions of all the larval stages concerned.

PARASITE COMMUNITY STRUCTURE IN MIGRATORY WHITEFISH *Coregonus lavaretus wartmanni* L. IN NE FINLAND: STOCHASTIC OR PREDICTABLE?

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Objective: The question whether parasite communities of freshwater fish are stochastic or predictable has been under debate for a long time. In Britain parasite communities in eel and trout have been stated to be stochastic. In Finland, however, some lakes have been shown to have similar parasite fauna despite the different trophic or pollution status of the lakes. These lakes form a chain and have been a part of post-glacial Lake Ancylus ca. 8,000 years ago. Because of this contradiction, the present study was aimed to determine whether parasite communities in whitefish were predictable in another Finnish lake system having a far longer isolation history, because it did not originally belong to Lake Ancylus.

Materials and methods: Eight lakes were grouped according to their location in three tributaries of the river Iijoki. Whitefish were examined for both endo- and ectoparasites, excluding protozoans. Differences in parasite communities between the lakes and the lake groups were analyzed using two similarity indices and Canonical Correspondence Analysis (CCA).

Results & Conclusion: Fourteen parasite species were found, a large proportion of which were specific to salmonids, leading to a high similarity in species composition. However, both similarity indices and CCA revealed significant differences in parasite communities between lakes and lake groups due to the great differences in the abun-

dances of a few key parasite species. These differences were mainly caused by ecological lake characteristics. Although these differences were observed, the fauna was predictable.

PATTERNS OF HELMINTH DISTRIBUTION WITHIN A SINGLE RIVER CATCHMENT

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Objective: A study of spatial patterns in the distribution of parasites of *Salmo trutta* in Norway led to the hypotheses that parasites may be grouped as common or rare, and that similarity in helminth community structure should decrease with distance between patches and increase with increasing connectivity among patches. These hypotheses were tested with respect to helminth parasites of *S. trutta* and *Anguilla anguilla* in a single catchment.

Materials and methods: In autumn 1997 samples of trout and eels were taken from two sites in the lower reaches of the River Exe, England, and from four of its main tributaries. Fish were examined by standard methods for helminth parasites and data analysed at component community level.

Results: Helminth communities of trout and eels varied in richness and, whilst there were qualitative similarities between the sites, there were also differences. Each river was dominated by a different helminth species and a species common in one river could be rare in another. For example: eels in all four tributaries harboured *Anguicolla crassus*, yet this species was absent from the R. Exe itself; and each tributary was characterised by a different species of acanthocephalan, one by invading *Pomphorhynchus laevis*.

Conclusion: Helminths could be characterised as common or rare in respect of each river, but a species common in one could be rare in another. Whilst there was some support for the hypotheses being tested, the major conclusion was that other factors were more important determinants of community differences, and landscape factors could overwhelm potential determinants of similarity.

SCHISTOSOMES IN ICELAND

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Objective: In Europe, causative agents of cercarial dermatitis are predominantly larval stages of schistosomes of the genus *Trichobilharzia*. The life-cycle includes various snails of the family Lymnaeidae and birds as intermediate and definitive hosts, respectively. During late summer in 1995 to 1997, repeated outbreaks of maculopapular skin eruptions were noted on legs of children after wading in the pond of the Family Park in Laugardalur, Reykjavík, Iceland. Clinical symptoms resembled those seen in cercarial dermatitis. The aim of the present study is to report human cercarial dermatitis in Iceland, describe the taxonomic status of the schistosome larvae and present recent investigation carried out on schistosome larvae and adult stages in Icelandic snails and birds, respectively.

Materials and methods: In 1997 and 1998 a total of 3,129 *Lymnaea peregra* from Laugardalur were examined for the presence of schistosome larvae, and in 1997, 6 *L. peregra* and 69 *Physa* cf. *integra* were examined from the adjacent Lake Tjörnin. In 1998, five whooper swans (*Cygnus cygnus*) were examined for adult schistosomes.

Results: A previously undescribed schistosome cercariae of the genus *Trichobilharzia* were found in both localities in 27 (0.9%) of the *L. peregra* snails examined. The penetration activity of the cercariae, confirmed by the observation of red maculae on legs, was observed on three bird species: *Anas platyrhynchos* f. dom., *Serinus canaria* and *Anser anser*; however, no parasites were detected in the inner organs of these birds. Infection of mice (*Mus musculus*) resulted in detection of schistosomula in the skin 30 minutes, 1h. and 4 h p.i. Later, post mortem examinations of inner organs were negative. The parasitological investigation of naturally infected whooper swans resulted in the detection of several *Trichobilharzia* sp. adults in three (60%) of the birds examined. Both sexes were found in the vein branches of the large intestine. Histological examination also showed the presence of both sexes in the venae, and eggs with fully developed miracidium which were surrounded by granulomatous host reaction.

Conclusion: Other records of adult bird schistosomes in Iceland include immature *Bilharziella polonica*, found in 1995 in a juvenile common eider (*Somateria mollissima*) and blood flukes belonging to the genus *Ornithobilharzia* sp. in three lesser black-backed gulls (*Larus fuscus*) (Eydal *et al.*, 1998). Thus, preliminary investigation indicate, that at least three different bird schistosome species of at least three different genera are known, so far, to occur in Iceland. A representative of one of these genera,

the previously undescribed *Trichobilharzia* cercaria (Kolarova *et al.*, 1999) is now, for the first time in Iceland, responsible for human cercarial dermatitis in the country. Further search for schistosome larvae and adults in Icelandic snail and bird species respectively, concurrent with taxonomic studies, are strongly recommended.

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THE EFFECT OF EYE FLUKES ON ATLANTIC SALMON PARR AFTER THE REGULATION OF THE ALTA RIVER

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Objective: Since 1987, the Alta river has been regulated for the purpose of hydro electric power production. In order to protect the Atlantic salmon stock, care is taken when regulating the river to not allow the water flow to deviate more than 10% from the natural water flow. Still, the annual salmon catch and the natural production of Atlantic salmon smolts in upper part of the river (below the dam) have declined since 1987. Preliminary studies have disclosed high intensities of eye flukes in the Atlantic salmon parr. In African regulated rivers, ecological changes with subsequent increase of the snail intermediate host populations have been responsible for increased prevalence and intensity of human flukes of the genus *Schistosoma*. Because a similar increase in the snail population is suspected to have occurred in the Alta river after the regulation, the present study was undertaken to investigate whether increased intensities of eye flukes might have contributed to the decline in the Atlantic salmon stock.

Materials and methods: Atlantic salmon parr were collected by electro fishing. Metacercaria of *Tylodelphys clavata* and *Diplostomum* spp. in the eyes (eye flukes) and protozoan ectoparasites were registered.

Results: The intensity of eye flukes in Atlantic salmon parr of the Alta river was higher than what was found in neighbouring rivers and than is reported from Atlantic salmon parr in rivers in Europe and North America. The results also suggest that the high intensities of eye flukes might have a negative effect on the population of larger parr (>2+) in the upper part of the river.

PARASITOLOGICAL ASPECTS OF GRAZING HEIFER CALVES AND SOWS ON THE SAME PASTURE

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Objective: To study the impact on gastrointestinal parasite transmission of grazing systems with heifer calves and pregnant sows sharing pasture in two different ways. Data related to bovine parasite transmission are presented.

Materials and methods: During their first grazing season, three groups of eight heifer calves each were grazing either together with sows (mixed grazing), alternating with sows (alternate grazing) or separately (control). The experiment was repeated over two consecutive grazing seasons. Current parameters, including body weights, herbage larval counts, faecal egg counts, and serum pepsinogen levels, were measured at two-week intervals throughout each grazing season.

Results and conclusions: Alternate grazing and mixed grazing, in particular, resulted in reduced bovine trichostrongylid transmission on the pasture. This was accompanied by significantly increased growth rates when compared with the control group. These effects were most pronounced during the second year of the experiment.

INTERACTION OF A *Cryptosporidium parvum* SPOROZOITE APICAL ANTIGEN, CP47, WITH EPITHELIAL CELLS

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Objective: Previous studies have shown manganese (Mn^{++}) to inhibit binding of *C. parvum* sporozoite membrane proteins to intact, fixed human ileoadenocarcinoma (HCT-8) cells in a dose-dependent manner. Sporozoite penetration of live HCT-8 cells is also affected similarly. Other cations such as Ca^{++} , Mg^{++} , and Zn^{++} have no effect, nor are they able to competitively reverse the effects of Mn^{++} . Studies herein were designed to examine whether a receptor/ligand interaction existed between *C. parvum* and the host cell and whether this interaction was Mn^{++} -sensitive.

Materials and methods: Sporozoite/HCT-8 cell binding assays, Mabs, FPLC, and affinity column chromatography were used to determine the type of interactions between the parasite and host cell and the sizes of the molecules involved. Immunoelec-

tron microscopy/colloidal gold labelling was used to visually identify the location of ligand within the parasite.

Results: Mn^{++} sensitivity was traced to the interaction of two proteins; a 47 kDa sporozoite ligand (CP47) and a 57 kDa host cell surface receptor (p57). Mn^{++} sensitivity was found to reside at the level of the receptor. Gut membrane preparations from ileal and duodenal scrapings of suckling mice and neonatal goats revealed the ileum, but not duodenum, to have affinity for CP47; neither region bound CP47 to any degree when scrapings were derived from adult mice. Immunoelectron microscopy revealed CP47 to occur at the apical end of *C. parvum* sporozoites, associated with the surface membranes, pellicle, and subpellicular microtubules. The molecule was not found within rhoptries, rhoptry ducts, micronemes, or other types of vesicles such as dense granules.

Conclusion: CP47 appears to represent a key sporozoite surface molecule critical for host cell invasion. Results imply that p57 may be a developmentally regulated antigen in most animals and the reason why infections can be so severe in neonates. Its apparent higher concentration along the ileum may explain why this portion of the intestine is the preferred site of infection.

Taenia solium IN THE NORTHERN HIGHLANDS OF TANZANIA

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Objective: Cysticercosis, caused by the zoonotic pork tapeworm *Taenia solium*, is emerging as an increasingly important agricultural and potential public health problem in the northern highlands of Tanzania. Thus far, no epidemiological studies have been conducted to investigate the situation. A research programme, involving veterinary parasitologists and veterinary public health specialists was recently initiated to examine the epidemiology of porcine cysticercosis in the northern highlands district of Mbulu, an area thought to be highly endemic.

Materials and methods: A randomized survey of pigs based on lingual examination for cysts was conducted in randomly selected communities. A questionnaire survey of pig owners was also conducted to collect information on awareness of the parasite, pig management, pork consumption and sanitation. The data were analyzed using Epi-Info version 6.

Results: Lingual inspection of pigs indicated a district-wide prevalence of 17.4% (n=770), with a range of 3.2 - 46.7% among individual villages. Questionnaires indicated poor pig management and hygiene practices among villagers. Many of the vil-

lagers questioned (58.2%) were aware of persons in their village exhibiting epileptic-like seizures - one of the distinctive symptoms of human cysticercosis. Also, 73.1% of the villagers knew of persons with tapeworms though none of them (0%) associated human taeniosis with porcine cysticercosis.

Conclusion: Cysticercosis is highly prevalent in the domestic pig population in Mbulu District. Questionnaire responses suggest that taeniosis/cysticercosis may also be prevalent in the District's human population. More comprehensive epidemiological studies are needed to collect baseline data on the prevalence of porcine cysticercosis and human taeniosis/cysticercosis in Mbulu District and other pork raising/consuming communities in Tanzania.

Acknowledgement: This research initiative is financially supported by DANIDA under the ENRECA Livestock Helminth Research Project coordinated by RVAU, Denmark.

Schistosoma mansoni CIRCULATING ANODIC ANTIGEN (CAA) RELATED TO PRAZIQUANTEL TREATMENT OF ENDEMIC POPULATIONS ON KOME ISLAND, LAKE VICTORIA, MWANZA REGION, TANZANIA. EFFECT OF VARIOUS INTERVENTION STRATEGIES.

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Objective: In order to assess the impact of different intervention activities and to define an optimal treatment strategy to control morbidity of schistosomiasis mansoni, various treatment schedules were applied in different villages on Kome island, Mwanza region, Tanzania. The following parameters were varied: a) Target population (whole village population vs. school children), b) Interval between treatments (6, 12 or 24 months) and c) Dose of Praziquantel (40 mg/kg vs. 20 mg/kg). In this presentation the effect on faecal egg excretion and serum levels of CAA, an intestinal antigen discharged from adult, live schistosome worms, will be reported.

Materials and methods: Blood and faeces samples were obtained at 6, 12, 18 and 24 months. Sera were analysed for CAA by means of ELISA. Egg excretion in faeces was determined by the quantitative Kato-Katz procedure. The study also included *in vivo* physical examination as well as ultrasonographic examination of the liver and spleen.

Results: Mean levels of CAA generally paralleled the infection intensities expressed as mean Kato Katz values, even if some disagreement between the two tests was noted. Although the effect of the intervention on prevalence was rather modest, there was a significant effect on infection intensity in all villages.

Conclusions: Determination of CAA was shown to be a valuable adjunct to conventional parasitological technique (Kato-Katz) for evaluation of infection intensity as well as for evaluation of the effect of the various intervention activities. Unexpected fluctuations in CAA levels could be related to immigration of previously untreated individuals, low dose of Praziquantel and/or insufficient compliance. The effect on morbidity as demonstrated by ultrasonography of the liver is still subject to analysis.

MALE BEHAVIOUR AND MALE HORMONES IN *Ascaridia galli*-INFECTED HENS

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Objective: Infections with *Ascaridia galli* are common in hens in deep-litter, free-range and organic production systems and observations have indicated that the infections may increase cannibalism and perhaps male behaviour.

Materials and methods: Two groups of 15 young Lohman Brown hens were weekly inoculated orally with 500 *A. galli* eggs per animal, while 2 comparable groups were kept as uninfected controls. During the course of infection behavioural observations were carried out by focal animal observations of 6 hens per group and male behaviour was recorded for all individuals. The intestinal worm burdens were recorded at slaughter 12 weeks after the start of the infections and individual sera were analysed for testosterone.

Results: The inoculated hens harboured 9.8 ± 8.4 worms ($0 \pm \text{SD}$) at slaughter, while no worms were found in the controls. Cannibalism was not observed, however, the *A. galli*-infected hens had increased frequency of male behaviour, including mating and aggression, and analyses revealed a significantly increased level of testosterone (0.117 ± 0.120 ng/ml) in the inoculated hens compared to the controls (0.036 ± 0.037 ng/ml) ($p=0.0014$).

Conclusion: The study revealed that *A. galli* cause an increase in male hormones and male behaviour of egg laying hens and a possible influence on egg production should be considered.

ENVIRONMENTAL SIDE-EFFECTS OF SYNTHETIC PYRETHROIDS GIVEN AS POUR-ONS TO CALVES

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Objective: To investigate lethal and sublethal effects of synthetic pyrethroids against the coprophagous fly *Orthellia cornicina* (Diptera: Muscidae).

Materials and methods: Larvae and adult flies were exposed to dung from untreated calves or to dung from calves treated at recommended doses with deltamethrin, flumetrin, cyfluthrin, or α -cypermethrin. 1) First stage larvae were seeded on dung collected 1, 3, 7, 14, or 28 days after treatment (d.a.t.) and the pre-adult mortality was recorded. 2) Adult mortality, excretion, and ovarian development were recorded over 5 days in nulliparous adult flies that were allowed access to dung collected 3 d.a.t. Excretion was estimated from faecal spots counted on a piece of filter paper (changed daily, together with dung). Follicular development in surviving flies was scored in 3 stages.

Results: 1) Pre-adult mortality was significantly increased in dung collected up to 7 d.a.t. with deltamethrin, α -cypermethrin, or cyfluthrin, but unaffected by exposure to flumetrin dung. 2) Only α -cypermethrin dung induced a significant increase in adult mortality. Follicular development was significantly delayed by α -cypermethrin and deltamethrin dung, but not by cyfluthrin or flumetrin dung. Excretion was significantly reduced by exposure to α -cypermethrin, deltamethrin, and flumetrin dung, but increased by exposure to cyfluthrin dung.

Conclusion: Dung from calves dosed with synthetic pyrethroids negatively influences the survival and reproduction of *O. cornicina*. The synthetic pyrethroids increase in their effects by the order: flumetrin < cyfluthrin < deltamethrin < α -cypermethrin. Use of synthetic pyrethroids may affect other elements of the decomposer fauna and the decomposition of dung organic matter.

INFLAMMATORY AND HUMORAL RESPONSES TO EXPERIMENTAL TRICHINELLOSIS IN THE RACCOON DOG

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Objective: In Finland, raccoon dogs (*Nyctereutes procyonoides*) are commonly infected with *Trichinella* spp. It is supposed that the species is a key player in *Trichinella* epidemiology in Finland, though nothing is known about the species' responses to trichinellosis.

Materials and methods: Three experimental groups of six raccoon dogs each were formed by placing one of the three male raccoon dogs (4 months old) from same litter in each group. One group was infected with pig origin *T. spiralis*, a second group with raccoon dog-origin *T. nativa* and the third served as a control group; infection dose was 1,000 larvae/kg of body weight. Serum samples were collected up to 12 weeks post infection. Every third week, biopsies from *M. triceps brachii* were sampled. Immunoblotting was done using both *T. spiralis* and *T. nativa* origin antigens. Parasite burden from muscle samples was analysed by trichinoscopy.

Results: At three weeks post infection, both free-living and encapsulated larvae were found in muscle samples; later larvae were encapsulated. The average parasite burden over the experiment was 376 larvae/g with *T. nativa* and 229 larvae/g with *T. spiralis*. Cellular inflammatory responses with eosinophilic granulocytes were present, but mild at three weeks, and increased thereafter up to twelve weeks, especially with *T. nativa*. Specific IgG responses with homologous antigen were evident as early as two weeks after and cross reactive antigens were found later with weaker reactivity.

Conclusion: Nurse cell formation completes between three and six weeks post infection with both *Trichinella* species. Stronger serological reactivity to homologous antigen indicates some species-specific immunological responses. Tissue reactions also differed between the parasite species.

NEMATODE PARASITE INFECTIONS IN ORGANICALLY PRODUCED YOUNG CATTLE GRAZING ON NATURAL PASTURES

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Objective: Organic cattle production is becoming common in Sweden. A recent survey showed that many such producers considered parasites to be of little significance and had no planned control strategies. However, a large number used various management procedures aimed at parasite control in young stock. This two-year study was designed to investigate these issues.

Materials and methods: Each year, 10 young cattle were randomly allocated to:

- Treatment 1. Untreated — Set—stocked
- Treatment 2. Untreated — Mid-summer move
- Treatment 3. Ivermectin bolus — Set-stocked.

Pastures in year 1 were virtually “worm free”, so animals received a “priming” dose of approximately 10,000 infective larvae prior to turn-out (mid May). At turn-out in the second year, animals in treatment 2 grazed on pastures spelled since mid-summer the previous year, but grazed by young cattle from turn-out until this time. The only difference between the two years was the presence of over-wintered larval infection.

Results: The first year showed that parasite infections were so low that there was no difference in weight gain between the three treatments. In contrast in the second year, animals in the treatments 1 and 2 were 30 kg lighter at the end of the season, compared with treatment 3.

Conclusion: This study showed that over-wintered larval survival from early contamination the previous year has a profound effect on the productivity of young cattle. Simply preventing pastures from being grazed by young cattle from mid-summer onwards will not provide satisfactory grazing for such animals the following year.

SUBMITTED PAPERS - POSTER PRESENTATIONS

ECOLOGICAL AND GEOGRAPHICAL ANALYSIS OF *Sebastes mentella*
PARASITIC FAUNA IN THE NORTH ATLANTIC

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Objective: Ecological and geographical analysis of *Sebastes mentella* parasite fauna variability in the North Atlantic.

Materials and methods: Ecological and geographical analysis of *S. mentella* parasite fauna variability was performed on the basis of set theory according to the method proposed by the Russian authors V. Andreev & Yu. Reshetnikov, 1978. The results of our examination of 1,349 fish specimens according to the method of complete parasitological dissection (Dogiel, 1933; Bykhovskaya-Pavlovskaya, 1985), as well as literature data (Sindermann, 1961; Williams, 1963; Bourgeois & Ni, 1983; Khan *et al.*, 1986; Moran *et al.*, 1996, and many others), were used to list the parasites of *S. mentella* from 16 North Atlantic areas ranging from the Barents Sea to the Canadian coast. Individual and group specificity of *S. mentella* parasite fauna composition in the mentioned areas were established and a dendrogram was plotted illustrating the similarity degree of *S. mentella* parasite fauna composition in the areas investigated.

Results: According to the similarity degree of the parasite fauna composition of *S. mentella* in the investigated part of its distribution range, five areas were identified. The first one unites the adjacent areas of the Norwegian and the Barents Seas. The second area comprises Icelandic and Greenlandic waters, including the pelagial of the Irminger Sea and the adjacent Labrador Sea. The third area is made up by the continental slope and shelf of Canada, from South Labrador to the Flemish Cap Bank, and the Gulf of St. Lawrence. The Faroe-Iceland Ridge (Rosengarten Bank) and the slopes of the Faroe Islands are the fourth and the fifth areas, respectively.

Conclusion: Ecological and geographical analysis of *S. mentella* parasite fauna allowed identification of five areas in the investigated part of the distribution range, which differ by the composition of *S. mentella* parasite fauna. Our further investigations will aim at definition of the status of intraspecific redfish groupings in those areas.

ACANTHOCEPHALAN INFECTIONS OF DUCKS IN DIFFERENT FEEDING AREAS IN CHUKOTKA

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Objective: To present and explain different *Filicollis anatis* and *Filicollis trophimenkoi* infections among four wild duck species examined from different lakes in North-West Chukotka (69.5 n.l.).

Materials and methods: In August 1987, acanthocephalans were obtained from 27 pintails (*Anas acuta*), 15 long-tailed ducks (*Clangula hyemalis*), 4 scaups (*Aythya marila*) and 17 eiders (*Somateria fishery*) collected at six isolated, different sized lakes on the Chaun lowland, North-West Chukotka (Paulauskas & Butkauskas, 1993). Only young birds were examined in order to evaluate the local transmission of the acanthocephalans.

Results: Four duck species were examined from one of the lakes. Infection prevalences of *F. anatis* were as follows: *A. acuta* - 76.2%, *C. hyemalis* - 16.6%, *A. marila* - 100% and *S. fishery* - 100%. Also, *F. trophimenkoi* was found in this area, but only *S. fishery* and *A. marila* were infected with prevalences of 50% and 25%, respectively. The highest intensity of *F. anatis* infection was found in *A. acuta*, 30 worms/bird. Other infected individuals were infected with 2-8 *F. anatis*/bird. In the eider, significant intensity differences, varying from zero to 240 acanthocephalans/bird, were observed in the birds studied from the six lakes. In eiders from one of these lakes, the third acanthocephalan species, *Polymorphus magnus*, was also confirmed.

Conclusion: Acanthocephalan infections of the four duck species examined obviously depended on the feeding possibilities in the study areas. Furthermore, the intensity of infection clearly depended upon the water reservoir size: the lower size - the higher intensity of infection. *F. anatis* was a common helminth in all the four duck species investigated. Our results confirmed the higher specificity of *F. trophimenkoi* than *F. anatis*, as also proposed by Atrashkevich, 1982.

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DOES DUNG BEETLE ACTIVITY INTERACT WITH THE DEVELOPMENT OF PARASITIC GASTROINTESTINAL HELMINTH LARVAE IN CATTLE DUNG?

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Objective: With respect to reduction in use of antiparasitic drugs, any interaction between different dung-breeding organisms becomes of interest. The objective of the present study was to determine whether dung beetle activity affected the development of parasite larvae in cattle dung.

Materials and methods: Faeces containing eggs of parasites (predominately *Cooperia* spp.) were pooled according to high (500 epg) and low (50 epg) egg count. Ten, 500g dung pats were formed for each epg and to half of these, dung beetles (20 *Aphodius rufipes*, 20 *A. scybalarius* syn. *rufus*) were added. Pats were incubated for 12 days at 21°C and approx. 85%RH. Then pats were divided where one half was baermannized for infective larvae recovery and the other half mixed with vermiculite and cultured for a further 12 days.

Results: A greater number of infective larvae were recovered from the dung subjected to beetle activity, compared with control dung ($P < 0.001$). However, following a further 12 days culturing, the numbers of larvae recovered from beetle-affected dung tended to remain the same, whereas there was a significantly greater number of larvae derived from the control dung ($P < 0.001$).

Conclusion: These results indicate that dung beetle activity in fresh faeces can optimise conditions for hatching of nematode eggs and development through to the infective larval stage. However, the results also suggest that this comes at a cost to the overall fate of the nematodes. This may be due to direct (mortality following beetle ingestion) or indirect (desiccation) effects on the nematode free-living stages in beetle-affected dung. In the control dung, development of eggs to infective larvae can be delayed due to relatively poorer development conditions, but ultimately can yield much greater numbers of infective larvae.

THE COCCIDIA OF INSECTIVORES; TREE SHREWS AND PRIMATES

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Objective: The coccidia (Apicomplexa: Eimeriidae) comprise a large group of obligate intracellular parasites that are reasonably host-specific; they are among the

most prevalent and specious of all parasite groups. This review provides the first taxonomic summation of the coccidia of these 3 mammalian lineages.

Methods: All species of coccidia that infect the orders Insectivora, Scandentia and Primates are evaluated.

Results: Insectivora (7 families, 66 genera, 428 species): 83 valid coccidia (6 *Cyclospora*, 23 *Isospora*, and 54 *Eimeria* spp.) are described from 36 (8%) species, 19 (29%) genera, 4 (57%) families. Three of the 4 families examined have only *Eimeria* (34 spp.) or *Isospora* (9 spp.) spp., whereas the Talpidae have 20 *Eimeria*, 14 *Isospora* and 6 *Cyclospora* spp.

Scandentia (1 family, 5 genera, 19 spp.): 4 *Eimeria* spp. are known from 2 (10.5%) species, 2 (40%) genera.

Primates (13 families, 60 genera, 233 spp.): 15 coccidia (1 *Cyclospora*, 7 *Eimeria* and 7 *Isospora* spp.) are described from 17 (7%) species, 15 (25%) genera, 8 (61.5%) families; of these, prosimians have 7 *Eimeria* and 1 *Isospora* spp., but anthropoids have 6 *Isospora* and 1 *Cyclospora* spp.

Conclusion: Given the intracellular nature of the coccidia, which suggests a long and finely-tuned association with their chosen host, there may be useful patterns of association that can provide clues to evolutionary relationships among selected host groups. However, there is still much work to be done since only 55 of 680 (4.4%) species in these 3 mammalian orders have been examined for coccidia.

METACERCARIAE (DIGENEA) IN ATLANTIC COD, HADDOCK, WHITING AND SAITHE AROUND ICELAND

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Objective: In 1994 we found *Proisorhynchoides* (syn. *Bucephaloides*) *gracilescens* metacercariae for the first time in cod and haddock (second intermediate hosts) which are common species around Iceland. Follow-up studies revealed the presence of the adult stage in the final host the anglerfish (*Lophius piscatorius*), but the mollusc serving as the first intermediate host has not yet been found in Icelandic waters. The anglerfish is common off the Southwest, South and Southeast coast of Iceland, but very rare off the North and East coast. The objective was to study infections of *P. gracilescens* metacercariae in four species of gadoid fish around Iceland.

Materials and methods: Sixty cod (*Gadus morhua*), 210 haddock (*Melanogrammus aeglefinus*), 33 whiting (*Merlangius merlangus*) and 10 saithe (*Pollachius virens*) caught in 1996 and 1998 at different localities around Iceland were examined. *P. gracilescens* metacercariae in the brain cavity, in nerves of selected fins and in nerves in the caudal part were counted.

Results: *P. gracilescens* metacercariae were very common in all four gadoid fish species examined. At almost every locality off the South, Southwest and Southeast

coast 100% prevalence was observed. Only in fish off the North and Northeast coast were no metacercariae detected (0% prevalence). Intensity varied between localities. At several localities the mean combined number of *P. gracilescens* metacercariae ranged from about 200 - 2,300 per fish. Numbers of metacercariae varied considerably between organs and a common pattern of distribution was observed in all fish species, the highest numbers being in the caudal part and in the caudal fin. Massive infections were observed in many fish (especially haddock) with up to 4,000 metacercariae in nerves of the caudal fin and 2,100 in the caudal part in certain individuals. Considerably lower numbers were in the brain cavity and in the anterior dorsal fin and very low numbers in the posterior anal fin.

Conclusions: The high prevalence and intensity of *P. gracilescens* metacercariae in gadoids, and almost 100% prevalence of the adult digenean in the anglerfish as shown in earlier studies, demonstrates that the parasite completes its life-cycle in Icelandic waters. The geographical distribution of infected gadoids around Iceland coincides with the main distribution of the final host, the anglerfish. The results also support the view that there must be a first intermediate host other than the bivalve *Abra alba*, the only hitherto described first intermediate host, which does not occur in Icelandic waters.

PREVALENCE OF *Borrelia burgdorferi* sensu lato IN *Ixodes uriae* TICKS IN ICELAND AND POSSIBLE OCCURRENCE OF HUMAN INFECTIVE GENOSPECIES

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Objective: To investigate whether the *Borrelia* species found in Iceland could be assigned to genospecies (i.e. species consisting of a specific genotype) known to cause human infection.

Materials and methods: A total of 93 *Ixodes uriae* ticks collected in the summers of 1996 and 1998 on the Westman Islands and Andriks Island were tested for *Borrelia* infection. Two methods for detecting *Borrelia* infection were applied: IFA (immunofluorescence assay) and PCR (polymerase chain reaction). The PCR included four different PCR procedures, based on Marconi & Garron, 1992. PCRa detects DNA sequences belonging to the *Borrelia burgdorferi* sensu lato group, while the three remaining PCR procedures (PCRb) detect DNA sequences specific for genospecies pathogenic to humans. Only ticks found to be infected with *B. burgdorferi* by PCRa were included in PCRb, since a positive result in both tests is required to confirm that the *B. burgdorferi* found belongs to genospecies with known pathogenicity to humans.

Results: The prevalence of *Borrelia* infection by IFA was found to be approximately 50% both in 1996 (29/50) and 1998 (19/43). In 1998, 8 ticks out of 43 were found to be infected according to PCRa, all of them belonging to the 19 IFA-positive. None of the PCRa-positive ticks were positive by PCRb.

Conclusion: The results indicate that *Borrelia* bacteria (or other bacteria which are cross-reactive in the IFA test) are quite common in *I. uriae* in Iceland. On the other hand, only approximately 40% of the IFA positive ticks could be confirmed as carrying *B. burgdorferi* sensu lato by the PCR method. *B. burgdorferi* genospecies known to be pathogenic to humans were not detected by the method used in this study.

NITRIC OXIDE AS NEURONAL MESSENGER IN FLATWORMS

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Nitric oxide (NO) is a new biological messenger, generated by nitric oxide synthase (NOS), from L-arginine and O₂. L-citrulline is formed as a byproduct in 1:1 relation. NOS can be detected by NADPH-diaphorase histochemistry.

Positive reaction was found in the nervous system of adult *Hymenolepis diminuta*, *Diphyllbothrium dendriticum*, *Eubothrium crassum*, *Azygia lucii*, *Haplometra cylindracea* and in *Mesocestoides vogae* tetrathyridia. The neuromuscular relationship was studied by staining with TRITC-labelled phalloidin.

NADPH-d staining occurs close to muscle fibres in all above-mentioned worms. The production of L-[H³] citrulline was measured in *H. diminuta* and the effect of NO was studied by incubating *M. vogae* with either L-arginine or NOS inhibitors.

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THE GREEN SPRUCE APHID - A EUROPEAN RESEARCH PROGRAMME TO IMPROVE FOREST PROTECTION. EC FAIR3-CT96 1792

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The green spruce aphid (*Elatobium abietinum* Walker) is a ubiquitous and potentially debilitating pest of spruce forests in Europe. The aphid causes widespread defoliation, reductions in tree growth and affects timber production. This European Community research project is aimed at minimising the impact of the pest through the integration of simple and cost-effective measures, such as improving plant resistance and insect biocontrol, while understanding more about the genetic variation in the aphid and regional variation in climate and forestry which will affect the interaction between insect and plant in future.

REDUCED INNATE RESISTANCE TO *Gyrodactylus salaris* IN SALMONIDS AFTER HYDROCORTISONE ADMINISTRATION

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Previous studies have shown that brook trout (*Salvelinus fontinalis*), which normally develop acquired resistance to *Gyrodactylus salaris*, remain infected for much longer following immunosuppression with hydrocortisone (Harris, Soleng & Bakke 1997). This study focuses on the effects of immunosuppression in two other salmonids with differing patterns of resistance. Brown trout (*Salmo trutta*) normally fail to support *G. salaris* population growth, and are considered innately resistant (Jansen & Bakke, 1996). Arctic charr (*Salvelinus alpinus*) show individual heterogeneity in response, and some fish are innately resistant. This resistance may have a genetic component (Bakke, Jansen & Harris, 1996).

Infections on brown trout: Batches of anadromous trout (Lierelva strain, 1+), were implanted with either hydrocortisol in cocoa butter, or with cocoa butter alone. After a week of acclimation, fishes were infected with *G. salaris* (range 5-9 parasites), and after 24h returned to the experimental tanks. On cocoa-butter injected controls, infections declined throughout the experimental period to a mean parasite burden of less than 0.5 parasites per fish after 6 weeks. On hydrocortisone injected fishes, however, the parasite burden increased throughout, attaining a mean of 16 parasites per fish (range 2-72) after 6 weeks.

Infections on charr: Anadromous charr (Alta strain, 1+) with a high level of innate resistance were identified by following the course of an initial *G. salaris* infection. They were separated from the susceptibles, disinfected and allowed 28 weeks to recover from their initial infection. Before challenge with *G. salaris*, half of the fish were implanted with hydrocortisone; the other half (controls) received cocoa butter alone. After a week of acclimation, fishes were infected with *G. salaris* (range 30-40 parasites), and after 24h returned to the experimental tanks. After a short period of initial increase (up to 14 days), their infections declined. However, the rate of decline was much slower in the hydrocortisone-treated fishes than in the controls. After six weeks, the parasite burdens on the control fishes had declined to c. 4 parasites per fish, while those on the hydrocortisone treated fishes remained at c. 35 parasites per fish.

Conclusions: In all cases, in brown trout and in previously exposed innately resistant Arctic charr, the hydrocortisone treated fishes proved significantly more susceptible, and supported *G. salaris* growth more readily, than did the controls. These findings extend our previous observations on brook trout, and allow us to draw the following conclusions:

- Hydrocortisone increases the susceptibility and population growth rate of *G. salaris* in all previously resistant species of salmonids tested (brook trout, brown trout, Arctic charr).
- A single group of hydrocortisone-sensitive mechanisms with different degrees of expression, may control *G. salaris* population growth in all tested salmonids.
- The results suggest that *G. salaris* could infect and be spread by any salmonid if levels of stress experienced are sufficient to induce immunosuppression.

DIET AND SEALWORM INFECTIONS OF SHORT SPINED SEA SCORPION (*Myoxocephalus scorpius*) AND ATLANTIC CATFISH (*Anarhichas lupus*) IN ICELANDIC WATERS

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Short spined sea scorpion (*Myoxocephalus scorpius*) and Atlantic catfish (*Anarhichas lupus*), are demersal fish species in Icelandic waters. Short spined sea scorpion is found in shallow waters at 2-20 meters depth, but Atlantic catfish at 10-300 meters. This paper compares the diet and sealworm (*Pseudoterranova decipiens*) infections of these two fish species, and evaluates their importance as intermediate hosts for the sealworm.

A total of 213 short spined sea scorpion were sampled from various coastal areas of Iceland. The majority, however, originated from the West-North-West coastal area, as did all the 118 Atlantic catfish studied.

The far most dominant food of the short spined sea scorpion were spider crabs (*Hyas* spp.). In the stomach of catfish, however, remains of bivalves, crabs and conchs were found. Sealworm larvae were the dominating nematode, but the short spined sea scorpion was far more infected than the catfish, prevalence being >90% and 46% respectively and density being 8.2-34.3/100 g and about 0.095/100 g fish, respectively.

Invertebrates such as crabs dominate the diet of short spined sea scorpion and catfish. Short spined sea scorpion is, however, much more infected with larvae of sealworm than catfish, and therefore more important intermediate host for the sealworm. This may be because it consumes mysids, the most important invertebrate intermediate host for the sealworm in Canadian waters (Marcogliese, 1992), which did not occur in catfish diet. More likely, however, this could be the result of different habits. Short spined sea scorpion inhabits more shallow waters, and therefore is closer to the seal colonies, than Atlantic catfish.

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A SIMPLE MODEL OF SEASONAL AND GEOGRAPHIC CHANGES IN THE ABUNDANCE OF SEALWORM (*Pseudoterranova decipiens*) IN THE STOMACH OF GREY SEAL (*Halichoerus grypus*) IN ICELANDIC WATERS

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Studies on the abundance of the sealworm (*Pseudoterranova decipiens*) in grey seals (*Halichoerus grypus*) in Icelandic waters have indicated seasonal variations (Ólafsdóttir & Hauksson, 1997). Furthermore, dietary studies of the grey seal have shown seasonal fluctuations (Hauksson & Bogason, 1997). Short spined sea scorpion (*Myoxocephalus scorpius*), which is heavily infected with larvae of sealworm (Hauksson, 1999, see previous abstract) was only eaten in considerable quantity during the autumn. Change in the diet in the autumn causes increased ingestion of sealworm larvae, especially in grey seals from the West and Northwest coastal waters. Off the South-coast the abundance of short spined sea scorpion in the diet of grey seals was much lower, and therefore less intake of sealworms there.

A simple model, of an annual life cycle of the sealworm in Icelandic waters, is presented, which could explain the seasonal and geographic variation observed.

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OBSERVATIONS ON FLOTATION OF *Balantidium coli* CYSTS IN McMASTER COUNTING CHAMBERS

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Objective: There have been reports of investigations using saturated NaCl as a flotation media for detecting cysts of *Balantidium coli* in pig faeces. We have not found this method suitable and have therefore performed further experiments with ZnSO₄ as recommended in the literature for flotation of protozoan cysts.

Materials and methods: The number of cysts in faeces was determined by a standard sedimentation method of washing, sieving, centrifuging and counting of the sediment in Sedgewick-Rafter chambers (S-R count). Flotation solution was added to sediment or faeces 14:1 (v/w) and the counting was performed in a McMaster counting chamber (McM).

Results: Results are expressed as % of S-R count. Samples taken at timed intervals after adding the flotation solution showed a maximum flotation of 94% in the first count within 1 minute after adding ZnSO₄ (1·H₂O) 331 g/l, but after 5 minutes this was reduced to 7%. In saturated NaCl only a maximum of 3% cysts floated. If the first McM sample was left undisturbed under the microscope, the cysts remained attached to the coverglass of the counting chamber and thus extended the period for optimal cyst counting. It was possible to float faecal samples directly, but only a maximum of 42% of the cysts were found floating. There was a slight increase in the number of floating cysts during the first 5 minutes and thereafter the number remained constant for at least 10 minutes.

Conclusion: It is possible to float and count cysts of *B. coli* adequately in ZnSO₄ from washed preparations of faeces, but many cysts are lost when using fresh faeces.

CARP LACTOFLORA AS A NATURAL DEFENSIVE RESOURCE

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Objective: Normal microflora of a living organism and lactoflora as its constituent part are known to prevent pathogenic bacteria from penetrating into the organism and their propagation in it. The aim of the work was to study the antagonistic properties of microorganisms of the genus *Lactobacillus* from the intestinal wall of carp (*Cyprinus carpio* L.) with respect to the stems of pathogenic bacteria belonging to the genera *Pseudomonas* and *Aeromonas*.

Materials and methods: The antagonistic properties of 72 bacteria stems isolated from the intestinal wall of 65 carp were studied. As test cultures, six stems pathogenic to fish (*Ps. fluorescens* 6, *Ps. fluorescens* 83, *Ps. fluorescens* 90, *A. hydrophila* subsp. *anaerogenes* ATCC 15,468, *A. sobria* cip. 7,433, *A. hydrophila* subsp. *hydrophila* ATCC 7,966) were used. The antagonistic properties of the cultures were studied by the method of Lenzner, 1989. The antagonistic properties of the *Lactobacillus* stems were evaluated by the size of the growth inhibition zone: 6-15 mm - weak (1 point), 16-30 mm - middle (2 points), > 30 mm - strong (3 points).

Results: All the studied lactoflora stems of the carp intestinal walls have been found to have a pronounced antibacterial effect on the test cultures. Strong antagonistic properties regarding the test cultures of the genus *Pseudomonas* have been revealed in 59.7% and regarding those of genus *Aeromonas* in 58.3% of the *Lactobacillus* stems. Of all stems of the genus *Lactobacillus* studied, four stems have been found to possess a strong antibacterial effect regarding all test cultures. Of these stems, two have been identified as *L. casei* subsp. *casei*, one as *L. brevis*, and one as *L. plantarum*.

Conclusion: The results show that the lactoflora of the intestinal walls in fish, like in warm-blooded animals, having an antibacterial effect on pathogenic microflora, participates in the formation of the natural ecological barrier of the organism and its performance.

BIOLOGICAL CONTROL PRINCIPLES OF ANIMAL ECTOPARASITES

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Objective: As pesticide application may have a negative impact on the environment and pest resistance to poisonous chemicals can develop after repeated applications, biological principles of pest control may play a major role. In order to control

parasitic diseases in agricultural practice, the possibilities for use of several natural pesticides and juvenile preparations were used against animal arthropod pests.

Materials and methods: About 350 pigs, 150 dogs and 135 cats showing clinical signs of ectoparasitic diseases were used in our trials. To control animal ectoparasites, our investigations were made to determine the effect of 5 insect juvenoids and 9 plant extracts. The basic dosage of juvenoids contained 1–2% of the active product and tests with plant extracts were carried out with 5–10% solutions.

Results: Recent surveys have shown an occurrence of clinical signs of swine sarcoptosis (caused by *Sarcoptes scabiei* var. *swis*) up to 25% of pigs and about 20% of dogs and cats were infected with mange mites, ticks or fleas. According to our results, the most active compounds were the mixtures of juvenile preparation ephoksen with several local plant extracts showing antiparasitic effects (cow parsley, tancy, wormwood etc.). After 2 weeks practically no live parasites were found on the treated animals. The synergistic effect was observed in co-action of different active substances.

Conclusion: Application of biological control principles in parasitology might be used against arthropod ectoparasites, despite their parasitic species and independent of their mode of adaptation and host-parasite interactions.

HELMINTHS OF GOATS IN ICELAND

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Introduction: Goats were probably imported to Iceland during the settlement of the country in the 8th to 10th century and there is no information on later imports. The earliest information on the number of goats is from 1703 when they were 818. Since then, their numbers have fluctuated from less than 100 to almost 3,000; their number now being just over 400 on 57 farms. Goats have always been grazed with sheep, the most common livestock in Iceland.

Objective: This is the first parasitological study on goats in Iceland and its objective was to determine the helminth species in goats, their prevalence and intensity.

Materials and methods: During the slaughtering period in September-October 1998, the gastro-intestinal tracts and lungs from a total of 15 kids, 4-6 months of age, from 8 farms in West- and North Iceland, were collected and examined for adult helminths. The faeces were examined for lung-worm larvae.

Results: The following species were found: *Teladorsagia circumcincta* (incl. *T. trifurcata* and *T. davtiani*) (prevalence 100%, range of parasite numbers 700-5,250); *Trichostrongylus capricola* (40%, 0-2,509); *Nematodirus filicollis* (87%, 0-450); *N. spathiger* (60%, 0-500); *Chabertia ovina* (27%, 0-16); *Oesophagostomum venulosum* (7%, 0-2); *Trichuris ovis* (53%, 0-7); *Muellerius capillaris* (only searched for larvae in faeces) (46%, larvae pr. g. faeces 0-53) and *Moniezia expansa* (20%, total length of worms in each kid 0-625 cm).

Discussion: The helminth species, their prevalence and intensity is similar to what would be expected in Icelandic lambs of the same age and at the same time of the year. Helminth species which have been found in sheep in Iceland, but were not found in this study are *Trichostrongylus axei*, *T. vitrinus*, *Cooperia oncophora*, *Bunostomum trigonocephalum*, *Dictyocaulus filaria* and *Protostrongylus rufescens*. These species are less common which is probably the main reason why they were not observed in this limited study.

GYRODACTYLIDS ON SALMONIDS FROM DANISH STREAMS - A PRELIMINARY REPORT

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Objective: The first country-wide survey of *Gyrodactylus* species of wild salmonids in Denmark is presented. The survey has been carried out as part of an EU-Fair project concerning improved diagnosis of *Gyrodactylus* species of aquacultured salmonid species. The project includes participants from Scotland, Norway and Denmark.

Materials and methods: Fish sampling took place in spring 1998 and included brooks and major streams from 10 of the 14 counties in Denmark. A total of 61 localities from 45 major streams were sampled by electro-fishing. In the field the fish were anaesthetized using MS222. Dissected fins and the body proper were preserved separately in 80% ethanol and brought to the laboratory where the fish were examined and gyrodactylids enumerated and isolated from the fins. The specimens were then mounted on slides with ammonium-picrate-glycerine for examination of opisthaptor structures.

Results: A total of 599 fish were recovered. Five different salmonid species were caught and included the following species: Atlantic salmon, *Salmo salar* (n = 13), grayling, *Thymallus thymallus* (n = 8), houting, *Coregonus oxyrhynchus* (n = 3), rainbow trout, *Oncorhynchus mykiss* (n = 38) and brown trout, *Salmo trutta* (n = 537). *Gyrodactylus derjavini*, *Gyrodactylus salaris* and *Gyrodactylus truttae* have thus far been identified. Another, not yet identified, morphotype has also been recovered. Infection prevalence of 100 seems to be the general pattern in the majority of the streams. In only 2 of the 61 localities were no infected fish recovered. Intensities of gyrodactylids on brown trout vary considerably ranging from 1 to 1,421 parasites. Likewise, mean intensities and abundances of *Gyrodactylus* spp. on *S. trutta* varies between the investigated streams and regions. The prevalence of *Gyrodactylus* spp. on all salmonids was 82% with an abundance of 71.5 and a mean intensity of 86.7. Concerning each salmonid species, the figures are as follows (prevalence — abundance — mean intensity): Brown trout: 83.9% - 76.8 - 91.5 ; salmon: 30.8% - 0.9 — 3.0;

rainbow trout: 92.1% - 38.0 - 41.3; grayling: 50% - 1.0 — 2.0. None of the three houting were infected.

NON-SYSTEMIC RESPONSE IN RAINBOW TROUT AGAINST *Gyrodactylus derjavini*.

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Objective: The present study was undertaken to investigate epidermal changes of host skin during infection and development of host response. Furthermore, it was studied whether the acquired protective immunity could be transferred by passive immunisation of naive hosts.

Materials and methods: Two groups (68 and 61 fish respectively) of rainbow trout (mean body weight 1.4 g) were infected with *G. derjavini* and parasite population dynamics followed weekly for a total of 70 days. After 34 days, 30 additional uninfected fish were transferred to each aquaria for infection by cohabitation. Infections on these subgroups of host were likewise followed. Mucous cell densities were determined on Alcian Blue stained fins from all fish including uninfected controls. The carbohydrate composition of these cells was determined by using a lectin assay. For passive immunisation, 15 fish (4-5 cm) received native immune sera (with complement) whereas another 15 fish received heat-inactivated sera (without complement). Finally 15 fish received decomplexed non-immune sera (control). The fish were then infected and the infections monitored for a total of 32 days.

Results: Parasite populations increased on all naive host groups and peaked 4-5 weeks p.i. after which infection levels decreased. Introduction of naive fish into responding host populations resulted in heavy infections of naive fish whereas immune fish remained infected at a very low level. Mucous cell density differed significantly during infection: Mucous cell hyperplasia was seen in one group 34 days p.i., but at the termination of the study a significant depletion was evident. Small changes in lectin binding patterns were seen during infection. Thus, a stronger expression of mannosyl, acetyllactosamine and glucosyl residues was evident in the infected fish. No differences in abundance between the experiment groups were detected in the passive immunisation trial at any time.

Conclusion: The observed immune response against *G. derjavini* in the present study was not caused by a systemic humoral component that can be transferred to naive hosts and confer protection. The exhaustion of mucous cells and the observed changes in carbohydrate epitopes indicate a role of these components in the response against *G. derjavini*. It is thus suggested that future effort in this particular parasite-host relation is laid in the study of localised responses.

INCREASE OF ANTAGONISTIC PROPERTIES OF BACTERIA OF GENUS *Vibrio* IN THE INTESTINAL TRACT OF FISH IN LAKE DRUKSIAI – COOLING RESERVOIR OF THE IGNALINA NUCLEAR POWER PLANT

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Objective: To evaluate the effect of radiation and chemical pollution on the antibiotic and bacteriocinogenic properties of microflora of the digestive tract of roach – the predominant fish species in the lake.

Materials and methods: The microflora species composition has been determined applying the Bergey's, 1994 determinative, the antibiotic and bacteriogenetic activities have been determined by the methods of Yegorov, Lielpetere & Jakobson.

Results: Microflora of the digestive tract performs an important role in the formation of the immunologic status of the organism. Microflora and its activity in particular are the first targets of the negative environmental factors. From the digestive tract of roach, 85 strains of bacteria belonging to 7 genera have been isolated, with the pronounced predominance of representatives of the genus *Vibrio*. This fact deserves most serious attention, the more so as the *Vibrio* bacteria have been found to occupy also the walls of the intestine. We have also been worried by the fact that in the bowels of roach the antibiotic activity (except *Vibrio* bacteria) has been found to be greatly reduced. The antibiotic activity of bacteria, which characterizes the resistance of bacterial biocenosis to pathogenic and conditionally pathogenic microorganisms, is one of the most important indices of the immunological reactivity of the organism. Of all the antibiotically active bacteria, 75% comprised the representatives of the genus *Vibrio*, which displayed a very high activity and the ability to inhibit the development of all test cultures studied. An analysis of the bacteriocinogenic activity revealed a very high (up to 75%) bacteriogenicity in normal microflora of the digestive tract, whereas the bacteriogenicity of bacteria of the genus *Vibrio* varied from 24 to 42.8%.

Conclusion: Analysis of bacterial activity in the digestive tract of the roach has revealed a disturbance of the normal microflora; its resistance to pathogens is damaged, conditions of its development are unfavourable, and the dominating position is occupied by representatives of genus *Vibrio*, characterized by high antagonistic activity. The results obtained allow a prognosis of the pathologic condition of the macro-organism.

Entamoeba gingivalis IN HUMAN IMMUNODEFICIENCY VIRUS TYPE 1-INFECTED PATIENTS WITH PERIODONTAL DISEASE.

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Objective: Necrotic periodontal disease is a progressive painful oral lesion in human immunodeficiency virus type 1 (HIV-1)-infected patients, and the etiology is unknown. Earlier studies of HIV-1-infected patients have shown significant changes in the viral and fungal oral microflora. The aim of this study was to relate the occurrence of protozoa to clinical symptoms and immunosuppression.

Materials and methods: Oral symptoms were registered in 45 patients at different stages of the HIV-1 infection and in 15 HIV-seronegative healthy controls. Saliva and dental plaque were analyzed for the presence of protozoa using trichrome staining.

Results: *Entamoeba gingivalis* was the only protozoa found in the oral cavities of HIV-1-infected patients with periodontal disease. Its presence was not related to the degree of immunodeficiency, but to the HIV diagnosis.

Conclusion: This study describes for the first time the findings of *E. gingivalis* in the oral cavities of HIV-1-infected patients.

MUSCULAR PREDILECTION OF *Trichinella* SP. IN EXPERIMENTALLY INFECTED RACCOON DOGS

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Objective: To determine predilection sites of *Trichinella spiralis* and *Trichinella nativa* larvae in muscles of raccoon dogs (*Nyctereutes procyonoides*). In Finland the raccoon dog is a common carnivore, which is often found to be infected with *Trichinella*. Because it is also suspected to be one of the links between wildlife and domestic animals, it is important to know their muscular predilection.

Materials and methods: In September 1998, 6 males were infected (1,000 larvae/kg) with *T. nativa*, 6 with *T. spiralis* and 6 served as a control group. After 12 weeks 5-10 g samples were analysed by the Stomacher digestion method. The muscles selected for examination were tongue, eye, masticatory muscles, diaphragm, back, abdominal muscles, front leg and hind leg. We also sampled the heart and intestine.

Results: Larvae were found in all samples, except in the heart and intestine, of the infected animals. The average intensity of infection in positive tissues varied between

142.4-788.2 larvae per gram (lpg) (an overall mean 320.1 lpg, excluding heart and intestine) in *T. spiralis*, but 217.7-748.8 lpg (mean 374.9 lpg) in the *T. nativa* group. The maximum amount of larvae in *T. spiralis* animals was 1,091 lpg and in *T. nativa* 1,152 lpg, both found in the front leg; *M. flexor carpi ulnaris*.

Conclusions: There were no obvious interspecific differences in predilection sites. Both *T. spiralis* and *T. nativa* showed the highest larvae numbers in the front leg, tongue and eyes. Animals infected with *T. nativa* had higher larval burdens probably because the raccoon dog is a natural host of this parasite.

INFLUENCE OF ENVIRONMENTAL TEMPERATURE ON THE DISTRIBUTION OF *Trichinella* SPP. IN ESTONIA

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Objective: To identify the *Trichinella* species occurring in Estonia and study the influence of environmental temperature on the distribution of the species in the country.

Materials and methods: From 1992-1998 muscle samples from 797 sylvatic and 1,181 domestic animals were collected from all parts of Estonia. From each animal a muscle sample of 5-10 g was examined by the digestion method. Muscle samples from 62 of the 89 *Trichinella*-positive samples were sent to the *Trichinella* Reference Centre for *Trichinella* spp. identification. Larvae were identified by RAPD analysis. Latitude and longitude of the sites where the animals were hunted were recorded. Isotherm lines used for Estonia are from Wallen, 1970.

Results: Three *Trichinella* species were identified in Estonia: *T. spiralis*, *T. britovi* and *T. nativa*. Sylvatic animals were either infected with *T. nativa* or *T. britovi* or both species. Domestic animals were infected with *T. britovi* (domestic pig) and *T. nativa* (fur-bearing farm animals). On a farm on the Western island of Hiiumaa, fur-bearing animals as well as brown rats were infected with *T. spiralis*. Animals infected with *T. nativa* were hunted in Middle and East Estonia, to the east of the -5 °C isotherm line in January. Animals infected with *T. britovi* were hunted on the western islands and west part of mainland of Estonia; west of the -4 °C isotherm line in January. Near the -5 °C isotherm line in January hunted animals were infected either with *T. nativa* or *T. britovi* or both.

Conclusion: The results suggest that a relation exists between the geographical distribution of *T. nativa*, *T. britovi* and *T. spiralis* and the environmental temperature in Estonia. The -6 °C isotherm line in January is a thermic barrier for the dispersion of *T. britovi* in cold area. The -4 °C isotherm line in January is a thermic barrier for the

dispersion of *T. nativa* in temperate area. Near the -5 °C isotherm line in January both *T. nativa* and *T. britovi* can be found.

CLINICAL PARAMETERS OF EXPERIMENTAL TRICHINELLOSIS IN THE RACCOON DOG

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Objective: In contrast to other EU-countries sylvatic and domestic trichinellosis is quite prevalent in Finland. The raccoon dog (*Nyctereutes procyonoides*) is a common host of sylvatic trichinellosis. We performed an experimental study to characterise clinical trichinellosis in this mammal.

Materials and methods: Of 18 male raccoon dogs, six were infected with *Trichinella spiralis*, six with *Trichinella nativa* and six served as a control group. Animals were divided in the groups so that in every group there was one of three siblings. Infection dose was 1,000 muscle larvae / kg body weight. EDTA-blood samples were collected before the infection and up to 12 weeks afterwards. The measured parameters were: WBC (tot. and differential count), RBC, HGB, PCV, MCV, MCH, MCHC, platelets. Body temperature was measured when collecting blood and the animals were weighed weekly.

Results: Two weeks after the infection, the body weights of the infected animals were significantly decreased. RBC, PCV and HGB decreased more in the infected groups than in the control group between two and eleven weeks post infection. No differences were noticed in total WBC dynamics, but eosinophilia was evident at the second and the third week. Animals were not febrile.

Conclusion: *Trichinella* infection caused significant weight loss and normochromic, normocytic anaemia soon after the infection. Short term eosinophilia was present at the larval circulating phase. Fever is an often-described symptom of human trichinellosis, but it was absent in the raccoon dogs.

TREATMENT OF REINDEER WITH IVERMECTIN HAS LITTLE EFFECT ON DUNG INSECT FAUNA

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Ivermectin is used against parasites in reindeer (*Rangifer tarandus* L.) in Fennoscandia. Most of the injected drug is excreted unchanged in the faeces, and several studies have shown that ivermectin in cattle dung disrupts survival of dung inhabiting insects. In this study, we investigated possible negative effects of ivermectin on the reindeer dung fauna.

Four reindeer calves were injected subcutaneously with standard doses of ivermectin (0.2 mg/kg body weight) in early December. The daily produced faeces was collected until day 30 after treatment, and the concentration of ivermectin was determined by high pressure liquid chromatography (HPLC) using fluorescence detection.

The highest concentration measured was on day 4 after treatment. The concentration approached zero on day 30. Field studies showed that the most important coprophilous beetles (*Aphodius* spp.) and flies (*Scathophaga* spp.) were not detected in dung from the winter season whether it contained ivermectin or not, probably because of the dry and pelleted faeces. Treatment against reindeer parasites as usually done in autumn/early winter implies that summer dung, which is the dung that can be utilized by the important coprophilous species, will not contain ivermectin.

The results indicate that the current treatment practice in autumn/early winter does not constitute a serious threat to the coprophilous fauna and the decomposition process of reindeer dung.

OUTBREAK OF TRICHINELLOSIS IN A WILDBOAR FARM IN SOUTHERN FINLAND

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Objective: In Finland, farming of game animals is increasing. Outbreak of trichinellosis in a wildboar farm in southern Finland is described.

Materials and methods: Wildboars were kept in a fenced forest area of about 20,000 m². The size of the herd varied between 30 and 60, consisting of 10 -15 sows, one older boar and young pigs. The animals were fed with grain without any food of animal origin. At meat inspection examination for trichinellosis was made by trichinascopy. Positive diagnosis was confirmed by artificial digestion. The farm is situated in the endemic area of wildlife trichinellosis. Foxes are known to have trichinellosis prevalence of 50% (1997, n=74) and raccoon dogs 35% (1997, n=142) in southern Finland. Situated nearby the farm is an ex-dumping area that was closed in the beginning of 1997, but left without proper after-care. The rat population increased and spread to the surrounding areas. According to a local opinion, the rats carried the *Trichinella* infection past the wildboar fence. Rats have trichinellosis prevalence of 25% in dumping areas of South-West Finland. However, samples of the rats from this area were not available.

Results: The first *Trichinella*-positive animal was detected in May 1997 and the last one in December 1998. Totally, 9 animals were found to be infected out of 21 slaughtered. The infection intensity varied considerably, median was 2.50 (range: 0.07 - 680 LPG).

Conclusion: Out door farmed wildboar can be infected by *Trichinella* species, although they are not offered feed of animal origin.

OCCURRENCE OF *Pneumocystis carinii* INFECTION IN AN ETHIOPIAN REGION

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Objective: *Pneumocystis carinii*, a well-known opportunistic infection in immunosuppressed patients in northern Europe and northern America, is said to be a rare infection in Africa. A suggested reason is masking of *P. carinii* infection by more prevalent *Mycobacteriae* infections; the chest radiographs of the patients with a suspect PCP are often difficult to differentiate from *Mycobacteriae* infection cases. Considering these patients often respond to cotrimoxazole treatment, suggesting *P. carinii* as a possible agent, the need to determine the prevalence of *P. carinii* infection in this region of Ethiopia is clear.

Materials and methods: DNA from frozen sputa of HIV-infected patients was amplified by a nested PCR assay, based on the mitochondrial LSU ribosomal RNA gene portion of *P. c. hominis*, the human form of *P. carinii*.

Results: The preliminary results indicate a rather high prevalence of *P. c. hominis* in this Ethiopian region, which fits with the previous clinical signs of a subgroup of the patients.

Conclusion: The present study reveal that *P. carinii* may have a higher prevalence than expected in this region. However, since the analytical sensitivity of the nested PCR is high, the detected *P. carinii* infections may include subclinical cases of PCP. By contrast, since the samples were ordinary sputa and not bronchial lavage, the nested PCR positives may reflect a pre-debute infection in some of the patients. In HIV-infected patients, with a progressively decreasing immune defence, these findings may predict a risk of an active infection later on.

METAZOAN PARASITES IN LONG ROUGH DAB (*Hippoglossoides platessoides limandoides* BLOCH, 1787) IN ICELANDIC WATERS

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Objective: The aim of the study was to improve the knowledge of parasite infections in marine fishes in Icelandic waters.

Materials and methods: Metazoan parasites were collected from fish flesh of 562 and intestines, gills and organs of 358 long rough dabs. The fish were caught in February and March from five locations around Iceland.

Results: Thirty-one parasite species were observed in long rough dab in the study: *Derogenes varicus*, *Brachiphallus crenatus*, *Lecithaster gibbosus*, *Steringotrema ovacutum*, *Steringophorus furciger*, *Stenacron vetustum*, *Podocotyle atomon*, *Zoogonoides viviparus*, *Pseudozoogonoides subaquephorus*, *Aporocotyle simplex*, *Stephanostomum baccatum*, *Otodistomum* sp., *Grillotia* sp., *Diphyllobothrium* sp., tetraphyllidean larvae, *Anisakis simplex*, *Pseudoterranova decipiens*, *Pseudoterranova* sp., *Hysterothylacium aduncum*, *H. rigidum*, *Contracaecum osculatum*, *Contracaecum* sp., *Cucullanus heterochrous*, *Spinitectus oviflagellis*, *Ascarophis* sp., acuarioid larvae, *Echinorhynchus gadi*, *Corynosoma strumosum*, *C. semerme*, *Lernaecera branchialis* and *Acanthochondria cornuta*. The results indicated some geographic variations in parasite distribution in Icelandic waters.

Conclusion: The parasite fauna in long rough dab in Icelandic waters appears similar to what has been observed in other areas in the North Atlantic. Distribution of necessary hosts in the parasites' life cycles is probably the most important factor affecting the observed geographic differences in parasite abundance. Different diet and migration of long rough dab or its prey species may also influence spatial variations in parasite burden.

INCREASED PARASITE ABUNDANCE ASSOCIATED WITH REPRODUCTIVE MATURITY OF THE CLAM *Anodonta piscinalis*

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Objective: Several studies on vertebrates have demonstrated that reproductive activities may increase the parasite load, but this has not been shown in invertebrate hosts.

Materials and methods: We studied abundance of a potentially harmful gill parasite, the ergasilid copepod *Paraergasilus rylovi*, from the freshwater bivalve host *Anodonta piscinalis* in relation to reproductive maturity of the host in the field.

Results: Prevalence of this previously unstudied parasite varied from 90 to 100%, and the mean parasite abundance from 16.3 to 28.8 among 3 study populations. Abundance of *P. rylovi* increased with host size. In the maturing age groups (3-5 yr.) the length-adjusted mean parasite abundance among mature, reproducing female clams that brooded glochidia larvae was 2 times higher than in non-reproducing females, the observed pattern being consistent among the 3 study lakes.

Conclusion: The present result indicates that maturation, and reproduction, is associated with increased parasite abundance in *A. piscinalis*, an invertebrate host. Alternative, mutually non-exclusive explanations may be found for the result. For example, changes in clam behaviour of filtration activity accompanying maturation can increase host exposure to parasites, or reproduction may decrease energy available to host immunologic defence.

HELMINTH PARASITES OF HORSES, COWS AND PIGS IN LITHUANIA DURING 1994-1997

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Objective: Until 1994, the parasitological situation in Lithuania was not investigated systematically. The main purpose of the present investigation was to evaluate the helminth fauna of cows, horses and pigs in Lithuania and study possible annual prevalence changes.

Materials and methods: We checked herds of horses, cows and pigs in all the 10 districts of Lithuania once per year. Helminth eggs were identified after concentration from faeces by the floatation and the sedimentation methods. On average, samples from 11,317 cows, 949 horses and 22,610 pigs were examined every year.

Results: In the cows, the prevalence of Trichostrongylidae was 6.9%, *Fasciola* sp. 5.5%, *Moniezia* spp. 3.5%, *Dicrocoelium* sp. 3.4%, *Toxocara vitulorum* 2.8% and *Dictyocaulus* sp. 2.5%. In the horses, the prevalence of Strongylidae was 41.8%, *Oxyuris equi* 14.9% and *Parascaris equorum* 4.7%. During the four-year period, Strongylidae in the horses increased from 48.7% to 60%. In the pigs examined, the prevalence of *Oesophagostomum dentatum* was 16.6%, *Ascaris suum* 9.8%, *Trichuris suis* 2.4% and other Trichostrongylidae 2.1%. The prevalence of *A. suum* decreased from 12.1% in 1994 to 5.5% in 1997.

Conclusion: During the four-year study period the helminths of cows did not change significantly. The number of horses infected by Strongylidae increased. *A. suum* infections decreased as a result of an effective antihelmintic control. In order to decrease the number of infected animals it is necessary to improve our parasitological control and prophylactic measures.

SCREENING FOR SURFACE-EXPRESSED ANTIGENS OF *Plasmodium falciparum* USING FRACTIONATED, POOLED MALARIA IMMUNE HUMAN IgG

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Objective: Malaria is one of the most infectious diseases in the world with yearly 300-500 million infected people and 1.5-2.7 million deaths. Mortality is mainly thought to be caused by cerebral capillary occlusions as *Plasmodium falciparum*-infected RBC's (pRBC) bind to endothelium, and to uninfected RBC's, termed rosetting. To further study pRBC surface antigens potentially involved in cell adherence, we screened a cDNA library with fractionated, pooled human immune IgG.

Materials and methods: Immunofluorescence and rosette reversion assays were used to study pRBC surface reactivities with the pooled human IgG from Malawi. Primary antibody fractionation was performed by binding IgG to purified FCR3S1 trophozoite pRBC, followed by elution of surface-bound antibodies. Secondary purification followed by binding this eluate to a nitrocellulose strip of the major reactivity, as observed after Western blotting. This final IgG enriched fraction was used in cDNA-expression library screening.

Results: Immunofluorescence showed specific surface IgG binding to pRBC. In rosette reversion, rosettes were inhibited up to 70%. From previous experiments we know that rosetting FCR3S1 has a 35 kDa surface antigen, besides the high molecular weight PfEMP1. Antibody fractionation gave 6-7 major reactivities in immunoblots of SDS-soluble proteins, with the highest relative increase around 35-36 kDa. After screening the cDNA library and isolating positive clones, 12 were studied more closely in Northern and Southern blots. One clone bound to chromosome 2 in PFGE-blots and has stretches of sequence identity with a novel sequence in the data bank.

Conclusion: Using a fractionated, pooled human IgG to screen a *P. falciparum* cDNA library, we have identified novel sequences potentially surface-located on the pRBC and are now pursuing further analyses of these clones for future over-expression of recombinant protein for potential vaccine production.

PARASITES OF TWO HOUSE MOUSE (*Mus musculus*) POPULATIONS IN ICELAND

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Objective: To study the parasites of the house mouse *Mus musculus* in Iceland.

Materials and methods: Two different populations were examined. In autumn 1995 a total of 20 mice (6 ad, 14 subad) were captured from a population which recently had colonized some outbuildings at the Institute for Experimental Pathology, Keldur, Reykjavík. In autumn 1995 and 1997, 19 and 4 individuals respectively (7 ad, 16 subad) were captured from an isolated population which has lived for decades in outbuildings at Stórhofdi on the Westman Islands, off the south coast.

Results: Based on morphological features, the former population is probably the subspecies *M. m. domesticus* and the latter one *M. m. musculus*, but the taxonomic status will be cleared later using genetic methods. Altogether 20 parasitic species were identified; 9 endoparasites and 11 ectoparasites. The endoparasites were three protozoans (*Entamoeba muris*, *Eimeria falciformis** and *Eimeria* cf. *hindley*), a cestode (*Vampirolepis* sp.) and five nematodes (*Aspicularis tetraptera*, *Syphacia obvelata*, *Heligmosomoides polygyrus**, *Trichuris muris* and *Capillaria* sp.). The ectoparasites were an unknown mallophagan, the fleas *Ctenophthalmus agyrtes* and *Nosopsyllus fasciatus*, and 8 parasitic mites. The most common mites were *Myobia musculi**, *Myocoptes musculus**, *Psorergates simplex** and *Echinonyssus latiscutatis*. Interestingly, also *Myonyssus decumani* was found at Stórhofdi. Previously, this mite has once been reported from the Shetland Islands, but does not occur in Scandinavia. Seven additional mite species were found in the coat of mice from Stórhofdi.

Conclusion: Striking differences were observed in the parasite fauna of the two populations. Only five parasites (*) were reported in both study areas.

SEASONAL VARIATIONS IN THE INFECTION OF *Anisakis simplex* L3-LARVAE

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Objective: Andersen, 1993, in a study of cod (*Gadus morhua*) throughout the year, found a clear peak in the abundance of *Hysterothylacium aduncum* L3 and L4 during late winter and early spring. The objective of the present study was to reveal whether these results reflected a more general infection pattern, including other marine nematodes such as *A. simplex*.

Materials and methods: The material was collected on the west coast of Norway by gill netting monthly during 1990. Twenty individuals from the three fish species, saithe (*Pollachius virens*), cod and redfish (*Sebastes marinus*), were normally caught each month. The fillets were examined on a light table, and the viscera by use of a dissecting microscope. A total of 571 fish were examined.

Results: In all the three host species investigated, the occurrence of *A. simplex* L3 peaks in April. However, in saithe the April infection, though indicating a tendency, is not significantly different from the rest of the months (Tukey-test). According to the present results, saithe seems to have a relatively steady, but high, infection throughout the year. In cod the April peak is very distinct, while this peak in redfish represents the maximum point in a successive raise of abundance starting in February. The high abundance of *A. simplex* in April corresponds with the peak of the spring bloom at this part of the Norwegian coast and the northward migration of Minke Whale (*Balenoptera acutorostrata*).

PARASITES OF ARCTIC CHARR *Salvelinus alpinus* (L.) FROM LAKE VIGDARVATN IN WESTERN NORWAY. DO NUMBERS OF DIFFERENT PARASITE SPECIES TEND TO COVARY?

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Are the differences in parasite numbers observed in individual hosts mainly caused by variability in exposure or by heterogeneity in susceptibility? If exposure is the main factor, infection levels may be unrelated to host genetics and parasites should play a minor role as selective agents. Exposure to parasites will usually show large temporal and spatial variations. The numbers of parasites found in a host may therefore mainly be a result of random events, overriding any differences in host susceptibility.

One approach to this problem is to search for co-variation of different parasite species within a host population. Any accumulation of parasites with different trans-

mission routes in the same host individuals would suggest that susceptibility does play a role, despite the variability of transmission. To study this problem, a sample of Arctic charr *Salvelinus alpinus* (L.) was caught by gillnets during one night in Lake Vigdarvatn in Western Norway. Blood samples were taken and the fish were frozen individually for later examination for parasites.

At present, the following parasite species have been found and identified: the ciliate *Capriniana* sp., the myxosporidians *Myxobolus* sp. and *Henneguya* sp., the monogenean *Discocotyle sagittata*, the digeneans *Phyllodistomum umblae* and *Crepidostomum* sp., the cestodes *Eubothrium salvelini*, *Proteocephalus* sp. and *Diphyllbothrium* spp. and the copepode *Salmincola edwardsii*. Many of these species have different transmission routes, and we could therefore not expect a significant covariation by chance alone. The statistical analysis of the results is currently in progress, and will be presented and discussed.

PRIMITIVE TOURISM AND SCHISTOSOMOSIS IN TRAVELLERS TO AFRICA

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Objective: Groups of Swedish travellers visiting Kenya, Tanzania, Malawi, Mozambique, Zimbabwe, Botswana, Namibia and South Africa by bus in 1996 and 1997, were tested for anti-schistosome antibodies.

Materials and methods: Indirect immunofluorescence test for antibody against gut associated antigen (GAA) was used to detect "early" infection with *Schistosoma mansoni* and *S. hematobium*. Sera was obtained from 80 individuals.

Results: 42/80 (52%) had serum anti-GAA activity indicating recent schistosomiasis. Of the positive cases, 19% were missed in the handling of returning travellers with symptoms or suspected of being infected for other reasons.

Conclusion: The observed risk of becoming infected with schistosomes was found to be surprisingly high. Thus, there is a need for better information and routine testing for this infection.

IMMUNOHISTOLOGICAL IDENTIFICATION OF TISSUE WORMS USING CROSS-REACTING MONOCLONAL ANTI-PARAMYOSIN ANTIBODY

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Objective: As identification of tissue-dwelling or -migrating parasites in tissue biopsies may be problematic, we wanted to generate an immunohistological marker for metazoan parasites in tissue sections.

Materials and methods: Monoclonal antibodies against *Lumbricus terrestris* paramyosin were tested for cross-reactivity by indirect immunofluorescence with frozen and paraffin sections of *Trichinella*, *Fasciola*, *Schistosoma* and other tissue-invading worms. Cross-reacting antibodies were affinity-purified to obtain IgG for biotinylation. The biotinylated anti-PM antibodies were then used to detect worm components in tissue sections.

Results: The biotinylated anti-PM antibodies, together with avidin-FITC or avidin-peroxidase reagents, could detect worm material in tissues and easily distinguish the parasite from the surrounding human or rodent tissues.

Conclusion: The developed anti-PM reagent could be a useful tool in diagnostic pathology.

APPLICATION OF MOLECULAR TECHNIQUES TO THE STUDY OF POPULATION VARIATION WITHIN THE ASCAROID NEMATODE *Hysterothylacium aduncum* (RUDOLPHI, 1802)

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It is generally accepted that *Hysterothylacium aduncum*, a nematode commonly occurring in the digestive tract of a wide range of marine teleosts, notably gadoids, represents a single species. Several synonyms have been ascribed to this nematode, confusing the systematics of the species. To investigate the validity of these synonyms and to contribute new information regarding the biology of this species, we applied two molecular approaches.

Samples of *H. aduncum* were obtained from *Pollachius virens*, *Gadus morhua*, *Merlangius merlangus* and *Melanogrammus aeglefinus* from nine Scottish and three Norwegian sites. The extent of genetic variation was determined by: a) scoring the allelic frequencies of polymorphic enzymes; and b) determination of the nucleotide sequence of selected target areas of the *H. aduncum* genome between hosts and col-

lection sites. Allozyme analysis applied to the Scottish material only, identified five polymorphic enzymes (EST, GPI, IDH, LDH, and PGM). It was possible to discriminate between *H. aduncum* from samples taken from the East and West Coast of Scotland and between *H. aduncum* from *P. virens* and *G. morhua* from West Coast samples. A partial sequence of the 18s ribosomal RNA gene and the internal transcribed spacer (ITS) region was determined as a means to discriminate the existence of sibling species and to detect differences at the population level. The variation in nucleotide sequences between samples for the target regions is presented and commented upon.

THE RELATIONSHIP BETWEEN PRESENCE OF FREE-LIVING AMOEBA AND *Legionella*: INCREASED SENSITIVITY OF *Legionella* DETECTION IN WATER

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Free-living amoebae are ubiquitous organisms inhabiting both natural and man-made aquatic environments where they are involved in complex interactions with bacteria. The role of environmental amoebae as 'reservoirs' and vectors of pathogenic bacteria such as *Legionella* is receiving increasing attention. In this study we examined tap water to which patients with legionellosis have been exposed. The objective of our study was to evaluate the correlation between the presence of amoebae and *Legionella* in the water distribution system. We examined also the ability of amoebae naturally infected with *L. pneumophila* to maintain their growth *in vitro*.

Material and methods: Warm-water samples were concentrated and cultivated in media specific for free-living amoebae. *L. pneumophila* was detected by immunofluorescence in amoeba-positive cultures using species-specific monoclonal antibody and the results were compared to those obtained by cultivation in *Legionella*-specific medium.

Results: Free-living amoebae were found in 19 out of 62 tested water samples. All but two amoeba-positive cultures contained intra- and extracellular *L. pneumophila*, demonstrated by the reaction with *L. pneumophila*-specific monoclonal antibody. After cultivation *Legionella* could be demonstrated in 8 out of 62 samples. Presence of *Legionella* was demonstrated in repeated examinations of amoebae cultured for 10 months.

Conclusions: 1. A close association was observed between *L. pneumophila* and amoebae in the water distribution system. 2. The sensitivity of *L. pneumophila* detection can be improved by immunocytological demonstration of bacteria associated with naturally infected amoeba isolated from water. 3. Amoebae present in water support the growth of *L. pneumophila in vitro* and the presence of bacteria is seen even after prolonged cultivation.

DISTRIBUTION OF TICK-BORNE SPIROCHETOSIS IN THE BALTIC REGION IN RELATION TO THEIR DISSEMINATION BY MIGRATORY BIRDS

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Introduction

The feasibility of eliminating natural foci of diseases is an important and debatable problem. Previous attempts to do this have been only temporarily successful. The importation or migration of infected vectors or reservoir hosts of pathogens into focal areas where disease circulation has been previously interrupted have often resulted in the re-establishment of disease in these foci. For example, tick-borne encephalitis virus circulation was fully suppressed following the application of DDT to thousands of hectares in the Kemerovo region of Russia which eliminated the tick *Ixodes persulcatus* Schulze, the primary vector of this pathogen. Nevertheless, this focus re-established 9 years later due to the transfer of infected ticks by birds (Chunikhin, 1966). A similar suggestion for controlling the spread of Lyme disease by sanitation of tick-infected rodents (Mather *et al.*, 1986) might be inappropriate because this measure would be only temporarily effective. Many authors have demonstrated that field-collected birds can infect ticks with the spirochete *Borrelia burgdorferi* sensu lato (Weisbrod, Johnson, 1989; Anderson *et al.*, 1986; Magnarelli *et al.*, 1992;

Humair *et al.*, 1993; Nakao *et al.*, 1994) and Piesman *et al.* (1996) showed that chickens experimentally infected by nymph bites can be infective for the native *Ixodes scapularis* Say larvae. It has also been proven that migratory birds act as phoretic hosts distributing infected ticks to new locations (Olsen *et al.*, 1995; Nicholls, Callister, 1996; Humair *et al.*, 1996). It is therefore clear that birds can infect ticks with *B. burgdorferi* s.l., conserving and mixing these pathogen populations within natural foci of hard tick-borne borreliosis.

Questions which remain to be answered are which birds species transfer infected ticks along their migratory routes and which spirochetes that they harbour are pathogenic to their bird hosts and to man.

Olsen *et al.* (1995) demonstrated that *Ixodes ricinus* (L.) transferred from northern Sweden harboured only one spirochete species pathogenic to man, *Borrelia garinii*, whereas ticks transferred from south-west and south-east Sweden contained all three tick-borne agents of borreliosis. Two years earlier Olsen *et al.* (1993) published data in which they demonstrated that *B.*

burgdorferi s.l. can be transmitted between *Ixodes* (*Ceratixodes*) *uriae* White and razorbills (*Alca torda*) on Bonden island (63°26' N, 20°03' E) in the Gulf of Bothnia. This island is devoid of mammals and the only vector and reservoir of the spirochete is *I. uriae*. Seabirds are a source of blood for the ticks and some of them probably serve as vertebrate hosts for the spirochete. The authors did not confirm the identity of the species as *B. burgdorferi* s.l. but their data strongly suggest that they had in fact discovered *B. garinii*. In neither of the aforementioned communications did the authors determine either the primary vertebrate host or the primary vector of the parasites which may have been circulating through different tick species.

The aim of this study was to determine whether the primary spirochete vectors may act as filters and accumulators of different *Borrelia* species in the different localities and indicate the degree to which birds function as disseminators of tick-borne agents of borreliosis.

Materials and methods

Based on data indicating that *B. garinii* is probably a pathogen of bird origin, we used the simple and inexpensive method of flagging to collect ticks from the vegetation along the main bird migratory routes connected the Baltic Region and southern Europe.

Ticks: *I. persulcatus* were collected from forests along the northern shore of the Gulf of Finland near St. Petersburg (Morskaya, Lisy Nos) and *I. ricinus* from the Curonian (Kurish) Spit in Kaliningrad Region. All specimens were checked under the stereomicroscope. The activity (Act.ind.) of adults and nymphs was assessed using the inclined

ticksdrome (Alekseev, 1996; Alekseev *et al.*, 1996). Ticksdrome inclined at 45° with a humid half with a strip of unbleached calico was used to check taxes to gravity, humidity, and scent for a tick crawling on its surface. A glass screen isolating it from the investigator allowed the paths of the tick to be traced on a transparency fixed to the screen. The activity index on this ticksdrome was obtained from equation using the speed (Sp), meander value (M – number of turns to the whole route length), the height (h) of the tick movement, the number of falls from or on the ticksdrome surface (NoF), portion of the upward path relative to the total path (WA), and the portion of the path above the humidity gradient (WB):

$$\text{Act.ind.} = \text{Sp} + h + \text{WA} + \text{WB} - \text{M} - \text{NoF}$$

Borrelia: All ticks were then dissected and borreliæ were detected and counted in 250 dark fields under the microscope (40x10). All spirochete-positive specimens were fixed in 70% ethanol in 0.5 ml Eppendorf tubes. The *Borrelia* species were identified by preparing the lysates for a polymerase chain reaction (PCR) by boiling in the presence of ammonium hydroxide (Rijpkema *et al.*, 1996). *Borrelia* DNA was detected by PCR which amplified the intergenic spacer region between 5S and 23S rRNA genes (Rijpkema *et al.*, 1995). Two species pathogenic for man were described from *I. persulcatus*: *B. garinii* and *B. afzelii* in single and mixed infections (Alekseev *et al.*, 1998). *Borrelia* spp identified from *I. ricinus* were *B. garinii*, *B. afzelii* and *B. valaisiana* (Alekseev *et al.*, 1998; Makroushina *et al.*, 1998).

This investigation was conducted between 1995-1997 and in total 911 *I.*

persulcatus and 1465 *I. ricinus* were examined. Chi-squared analysis was performed on the data using Systat (Wilkinson, 1990).

Results

Both localities in this survey lie on the Italian-Spanish bird migratory route (Payevsky, 1971). Birds are not only the reservoir but also the disseminators of pre-imaginal stages of ticks attached to them.

I. persulcatus from the northern shore of the Gulf of Finland mostly harboured *B. garinii* (prevalence 64.6%, 1995-1997 data) with a high percentage of dual infections of *B. garinii* and *B. afzelii* (25.5%) (Aleksseev *et al.*, 1998; Mak-

roushina *et al.*, 1998). In *I. ricinus* from the Curonian Spit the most prevalent *Borrelia* species was *B. afzelii* (44.6%) whereas *B. garinii* was recorded from only 8% of ticks. One third of *B. garinii*-infected specimens concurrently harboured *B. afzelii*.

Nymphs of *I. persulcatus* were found to be much more active than those of *I. ricinus*, especially among specimens collected in autumn (Fig. 1), and had a 1.5–2.3 times greater Activity Index (Fig. 1, A-1: $p=0.01$ and 0.001). *I. persulcatus* nymphs moved faster and higher which most probably permitted them to be in closer contact with ground-feeding birds.

There was a considerable difference between spring and autumn in the prevalence of *Borrelia* species in *I. ricinus* during the first and second seasonal peaks of nymph and adult tick abundance. Using the dark-field technique to detect *Borrelia*, we found spirochete prevalence to be low in spring ($100/1211 = 8.3\%$) whereas in the autumn it was 2.65 fold greater ($65/296 = 22\%$). The difference in prevalence between these two seasons was significant (chi-squared=34.27, $p<0.001$). However, the prevalence of the *Borrelia* species pathogenic to man, as detected by PCR from dark-field positive specimens, displayed the opposite pattern. Of the spirochete-positive ticks collected in the spring 55% (55/100) harboured *B. afzelii* or *B. garinii* whereas only 9.8% (5/51) of ticks collected in the autumn were infected by *Borrelia* species pathogenic to man. This seasonal difference was also significant (chi-squared=14.26, $p<0.001$, Fig. 2).

On the northern shore of the Gulf of Finland, which lies within the northern

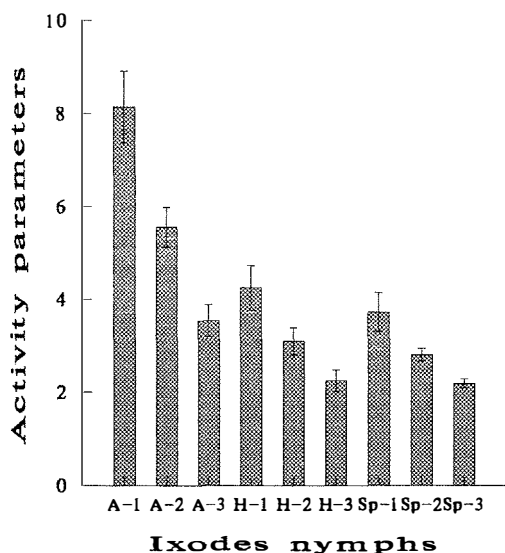


Fig. 1. Comparison of activity parameters of nymphs *Ixodes persulcatus* (1) and *Ixodes ricinus* (2, 3).

A - value of activity index; H - mean height to which nymphs climbed; Sp - mean speed of movement (cm/min); 2 - nymphs collected during the spring peak of activity; 3 - nymphs collected during the autumn peak of activity.

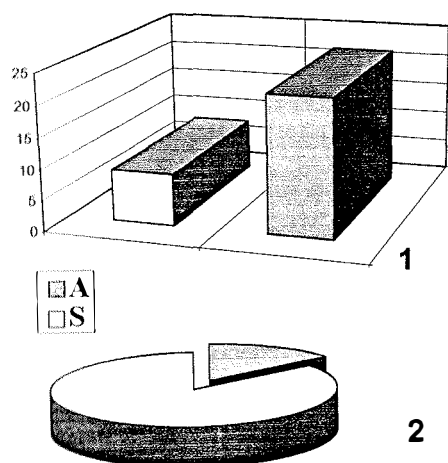


Fig. 2. Prevalence of spirochete and *Borrelia* species in *Ixodes ricinus* ticks within different seasons (1995-1997).

A - autumn data; S - spring data. 1 - spirochete prevalence according to dark-field microscopy; 2 - pathogenic *Borrelia* species prevalence determined by PCR.

part of the bird migratory route, *B. garinii* was prevalent in *I. persulcatus* (64%) whereas in the middle section of this route (Curonian Spit) only 8% of *I. ricinus* were infected with *B. garinii*.

These results might be explained by differences in the source of spirochetes which can be transferred by birds during both the spring and autumn waves of their migration.

Discussion

It is not surprising that in the northern parts of bird migratory routes *I. persulcatus* were mainly infected with *B. garinii*. This *Borrelia* species might be transferred from the northern Europe where foci of *Borrelia* infective purely to birds exist (Olsen *et al.*, 1993, 1996). The greater activity of *I. persulcatus* nymphs allows them to feed on birds and thereby become infected with *B. garinii*.

Less active *I. ricinus* nymphs are better adapted to feed on small animals, e.g. rodents, than on birds and are thus more frequently infected with *B. afzelii*.

Our data is consistent with that from the literature in demonstrating the important role of birds as disseminators both of *Borrelia* species and of spirochete-infected ticks that may attach to them. They strongly suggest that of the species pathogenic to man it is mainly *B. garinii* which is carried by birds from North to South (e.g. from the Baltic to southern Europe) together with undetermined *B. burgdorferi* s.l. and other spirochete species reported to be pathogenic to birds. This explains why *I. persulcatus* harboured mainly *B. garinii* and why *I. ricinus* in autumn are frequently infected with unidentified *B. burgdorferi* s.l.

In contrast, *Borrelia* species causing human disease are mostly transferred from the southern Europe to the Baltic while the number of specific bird pathogens decreases. This most probably results from the death of infected hosts as shown by Valkiunas (1997) for birds infected by Hemosporida. Our data also suggest that *B. garinii* and *B. afzelii* do not cause disease in birds as they quickly become immune to the spirochetes following *Borrelia* inoculation by ticks cannot then become re-infected (Piesman *et al.*, 1996). This emphasises the significance of ticks as a reservoir of borreliae and the phoretic role of birds in the re-establishment of borrelioses (and other infection) foci.

Resume

Differences in the occurrence of *Borrelia* species in *Ixodes persulcatus* Schulze and *Ixodes ricinus* (L.) were

used to show that these vectors might be accumulators of different species of spirochetes and can serve as markers of those pathogens transferred northwards and southwards along the Baltic-Italian-Spanish bird migration route. *I. persulcatus* is mainly an accumulator of *Borrelia garinii* whereas *I. ricinus* mainly harbours *B. afzelii* and some other undetermined spirochete species. *Borrelia* species prevalence varied seasonally most probably in accordance with the spring and autumn waves of bird migration.

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