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Abstracts of Papers

Prevalence of intestinal parasites in children at 44 schools in the Boland/Overberg Health Region, Western Cape Province, South Africa

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This study was a survey to determine the prevalence of helminths and protozoa in children at primary schools in the Boland/Overberg Health Region. A random sample of 44 schools was drawn from 210 schools serving relatively disadvantaged communities ($n = 1260$ children). At each school the Boland/Overberg parasite control team of the Department of Health collected stool samples of about 30 g per child from approximately 15 boys and 15 girls in Grade 3. Stools were processed for microscopy using the formal-ether concentration technique. Overall prevalence of helminths, expressed as eggs per gram, was 43%. *Trichuris trichiura* was the most frequently occurring helminth (31%). Protozoal cysts were expressed semi-quantitatively. Prevalences were: *Giardia duodenalis* (20.5%), *Entamoeba coli* (48.7%) and *Entamoeba histolytica/dispar* (11.3%). Small, unidentifiable operculated eggs were present in stools of 6 children. *Fasciola*, *Trichostrongylus*, hookworm and *Strongyloides* eggs/larvae were identified in the stool of a single, different child for each parasite — 30% of schools need intervention by WHO criteria for helminths alone; 43% of schools had high prevalences of helminths and/or protozoa. Intervention should include improvement of sanitation, intensive health education and a deworming programme when prevalence exceeds 50%. The distribution of schools according to prevalence of helminths was mapped using GIS to identify spatial pattern in the data, as well as for use in health management.

Unusual protozoans found in the oesophageal folds of the abalone *Haliotis kamtschatkana kamtschatkana* Jonas, 1845

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In the cold Pacific waters off California, red abalone (*Haliotis rufescens* Swainson, 1822) has traditionally been the most popular and commercially important species (Hahn 1989). Six other slightly less desirable species also occur in this area, namely *H. kamtschatkana kamtschatkana* Jonas, 1845; *H. cracherodii cracherodii* Leach, 1814; *H. corrugata* Wood, 1828; *H. fulgens fulgens* Philippi, 1845; *H. sorenseni* Bartsch, 1940, and *H. walallensis* Stearns, 1899. Histological sections were made of *H. kamtschatkana kamtschatkana* at the Pacific Biological Station in Nanaimo, British Columbia, Canada. These were examined in the laboratory in Bloemfontein, and the presence of several unidentified protozoans in the folds of the haliotid's oesophageal pouch was confirmed. Some of these protozoans resemble scyphidiid peritrichs found associated with abalone gills from the South African coast. Studies of whole mounts as well as scanning electron microscopy are necessary to describe these protozoans.

The development of Haller's organ in the East African tick *Amblyomma gemma*

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Amblyomma gemma, a 3-host tick is found exclusively in the arid parts of East Africa. The adults parasitise large ungulates (including livestock) and carnivores, while the immature stages may also infest small mammals, birds and reptiles. This tick is of economic importance as it acts as a vector of heartwater, theileriosis and human-tick bite fever. Larvae, nymphs and adult ticks were preserved in 70% ethanol, after submersing them in hot water. The micromorphology of Haller's organ was described using scanning electron microscopy. The Haller's organ of the larvae had 2 rows of paired socketed sensory setae proximal to the capsule, which had a very thin slit-like opening. The anterior pit contained 6 sensillae of different types. A pair of long sensory setae was located distal to the anterior pit with the lateral seta pitted. The composition and grouping of the different sensory organs changed with each moult. A group of 4 setae appeared between the 2 pairs of proximal setae in the adult, while the pitted sensilla in the anterior pit increased greatly in size. The selection of different hosts by the instars as well pheromonal attraction in the adults may be reflected by these morphological changes in Haller's organ.

Automated detection of malaria during full blood count analysis

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Effective treatment of malaria requires an accurate and timely diagnosis since unsuspected cases of *Plasmodium falciparum* malaria are often fatal. This study evaluated the reliability of an automated diagnostic procedure for malaria, which relies on the detection of malaria pigment during routine full blood count (FBC) analysis. A total of 651 blood samples were evaluated by microscopy and rapid tests for HRP-2 protein (ICT Malaria Pf) and parasite pLDH (Optimal). Positive samples ($n = 247$) had microscopically detectable parasites; *P. falciparum* ($n = 232$), non-*P. falciparum* ($n = 9$) and mixed infections ($n = 4$). Malaria-negative samples ($n = 359$) were negative on microscopy and rapid tests. Convalescent cases ($n = 45$) were negative microscopically, reacted positively on at least one positive rapid test and had a recent documented history of positive microscopy. All samples were processed with the Abbott Cell-Dyn CD3200 analyser and screen displays visually examined for abnormal depolarising white blood cell events (BJH 104: 499, 1999). A total of 236 from 247 malaria positive cases were CD3200 positive, and 340/359 malaria negative cases were CD3200 negative. All 45 convalescent malaria cases were CD3200 positive. We conclude that abnormal CD3200 depolarising events are very sensitive (96%) and specific (95%) for detecting malaria. This technology will be particularly valuable for patients with unsuspected malaria who have FBC analysis performed for unexplained pyrexia.

Application of a Parasite Index correlated with water quality on the Vaal River System

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The Fish Health Assessment Index (HAI) and associated Parasite Index (PI) has been successfully applied to the Olifants River system. This study was undertaken to test whether the index can demonstrate a negative correlation between water quality and fish health in the Vaal River system. The localities sampled were the Vaal Dam (good water quality) and Vaal River Barrage (poorer water quality). Twenty sharp-toothed catfish were collected bimonthly at each locality using gill nets. While performing the HAI, fish were examined for parasites and numbers recorded as 'ectoparasites' or 'endoparasites'.

Ectoparasites are presumed to be more exposed to effects of poor water quality than endoparasites. Percentage of infected hosts and mean intensity of ectoparasitic infection were greater at the less polluted locality. The converse was true with regard to endoparasite numbers. *Trypanosome* infection is an exception. It is transmitted by ectoparasites and conforms to values recorded for the latter. Data collected were applied to 3 different variations of the Parasite Index and results compared. For all 3, higher HAI values correlate with poorer water quality, with none producing superior results.

Aspects of the life cycle of *Lamproglena clariae* (Crustacea: Copepoda)

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The genus *Lamproglena* was created in 1832 by von Nordmann and consists of 41 species and 1 subspecies. To date very little has been published on the ecology of the genus *Lamproglena*, including *L. clariae*, described by Fryer in 1956. The aim of the current study was to establish the life cycle of *L. clariae*. The African sharptooth catfish, *Clarias gariepinus*, is the natural host for *L. clariae*. The fish were collected bimonthly from the Vaal Dam, using gill nets of various sizes. The host's gills were removed and examined for parasites. Various larval stages were found along with the male and female adult attached to the gill filaments. The life cycle apparently consists of 3 naupliar, 5 copepodite and 2 cyclopid stages. The number and size of appendages can be used for the identification of each of the life stages. Furthermore, the number and size of the thoracic and abdominal somites are used to distinguish different stages, particularly those of the copepodite and cyclopid stages. The life cycle of *Lamproglena clariae* was established. It resembles the life cycle of other parasites in the family Lernaeidae, such as *Lernaea cyprinacea*.

On a seemingly aberrant *Kroyeria sphyrnae* (Copepoda: Siphonostomatoida) from the Indian Ocean, South Africa

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Kroyeria sphyrnae was originally described by Rangnekar (1957) from an unknown hammerhead shark (*Sphyrna* sp.). *K. sphyrnae* can easily be distinguished from other *Kroyeria* species by the long, acute, lissome dorsal stylets, the relatively short interpodal stylets of legs 2, 3 and 4 and the mandible with 7 teeth of different sizes. The copepods were fixed and preserved in 70 % ethanol. In the laboratory they were studied by bright field and phase contrast microscopy using the wooden slide technique. Before dissection, copepods were cleared in lactic acid in which a pinch of lignin pink had been dissolved. Some atypical features of the collected *K. sphyrnae* specimens from smooth hammerhead sharks (*Sphyrna zygaena*) are a swollen anterior part of the genital complex and a dorsal stylet with a bifid tip. Nevertheless, based on these aberrant characteristic differences only, these specimens are not considered to be a new species, but simply illustrate variation in the morphology of *K. sphyrnae*. The general suit of sexually dimorphic characteristics of the male, to date unknown, as well as differences pertaining to the armature of the first antennae and some aspects of the armature of legs 1–4 are described.

Polystomatid biogeography

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Monogeneans are mainly parasitic in fish but one family, the Polystomatidae, has radiated and infects anurans, chelonians, the Australian lungfish, the Japanese salamander and the hippopotamus. Polystomatids are known from all continents and most of the larger islands inhabited by their hosts. The family is represented by 19 genera. *Diplorchis* Ozaki, 1931, *Eupolystoma kaw*, 1950, *Mesopolystoma* Vaucher, 1981, *Metapolystoma* Combes, 1976, *Neodiplorchis* Yamaguti, 1963 *Parapolystoma* Ozaki, 1935,

Parapseudopolystoma Nasir & Fuentes Zambrano, 1983, *Polystoma* Zeder, 1800, *Protopolystoma* Bychowsky, 1957, *Pseudodiplorchis* Yamaguti, 1963, *Pseudopolystoma* Yamaguti, 1963, *Riojatrema* Lamothe-Argumento, 1964, *Sundapolystoma*, 2001, and *Wetapolystoma* Gray, 1983, are known from anuran hosts, *Neopolystoma* Price, 1939, *Polystomoidella* Price, 1939, and *Polystomoides* Ward, 1917, from chelonians, *Concinocotyle* (Pichelin, Whittington & Pearson, 1991) from the Australian lungfish and *Oculotrema* Stunkard, 1924, from a mammal, the hippopotamus. *Polystoma* has a cosmopolitan distribution except that it is not known from Australia; *Protopolystoma* and *Metapolystoma* are restricted to the Afrotropical Region while *Eupolystoma* occurs in both the Afrotropical and the Oriental Regions; *Mesopolystoma*, *Parapseudopolystoma* and *Wetapolystoma* are known from the Neotropical Region; *Neodiplorchis*, *Pseudodiplorchis* and *Riojatrema* are Nearctic species; *Parapolystoma* and *Sundapolystoma* are known from the Australian Region while *Diplorchis* is known only from the Palaearctic Region. Among the chelonian parasites *Polystomoides* and *Neopolystoma* have a wide distribution while *Polystomoidella* is largely restricted to the Nearctic Region. *Concinocotyle* is restricted to the Australian Region, *Pseudopolystoma* is restricted to the far-eastern Palaearctic Region, and *Oculotrema* is restricted to the Afrotropical Region. Polystomatids constitute a very old group of parasites that radiated with their vertebrate hosts. The close link between host and parasite makes this an ideal group to study host-parasite relationships. Furthermore, the apparently wide distribution of some groups as opposed to the restricted distribution of others present interesting opportunities to study the phylogeny of this group.

The influence of *Dipylidium caninum* on the life cycle of *Ctenocephalides felis*

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The dog tapeworm, *Dipylidium caninum*, occurs throughout South Africa and is cosmopolitan in its distribution. This tapeworm is one of the most common intestinal parasites of dogs, cats or other small carnivores as the definitive host, and arthropods such as fleas or lice as intermediate hosts. It is occasionally found as a human parasite, usually in children. The cat flea, *Ctenocephalides felis*, becomes an intermediate host of *D. caninum* when flea larvae become infected by eating organic debris contaminated with tapeworm eggs. The definitive host is infected when ingesting an infected flea. During this study flea larvae were exposed to different concentrations of tapeworm eggs to determine the effect of *D. caninum* on the completion of the flea's life cycle. Where flea larvae of different instars were exposed to tapeworm eggs, hexacanth tapeworm larvae were found in all 3 instars, which indicated that *C. felis* can become contaminated during any period of the larval stage. Only about 63.2 % of the larvae developed into adult fleas after being exposed to a high concentration of tapeworm eggs, whereas 94.7 % of the larvae completed the life cycle at a low egg concentration. No less than 96 % of the flea larvae that fed on standard rearing medium and remained uninfected, developed into adults. The results indicated that *D. caninum* is harmful to its larval host.

Preliminary report on the structure and function of the salivary glands of flat female *Amblyomma hebraeum* and *Rhipicephalus appendiculatus*

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Salivary glands were collected from flat (unengorged) females of both *Amblyomma hebraeum* and *Rhipicephalus appendiculatus*. Samples of each species were fixed in Bouin's solution, 7-micrometre thick sections were cut and stained in haematoxylin and eosin, and were used for examining the structure of the glands. One sample of each species was used for TEM to also study the structure of the glands. More salivary gland samples of each species were rapidly frozen over liquid nitrogen, sectioned in a cryotome at 8-micrometre thickness, and used in histochemical experiments testing for two diaphorases, one dehydrogenase, fatty acids and phospholipids.

The aim of this project was to determine the feasibility of using histochemistry to demonstrate some important parameters indicative of physiological activities in the salivary glands of unfed female ticks with long and ticks with short mouthparts. Preliminary results show that the histochemical tests performed were positive.

Are human intestinal parasites significant covariables of child growth and water quality in the Boland/Overberg Health Region?

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The objective of an ongoing study is to test whether prevalence of helminths and protozoa can be associated with impaired growth of children and lack of clean water and sanitation in the region. There are 210 primary schools and a random sample of 44 was used in a cross-sectional survey. Faecal samples from 1260 children were examined for helminth eggs and protozoan cysts. Results are analysed statistically and mapped by Geographic Information System methods (GIS) to define spatial relationships between prevalence of parasites, growth of children, sanitation and water supply. There are significant differences in mean height-for-age between schools and health wards in the Region. Prevalence of *Giardia*, an indicator of water pollution, exceeded 14 % in 31 schools. A piped water supply was not available in 68 % of these high-*Giardia*-prevalence schools. Preliminary indications are that intestinal parasites probably reflect conditions under which communities live. Analysis of faecal surveys and spatial relationships (GIS) could be cost-effective tools for the formulation of health policy, as well as for health management, delivery and education, under local conditions.

Ticks biting humans in South Africa

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South Africa is a popular tourist destination with an influx of international travellers exploring nature on private land and in provincial and national parks. With greater use of these facilities by South Africans, the risk of being bitten by disease-carrying ticks has increased. Information for travellers about the risks of tick bites and tick-borne diseases is generally sparse. We report on results collected both from the literature and personal records that provide data on which ticks bite and transmit disease to humans. In a total of 200 reports over a period of 24 years, 19 tick species in 5 ixodid genera were recorded, namely: *Rhipicephalus* (58.5 %), *Amblyomma* (18.5 %), *Hyalomma* (10.5 %), *Haemaphysalis* (8 %) and *Ixodes* (4 %) and one record of an argasid tick (*Otobius* sp.). *Rhipicephalus simus* (17 %), *Amblyomma hebraeum* (16.5 %), *Rhipicephalus gertrudae* (12.5 %), *Rhipicephalus appendiculatus* (10 %), *Rhipicephalus maculatus* (9.5 %), *Hyalomma truncatum* Koch 1844 (8.5 %) and *Haemaphysalis leachi* (7.5 %) were the most common ticks biting humans.

Humoral immune response assessments in Saanen and Indigenous goats against adult *Rhipicephalus appendiculatus* infestation

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Changes in serum gamma-globulin levels, replete female tick mass and numbers recovered during 3 consecutive tick infestations were used as parameters to monitor humoral immune response in Saanen and indigenous goat breeds against adult *Rhipicephalus*

appendiculatus infestation under laboratory conditions. As stress is regarded as a factor contributing to immunosuppression in hosts exposed to tick infestation, the chronic stress levels of Saanen and indigenous goats were assessed under different housing conditions and during 3 consecutive tick infestations by monitoring the serum cortisol levels. Under different housing conditions as well as during tick infestations the mean of all cortisol values (consistently less than 250 nmol/l) was negligibly low. During the 3 consecutive infestations the serum gamma-globulin levels increased for both Saanens and indigenous goat breeds and correlated negatively with the mean replete female mass and numbers recovered. Both the breeds exhibited a humoral immune response. The humoral immune response of the indigenous goats was high compared to that of the Saanens.

The micromorphology of the long-nosed cattle louse *Linognathus vituli* (L.)

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These bloodsucking lice are important ectoparasites of calves that cause not only irritation, but also severe anaemia during heavy infestation. The lice were collected from infested calves and fixed in 70 % ethanol. After ultrasonic cleaning, they were routinely prepared for scanning electron microscopy (SEM) and viewed in a Leica Stereoscan 420 SEM. The SEM studies revealed specialisation of the legs as grasping organs. The stout tarsal claws vary in size from the small 1st pair to the robust claws of the 2nd and 3rd pair of legs, which close against large pretarsal sclerites consisting of pads that are ridged to improve the grip on the hair. The abdominal exoskeleton is covered by protective scales and posteriorly-directed spinous setae. The lumen of the large thoracic and 6 pairs of smaller abdominal spiracles are lined with circular rims. The anterior haustellum consists of a pair of vertical flaps that protects the opening of the hypopharynx and a pair of ventral openings from the salivarium. The distal segment of the antenna ends in a 'peg organ' which consisted of 11 sensory setae, while sensory 'pore organs' and a 'plate organ' were observed on the lateral surfaces of the 2 distal antennal segments that are presumably used for orientation on the host.

Ergasilid parasites found on mullet species along the South African coast

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The flathead mullet, *Mugil cephalus*, has a wide geographical distribution and is parasitised by more than 40 different species of parasitic copepods. It acts as host for 9 species of Ergasilidae. The southern mullet, *Liza richardsonii*, is common in estuaries as well as intertidal pools along the south coast of South Africa. During field trips to the east coast (Bazley) and south coast (De Mond Nature Reserve and Jeffreys Bay) of South Africa, more than 100 specimens of *Mugil cephalus* and *Liza richardsonii* were collected and examined for parasitic copepods. Collection methods included using hand nets and cast nets. After collection the fishes were taken to a fully-equipped field laboratory where each specimen was examined for different groups of parasites occurring in and on the host. All parasites found were removed and fixed according to standard methods for that specific group, and all the parasitic copepods were fixed in 70 % ethanol. *Dermostergasilus mugilis* was collected from the gills of *Liza richardsonii* at the De Mond Nature Reserve as well as from the gills of *Mugil cephalus* at Bazley, whereas *Ergasilus mugilis* was collected from the gills of *Mugil cephalus* at Jeffreys Bay.

Aspects of the ecology of a *Paradiplozoon* sp. from *Barbus aeneus* collected in the Vaal Dam

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Representatives of the Diplozoidae are specific to cyprinid freshwater fishes and have a life cycle dependent on seasonal changes.

Incidence of infection in *Paradiplozoon* is therefore expected to vary seasonally. Fishes from various species were collected bimonthly from December 1998 to February 2000 with gill nets in the Vaal Dam. Fish were killed by a cut through the spinal cord and age, sex and size determined. Gills were removed and examined under a dissection microscope and distribution of parasites recorded. Parasites were removed, fixed in warm aceto-formaldehyde-alcohol and preserved in 70 % ethanol. *Paradiplozoon* sp. was found only on *Barbus aeneus* and is thus strictly host-specific. No preference for sex, age or size groups were shown. Distribution records showed no preference for gills on either left or right side of the host. However, a higher incidence of adult worms was recorded on the first and third gill arches in the median region, suggesting preference for these sites. The highest prevalence (76.9 %), mean intensity (1.45) and abundance (1.12) was recorded during summer (October 1999). It can be concluded that *Paradiplozoon* is highly host specific, shows some site preference, and is variable with regard to seasonal occurrence.

The seasonal abundance of adult ixodid ticks infesting cattle belonging to resource-poor farmers in the north-eastern Free State Province

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A study was undertaken to identify tick species infesting cattle owned by resource-poor farmers in the north-eastern Free State Province of South Africa. Infestations by *Boophilus decoloratus* (Koch, 1844), *Rhipicephalus evertsi evertsi* (Neumann, 1897), *Rhipicephalus follis* (Donitz, 1910), *Rhipicephalus gertrudae* (Feldman-Muhsam, 1960) and *Rhipicephalus punctatus* (Warburton, 1912) were monitored on cattle of mixed breeds at monthly intervals from May 1998 to April 1999. These cattle were of poor quality stock and bred under traditional husbandry practices where tick control is minimal. High tick intensity on cattle was observed between March and June, with a peak in April/May for *B. decoloratus* and in May for *R. e. evertsi*. Few ticks of the other 3 species were recovered from cattle, with small peaks in October for *R. follis*, in November for *R. gertrudae* and *R. punctatus*. At the end of winter in this region, tick numbers (*B. decoloratus*) declined markedly, with those of *R. e. evertsi* and other rhipicephalids increasing. *B. decoloratus* and *R. e. evertsi* are vectors of economically-important tick-borne diseases in South Africa; *B. decoloratus* is strongly associated with bovine babesiosis, anaplasmosis and spirochaetosis, whereas *R. e. evertsi* is associated with equine babesiosis, anaplasmosis, ovine theileriosis, spirochaetosis and paralysis. Results obtained in this study provide valuable information needed for tick control strategies in the area.

Varroa jacobsoni as a possible mechanical vector for *Paenibacillus larvae larvae* pathogenic to *Apis mellifera*

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Paenibacillus larvae larvae, the causative agent of American foulbrood of the honeybee *Apis mellifera*, is a Gram-positive bacterium forming extremely resistant endospores. Only the larval forms of the honeybee are affected. They are infected by spores transmitted by adult worker bees. *Varroa jacobsoni* is a parasitic mite feeding on larvae as well as on adult bees. In bee colonies these mites are usually transmitted by robbing and drifting individual bees. The aim of this investigation was to establish whether *V. jacobsoni* can play a role in the transmission of *P. l. larvae* spores from infected to healthy bee colonies. *Varroa jacobsoni* that had been collected from colonies heavily infected with American foulbrood were homogenised, the resulting suspension plated out on the surface of a semi-selective MYPGP agar medium and incubated at 35 °C. Colonies of bacteria that could be identified as *P. l. larvae* were visible after 4 days. It is concluded that American foulbrood may be transported from

infected to healthy bee colonies by spores of *P. l. larvae* carried in or on *V. jacobsoni*.

Cercariae shed by freshwater snails from the Okavango Delta, Botswana

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In Africa, millions of people and their livestock are infected with *Schistosoma* and other trematode parasites that could be detrimental to their health. It is therefore necessary to obtain information about the freshwater snails that are intermediate hosts to these parasites. During an extensive field trip to the Okavango Delta, Botswana, 264 freshwater snails of 8 different genera were collected. Eight different cercariae were found to infect freshwater snails as intermediate hosts: a vivax cercaria infecting *Pila occidentalis*, an echinostome and strigeid cercaria from *Lymnaea natalensis*, a xiphido cercariae from *Lanistes ouum*, a schistosome and strigeid cercariae from *Biomphalaria pfeifferi*, a parapleurolophocercous cercaria from *Cleopatra elata* and a paramphistome cercaria from *Bulinus globosus*. Freshwater snails must be considered part of the freshwater community, particularly as they play a major role in the food chain that produces edible fish, which constitute an important source of protein for human populations in certain regions in Africa.

Occurrence and source of *Cryptosporidium* and *Giardia* in catchment areas and wastewater works in KwaZulu-Natal

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The enteric protozoa *Cryptosporidium parvum* and *Giardia lamblia* have been recognised as important causes of both outbreak-related and sporadic diarrhoea in humans. The aims of this study were to investigate the occurrence of *Cryptosporidium* and *Giardia* in catchment areas and wastewater works. Water sampling sites were chosen along the uMsunduzi River and its tributaries as they flow through the rural and peri-urban areas of Vulindlela and through the city of Pietermaritzburg. *Cryptosporidium* was detected in 3 % while *Giardia* was detected in 23 % of samples analysed. High numbers of (oo)cysts were detected in rivers following increased rainfall possibly owing to sewer blockages or breakages or run-off from faecally contaminated land. *Cryptosporidium* and *Giardia* were also detected in the treated effluent and sludge samples from Darvill wastewater works. Viable *Cryptosporidium* oocysts and *Giardia* cysts were detected in the effluent and sludge samples, indicating that neither the activated sludge process nor anaerobic digestion are able to inactivate these organisms. As *Cryptosporidium* oocysts and *Giardia* cysts have been found in surface water and wastewater in KwaZulu-Natal, it is necessary to determine the source of these protists so that they can be monitored as part of an integrated catchment management plan.

Migration and urban schistosomiasis in Pietermaritzburg, South Africa

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Rural-urban migration to Pietermaritzburg has led to the establishment of informal settlements in and around its centre. The rapid growth of these settlements has public health consequences, which need to be addressed and monitored. Urinary schistosomiasis is present at low prevalence (7.2 %) and intensity levels (mostly <200 eggs/ml) amongst settlement dwellers but these may be underestimated owing to surveillance difficulties. Transmission was patchily distributed, perhaps as a result of migration and because infections were mostly light. Infected snail hosts (*Bulinus africanus*) were found in nearby rivers and dams and an active focus of *Schistosoma haematobium* transmission was discovered in a city park – the first example of urban schistosomiasis from South Africa. A structured

interview questionnaire administered to settlement occupants revealed that violence caused 52 % of the migration cases and that the migrants with moderate and high-intensity infections came mostly from three areas. Few migrants had any knowledge of schistosomiasis. These findings have implications for urban health monitoring programmes. Surveillance and monitoring programmes are important as they play a central role in better understanding the relationship between disease and mobility, which are 2 neglected factors in epidemiology. Multi-sectoral routine monitoring and surveillance programmes need to be established in areas where rural-urban migration is occurring. These programmes should monitor the distribution of freshwater snails and determine the prevalence of *Schistosoma haematobium* amongst them. Water contact studies are required to establish possible points of transmission. The migration patterns of informal settlement dwellers need to be described as well as establishing the prevalence of *Schistosoma haematobium* infection amongst them. It is important that the epidemiology of *Schistosoma haematobium* be described for the migrants from the area under surveillance and the area from which they originate. Education programmes need to run parallel with these respective surveillance programmes.

The potential of pathogenic fungi for tick control

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Entomogenous fungi *Beauveria bassiana* and *Metarhizium anisopliae* were cultured on Sabourauds Dextrose agar and conidia harvested and suspended in water or oil emulsions. They were sprayed on ticks (*Rhipicephalus appendiculatus* and *Boophilus decoloratus*) engorging on cattle or applied by immersion. The ticks were incubated in the laboratory or sealed in nylon tetra packs, left in the field in shade and pathological effects recorded. In field paddock experiments, conidia were sprayed on tick-seeded pastures once per month on 5-acre paddocks each carrying 5 zebu cattle and on-host ticks counted monthly for 6 months. Both formulations of the 2 fungi induced mortalities in all ticks tested. Generally, mortalities were higher with the oil formulation. Aqueous formulations of both fungi induced mortalities in adult *R. appendiculatus* (36–64 %) and in *B. decoloratus* (40–50 %) engorging on cattle and reduced fecundity in *R. appendiculatus* by 70–80 %. The fungi also reduced egg hatchability in both ticks; *R. appendiculatus* (80–90 %) and *B. decoloratus* (48–68 %). In potted-grass experiments, mortality in nymphs of both *R. appendiculatus* and *A. variegatum* was about 100 % with oil and 80 % with aqueous formulations, whereas in adults, mortality was 20–40 % (aqueous) and 65–100 % (oil), respectively. In field paddocks sprayed with fungi, tick populations were significantly lower than in the control. After 6 months of spraying, the mean numbers of adult *R. appendiculatus* on cattle were reduced by 80 % in *B. bassiana* and 92 % in *M. anisopliae*-treated paddocks. These fungi have potential as biological control agents for ticks and therefore merit further studies.

Xiphidio cercariae in the Free State and North West Province

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Freshwater snails serve as intermediate hosts for a variety of parasites, for example *Bulinus africanus* for *Schistosoma haematobium* and *Biomphalaria pfeifferi* for *S. mansoni*. Less well known is the xiphidio cercariae characterised by its xiphidio-spine at the anterior end of the body. Six different xiphidio cercariae were found in these areas and studied. Freshwater snails were collected from rivers, ponds and farm dams using metal scoops. Cercariae spontaneously shed by these snails were collected and studied by light microscopy using Nile blue sulphate and neutral red as vital stains, and routine scanning electron microscopy techniques. Two freshwater snail species were found to serve as first intermediate hosts for xiphidio cercariae. *Bulinus tropicus* secreted 3 xiphidio types in the Free State and 2 xiphidio types in the North West Province. *Lymnaea natalensis* secreted 1 xiphidio type in the North West Province. These cercariae

do not only differ internally from each other, but also in their external surface covering. The life cycles of these parasitic stages are unknown. They display interesting survival possibilities, especially in areas with only temporary water bodies.

A new gyrodactylid (Monogenea; Gyrodactylidae) from the mormyrid, *Marcusenius macrolepidotus* in the Northern Province, South Africa

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The family Gyrodactylidae comprises 25 genera and parasitises a variety of hosts. They have a worldwide distribution with 4 genera recorded from Africa, i.e. *Gyrodactylus* Nordmann, 1832; *Macroglyrodactylus* Malmberg, 1957; *Gyrdicotylus* Vercammen-Grandjean, 1960, and *Afrogyrodactylus* Paperna, 1968. During a parasitological survey of mormyrid fishes in the Northern Province, a viviparous monogenean was found on the external surface of *Marcusenius macrolepidotus*. This monogenean could not be classified into any known genus within the Gyrodactylidae. Monogenetic parasites were fixed with hot ($\pm 70^{\circ}\text{C}$) AFA fixative and stored in 70 % ethyl alcohol. Unstained specimens used for measurement of the hamuli and marginal hooks were mounted in glycerin jelly. Specimens used for detailed anatomical studies were stained in Horen's Trichrome and counterstained in acetocarmine. Conventional methods were used for scanning electron microscopy. The following characteristics were recorded: Cirrus large, with 1 large needle-like spine and numerous small spinelets. Peduncular bar present. Haptor clearly demarcated from body and situated on a small pedicle. Haptor with 1 pair of large anchors, ventral bar, small dorsal bar and 16 marginal hooks of equal size. This appears to be a new genus that is closely related to *Swingleus* Rogers, 1969, and resembles the general body shape of *Polyclithrum mugilini* Rogers, 1967, but differs from the abovementioned genera and all genera of the Gyrodactylidae by the morphology of the haptor armament and cirrus and the presence of a haptor pedicle.

Alternative helminth control methods and their effect on the internal parasite burdens in donkeys

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Twenty-three southern African working donkeys (*Equus asinus*) were allocated to 8 experimental groups. The aim of the study was to determine what effect alternative helminth control methods will have on their internal parasite burdens. Three alternative control methods (monthly removal of faeces from the pasture, pre-winter treatment with moxidectin, and a combination of these 2 methods) and a replicate of each were compared with 2 controls. *Post mortem* examinations were performed on 9 of the donkeys at the end of the 16-month study. Estimated worm burdens ranged from 3831 to 29 501 and 38 helminth species were recorded. The latter included a previously unknown cyathostome species that is being described. *Cyathostomum montgomeryi* was the most abundant cyathostome and *Triodontophorus hartmannae* the most abundant large strongyle. The large strongyles were less abundant compared to the cyathostomes, and both groups preferred the ventral colon. Both the monthly removal of faeces from the camps and pre-winter treatment resulted in a reduction in the total number of mucosal larvae and the strongyle parasites in the lumen, but the combination of these 2 interventions had the largest impact.

Refractoriness of Leishmaniosis in experimentally-infected domestic and wild birds

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In the first of a series of experiments, a wild rock pigeon (*Columba guinea*), a wild guinea fowl (*Numidia meleagris*), a domestic chicken

(*Gallus gallus*) and a domestic feral pigeon (*Columbia livia*) were subcutaneously challenged each with 1×10^7 culture-derived stationary phase *Leishmania major* (NLB-144) promastigotes. No parasites were recovered from any of the birds during weekly sampling over a period of 11 weeks using both smears and cultures, and nor were lesions or nodules suggestive of a cutaneous infection observed. All the birds except the guinea fowl were sacrificed at 12 weeks post *L. major*-infection, and cultures and smears prepared from liver, heart, bone marrow and blood were negative for parasites. In a 2nd series of experiments, uninfected birds, a rock pigeon, a chicken, and a feral pigeon, as well as the guinea fowl previously infected with *L. major*, were challenged subcutaneously with an inoculum dose of 1×10^8 culture derived *L. donovani* promastigotes. The animals were sacrificed 4 weeks post challenge. Impression smears and cultures prepared from the liver, heart, bone marrow and blood were negative for parasites. Serum from all the experimental groups when assayed for *Leishmania*-specific antibodies using the direct agglutination assay showed low evidence of humoral response to infection. As it is not possible to experimentally infect these birds even with high doses of either *L. major* or *L. donovani* parasites, it is concluded that domestic and wild birds cannot be reservoirs of *Leishmania* infection.

A geographic information system's model for schistosomiasis in South Africa using temperature and rainfall

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Data were obtained from a hard copy 'Atlas of Bilharzia in South Africa', and incorporated digitally into a geographic information system (GIS) together with national environmental data. The resulting easily manipulated, dynamic database was used to illustrate the occurrence of both urinary and intestinal schistosomiasis in South Africa, and then to assess the relationship between temperature, rainfall and schistosomiasis. The results of 2 models are presented. The 1st is a descriptive model that translated published field experimental data on temperature and schistosome distribution in South Africa onto maps using underlying spatial minimum and maximum temperature parameters. The 2nd is a statistical model that used the spatial tools within a raster GIS software package (Idrisi) and ordered logistical analysis to determine the relationship between the occurrence of schistosomiasis and underlying climate data. The results of the descriptive model indicate that minimum temperature regimes and temperature range effectively explained the distribution of schistosomiasis. The regression analysis showed that temperature and rainfall are significant in describing the occurrence of *Schistosoma haematobium* in South Africa.

The prevalence of *Toxoplasma gondii* infections in KwaZulu-Natal, South Africa

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Congenital toxoplasmosis normally arises from primary infection in immunocompetent women during pregnancy. However, reactivation of latent *Toxoplasma* infections could occur in women who are severely immunocompromised, resulting in the materno-foetal transmission of the parasite. A cross-sectional observational study involving 1500 women (HIV-infected and HIV-uninfected) was conducted in order to determine the prevalence of *Toxoplasma gondii* infections. Residual sera were collected from King Edward VIII Hospital (Virology) over a 2-month period. The samples were tested for IgG specific anti-*Toxoplasma* antibodies using an in-house ELISA system. Univariate statistical analysis was performed to determine the prevalence of toxoplasmosis in different demographic groups and in the HIV-positive and negative subjects. The overall prevalence of *Toxoplasma gondii* infection in pregnant women was 39 % (95 % CI 37–42 %). In HIV-negative women the prevalence of toxoplasmosis was 38 % (95 % CI 35–40 %) compared to a prevalence of

43 % (95 % CI 38–48 %) in the HIV-positive group. Our findings show that the overall prevalence of toxoplasmosis among pregnant women in KwaZulu-Natal is higher than previously reported. *Toxoplasma*-specific IgM antibody testing is currently being conducted to determine the number of acute infections. These data will be useful to predict the number of babies at risk of congenital infection.

A sero-epidemiological survey of parasites in cattle in the north eastern Free State

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A survey to determine the incidence of parasites in cattle ($n = 383$) was conducted in the northeastern Free State between August 1999 and July 2000. Giemsa-stained blood smears were negative for blood parasites. In total, 94 % of the cattle were sero-positive for *Babesia bigemina* by indirect fluorescent antibody (IFA) test and 87 % were sero-positive for *Anaplasma* by enzyme-linked immunosorbent assay (ELISA). There was no significant difference in the incidence of either anaplasmosis or babesiosis between the seasons. Faecal samples from these animals were analysed for the presence of helminths and protozoa using the McMaster and Visser sieve techniques. The average egg per gram calculations was found to be low. Typical helminths that were found in this area included *Haemonchus placei*, *Nematodirus* spp., *Oesophagostomum radiatum*, *Cooperia* spp., and *Ostertagia ostertagi*. *Eimeria* sp., which is a protozoan, was also found. The dominant parasites are *Paramphistomum cervi*, *Dicrocoelium lanceatum* and a protozoan, *Buxtonella* sp. Results from this study generally indicate low prevalence of helminthic infections in the study area. The observation of negative blood smears but high incidence of positive serological results for *Anaplasma* and *Babesia* for the same group of cattle indicates that this area is endemic for these diseases but with a stable disease condition.

Factors influencing vector-parasite transmission of theileriosis

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The factors that influence transmission by the tick *Rhipicephalus appendiculatus*, the vector for the Protozoan parasite *Theileria parva*, which causes severe theileriosis, were investigated. The disease, known as East Coast fever, which causes 1.1 million deaths of cattle per year and costs over 168 million US dollars per annum, is a major problem in 11 countries in sub-Saharan, East and Central Africa. *In vivo* and *in vitro* feeding methods were used to study the role of lectins, enzymes, hormones and blood-meal components on the development and transmission of *T. parva*. The potential role of these factors is discussed in relation to the current methods for the control of theileriosis. The control of East Coast fever, like many other vector-borne diseases, is based on elimination of the tick vector and the parasite within the mammalian host. Currently, an integrated approach is being sought that encompasses tick control by acaricides, biological control, infection and treatment, chemotherapy and breeding of resistant breeds of cattle, which is expected to reduce the disease incidences to below the economic threshold. The high cost of acaricides, tick resistance, environmental pollution and levels of water in arid and semi-arid regions have all rendered the use of acaricides unpopular. Early, rapid and accurate diagnosis is a prerequisite to effective chemotherapy, since none of the drugs are effective when respiratory signs have developed in East Coast fever. The extreme cost of chemotherapy (\$40) per head has led to limited use of this method. An infection and treatment method that included administering a single dose of oxytetracycline at the time of infection with sporozoite stabilate results in the development of

immunity in cattle and is currently being used in East and Central Africa. Vaccination against theileriosis is of high priority in the control of this disease, but as yet no effective vaccine has been developed. The search for new antigens continues. The study had demonstrated that lectins should be investigated further as potential immunogens in blocking the transmission of *T. parva* as a new method of controlling theileriosis.

Severe manifestations of rickettsial disease in children

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The spotted fever group (SFG) rickettsiae are obligate Gram-negative bacteria that are transmitted to their vertebrate hosts by ticks. Infections with SFG rickettsiae are very prevalent in sub-Saharan Africa and there are now known to be at least 2 pathogenic SFG rickettsiae on the continent. African tick-bite fever (ATBF) is caused by *R. africae* which is transmitted to man by *Amblyomma* spp., ticks of domestic ruminants and wild ungulates. Mediterranean spotted fever (MSF) is caused by *R. conorii*, which is transmitted to humans by various dog ticks. Complications due to tick-bite fever (TBF) in South Africa are considered to be rare, especially in children. There are no published reports of severe irreversible complications in children in South Africa. Recently we treated a number of children with severe, complicated rickettsial disease. Most of these children did not present with classical symptoms of TBF, but with cerebral, cardiac or pulmonary disease characterised by signs of encephalopathy, cardiomyopathy, interstitial pneumonia or pulmonary vascular disease. Morbidity and mortality in these cases were high. Diagnosis was made with routine serological tests. In a number of patients we could confirm the serological diagnosis with immunofluorescent antibody staining or polymerase chain reaction of biopsy- or post-mortem material. We conclude that in our area SFG rickettsiae can cause severe complications in childhood. The impression is that these complications are not uncommon and therefore a high index of suspicion should be maintained.

Preliminary report on myxosporean parasites infecting fishes in the Okavango River and Delta, Botswana

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The phylum Myxozoa Grassé, 1960 comprises an immensely diverse and intricate group of spore-forming obligatory parasites. Over 1300 known species are known throughout the world. Some myxosporeans have an undeniable pathogenic effect and can weaken or even kill the hosts they parasitise. The present work on myxosporeans parasitising fish in Botswana is the first recent study to be undertaken in southern Africa and the first to be initiated in Botswana. During 2 extensive field trips to the Okavango River system in June and July 1998 and 1999, 275 fish representing 31 species of 9 families were examined. Fishes were collected using a series of gill nets as well as small hand nets. The live fish were taken to a fully-equipped field laboratory, where they were kept alive in aerated aquaria. The fishes were subsequently examined for the presence of myxosporean parasites and any myxosporeans found were fixed using standard techniques for the group. Five myxosporean species from the genus *Henneguya* Thélohan, 1892, were found parasitising 5 different fish hosts, while 8 myxosporeans from the genus *Myxobolus* Bütschli, 1882, were found parasitising 9 different host fish species.

Cloning of a *Haemonchus contortus* P-glycoprotein gene (PGP-A) promoter region

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Helminth infections are a major constraint for livestock production in many parts of the world. Control of these infections relies mostly on the use of anthelmintics. However, resistance against anthelmintics has severely limited their effectiveness in the control

of helminthoses. P-glycoproteins (Pgp) are transmembrane proteins associated with acquired multi-drug resistance in mammalian cells and some protozoan parasites by a process of active drug export. There is evidence of *Pgp* genes in nematodes, both in the free living *C. elegans* and in the sheep parasite *Haemonchus contortus*. The findings of over-expression of P-glycoprotein gene in anthelmintic-resistant parasites forms the basis of our research, which focuses on studying the regulation of expression of this gene. A homologue of *Pgp* gene, *Pgp-A*, has been isolated and shown to be over-expressed in anthelmintic-resistant *H. contortus*. Since over-expression of genes can be a result of gene amplification or changes at the promoter region of the gene, we are attempting to map out the *Pgp-A* promoter region in *H. contortus*. An approximately 1 kb fragment from the 5' end of the *Pgp-A* gene has been amplified from the *H. contortus* cDNA obtained by reverse-transcription polymerase chain reaction (RT-PCR) and sequenced. The sequence is yet to be analysed for the presence of a promoter.

Further information on the life cycle stages of *Haemogregarina bigemina* Laveran & Mesnil, 1901, in South African hosts

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Over the past 3 years an in-depth study of the blood parasite *Haemogregarina bigemina* Laveran & Mesnil, 1901 (Protozoa; Apicomplexa), which is parasitic in some South African marine intertidal fishes, was undertaken. Host fish (clinids and rocksuckers) were captured in rock pools at De Hoop Nature Reserve during April 1998 and 2000, and at Jeffreys Bay in January 1999. All of these fishes were found to be parasitised by *H. bigemina* as well as by ectoparasitic gnathiid isopods, *Gnathia africana* Barnard, 1914 (Isopoda; Gnathiidae). In order to investigate the possible presence of blood protozoan life stages in the fish hosts as well as its ectoparasites, captured fish were maintained in buckets of fresh, aerated seawater until the gnathiids had fed to repletion. Fishes were then measured and blood smears were prepared and screened for *H. bigemina*. Fully-fed gnathiids were kept in seawater at between 18–22 °C for periods up to 28 days post feeding (d.p.f.). Gnathiid larvae were then smeared and screened for stages of *H. bigemina*, either immediately or on each of 1–28 d.p.f. A great variety of development stages, presumed to be those of *H. bigemina*, were found in the gut of some of these gnathiid larvae that had fed on fish infected with *H. bigemina*. This study presents strong evidence that gnathiids can act as vectors of fish haemogregarines.

Morphology of twisted-wing parasites (Strepsiptera: Halictophagidae)

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The twisted-wing parasites constitute an unusual order of insects with respect to their morphology and biology. Their taxonomic position is controversial. While some authors place them in the order Coleoptera, recent molecular studies indicate that this group is more closely related to the Diptera. The Strepsiptera are comprised of 10 families that are all highly host-specific. The present study focused on the family Halictophagidae, which parasitises leafhoppers (Homoptera: Cicadellidae). Material was collected by sweeping grass with a sweepnet and selecting the parasitised leafhoppers. A head of the parasite protruding from the host's abdomen indicates infection. Pupating males were dissected out, critical-point dried, and examined in a Jeol 6400 scanning-electron microscope. The aim of this study was to study the morphology of the parasite, as well as the host. Sexual dimorphism was apparent. The female lacks antennae and legs, and never leaves the host. Males upon emerging are free-living, have a life span of only a few hours, raspberry-like protruding eyes, and rudimentary front wings resembling the club-shaped halteres of dipterans. The hind wings are fan-shaped with a few reduced veins.

Polystomatid sclerite morphology as a taxonomic character

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Polystomatid flatworms (Monogenea: Polystomatidae) are soft-bodied organisms with hooks and suckers for attachment. High intraspecific and low interspecific variation as well as limited morphological characters complicates identification for these parasites. The only stable body parts not subject to deformation are the marginal hooks and hamuli. These hooks show variation between genera and species. The aim of this study was to characterise these marginal hooks by means of digital imaging in order to evaluate the taxonomic value of these sclerites. This was done by a process of Fourier transformation. Parasites were cleared in a Bouin-glycerin mixture and mounted temporarily. Cleared parasites were studied on a Nikon E800 compound microscope, and photographs were taken with the camera fitted to the microscope. The micrographs were scanned and stored on the computer. Images were mathematically digitised in Matlab. Fourier coefficients were calculated from the digitised points, in order to remove the rotation and specific position of the hooks. The Fourier coefficients for each image were correlated with measurements of the hooks. Results from the present study could be beneficial for the development of a protocol to be followed when new polystomatid parasites are described.

An unusual argulid from the northern coast of KwaZulu-Natal, South Africa

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During fish parasitological surveys along the coast of northern KwaZulu-Natal, male and female specimens of an argulid were found, which at first sight resembled a branchiuran of the genus *Chonopeltis*. Upon closer examination of the material collected from the kelee shad, features such as the antennae and antennule, pre-oral spine, mouth tube and the second maxilla typical of the genus *Argulus* were noted. The carapace of this argulid is small and trifoliate, and the thorax and abdomen are elongated. The specimens were described as a new species, *Argulus izintwala* Van As & Van As, 2000. *Argulus izintwala* shows convergent similarities to the genus *Chonopeltis*, not only morphologically, but also on host specificity. All 14 known species of the genus *Chonopeltis* are host-specific. During the survey in KwaZulu-Natal, specimens of *A. kosus* Avenant-Oldewage, 1994, were also collected, but from 8 different fish species. In total, 14 different fish species were examined for all

types of external fish parasites, and *Argulus izintwala* was found only on *Hilsa kelee* (Cuvier).

Life cycle and morphology of *Xenopacarus africanus* (Acari: Ereyinetidae)

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Xenopacarus africanus belongs to the subfamily Lawrencarinae that contains all acarine species parasitic in the nasal cavities of amphibians. *Xenopacarus* is parasitic in the nasal cavities of *Xenopus laevis*. The biology of *Xenopacarus* is not well known, and no SEM studies have been conducted on the morphology. This study aimed to clarify biological aspects such as the life cycle and seasonal fluctuation, as well as morphological characters by SEM. Monthly dissections were made and numbers and positions of larvae, nymphs and adults were made. Several experimental studies were conducted to determine the life cycle and method of infection. It was found that the larvae leave the host when it is resting at the surface of the water. Parasites then travel on the water surface in search of a new host. They enter the new host through the nasal openings where they remain until they are in the nymph stage. They then move to the Eustachian passages and remain here until adult. The SEM studies revealed that the mites display typical acarine morphology, with an ereynetal organ that is unique to the Ereyinetidae.

Seasonal infection of *Protopolystoma xenopodis* (Monogenea: Polystomatidae) and the relationship with its anuran host, *Xenopus laevis*

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Infection levels and egg production of *Protopolystoma xenopodis* were monitored over a 12-month period. The study was performed on a natural population of the African clawed frog, *Xenopus laevis* (Daudin 1803). A freeze-branding technique was used to number the frogs to recognise recaptures, which enabled monitoring of the infection history of individual frogs. Harris-hematoxylin-stained cross-sections of phalanges were used to age the frogs. The mean prevalence of *P. xenopodis* for the 12 months was 39 % and the mean rate of egg production, 7.6 eggs/host/24 h. Longevity of the hosts was determined at 4 years. Frogs are exposed to infection throughout their life span and infection does not seem to be related to host age. Environmental changes and host breeding season together play an important role in parasite transmission.