



Review

A review of parasitic zoonoses in a changing Southeast Asia

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ABSTRACT

Parasitic zoonoses are common and widely distributed in the Southeast Asian region. However, the interactions between parasites, hosts and vectors are influenced by environmental, socio-cultural and livestock production changes that impact on the distribution, prevalence and severity of disease. In this review we provide an update on new knowledge in the context of ongoing changes for the food-borne pig associated zoonoses *Taenia solium* and *Trichinella* spp., the food-borne trematodes *Opisthorchis viverrini* and *Clonorchis sinensis*, the water-borne trematodes *Schistosoma* spp., the vector-borne zoonotic protozoa *Plasmodium knowlesi* and *Leishmania* spp. and the soil-borne zoonotic hookworm *Ancylostoma ceylanicum*. These various changes need to be considered when assessing or developing regional control programs or devising new research initiatives in a changing SE Asia.

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1. General introduction

The zoonotic parasites circulating in Southeast (SE) Asia are a significant burden on human health and wellbeing and there are multiple transmission pathways that place people at risk. Here we discuss the food-borne pig associated helminths *Taenia solium* and *Trichinella* spp.; the small food-borne trematodes *Opisthorchis viverrini* and *Clonorchis sinensis*; the water-borne trematodes belonging to the genus *Schistosoma*; the vector-borne protozoa *Plasmodium knowlesi* and *Leishmania* spp. and the soil-borne zoonotic hookworm *Ancylostoma ceylanicum*. All but *P. knowlesi* and trichinellosis have recently been designated neglected tropical diseases (NTDs) by the World Health Organisation (WHO, 2010). Worldwide, NTDs predominantly affect the poor with more than 40 million people infected and 750 million at risk (Keiser and Utzinger, 2005; Hotez et al., 2008), furthermore zoonotic neglected diseases make a significant contribution to the entrenchment of poverty in poor rural communities who derive income from livestock production (WHO, 2010). Vector-borne protozoan pathogens cause relatively few public health problems in SE Asia in comparison to Latin America and Africa, however, the recent discovery of a simian malaria parasite, *P. knowlesi*, infecting humans has reawakened interest, as this may have been an undetected cause of disease for many years in people who derive their living from the forest.

Southeast Asia is currently under going changes with respect to climate change, environmental degradation, deforestation and river basin management, socio-economic development and the industrialisation of livestock production. These complex ecological changes have the potential to modify the interactions between hosts, vectors and parasites and these altered interactions impact on the distribution, prevalence and severity of disease. In this review we provide an update of new knowledge in the context of ecological changes in SE Asia, and we briefly discuss the implications for the design and implementation of control programs or research initiatives.

2. Food-borne pig associated zoonotic helminthiasis

The traditional practice of consuming uncooked or partially cooked meat in some SE Asian nations places many people at risk of acquiring food-borne parasitic zoonoses, particularly *T. solium* and members of the genus *Trichinella*. Many of the changes currently taking place in SE Asia have

the potential to directly impact on the transmission of these medically important parasites to pigs and by extension to people.

2.1. *Taeniasis and cysticercosis* lifecycle, epidemiology and distribution in the region

The *T. solium* taeniasis and cysticercosis infection complex involves two distinct disease transmission processes and requires both humans and pigs to maintain the lifecycle. Humans are the definitive host, acquiring the adult tapeworm (taeniasis) following ingestion of viable larvae (cysticerci) in contaminated pork. Eggs are shed into the environment by the adult worm via faeces; pigs become infected following ingestion of contaminated feed or water or through direct coprophagia, thus completing the lifecycle. *T. solium* has public health significance because humans can also be inadvertently infected with cysticerci following the ingestion of eggs through poor hygiene or contaminated food and water. Human cysticercosis cases are not involved in perpetuating the lifecycle but are clinically important since cysticerci may form in the brain causing neurocysticercosis, leading to seizures, epilepsy, neurological sequelae or death. Taeniasis and cysticercosis caused by *T. solium* has been the subject of a number of recently published reviews with an Asian focus (Ito et al., 2003; Rajshekhar et al., 2003; Willingham et al., 2003, 2010; Dorny et al., 2004; Wandra et al., 2007; Conlan et al., 2008, 2009). Perhaps the most consistent underlying element of these reviews is the distinct lack of high quality data from community level studies describing the epidemiology and distribution of *T. solium* in SE Asia. We will not replicate these reviews here; rather we seek to provide an update of new knowledge in the context of the changes taking place in much of SE Asia.

The distribution and epidemiology of *T. solium* in Thailand, Vietnam, Laos and Cambodia are described in detail by Willingham et al. (2010) and the distribution of *T. asiatica* in SE Asia has recently been described by Eom et al. (2009). The most recent data comes from Laos where surveys were conducted in 24 village communities in four northern provinces and among pigs at slaughter. Human cysticercosis prevalence was determined to be 2.2% by antigen capture ELISA and there was strong evidence of a focal distribution with just over half of the cases detected residing in three villages in Oudomxay province (Conlan et al., in preparation). No significant risk factors for cysticercosis were found, and although infection was rare, the highest prevalence was observed in people of the Mon-Khmer

ethnic group, the poorest households and people from Oudomxay province. The prevalence of taeniasis was estimated to be 8.4% (110/1306) based on copro-microscopy and self-reported proglottids in stool; prevalence ranged from zero to 17% at the village level and was significantly associated with consumption of uncooked beef (*laap ngeua*), age, gender, province and ethnicity. *T. saginata* was the most frequently detected tapeworm, 94% (33/55) and 6% (2/35) of recovered worms were identified by PCR as *T. saginata* and *T. solium*, respectively. In Lao pigs, *T. solium* cysts were infrequently detected, 0.8% (5/590) of pigs at slaughter had visible cysts and all were heavy infections. *T. hydatigena* cysts were detected in 22.4% of pigs (132/590) and *T. asiatica* cysts were detected in 0.2% (1/590). Seroprevalence of swine cysticercosis by antigen ELISA was 68.5% (404/590) and was disproportionate to the prevalence of all *Taenia* cysts detected by inspection. All but three of the inspection positive animals (three *T. hydatigena* positive pigs) were reactive in the ELISA; thereby providing evidence that the inspection results were specific, but not at all sensitive. Intestinal helminth studies in the Laos rarely describe the species of *Taenia* detected in stool (Conlan et al., 2008), however *Taenia* tapeworms were partially identified to the species level in three recent studies. In a study on *Opisthorchis viverrini* infection (Sayasone et al., 2009) in southern Laos, 23 tapeworms were recovered following praziquantel treatment and 18 were identified as *T. saginata* and the remaining five were not identified to the species level. The methods used by Sayasone et al. (2009) to characterise the tapeworms were not reported. A similar study in Khammuane province in central Laos recovered 15 worms from 12 patients and all were morphologically identified as *T. saginata* (Chai et al., 2009). *T. asiatica* can be misclassified as *T. saginata* in the absence of molecular confirmation (Anantaphruti et al., 2007) and caution should be exercised in interpreting data from the studies of Sayasone et al. (2009) and Chai et al. (2009) without the exclusion of *T. asiatica* from the differential diagnosis. The third study, conducted nationally, reported an overall taeniasis prevalence of 1.1% (408/37,090) from which 120 tapeworms were genetically and morphologically typed; three *T. solium* cases were identified from Luangprabang province in northern Laos (Eom et al., 2009). To date, *T. solium* has only been reported in the north of the country whereas *T. saginata* has a national distribution, *T. asiatica* taeniasis has not yet been detected in Laos.

T. solium taeniasis and cysticercosis have been confirmed in two regions of the Indonesian archipelago (see Willingham et al., 2010). In Bali, *T. solium* is endemic but transmission and prevalence seems to be on the decline (Wandra et al., 2006, 2007; Sudewi et al., 2008). In Papua, *T. solium* appears to be hyperendemic in at least two districts where seroprevalence levels in the human population are some of the highest in the world and endemic in a further two districts (Salim et al., 2009). No other human *Taenia* species are known to be co-endemic in Papua (Wandra et al., 2007). North Sumatra, Flores, Sulawesi and other regions are often reported in the literature to be endemic for *T. solium*, but there are no verifiable contemporary published data to support this assertion. *T. solium* taeniasis in humans or cysticercosis in pigs has not been reported

from North Sumatra; rather, evidence indicates the area is endemic for *T. asiatica* (Wandra et al., 2006, 2007) although this seems to be on the decline (Ito et al., 2003). In Flores, published literature (Simanjuntak et al., 1997) does not provide evidence of *T. solium* endemicity. The most reliable and conclusive data come from studies conducted in Bali and Papua (Wandra et al., 2007; Sudewi et al., 2008; Salim et al., 2009). There is evidence, albeit limited, that *T. solium* is present in Timor-Leste and Indonesian West Timor based on reports of suspected cases by district health officials (see Willingham et al., 2010) and a case report of several *T. solium* worms being extracted from a patient presenting with a perforated intestine after blunt trauma to the abdomen (Abu-Salem and Hassan, 2003). Further studies of the human and pig populations are required to understand better the epidemiology of *Taenia* spp. on Timor Island.

Limited data are available for taeniasis and cysticercosis in countries such as Malaysia and the Philippines although evidence presented by Willingham et al. (2010) indicates endemicity. Sporadic human neurocysticercosis cases are infrequently observed in Malaysia (Arasu et al., 2005; Nor Zainura et al., 2005) and typically detected in migrant workers (Arasu et al., 2005). However, a recent survey of 135 people from a single rural village in Ranau district, Sabah, East Malaysia found 2.2% seroprevalence for antibodies against cysticercosis (Noor Azian et al., 2006). These authors used a cut-off calculated as the mean of the 135 serum samples tested plus three standard deviations rather than a more robust use of a panel of negative control sera. It is not clear why this 'arbitrary' cut-off was used and as such Noor Azian et al. (2006) may have underestimated the seroprevalence in the Ranau community by at least four-fold. A conservative estimate based on the data presented in the Noor Azian et al. (2006) study indicates that seroprevalence could be greater than 10% in Ranau village. It is difficult to draw conclusions from this study, but *T. solium* cysticercosis in non-Muslim indigenous communities of Malaysia may be an unrecognised problem. To date, no surveys of swine cysticercosis or human taeniasis have been reported in the scientific literature in Malaysia. In the Philippines, *T. solium* cysticercosis has been detected in swine (see Martinez-Hernandez et al., 2009) and a single seroprevalence survey for human cysticercosis found that 24.6% of the Macanip community in Eastern Visayas had antibodies. As with other regions in SE Asia, human cysticercosis may cluster in poor, remote communities of Malaysia and the Philippines.

In addition to *T. solium*, two other taeniid species cause human taeniasis in SE Asia. *T. saginata* and *T. asiatica*, which are associated with bovines and pigs as intermediate hosts, respectively, are also prevalent in the region with variable distribution (see Ito et al., 2003; Eom et al., 2009). Neither *T. saginata* or *T. asiatica* are associated with human cysticercosis, but they could potentially influence the transmission dynamics of *T. solium* through competitive mechanisms associated with crowding in the human gut (Conlan et al., 2009). In addition to *T. solium* and *T. asiatica*, pigs are also the intermediate host for the dog tapeworm *T. hydatigena* and through immune-mediated processes in the intermediate host this canine taeniid may limit the reproductive

potential of related species, including *T. solium* (Conlan et al., 2009).

Kanchanaburi province in western Thailand appears to be the only locality where the sympatric occurrence of all three human *Taenia* species has been definitively established in a single geographically restricted area (Anantaphruti et al., 2007, 2010). All three human *Taenia* species are endemic in the vast Indonesian archipelago (Wandra et al., 2007) but there appears to be geographic partitioning of the three tapeworms. *T. asiatica* has been reported from Bali (Simanjuntak et al., 1997), but there are no contemporary data to verify this assertion and recent reviews indicate that only *T. saginata* and *T. solium* are endemic (Wandra et al., 2006, 2007). A hospital based study in Vietnam detected all three species (Somers et al., 2007), but it is not clear if this constituted sympatric occurrence or if the patients were from geographically distinct areas. Likewise, in the Philippines all three human *Taenia* worms have been detected (Eom et al., 2009; Martinez-Hernandez et al., 2009) but sympatric distribution cannot be determined from the limited data.

The co-distribution of canine *Taenia* is difficult to determine since there is scarce literature on *T. hydatigena* infecting pigs or dogs in SE Asia. As far as we are aware, *T. hydatigena* has only been reported in pigs in Vietnam (Willingham et al., 2003) and Laos (Conlan et al., in preparation) and that four *Taenia* species of humans, dogs, pigs and bovines are co-endemic in both countries, and are likely to occur sympatrically. Conlan et al. (in preparation) observed that in this multi-species co-endemic environment, one *Taenia* species predominated in the human host and one in the pig host. *T. saginata* was the predominant adult-stage worm infecting people in northern Laos and *T. hydatigena* accounted for the majority of cysts detected in pigs at slaughter (Conlan et al., in preparation). These authors used a simple maximum likelihood estimator to predict true prevalence in pigs and estimated 56% were infected with *T. hydatigena* in comparison to 4% and 1% of pigs infected with *T. solium* and *T. asiatica*, respectively (Conlan et al., in preparation). The results from Laos provide indirect evidence that immune-mediated competitive mechanisms in the intermediate host may suppress the transmission potential of *T. solium*. Consumption of uncooked beef in Laos was highly prevalent (Conlan et al., in preparation) and was probably the strongest factor controlling human taeniasis; this in turn reduced the infection pressure of *T. solium* on pigs. In contrast to the Lao study, there was no clearly dominant species in west Thailand where three human *Taenia* species are co-endemic (Anantaphruti et al., 2007, 2010), providing evidence that crowding in the definitive host may not be a density-dependent constraint. At this stage there are no published data on *T. solium*, *T. asiatica* or *T. hydatigena* prevalence in the pig population in this endemic region of west Thailand. From the human data though, it seems *T. solium* and *T. asiatica* co-exist in the pig population in Kanchanaburi province without immune-mediated competitive interference. Research is needed to understand the immune-mediated interactions of related *Taenia* species in pigs as was undertaken for ovine cysticercosis more than 30 years ago in New Zealand (Gemmell et al., 1987).

2.2. Trichinellosis epidemiology and distribution in the region

Trichinellosis is a direct zoonosis caused by infection with nematodes of the genus *Trichinella* and is one of the most widely distributed parasitic zoonoses worldwide (Dupouy-Camet, 2000; Pozio and Murrell, 2006). Infection occurs via the consumption of encysted larvae in the muscle of infected animals and involves an enteral phase associated with excystment, sexual maturation, reproduction and larval penetration of the intestinal wall and a parenteral phase associated with the migration of larvae, via lymphatic and blood vessels, to striated muscles where they encyst in a nurse cell complex. Clinical symptoms in humans are related to the number of viable larvae consumed and are typically associated with the parenteral phase (Dupouy-Camet et al., 2002). Humans are a dead-end host and not involved in perpetuating the lifecycle.

Three species of *Trichinella* have been documented in the SE Asian region, the encapsulated *T. spiralis* and the non-encapsulated *T. pseudospiralis* and *T. papuae*, and all have been associated with human disease (Pozio et al., 2009). *T. spiralis* has a regional distribution (Pozio, 2001) with the majority of outbreaks recorded in the ethnically diverse regions of central and northern Laos, northern Thailand and northwest Vietnam where consumption of uncooked pork is common (Barennes et al., 2008; Kaewpitoon et al., 2008; Taylor et al., 2009). Recent outbreaks of *T. papuae* originating from wild pigs in Thailand (Khumjui et al., 2008; Kusolsuk et al., 2010) together with cases from Papua New Guinea (PNG) (Pozio et al., 1999, 2004) suggests the geographic range of this sylvatic species encompasses continental SE Asia and all the main islands to PNG (Kusolsuk et al., 2010). *T. pseudospiralis* was detected in southern Thailand where villagers were infected after consuming wild pig meat in 1994/1995 (Jongwutiwes et al., 1998).

Data on trichinellosis of wildlife and domestic animals in SE Asia are scarce. Surveys of pigs in SE Asia, specifically addressing trichinellosis prevalence and burden of infection, are limited and contemporary data are documented in two small research studies in Vietnam and Laos. In Son La province in northwest Vietnam, an outbreak of human trichinellosis occurred in 2008 (Taylor et al., 2009) leading to an investigation of swine trichinellosis from December 2008 to April 2009 (Vu Thi et al., 2010). Vu Thi et al. (2010) found almost one fifth of pigs in the survey area had serological evidence of *Trichinella* infection as determined by the excretory–secretory (ES)-ELISA and 15% of these serologically reactive animals had evidence of muscle larvae. *T. spiralis* was the only species detected and the muscle burden ranged from 0.04 to 0.38 larvae per gram (lpg) of muscle (Vu Thi et al., 2010), indicating a relatively low burden of infection but still posing a risk for human disease. The disproportionate serological and muscle digestion results in this study were interpreted as being due to low sensitivity of muscle digestion or lack of ES-ELISA specificity (Vu Thi et al., 2010). Since 50 grams of muscle per animal was digested, it seems reasonable to assume that poor test specificity was the strongest controlling factor in a study environment where polyparasitism in the pig population is common. In Laos, a recent study of swine

trichinellosis was conducted in four northern provinces, three bordering Vietnam in the northeast and one sharing a border with China in the north (Conlan et al., in preparation). Muscle digestion of tongue and diaphragm was the only method used and less than 2% of slaughter pigs were infected, ranging from zero to 4% for the four provinces. A subset of larvae were speciated and all identified as *T. spiralis*. Ten animals had 0.1–0.9 lpg, three animals had 1–10 lpg and two animals had >10 lpg, the highest recorded burden of infection was 69 lpg (Conlan et al., in preparation). A slaughterhouse survey in Cambodia in 2005 found a very low seroprevalence of swine trichinellosis (1.13%, 5/440) and there was no difference between intensively produced and free-range pigs (Sovyra, T., unpublished thesis, Chiang Mai University, Thailand and Frei University, Berlin, Germany).

The majority of reports of trichinellosis arise from outbreaks in human populations (Pozio, 2007) and for the most part these have been discussed in detail elsewhere (Pozio, 2001, 2007; Kaewpitoon et al., 2008; Odermatt et al., 2010). Community level surveys of trichinellosis in SE Asia specifically addressing prevalence and risk factors of exposure to this food-borne nematode are scarce. In part this is a consequence of the difficulty of interpreting serological data based on the ES-ELISA and the excessive cost of western blot analysis. However, a recent study has sought to investigate human trichinellosis at the community level in four provinces in northern Laos (Conlan et al., in preparation), including Oudomxay province where an outbreak in 2005 affected more than 600 people (Barnes et al., 2008). Almost one fifth of the survey population had antibodies to *Trichinella* detected by the ES-ELISA. Antibody positivity was significantly associated with consumption of uncooked pork (particularly fermented pork sausage, “*Som moo*”), increasing age, increasing wealth status and ethnicity; people of the ethnic minority Hmong and Mon-Khmer groups were less likely to have ES-ELISA reactive sera compared to people from the majority Lao-Tai ethnic group. The consumption of uncooked fermented pork was most common in the Lao-Tai ethnic group and increased with increasing age with almost 50% of people aged 25–54 years reporting consumption of fermented pork. The interpretation of the serological data presented problems since many of a subset of the ES-ELISA positive sera were negative by western blot analysis and may have represented poor specificity and false positives. False positive results have been associated with polyparasitism and infections with other nematodes (Gomez-Morales et al., 2008) and these were common in the Lao study population (Conlan et al., in preparation), indicating trichinellosis seroprevalence may have been overestimated.

Even with an apparent decline in the number of outbreaks in northern Thailand (Kaewpitoon et al., 2008) and an apparent increase in northwest Vietnam (Taylor et al., 2009), there is insufficient evidence to suggest that trichinellosis is emerging or re-emerging in the SE Asian region. The evidence to date indicates that trichinellosis may be endemically stable. The minimum number of larvae required to cause clinical disease has been estimated to be between 70 and 150 larvae (Dupouy-Camet et al., 2002) and in Laos the volume of fermented sausage consumed in

a sitting is most often less than 50 grams (Conlan et al., in preparation). The prevalence of *T. spiralis* larvae in backyard and free-range pigs is relatively low and the majority harbour a low worm burden (<1 lpg) (Vu Thi et al., 2010) (Conlan et al., in preparation) suggesting that in a community where uncooked pork is consumed, most infections will be subclinical. Severe clinical cases predominantly occur as sporadic point source outbreaks or sporadic isolated cases (Odermatt et al., 2010). Trichinellosis endemic stability requires verification by well-designed and comprehensive epidemiological studies of pigs and people but it could provide important insights for the implementation of disease control initiatives.

2.3. Southeast Asia's livestock revolution

Southeast Asia is currently in the midst of a livestock revolution driven by a high demand for animal derived protein, to meet this demand livestock production has increased in terms of absolute numbers, but most dramatically in production output. Official pig production data published by the United Nations Food and Agricultural Organisation (FAO) clearly demonstrates this trend (FAO, 2010a,b); in the 11 ASEAN nations, the number of pigs produced in 1998 rose from 53.9 million to 69.4 million in 2008, representing an increase of 28.7%. Whereas, the volume of pork produced in the same period rose from 4 million tonnes to 6.4 million tonnes, representing an increase of 58.9%. The numbers are even more telling for the three countries producing the most pork, Vietnam, Philippines and Thailand, where pig numbers have increased by 34% while pork output has increased by 75.1% (FAO, 2010a,b).

Increased productivity has principally been achieved by improved pig production practices, including the introduction of new genetics, better feeding, improved husbandry and management and disease control. Thai pig production is firmly entrenched in the intensive commercial sector using Western pig breeds (Kunavongkrit and Heard, 2000). After the introduction of commercial breeds in the 1960s (FAO, 2002) the intensive commercial sector grew rapidly and accounted for 80% of all Thai pig production by 2000 (Kunavongkrit and Heard, 2000). However, increased productivity has not been achieved by intensification of the industry across the region; in Vietnam the majority of pigs are still produced by smallholder farmers but intensification of the industry continues (Tisdell, 2009). Western breed pigs are ill suited to harsh free-range systems and are not common in remote mountainous areas (Dang-Nguyen et al., 2010). Indigenous breed pigs in Thailand and Vietnam are mostly produced in remote, mountainous ethnic minority communities but they now make up only a small portion of the genetic pool in the respective countries (Charoensook et al., 2009; Dang-Nguyen et al., 2010). In other Mekong countries where trichinellosis and *T. solium* are also a concern, the change has not been so great. For the period 1998–2008, pork output in Laos rose by 77% being mirrored by a 71% increase in the number of pigs produced, a similar situation was observed in Cambodia where pork production increased by 10% while the pig population declined by 5% (FAO, 2010a,b). Western pig breeds have

increased in number over the past 10 years in these two Mekong countries, however the majority of production still takes place in the smallholder-backyard farm sector using 'indigenous' breeds.

The dramatic changes to pig production in Vietnam and Thailand over the past 20 years have important implications for zoonotic parasite transmission. In Thailand, outbreaks of trichinellosis and *T. solium* taeniasis and cysticercosis are concentrated in remote areas populated by ethnic minority groups who practice traditional pig production methods and consume uncooked or improperly cooked pork (Waikagul et al., 2006; Anantaphruti et al., 2007, 2010; Kaewpitoon et al., 2008). In the 30-year period 1962–1991 there were 118 documented outbreaks of trichinellosis involving 5400 patients, averaging four outbreaks and 180 patients per year (Khamboonruang, 1991). In contrast, in the 16-year period 1991–2006 there were 17 outbreaks involving 1920 patients, averaging one outbreak and 121 patients per year (Kaewpitoon et al., 2008). Significantly, many of the outbreaks since 1991 have been attributed to the sylvatic cycle after consuming wild pig meat (Kaewpitoon et al., 2008; Khumjui et al., 2008; Kusolsuk et al., 2010), indicating that human trichinellosis contracted via domestic pigs is now rare. Likewise, *T. solium* taeniasis and cysticercosis cases in Thailand are rare in the general population (Waikagul et al., 2006). There are insufficient longitudinal pig production and corresponding human disease data from other countries in the Mekong region to make similar comparisons, but we could reasonably expect incidence in human populations to decline with the continued expansion of improved pig husbandry practices.

3. Food-borne trematodes

Clinically important trematodes in SE Asia include *Opisthorchis viverrini*, *Clonorchis sinensis*, *Fasciola* spp. and *Paragonimus* spp. (Sripa et al., 2010) and the most prevalent trematodes with severe clinical complications are the small food-borne liver flukes, *O. viverrini* and *C. sinensis*. The complex life cycle of *O. viverrini* and *C. sinensis* includes two intermediate hosts, *Bithynia* snails and fish predominantly belonging to the family Cyprinidae. Final definitive hosts including humans, dogs and cats become infected after eating raw or inadequately cooked fish harbouring infective metacercariae (see Sripa et al. (2007) for a detailed description of the life cycle). The adult liver fluke infections induce several hepatobiliary diseases in humans including hepatomegaly, cholangitis, gallstones, and cholangiocarcinoma, a subtype of primary liver cancer arising from the bile ducts (Sripa et al., 2007; Sripa and Pairojkul, 2008). Liver fluke endemic countries, including Laos, Thailand and Vietnam, are among the top six countries worldwide with the highest incidence of liver cancer (Ferlay et al., 2010). Since food-borne parasitic zoonoses involve complex interactions between several diverse hosts control of these infections is difficult. With a paucity of parasite control campaigns in certain countries in the SE Asian region and possibly changes to the environment and climate, re-emergence of these food-borne diseases may be increasing, as is the case for other trematodes (Yang et al., 2005; Mas-

Coma et al., 2009). We highlight here the epidemiology, life cycle, climate and environmental changes and potential impact on emergence of opisthorchiasis and clonorchiasis.

3.1. *Opisthorchis* epidemiology and distribution in the region

The first human cases of *O. viverrini* infection were reported in Thailand, nearly 100 years ago (Leiper, 1915). Later, opisthorchiasis has been reported from other countries including Laos, Cambodia and Vietnam with sporadic case reports from Malaysia, Singapore and the Philippines (Sripa et al., 2010). Over 10 million people in Southeast Asia are infected with *O. viverrini* (Sripa et al., 2010). In Thailand, an early study reported a high prevalence of up to 100% in certain villages of Northeast Thailand (Sadun, 1955) and the first nationwide survey in 1980–1981 revealed an overall prevalence of *O. viverrini* infection of 14%; northeast (34.6%), central (6.3%), the north (5.6%) and the south (0.01%). Due to intensive and continuous control activities, the national prevalence of infection declined to 9.4% or about 6 million people infected in the year 2000 (Jongsuksuntigul and Imsomboon, 2003). However, contemporary data indicates local endemic areas in northeast Thailand still have high prevalence approaching 70%, possibly due to re-emergence (Sripa, 2008).

In Laos, it is estimated that between 1.5 and 2 million people are infected with *O. viverrini* (WHO, 2008), representing almost a third of the Lao human population. A nationwide survey of 29,846 primary schoolchildren from 17 provinces and Vientiane Municipality showed an average prevalence of *O. viverrini* infection of 10.9% with high prevalence in Khammuane, Saravane and Savannakhet province (32.2%, 21.5% and 25.9%, respectively) (Rim et al., 2003). A recent survey in Saravane district revealed *O. viverrini* prevalence of 58.5% among 814 persons from 13 villages (Sayasone et al., 2007). There are few reports on *O. viverrini* infection in Cambodia. A small survey in primary schoolchildren from Kampongcham province demonstrated *O. viverrini* prevalence of 4.0% from 251 subjects (Lee et al., 2002). A high prevalence of opisthorchiasis (40%) was recently observed in the human population of Kratie province in northeastern Cambodia (Sinoun M., personal communication). The geographical range of *O. viverrini* extends to southern Cambodia with a recent survey detecting metacercariae in 10 species of freshwater cyprinoids with prevalence ranging from 2.1% to 66.7% of captured fish (Touch et al., 2009). In Vietnam, both human liver fluke infections have been reported; *C. sinensis* in the northern region and *O. viverrini* in the central and southern regions. *O. viverrini* prevalence in three endemic southern provinces range from 15.2% to 36.9% (De et al., 2003) and prevalence of up to 40% has been reported in six endemic districts in central Vietnam, namely Nui Thanh, Mo Duc, Mhu My, Song Cau, Tuy An and Buon Don (WHO, 2008).

3.2. *Clonorchis* epidemiology and distribution in the region

C. sinensis infection has been reported from many parts of east Asia; eastern Siberia, Japan, the Republic of

Korea, China, Taiwan and Vietnam. Sporadic cases have been reported in Malaysia, Singapore and the Philippines (IARC, 1994). In China, *C. sinensis* is endemic in southern and northeastern provinces, i.e. Guangxi, Guangdong, Heilongjiang, Jilin (Lun et al., 2005). In Southeast Asia, clonorchiasis has been reported in northern Vietnam. An epidemiological survey carried out in 12 of 61 provinces of Vietnam showed that *C. sinensis* was prevalent in nine northern provinces with prevalence ranging from 0.2% to 26.0%, mainly in the Red River delta region (De et al., 2003; WHO, 2008). A recent study in northern Vietnam showed human *C. sinensis* prevalence of 26% (Dang et al., 2008). However, the prevalence of *C. sinensis* infection in fish was quite low (1.9–5.1%) (Thu et al., 2007). *C. sinensis* cases have been detected by molecular diagnosis in central Thailand, indicating the geographic range may extend beyond current knowledge (Traub et al., 2009), but no other reports of clonorchiasis in SE Asia other than northern Vietnam have been published. More work is therefore required to confirm the geographic range and abundance of *C. sinensis* in other countries including northern Laos.

3.3. Ecology of *O. viverrini* and *C. sinensis*

Three species of *Bithynia* snails namely *Bithynia funiculata*, *B. siamensis siamensis* and *B. siamensis goniomphalos* have been reported as the first intermediate host for *O. viverrini* in Thailand (Brandt, 1974). Over 80 species of the Cyprinoid family, and at least 13 species of other families can serve as the second intermediate host (Komiya, 1966; Vichasri et al., 1982; Rim, 1986; WHO, 1995). The prevalence of liver fluke infection in snails is typically low, 0.05–1.6% (Harinasuta, 1969; Brockelman et al., 1986; Adam et al., 1993), whereas the prevalence in several species of cyprinoid fish may be as high as 90–95% (Harinasuta and Vajrasthira, 1960; Vichasri et al., 1982). The most common susceptible species of cyprinoid fish are in the genus of *Puntius*, *Cyclocheilichthys* and *Hampala* (Wykoff et al., 1965; Vichasri et al., 1982). The disproportion of prevalence of the liver fluke in snail hosts and fish reflects the effective infection and abundance of cercariae. The release of up to 1728 cercariae per day has been reported from one *O. viverrini* infected snail (Phongsasakulchoti et al., 2005). The intensity of liver fluke infection in fish varies by season, type of water body, species and individual (Vichasri et al., 1982; Rim, 1986; Sithithaworn et al., 1997). The number of metacercariae per fish generally ranges from one to hundreds. However, over 30,000 metacercariae per fish and more than 6000 metacercariae per gram have been reported in *Pseudorasbora parva* from China and Korea (Rim, 1986; Chen et al., 1994). In Thailand, metacercarial burdens peak in the dry winter months October to February and decreases in the summer and wet season months March to September (Vichasri et al., 1982; Sithithaworn et al., 1997). Snail populations also exhibit strong seasonal dependent variation, being highly abundant in the wet season (Brockelman et al., 1986). *Bithynia* spp. are distributed mainly in shallow, clear water reservoirs with a depth of less than 30 cm (Ngern-klun et al., 2006). The breeding grounds of snails are paddy fields and the environs of major river basins

and lakes. Therefore, wetland areas with small lakes along rivers are good habitats for snails and fish intermediate hosts and subsequent liver fluke transmission.

Transmission of *Opisthorchis* and *Clonorchis* among hosts is seasonal (Wykoff et al., 1965; Brockelman et al., 1986; Chen et al., 1994; Rim, 2005). In tropical countries, such as Thailand, it is likely that the peak time of faecal contamination of water reservoirs and snail infection occurs in the wet season through household drainage and open defecation during planting (Wykoff et al., 1965; Brockelman et al., 1986). Transmission of cercariae to fish and subsequently metacercariae to humans may be highest at the end of the wet season when snails are concentrated and fish are still abundant (Vichasri et al., 1982). Despite the complex host finding mechanism (Haas et al., 1990), the free swimming cercaria can locate appropriate species of fish in reservoirs. For *C. sinensis*, the shedding of cercariae from snails is governed by water temperature. Flukes may over winter as rediae in the snail host and erupt in spring, or new infections may re-establish each year from faecal contamination. In either case, peak transmission would occur in summer months (Rim, 1986).

Prevalence of liver fluke in reservoir hosts such as pigs, cats, and dogs, varies considerably by area (Scholz et al., 2003; Sithithaworn and Haswell-Elkins, 2003; Lin et al., 2005; Nguyen et al., 2009). A relatively high prevalence of liver fluke infection has been reported in cats (36.4%) and to a lesser extent in dogs (3.8%) in the Chi River basin of Northeast Thailand (Enes et al., 2010) where the prevalence of *O. viverrini* infection is high (Sripa, 2008). Similarly, high prevalence of *C. sinensis* in cats (70%), dogs (50%) and pigs (27%) correlates with human prevalence (31.6%) in southern China (Yu et al., 2003; Lin et al., 2005). Faecal contamination from infected animals undoubtedly contributes to transmission to snails in liver fluke endemic areas, particularly during flooding. Therefore, control of reservoir host transmission by anthelmintic treatment, concurrent with human treatment, is recommended to prevent re-emergence after liver fluke elimination in humans.

3.4. Environmental changes and potential impacts on FBT ecology and future control

Despite control campaigns over the past three decades in Thailand and Laos, food-borne zoonotic trematodes remain major health problems in the region. Recent evidence suggests that climate change may affect geographical distribution of certain parasitic diseases (Poulin, 2006; Yang et al., 2010). Reinfection or re-emergence is common due to the persistence of environmental risk factors including infected snails and fish intermediate hosts, reservoir hosts (cats and dogs) and humans. Transmission occurs in both natural habitats and in aquaculture ponds, and is most variable both geographically and temporally, with the variability related to climatic conditions. In SE Asia, climate change is a real phenomenon, causing more frequent intense events such as storms and flooding (ADB, 2009). Climate change is expected to have a significant effect on the food-borne zoonoses (Mas-Coma et al., 2009). More frequent extreme weather conditions, mainly heavy rainfall, can readily change the transmission pattern

through different mechanisms. For example, flooding can quickly change habitats affecting the density of intermediate snail host species, and transport infected snails to new areas. Moreover, runoff from human settlements and animal keeping areas can carry liver fluke eggs into snail habitats and thereby increase infection pressure on the first intermediate hosts.

Warmer conditions generally promote the transmission of parasites and raise their local abundance (Poulin, 2006). The Intergovernmental Panel on Climate Change (IPCC, 2007) reports an increasing trend in mean surface air temperature in Southeast Asia over the past several decades, with a 0.1–0.3 °C increase per decade recorded between 1951 and 2000. As mentioned earlier, all trematodes have complex life cycles and use snails as their first intermediate hosts. Asexual multiplication of the trematodes in snails produces a large number of infective cercariae. The cercarial production rate in snails is fundamental for overall parasite transmission success and this process is relatively temperature dependent, in that an increase in temperature is coupled with an increase in cercarial output (Lo and Lee, 1996; Umadevi and Madhavi, 1997; Mouritsen, 2002). Temperature-mediated changes in cercarial output also vary among trematode species, from small reductions to 200-fold increases in response to a 10 °C rise in temperature (Poulin, 2006). In addition, geographical latitude may also affect the production of cercariae by snails. Within the latitude range of 20–55°, trematodes from lower latitudes showed more pronounced temperature-driven increases in cercarial output than those from higher latitudes. The net outcome of increasing temperature will be a greater number of cercarial infective stages in aquatic habitats. A few reports have mentioned the effects of climate and environment changes on *O. viverrini* and *C. sinensis* emergence, albeit indirectly (Sithithaworn and Haswell-Elkins, 2003; Andrews et al., 2008). By the nature of their life cycle, it is possible that climate change in SE Asia, including intense rainfall and flooding and warmer temperatures, may enhance liver fluke abundance and transmission.

4. Schistosomiasis

Schistosomiasis is caused by infection with species of the blood-fluke *Schistosoma*. The life-cycle includes a single intermediate host, a freshwater snail, which for the endemic Chinese and Southeast Asian *Schistosoma*, is always of the family Pomatiopsidae. Three *Schistosoma* species are recognized as infecting humans in Southeast Asia, namely *S. japonicum*, *S. malayensis* and *S. mekongi*. Phylogenetically all three species belong to the *Schistosoma sinensium* clade (Attwood et al., 2008), so named because of the basal (ancestral) position of *S. sinensium* in the clade comprising these four *Schistosoma* species and *S. ovuncatum*. *S. sinensium* and its sister taxon *S. ovuncatum* are both transmitted by snails of the Triculinae and both are exclusively parasites of rodents; these two characters are regarded as plesiomorphic (ancestral) in Asian *Schistosoma* (see Davis, 1992; Attwood et al., 2002). These taxa have also been referred to as the “*Schistosoma japonicum*-group”

because all have a minutely spined egg as first described for *S. japonicum* (see Rollinson and Southgate, 1987).

4.1. *Schistosoma japonicum* epidemiology and distribution in the region

Schistosoma japonicum is often described as a “true zoonosis” (Ross et al., 1997; Gan et al., 2006; McGarvey et al., 2006), because 46 definitive host species representing 28 genera and 7 orders have been found to be naturally infected with *S. japonicum* (Ross et al., 2001; Shi et al., 2001; McGarvey et al., 2006); however, only one third of these species are thought to have a potentially significant role in transmission (Chen, 1993). Schistosomiasis japonica causes major public health problems in China and the Philippines (McGarvey et al., 2006) and *S. japonicum* has also been reported from a few isolated foci in central Sulawesi (Indonesia) where some human infections have been reported (Cross, 1976). Approximately 6.7 million people live in areas of endemic schistosomiasis japonica in the Philippines with 200,000 people estimated to be infected (Coutinho et al., 2006). In China, despite over 45 years of integrated control efforts, approximately one million people, and more than 1.7 million bovines and other mammals, are currently infected (Zhou et al., 2005). The resilience of schistosomiasis japonica to control efforts can be attributed to the existence of significant animal reservoirs of disease. Much work has been devoted to determining which species are involved as reservoirs for disease affecting the human population in endemic areas.

Although 19 species of Rodentia are found naturally infected in China and the Philippines, many of these (particularly the field rats) may not be epidemiologically significant. In the Philippines, prevalences of 85% and 56.5–95.5% have been recorded in natural populations of *Rattus norvegicus* and *R. rattus*, respectively, but in most cases the adult worms were found trapped in the lungs and few produced viable eggs (He et al., 2001). Studies in Anhui Province, China, estimated Relative Transmission Indices (RTIs) for different definitive host groups; these data, which combined estimates of prevalence, intensities of infection, and fecal production, to determine relative contributions to transmission, indicated that 89.8% of eggs originated from water buffalo (*Bubalus bubalus*), 5.4% from goats (*Capra hircus*), 4.4% from humans (*Homo sapiens*) and only 0.2% from dogs and pigs (*Canis familiaris* and *Felis domestica*) (Wang et al., 2005). However, there appears to be marked inter-village variation (even among ecologically similar villages) and the same authors reported an RTI of 80.4% for humans and only 4.5% for water buffalo in the second of the two villages in their study. In contrast, in the Philippines, Riley et al. (2008) used goodness-of-fit testing for mathematical transmission models, with an AIC approximation, to explain variation in the prevalence of human infections among 50 villages of western Samar. Their findings suggested no significant role for water buffalo in the *S. japonicum* transmission cycle that affects humans, but some association was indicated between transmission to snails from rodents and prevalence in humans (Riley et al., 2008). A cross-sectional survey of the same 50 villages was also undertaken in order to investigate any

association between infection in humans and in animals in the Philippines (McGarvey et al., 2006). The study found strong associations between the intensity of infections (as eggs per gram, epg) in cats, dogs and humans; this is in contrast to work done in China, which found little role for dogs and cats in the maintenance of infections in human populations (Wang et al., 2005). In western Samar the prevalence in the different host groups were; rats 30%, dogs 19%, water buffalo 3%, cats 3% and pigs 2%. It should be noted that the relatively low prevalence in the buffalo population could be an effect of the age of the animals sampled, it is noted that buffalo under 18 months of age tend to pass more eggs than older animals (Ross et al., 2001). The low prevalence in pigs may be attributed to the fact that they are mostly kept penned. Goats and sheep were not included in the Samar study, but these animals are highly permissive to *S. japonicum* and are often allowed to graze freely, so that they may be becoming increasingly significant in China (Wang et al., 2005).

Epidemiological assessments based on RTI values assume that there is no parasite sub-structuring by definitive host type, such that all parasites are equally likely to be transmitted by either definitive host group. Recent work in China and the Philippines suggests that different parasite lineages may be more compatible with specific host groups; this implies that parasites circulating in some animal reservoirs maybe less important in the maintenance of infection in human populations than others. Recent work, also in western Samar of the Philippines, has shed some light on this question. Rudge et al. (2008) used microsatellite markers to genotype adult worms and larval stages at multiple loci; they then estimated Wright's F-statistics (by AMOVA) and investigated geographical and among definitive-host group structuring of parasite genetic variation. The variation among the different host groups accounted for only around 1% of the total variation, with variation among individual host animals accounting for 92% of the total. However, alleles at two loci were exclusive to rats and all of these private alleles occurred at frequencies around 10%; this suggests some degree of isolation of the parasite population in rats from those in other host groups. Estimates of population phylogenies clustered the parasites from dogs and humans relative to those from rats and pigs. The authors suggested that the clustering of parasites of dogs and humans reflects the overlapping range of these two groups; they also noted that the population of dogs was three times that of water buffalo in this region and that *S. japonicum* may be evolving to infect dogs more efficiently in this area (Rudge et al., 2008). These findings differ from a study in China where clustering of worms from humans and bovines was observed relative to those from goats, dogs, cats and pigs (Wang et al., 2006). One could explain this as an effect of parasite strain sub-structuring leading to differential transmission among definitive host species. In contrast, Rudge et al. (2009) reported a clustering of isolates from dogs and bovines in marshland areas and humans, rodents and dogs in highland areas of China, but often found little differentiation among parasite sub-populations of different host types in sympatry. These authors suggested that patterns may differ even among local villages or between years (Rudge et al., 2009).

4.2. *Schistosoma mekongi* epidemiology and distribution in the region

In contrast to *S. japonicum*, *S. mekongi* is regarded as a parasite maintained mostly through transmission via human populations. *S. mekongi* uses the snail *Neotricula aperta* as its intermediate host and published records identify the following foci of *S. mekongi* transmission: Ban Hat-Xai-Khoun, Khong Island, southern Laos (Harinasuta and Kruatrachue, 1962); Kratie in Kratie Province, north-eastern Cambodia, approximately 180 km downstream of Khong Island (Audebaud et al., 1968); and San Dan, Sambour District, also in Kratie Province (Biays et al., 1999). All the aforementioned sites lie along the lower Mekong River. More recently, transmission of the parasite has been discovered in tributaries of the Mekong river, but also within the Mekong Basin, namely at Sa Dao in the Xe Kong river of Cambodia (Attwood et al., 2004). The potential human population at risk from Mekong schistosomiasis is currently over 1.5 million (Attwood et al., 2008), with around 800 people infected in Laos and around 2000 in Cambodia (crude estimates based on prevalence data in Urbani et al., 2002; Muth et al., 2010). At Sa Dao, no human infections were detected in 2004, but the disease re-emerged in 2005 (Sinuon et al., 2007). In addition, in 2004 the prevalence of infection among *N. aperta* collected at Sa Dao was 0.14% (Attwood et al., 2004). Similarly, despite an almost eight-fold reduction in the prevalence in the human population at Khong Island in Laos (1969–2003), the estimated prevalence in the local *N. aperta* populations had changed little (Attwood et al., 2001). These observations suggest that there may be a significant zoonotic component to the transmission of *S. mekongi*. Prevalences of 12.2% and 3.6% for pigs (in Laos) and dogs (in Cambodia), respectively, provide direct evidence for the importance of reservoir hosts (Strandgaard et al., 2001; Matsumoto et al., 2002). Surveys of cows, water buffalo, pigs, horses and field rats in 5 villages of Kratié Province (Cambodia) failed to detect any infection with *S. mekongi* (see (Matsumoto et al., 2002). Historical surveys have detected no natural infections in water buffalo (Schneider, 1976) and reported a similar prevalence in dogs at Kratié (Iijima et al., 1971), suggesting that the recent findings represent a long term equilibrium state.

4.3. *Schistosoma malayensis* epidemiology and distribution in the region

Human schistosomiasis in Malaysia is caused by *S. malayensis*. To date, human infections in Malaysia appear to be restricted to West (peninsular) Malaysia and more specifically to the Jelai and Tembeling river systems that drain into the Pahang river in Pahang State. The snail intermediate hosts are three species of *Robertsiella*, again snails of the family Pomatiopsidae (see Attwood et al., 2005). Humans and rats are the only known natural hosts for *S. malayensis* (see Ambu et al., 1984), with *Rattus muelleri* and *Rattus tiomanicus* recorded as the main definitive hosts (Greer et al., 1988; Attwood et al., 2005); however, the low prevalence in humans, combined with the failure to recover eggs from the stool of a biopsy-positive patient (Murugasu et al., 1978) or from serologically positive patients (Greer

and Anuar, 1984), suggests that humans are not an important host for this parasite. A small number of experimental infections also indicated that dogs are not permissive hosts (Ambu et al., 1984).

Consideration of the data currently available suggests varying but significant animal reservoirs of infection for all three species of Asian *Schistosoma* infecting humans. The major zoonotic element in transmission of human disease is attributable to *S. japonicum*; however, it is not clear if differences in host group utilization (e.g., the differences in the involvement of dogs, bovines and rodents between the Philippines and China) result from small sample sizes (in some cases only two villages were sampled), differences in land form (highland or flat marshland), different definitive host population sizes and behaviour, or different parasite strains. The parasite in the Philippines is transmitted by *Oncomelania hupensis quadrasi* whilst that in the lower Yangtze basin in China is transmitted by *O. h. hupensis*; these two snails may have different ecological habit and such differences could affect definitive host usage. One most obvious difference is that transmission (and snail activity) in China is much more seasonal than in the Philippines. Clearly more villages, host animals and ecological situations must be sampled in order to determine which host groups are most epidemiologically significant for disease in humans so that these can be targeted by control programs (the China National Control Program currently includes only cattle and water buffalo, Wang et al., 2005). In this way inter-village variation can be assessed and any important and stable patterns identified. In the case of Mekong schistosomiasis there is indirect evidence for a major animal reservoir (i.e., prevalences in, and densities of, snail populations remain stable in the face of marked reductions in human infections), but prevalences in dogs in Cambodia are relatively low (e.g., one dog in 310 sampled in 2001 was found to be infected) and it is likely that additional species are involved. Successful control of *S. mekongi* is unlikely to be achieved until all reservoir host species are known and their roles characterized. Of the three species, *S. malayensis* is most markedly a zoonosis, with transmission apparently entirely maintained by sylvatic rodents. In this sense *S. malayensis* has retained the ancestral character state being, like *S. sinensium*, a parasite primarily of rodents (Attwood et al., 2008). Consequently, studies of *S. malayensis* may be useful in understanding the processes of host-switching in the evolution of *Schistosoma*.

5. Vector-borne protozoan zoonoses in SE Asia

SE Asia has been spared devastating vector borne zoonotic protozoa such as trypanosoma which cause enormous harm to public health in Africa and South America. Indeed, the most important vector-borne zoonotic protozoan in SE Asia, *Plasmodium knowlesi*, was only discovered as a common human pathogen in 2004. Other vector-borne protozoa, which have not yet been recorded in SE Asia but occur in adjacent regions, include *Babesia* spp. in Japan, Taiwan, Korea and China (Shih et al., 1997; Homer et al., 2000; Marathe et al., 2005) and *Trypanosoma evansi*

in central India (Joshi et al., 2005). Further research may demonstrate these pathogens in SE Asia.

5.1. *Plasmodium knowlesi* malaria

P. knowlesi was identified in 1931, by Professor R. Knowles and Assistant Surgeon B.M. Das Gupta, of the Calcutta School of Tropical Medicine and Hygiene (Knowles and Das Gupta, 1932). There was confusion as to the species of primate in whose blood the parasite was found (initially said to be the African species *Cercopithecus pygerythrus*), but it was eventually determined to be the long-tailed macaque *Macaca fascicularis* (Eyles, 1963; Singh et al., 2004). It is the only primate malaria with a 24-h asexual blood-stage cycle and, unlike *P. vivax* and *P. ovale*, *P. knowlesi* is not known to have a hypnozoite stage. The genome has been sequenced (Pain et al., 2008). The parasite was used in the 1930s as a fever-inducing agent for the treatment of neurosyphilis (Knowles and Das Gupta, 1932). It was recognized that *P. knowlesi* parasites at different stages in human erythrocytes were difficult to distinguish from *P. falciparum* and *P. malariae* by microscopy. The first natural human infection of *P. knowlesi* was reported in 1965 in a man who returned to the USA from peninsular Malaysia. A further report in 1971 described human *P. knowlesi* infection, also contracted in peninsular Malaysia. Mosquito borne monkey-to-human and human-to-human transmission of *P. knowlesi* can occur under experimental conditions.

Singh et al. (2004) demonstrated, for the first time, the public health importance of *P. knowlesi* during an investigation of what was thought to be human *P. malariae* infection in Sarawak, eastern Malaysia. On finding that the laboratory and clinical features of these infections were atypical and a nested PCR assay failed to identify *P. malariae* DNA they demonstrated the parasites to be *P. knowlesi*. By PCR assay, 120 (58%) of 208 malaria patients tested positive for *P. knowlesi*, whereas none were positive for *P. malariae*. Most of the *P. knowlesi* infections were in rural adults without clustering within communities. The sequences for the isolates showed within species polymorphisms, suggesting that the infections were unlikely to be caused by a clonal outbreak.

Since this revelation of a fifth human malaria parasite (White, 2008), human *P. knowlesi* infections have been reported from wide areas of Southeast Asia including Thailand (Jongwutiwes et al., 2004), Myanmar (Burma) and the Burma/China border (Zhu et al., 2006; Figtree et al., 2010), the Philippines (Luchavez et al., 2008), Singapore (Ng et al., 2008), Sabah (Cox-Singh et al., 2008), Peninsular Malaysia (Cox-Singh et al., 2008), Kalimantan (Sulistyaningsih et al., 2010) and Vietnam (Van den Eede et al., 2009, 2010). It has not, as far as we are aware, been described from Cambodia, Laos or South Asia. *P. knowlesi* has been described from Formosan macaques (*Macaca cyclopis*) on Taiwan (Garnham, 1963) but the identity of the parasite is in doubt and it is likely to have been *P. inui*, which is not known to infect humans (Huang et al., 2010). Recent malaria surveys of *M. cyclopis* in Taiwan revealed *P. inui* but no *P. knowlesi* (Huang et al., 2010). Taiwan has been malaria free since 1965 and there is no

contemporary evidence of *P. knowlesi* infecting the human or simian populations, even though the vectors of simian malaria, the *Anopheles leucosphyrus* group, are present.

An important consideration for the detection of *P. knowlesi* in new environments is that the *Pmk8–Pmk9* primers for PCR detection of *P. knowlesi* cross-react with *P. vivax* (Imwong et al., 2009; Sulistyarningsih et al., 2010). It is very likely that the rapid increase in frequency of reports do not represent an 'emerging' disease but represent emerging *P. knowlesi* awareness. Indeed, an archival study of blood films collected in Sarawak in 1996 demonstrated that 97.2% (35/36) of those diagnosed morphologically as containing *P. malariae* parasites contained *P. knowlesi* and not *P. malariae* DNA (Lee et al., 2009). It is likely that many previous reports of *P. malariae* in SE Asia were *P. knowlesi*. Similarly, with the increasing sophistication of molecular assays and increased access to health care of forest dwellers in SE Asia other simian malarias may be discovered in humans. Small studies suggest that pan-malaria lactate dehydrogenase and pan-malarial aldolase antigen, but not histidine rich protein, based rapid tests for malaria will detect *P. knowlesi* (van Hellemond et al., 2009).

P. knowlesi infection can cause a wide spectrum of illness and severe disease (Daneshvar et al., 2009). In a prospective study of the clinical features of 152 patients with PCR-confirmed malaria in Sarawak, 70% had *P. knowlesi* infection and 93.5% of these had uncomplicated malaria and responded to chloroquine and primaquine. The remaining 6.5% had severe disease, and the most frequent complication was respiratory distress. *P. knowlesi* parasitaemia, serum creatinine level, serum bilirubin, and platelet count at admission were independent determinants of respiratory distress. Two patients with *knowlesi* malaria died, representing a case fatality rate of 1.8% (Daneshvar et al., 2009). In a recent post-mortem analysis of a patient who died of *P. knowlesi*, some evidence for parasite sequestration in the brain, as described for *P. falciparum*, was found (Cox-Singh et al., 2010). *In vivo*, *P. knowlesi* responds to chloroquine (Daneshvar et al., 2010). In a prospective evaluation of oral chloroquine and primaquine therapy in patients admitted in Sarawak, with PCR-confirmed single *P. knowlesi* infection, oral chloroquine was given for three days followed by, at 24 h, oral primaquine for two consecutive days. Of 73 patients recruited, 60 completed follow-up over 28 days. The median fever clearance time was 26.5 h (inter-quartile range: 16–34). The mean parasite clearance time to 50% (PCT50) and 90% (PCT90) were 3.1 h (95% confidence interval (CI): 2.8–3.4) and 10.3 h (95% CI: 9.4–11.4), respectively. These clearance times were more rapid than in a comparison group of 23 patients with *vivax* malaria. No *P. knowlesi* recrudescences or re-infections were detected by PCR. Therefore, in Sarawak chloroquine plus/minus primaquine is an inexpensive and highly effective treatment for uncomplicated *P. knowlesi* malaria infections. Primaquine is used as a gametocytocidal agent to reduce transmission. However, with both chloroquine resistant *P. falciparum* and *P. vivax* in Borneo, misidentification of *P. falciparum* and *P. vivax* as *P. knowlesi*, or cryptic mixed infection could have dire consequences for the patient. Other antimalarials that have been used successfully in *P. knowlesi* malaria include mefloquine, quinine, atovaquone/proguanil and

sulphadoxine-pyrimethamine (Daneshvar et al., 2010). The artemisinin derivatives are likely to be highly effective but formal proof of this is awaited.

In Peninsular Malaysia in the 1960s *Anopheles hackeri* was identified as the vector for *P. knowlesi*. As this mosquito is predominantly zoophagic and feeds mainly at the canopy level (Cox-Singh and Singh, 2008) it was not thought to be important for transmission to humans—who rarely visit the forest canopy. However, recent work from Sarawak suggests that *P. knowlesi* malaria is transmitted to humans from long-tailed (*Macaca fascicularis*) and pig-tailed (*M. nemestrina*) macaques by *Anopheles latens* mosquitoes when humans visit forested areas (Vythilingam et al., 2006; Tan et al., 2008). Tan et al. (2008) demonstrated that *A. latens* mosquitoes were attracted to both humans and caged monkeys (probably *Macaca fascicularis*) and that forest-caught *A. latens* contained *P. knowlesi* sporozoites.

Old World monkeys are conventionally divided into two subfamilies, the Colobinae and Cercopithecinae and both taxa contain diverse species in SE Asia. *P. knowlesi* has been found in the cercopithecline monkeys *M. fascicularis* and *M. nemestrina* and in a colobine monkey—the banded leaf monkey (*Presbytis melalophos*). However, there appears to be only one report of *P. knowlesi* in the banded leaf monkey and this does not state whether the primates were wild or describe the malaria parasites (Eyles et al., 1962). As *P. knowlesi* is lethal for rhesus monkeys (*M. mulatta*) and the hanuman langur (*Semnopithecus = Presbytis entellus*), the two most abundant non-human primates in India (Garnham, 1963), these primates are less likely to be important in transmission to humans. If this is correct, *P. knowlesi* is unlikely to be common in the large areas of south Asia where these two species are the predominant non-human primates. In *M. fascicularis*, infection results in prolonged low-level parasitaemia. Whether *P. knowlesi* infections in Malaysian Borneo is mostly due to transmission between humans or between monkeys and humans by mosquitoes is uncertain. However, the lack of clustering of cases within longhouses suggests that transmission occurs away from the vicinity of longhouses and that monkey-to-human rather than human-to-human transmission is taking place. Urban *P. knowlesi* has not been described, and despite macaques being kept as pets and in zoos, transmission is unlikely as the known vectors are predominantly forest mosquitoes.

M. fascicularis and *M. nemestrina* are found in the Philippines and Indonesia, throughout Malaysia, Thailand, Vietnam, Laos and Cambodia through to Burma, the Nicobar Islands and Bangladesh (Cox-Singh and Singh, 2008). *M. fascicularis* has also been introduced to Mauritius, Palau and Papua New Guinea (IUCN, 2010b), raising the possibility of transmission there if vectors are present. *P. melalophos* occurs on Sumatra (IUCN, 2010a) but the taxonomy of these primates is confusing, with diverse related *Presbytis* species throughout south and SE Asia and, as far as we are aware, *P. knowlesi* has not been described from Sumatra.

The social organisation of these primates differ, in terms of ranging patterns, relationships to humans and time spent on the ground versus the canopy and these factors may have important influences on their relevance as reservoirs for transmission of *P. knowlesi* to humans. There is

also evidence that primates have evolved medical plant use (Newton, 1991) and it is possible that they consume plant secondary compounds as antimalarials.

The finding of humans commonly afflicted by simian malaria is important for malaria elimination. With humans encountering infected mosquitos in forests, *P. knowlesi* cannot realistically be eliminated. However, so far the areas where it is known to commonly cause clinical problems are relatively few.

5.2. Leishmaniasis

Leishmaniasis, named after the Scottish pathologist William Leishman, is caused by obligate intracellular protozoa of the genus *Leishmania*. It is transmitted by phlebotomine sandflies and occurs in tropical and subtropical regions of the Middle East, India, China, Africa, and southern and central America. Although described from 62 countries with an estimated 500,000 new cases/year (Guerin et al., 2002) it has very rarely been described from SE Asia. The nearest region to SE Asia with a high incidence of visceral leishmaniasis is Northeast India.

Phlebotomus species of sandflies breed in organic debris and feed mostly on plant juices but parous females need at least two vertebrate blood meals to permit egg maturation. Species vary in their degree of anthropophilia and hence transmission of leishmaniasis to humans. Leishmaniasis has been reported in SE Asia as an imported disease amongst those returning from residence in endemic areas (Suttinont et al., 1987; Suankratay et al., 2010). Cutaneous leishmaniasis has been reported from East Timor, the only record we are aware of from the Indonesian archipelago. However, clinical descriptions or laboratory confirmation were not provided (Chevalier et al., 2000). In 1999 visceral leishmaniasis was reported, for the first time in Thailand, in a 3-year-old girl living in Suratthani Province (Thisyakorn et al., 1999). Apparent autochthonous transmission of visceral leishmaniasis has since been described in four additional patients from northern, central, and southern Thailand, but not in the northeast or elsewhere in SE Asia. As far as we are aware locally acquired cutaneous leishmaniasis has not been described from Thailand, only imported cases have been reported (Viriyavejakul et al., 1997). Recently, leishmaniasis has been described in a human immunodeficiency virus infected Thai fisherman who presented with nephrotic syndrome, fever, anemia, and thrombocytopenia. It seems most likely that he contracted the disease in Thailand although Indonesia is a remote possibility. Polymerase chain reaction and nucleotide sequence analysis of the internal transcribed spacer 1 of the small subunit ribosomal RNA gene in blood and kidney biopsy specimens showed a putative new *Leishmania* species, of similar genotype to that described in a patient from southern Thailand with visceral leishmaniasis (Suankratay et al., 2010). The ITS1 sequence of the rRNA gene suggested similarity to *Leishmania brasiliensis* and *Leishmania guyanensis*, which are the causative agents of New World visceral and cutaneous leishmaniasis, respectively. *Leishmania infantum*, the cause of visceral leishmaniasis in the Mediterranean, western Asia and cen-

tral China, was described in a patient in Bangkok (Maharom et al., 2008).

Phlebotomine sandflies are widely distributed in Thailand but collections in the villages of leishmaniasis patients yielded no known potential vector sandfly species of *Leishmania*. However, cow-biting and bat-biting cave-dwelling sandflies, vectors of Old World visceral leishmaniasis, have been described elsewhere in Thailand (Apiwathnasorn et al., 1989). Positive direct agglutination tests (DAT) for *Leishmania* antibody have been reported from Thai cats and cows but there seem to have been no investigations of dogs as potential reservoirs apart from one investigation in Bangkok (Sukmee et al., 2008).

It remains unclear whether these very rare reports of visceral leishmaniasis represent clinical manifestations of very low levels of transmission in Thailand and further investigations of potential vectors and reservoirs are required. An alternative explanation could be unsustainable transmission via sandflies from index patients with visceral leishmaniasis who acquired their infection elsewhere. Wild vertebrates may also be reservoirs. Given the rarity of the disease it will not be on the differential diagnosis of the majority of health workers in SE Asia and laboratory diagnostic techniques are rarely available—suggesting that the incidence could be underestimated. On the other hand, the relatively high incidence of HIV infection in Thailand would have been expected to increase the incidence and clinical ‘visibility’ of this disease (Guerin et al., 2002).

6. Zoonotic hookworm disease in SE Asia

Human hookworm infections continue to be a major public health problem in SE Asia with approximately one quarter of the 563 million inhabitants infected (Hotez and Ehrenberg, 2010). Worldwide, enteric human hookworm infections are predominantly associated with two species, *Ancylostoma duodenale* and *Necator americanus* (Brooker et al., 2004), and neither is considered zoonotic. However, pigs have been implicated as transport hosts of *N. americanus* (Steenhard et al., 2000) and may have an important role in the natural history of human disease. Of the zoonotic hookworm species that cause human disease, *A. ceylanicum* is the only species capable of establishing a patent enteric infection in humans, canines and felines (Anten and Zuidema, 1964; Wijers and Smit, 1966; Yoshida et al., 1968; Carroll and Grove, 1984, 1986). Historically, *A. ceylanicum* has received little attention despite it being known to cause human disease for at least the past 40–50 years (Anten and Zuidema, 1964; Wijers and Smit, 1966; Carroll and Grove, 1986; Traub et al., 2008).

6.1. Historical perspectives on *Ancylostoma ceylanicum*

Zoonotic hookworm disease resulting in anaemia was first described in 1964 in Dutch marines returning from service in West New Guinea (now Indonesian West Papua) (Anten and Zuidema, 1964). Nine of eleven (82%) returning marines were found to have a patent enteric infection with *A. braziliense* (Anten and Zuidema, 1964) which was later referred to as *A. ceylanicum* (Wijers and Smit, 1966; Chowdhury and Schad, 1972). Three marines were infected

with more than 100 adult worms and two of these otherwise healthy well-fed marines were anaemic (Anten and Zuidema, 1964) which is in stark contrast to most reports where only a few adult *A. ceylanicum* worms have been recovered (Kian Joe and Kok Siang, 1959; Yoshida et al., 1968; Chowdhury and Schad, 1972). A follow-up study using *A. ceylanicum* worms originating in West New Guinea and passaged through dogs showed that infection in healthy volunteers produced severe clinical symptoms within 15–20 days after cutaneous exposure to L3 larvae, including severe abdominal pain and epigastric spasms (Wijers and Smit, 1966). All volunteers were exposed to relatively few larvae and patent infections with low egg counts were detected in all cases, of particular note was the finding that a small worm burden resulted in cognitive impairment and difficulty concentrating (Wijers and Smit, 1966). A second experimental infection study involving two well-fed healthy volunteers in Australia (Carroll and Grove, 1986) reported similar severe abdominal pain 5 weeks after infection with associated diarrhoea in one case; Carroll and Grove (1986) were also able to demonstrate recurrent bouts of abdominal disturbance over several months.

A. ceylanicum is the most neglected of all human hookworm species, typically considered to be an unimportant pathogen (Chowdhury and Schad, 1972; Brooker et al., 2004; Hotez et al., 2004) due to an absence of demonstrated heavy infections and subsequent anaemia (Brooker et al., 2004). *A. ceylanicum* is described as a poorly adapted human hookworm (Chowdhury and Schad, 1972) and ill-suited to the human gastrointestinal tract, resulting in patent infections with low fecundity. The evidence for clinical insignificance however comes from experimental studies involving healthy well-fed adults (Wijers and Smit, 1966; Carroll and Grove, 1986) and urban inhabitants (Kian Joe and Kok Siang, 1959; Chowdhury and Schad, 1972). The clinically significant findings from West New Guinea (Anten and Zuidema, 1964), with vastly different environmental exposures, has been largely overlooked for 45 years. In addition, the non-blood loss symptoms associated with *A. ceylanicum* infection, including cognitive impairment from light infections (Wijers and Smit, 1966), rarely receive a mention. Furthermore, there is a distinct similarity between acute clinical presentation caused by *A. ceylanicum*, including severe abdominal pain (Wijers and Smit, 1966; Carroll and Grove, 1986; Traub et al., 2008) and recurrent abdominal disturbance (Carroll and Grove, 1986), and eosinophilic enteritis caused by *A. caninum* that is indicative of intestinal hypersensitivity (Prociv and Croese, 1996).

6.2. Contemporary studies on enteric hookworm prevalence

Three community surveys in SE Asia in the past 45 years report hookworm to the species level and *A. ceylanicum* is prevalent, to varying degrees in all studies (Traub et al., 2008; Sato et al., 2010) (Conlan et al., In preparation). In a recent study in northern Laos, 46% of the human survey population from 24 villages were found to have hookworm infections and a randomly selected sub-

set of samples showed that up to one third of infections were *A. ceylanicum* and two thirds *N. americanus* (Conlan et al., in preparation). Furthermore, almost all village dogs in northern Laos had hookworm infection and molecular analysis of a subset of samples detected *A. ceylanicum* in 85% of infected dogs; *A. caninum*, *A. braziliense* and *N. americanus* eggs were also detected in Lao village dogs (Conlan et al., in preparation). Hookworm ecology in southern Laos may be different from the north, where *A. duodenale* may be the predominant *Ancylostoma* species in people (Sato et al., 2010), but this study was only conducted in a single village and no dog data were reported. A survey of humans and dogs in Bangkok recorded *A. ceylanicum* as the predominant hookworm species in dogs and almost a third of human hookworm carriers in the study population (2/7) harboured *A. ceylanicum* (Traub et al., 2008). Notably, only the *A. ceylanicum* cases suffered chronic abdominal disturbance (Traub et al., 2008). These recent surveys from Thailand and Laos indicate that dogs have an important role in the natural history of human infection. Unfortunately, no detailed clinical or worm burden data were reported in these studies but the high prevalence of *A. ceylanicum* in humans and dogs warrants further investigation.

6.3. Cutaneous larva migrans

Zoonotic infections caused by dog and cat hookworm species, *A. caninum*, *A. braziliense* and *A. tubaeforme* can also occur and the pathogenic nature of the infection is dependent on the migration of larvae to ectopic sites in the paratenic human host (see Bowman et al., 2010). Cutaneous larva migrans (CLM) is the most common disease described (Bowman et al., 2010), other clinical manifestations include eosinophilic enteritis (Croese, 1988; Prociv and Croese, 1990, 1996; Croese et al., 1994), eosinophilic pneumonia (Löffler's syndrome), myositis, folliculitis, erythema multiforme or ophthalmological manifestations (see Bowman et al., 2010). Cutaneous larva migrans is predominantly associated with *A. braziliense* (Bowman et al., 2010) and published reports of CLM from SE Asia tend to be limited to tourists returning home (Jelinek et al., 1994; Malvy et al., 2006). Since *A. braziliense* is rarely reported in SE Asia, with just a few reports from Malaysia, Indonesia and Laos (Conlan et al., 2010, in preparation; Yoshida et al., 1973; Margono et al., 1979), it is not clear what hookworm species were the cause of these CLM cases, possibly *A. ceylanicum* or *A. caninum*. In light of the advances in *Ancylostoma* molecular diagnostics (Traub et al., 2004, 2007, 2008; Palmer et al., 2007), the geographic range and prevalence of *A. braziliense* in SE Asia should be reappraised. *Ancylostoma ceylanicum* on the other hand is endemic in SE Asia with a wide geographic range, encompassing Indonesia, Borneo, Malaysia, Philippines, Thailand and Laos (Kian Joe and Kok Siang, 1959; Anten and Zuidema, 1964; Velasquez and Cabrera, 1968; Yoshida et al., 1968, 1973; Setasuban et al., 1976; Margono et al., 1979; Choo et al., 2000; Scholz et al., 2003; Traub et al., 2008; Sato et al., 2010; Conlan et al., in preparation) and can cause CLM, presenting as a maculopapular 'ground itch' (Haydon and Bearup, 1963; Wijers and Smit, 1966).

6.4. *Eosinophilic enteritis*

Eosinophilic enteritis has been well described for *A. caninum* infections in northeastern Australia (Croese, 1988; Procriv and Croese, 1990, 1996; Croese et al., 1994) with sporadic case reports from the United States of America (Khoshoo et al., 1994, 1995) and Egypt (Bahgat et al., 1999). It is notoriously difficult to make a definitive diagnosis of *A. caninum* eosinophilic enteritis due to the vagaries of clinical symptoms, the variability of serological results and the difficulties of recovering worms (Procriv and Croese, 1996). While the symptoms are non-specific, abdominal pain is almost invariably observed and can range from severe acute pain mimicking appendicitis to more mild discomfort; pain can become chronic or recurrent and in rare cases bowel obstruction or bleeding can occur (Procriv and Croese, 1996). Other symptoms may include anorexia, nausea and diarrhoea (Procriv and Croese, 1996). *Ancylostoma caninum* eosinophilic enteritis has not been documented in SE Asia even though this hookworm is prevalent with a wide geographic range (Setasuban et al., 1976; Margono et al., 1979; Traub et al., 2008; Conlan et al., in preparation). In part, this may be due to the difficulty of establishing hookworm as a cause of obscure and/or recurrent abdominal pain or eosinophilic enteritis.

6.5. *Ancylostoma ceylanicum* in the context of regional programs for the control of soil transmitted Helminths

Mass drug administration (MDA) using a single dose of mebendazole or albendazole for the control of soil-transmitted helminths is widespread in SE Asia with greater than 90% of school age children in Laos and Cambodia and greater than 70% in Vietnam treated (Montresor et al., 2008; WHO, 2009). Mebendazole is commonly administered in SE Asia due to safety and low cost (Flohr et al., 2007; Phommasack et al., 2008). However, hookworm disease continues to be an important public health problem throughout the region and there is little evidence that MDA is effectively reducing the burden of hookworm disease. There are multiple reasons for this issue; foremost among them is the low efficacy of a single dose of mebendazole in reducing egg output (Flohr et al., 2007; Keiser and Utzinger, 2008), but there is no data describing the efficacy of mebendazole in clearing or reducing egg counts for *A. ceylanicum*.

7. Concluding comments

The parasitic zoonoses circulating in SE Asia are a major burden on public health and wellbeing. The magnitude and scope of this burden varies for each of the parasites we have discussed. For the medically important trematodes, the ecological changes currently taking place have the potential to increase the abundance and distribution, thereby placing far more people at risk of infection. The impacts of climate change on food-borne trematodiasis have been discussed for some time now (Mas-Coma et al., 2009), however the planned hydropower development on the Mekong River mainstream and its tributaries in Laos and Cambodia (MRC, 2005; ICEM, 2010) may have an impact on

trematode distribution and abundance by altering snail and fish ecology and human interaction with the river environment. As such, these potential impacts need to be monitored.

In contrast, the trend towards more commercially oriented livestock production has the potential to reduce the abundance and burden of both *T. solium* and *Trichinella* spp. In the SE Asian nations where *T. solium* is endemic, there exists a window of opportunity for a concerted effort to identify hot-spots of endemicity and pin-point control programs designed in consultation with effected communities. However, for this to be achieved, improved diagnostic methods for use in a multi-*Taenia* species environment are urgently required, a thorough understanding of pork supply networks and the political will to deliver services to poor marginalised communities will also be important (Conlan et al., 2009; Willingham et al., 2010). In an environment where people have a preference for consuming uncooked or partially cooked meat from domestic or wild pigs, the control of trichinellosis will remain problematic. *T. spiralis* incidence will most likely continue to decline in the more developed countries such as Thailand, but sporadic cases and outbreaks will continue to occur in Laos and Vietnam. Sensitisation of health officials to trichinellosis is required so that cases and outbreaks can be more thoroughly investigated and documented and the *Trichinella* worms circulating and causing disease in a population can be accurately identified (Odermatt et al., 2010).

The relative rarity of vector-borne protozoan diseases in SE Asia make it difficult to predict what impact environmental and socio-cultural changes will have on the distribution and incidence of *P. knowlesi* and *Leishmania* spp. infection. Like trichinellosis, health officials will need to be sensitised to these parasitic zoonoses in the differential diagnosis for patients. In addition, vector competence studies will be important to effectively monitor the emergence of these medically important parasites.

It is apparent that *A. ceylanicum* is highly endemic in the dog population of some SE Asian nations with spill over into the human population. At this stage we do not have a good understanding of the clinical significance of *A. ceylanicum*, but historical data indicates that it does cause clinical disease. In areas where this zoonotic hookworm is prevalent in humans and dogs, the wide spread use of anthelmintics such as mebendazole in the human population will have limited impact on *A. ceylanicum* distribution and it may even provide a niche environment for *A. ceylanicum* to thrive (Thompson and Conlan, in press). Therefore, the use of anthelmintics to control hookworm in dogs will need careful consideration to avoid clearing *T. hydatigena* from dogs and consequently altering the infection pressure on pigs, potentially creating a pig population with greater susceptibility to *T. solium* infection (Thompson and Conlan, in press).

The ecological changes currently taking place in SE Asia present new risks for the emergence or re-emergence of clinically important parasitic zoonoses while at the same time presenting new opportunities for disease control. To address these issues, we will need to confront all aspects of parasite ecology as a collaborative international research community, employing the latest diagnostic and

risk assessment technologies together with effective consultative community-led health programs.

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