

Principle

Strongyloidiasis, a soil-transmitted helminthiasis caused by *Strongyloides stercoralis*, remains an important public health problem worldwide (Neva, 1986). The infection is endemic in tropical and subtropical regions, but is seen rarely in temperate zone, some parts of Europe, the southeastern United States and Puerto Rico. Clinically, strongyloidiasis is significant because infection persists for many years in the human host, usually as an undetected, asymptomatic condition. Under specific conditions such as immunosuppression, this undetected larvae can produce extensive tissue infection, so called hyperinfection syndrome (Mahmoud, 1996). There is some dispute whether *S. stercoralis* is an opportunist in HIV patients. Because hyperinfection with *S. stercoralis* is a well-documented feature in immunosuppressed patients, HIV infection is considered an inclining factor for severe strongyloidiasis. However, observations from the tropics do not suggest that this parasite is an opportunist in HIV patients. *S. stercoralis* larvae can be activated to produce hyperinfection in patients who have been immunosuppressed in various ways, especially by the administration of steroids (Neva, 1986). It was thought that impaired cell-mediated immune responses due to immunosuppression, malignancy, miliary tuberculosis, malnutrition and leprosy were primarily responsible for hyperinfection (Zumla and Croft, 1992).

Human infections by *S. stercoralis* are often chronic and persistent with low level or fluctuations of larval excretion. Persistent infections are caused by well-documented internal autoinfection. There must be at least three anatomic levels at

which immune responses of the host are ineffective leading to persistent infections: 1) inability to eliminate adult worms from the submucosa of the small bowel, 2) failure to prevent reinvasion of the colon by infective larvae, and 3) inability to destroy larvae that have already reinvade and are in transit back to the intestine (Neva, 1986).

The most simple method of parasitological diagnosis is microscopic demonstration of larvae in a direct smear of a fecal specimen. But this may require a combination of luck and persistence in searching through multiple smears since the excretion of larvae is often sporadic and scanty in numbers (Neva, 1986). Three traditional methods to detect the larvae from stool are the direct smear, filter paper culture, and formalin-ether concentration methods but these methods require skill, time, and often give unreliable results (Arakaki *et al.*, 1990). Agar plate method appears to be more efficient than traditional method and it is necessary to discriminate *Strongyloides* microscopically from other similar nematode larvae by skillful personnel (Koga *et al.*, 1990). However when compared with other methods, the agar plate method has the highest cost (de Kaminsky, 1993). For the past several years, a number of reports on serological diagnosis have relied on the use of antigens prepared from larvae of either the human parasite, *S. stercoralis*, or the rat parasite, *S. ratti*. Most of these studies have employed the enzyme-linked immunosorbent assay (ELISA), the indirect immunofluorescent (IFA) test, the indirect hemagglutination (IHA) test, and the immunoblot (Carroll *et al.*, 1981; Grove and Blair, 1981; Neva *et al.*, 1981; Sato *et al.*, 1985; Gam *et al.*, 1987; Bailey, 1989; Conway *et al.*, 1993a; Conway *et al.*, 1993b; Conway *et al.*, 1994; Lindo *et al.*, 1994). Two methods that have high sensitivity for detection of antigens and antibodies were

immunofluorescence and radioimmunoassay. However, in practice immunofluorescence is not easy to interpret since it depends on subjective visual assessments of fluorescence and the results are usually expressed as the serial dilution of serum that gives the least fluorescence. About the radioimmunoassay, the isotope labels may decay rapidly, so that the conjugates have a short shelf-life. Both the immunofluorescence and radioimmunoassay require complex equipment for the assessment. The ELISA has been adopted although it is not quite as precise as radioimmunoassay. But the reagents appear to show no health hazards, are stable and have long shelf-lives. Moreover, the estimation of results can either be visual or be made with a rather simple spectrophotometer of the type found in most laboratories (Voller *et al.*, 1976). Even though the serodiagnosis by ELISA has been reported to be more sensitive than parasitological methods, it cannot differentiate an ongoing infection from a past infection and it is helpful in the diagnosis of disseminated infection. This problem could be overcome by the demonstration of parasite antigen in infected humans as this would be indicative of current infection (Nageswaran *et al.*, 1994).

Coproantigen detection by ELISA has been successfully developed for the diagnosis of fascioliasis, opisthorchiasis, taeniasis, echinococcosis, amebiasis (Youssef *et al.*, 1991; Allan *et al.*, 1992; Espino and Finlay, 1994; Sirisinha *et al.*, 1995; Nonaka *et al.*, 1996; Malgor *et al.*, 1997; Espino *et al.*, 1998; Singh *et al.*, 1999). In each of these infections, antigen detection was found to be more sensitive than parasitological examination because antigens could be detected in microscopically negative feces. Recently there was a report on the detection of coproantigen in *Strongyloides ratti* infection based on a sandwich capture ELISA

using polyclonal antibodies prepared against adult and filariform larval somatic antigens. It was shown that both antibodies were equally effective in detecting *Strongyloides* coproantigens. The detection of coproantigens coincided with the appearance of larvae in the stool and the coproantigen level remained elevated even when the parasite count was low (Nageswaran *et al.*, 1994).

Lindo *et al.* (1994) showed by Western blot analysis that the 41-kDa protein of *S. stercoralis* third-stage or filariform larvae (L3) gave 100% sensitivity and 94% specificity. They showed this protein was the most important antigen and should be subjected for further study. In this study we are interested in producing monoclonal antibodies (mAb) against 41-kDa protein of *S. stercoralis* L3 and the mAb will be used in the development of coproantigen assay for diagnosis of human strongyloidiasis.