Introductory remarks on microsporidia in the AIDS era

J. Lom

Institute of Parasitology, Academy of Sciences of the Czech Republic, Branišovská 31, 370 05 České Budějovice, Czech Republic

The research of microsporidia, which actually dates from the description of Nosema bombycis in 1857, is marked by several milestones. Kudo's classical monograph from 1924 has summarized the first era of accumulation of data on the diversity of species and genera. One year earlier, Encephalitozoon cuniculi, the first mammalian species was described (Levaditi C. et al. 1923: C. R. Acad. Sci. 177: 985–988), although later its microsporidian nature was not unambiguously accepted. In 1959, the first undisputed discovery of microsporidiosis in a Japanese boy (Matsubayashi H. et al. 1959: Arch. Pathol. 67: 181–187) started the series of findings of rare cases of human microsporidia preceding the outbreaks of HIV-induced immunosuppression. In the sixties, studies of several authors on the mode of spore hatching eventually revealed the process of polar tube eversion with the resulting injection of the sporoplasm into the host cell (see the short summary in Canning E. U. and Lom J. 1986: The Microsporidia of Vertebrates, Academic Press, London, 289 pp.) In the eighties, a series of papers published by several authors over several years resulted in safe proof of sexual process and alternation of hosts in some arthropod microsporidia. In 1987, a paper by Vossbrinck et al. (Vossbrinck C. R. et al. 1987: Nature 326: 411–414), completing earlier studies, presented the claim of the extraordinary low phylogenetic position of microsporidia.

The last milestone which I shall mention here is the discovery that microsporidia appear as serious pathogens in clinical cases of AIDS; this started a new era in microsporidian research. This disease, by drastically reducing the immune competence of infected people, revealed like a ghastly experimentalist the previously unanticipated existence of several species of microsporidia, which were found to flourish in patients with collapsing immune system. Microsporidia were found to be severe pathogens in AIDS patients and consequently, attracted wide attention of clinicians, parasitologists and protozoologists. In terms of practical impact, the significance of microsporidia as pathogens of economically or medically important arthropods dramatically shifted in favour of the interest which they have as parasites of man.

The most widely distributed is Enterocytozoon bieneusi, described in 1985 by Desportes et al. (Desportes L. et al. 1985: J. Protozool. 32: 250–254); the other species, found in a steadily increasing prevalence are Septata intestinalis Cali et al., 1992, Encephalitozoon hellem Didier et al., 1991 and E. cuniculi Levaditi et al., 1923 the existence of which in humans was eventually confirmed by molecular biology methods (N. Pieniazek, pers. comm.). Along with the other rare species found mostly just once - Nosema comorii Sprague, 1974, N. corneum Shadduck et al., 1990, N. ocularum Cali et al., 1991, Pleistophora sp. of Ledford et al., 1985, Microsporidium africanum Canning, 1986 and M. ceylonensis Canning, 1986 - they raised the number of named microsporidia infecting man to ten.

By now, a remarkable amount of data has been accumulated mainly on pathogenicity, clinical course of inflicted infection, diagnostics, life cycle and cell structure of human microsporidia. This growing knowledge, however, opens an equally increasing number of questions.

The identity of the most widely distributed agent Enterocytozoon bieneusi poses no problems. There is no evidence that there could be any biological or epidemiological ties with E. salmonis, its only congeneric species. The genus Encephalitozoon comprises species which seem to be closely related. Besides E. cuniculi and E. hellem there probably will be additional species both in man and animals, and several authors have unpublished evidence suggesting that it is so.

In view of the amazingly large host range of E. cuniculi it would be worthwhile to examine to what extent the isolates from various rodents, lagomorphs or carnivores and man may differ. This may have bearing to varying degree of susceptibility in particular hosts and thus to the epidemiology. The differences between the genus Septata and Encephalitozoon seem not very pronounced and further research will prove whether its distinctness should be upheld or not.

Studies on the life cycle will certainly be crucial for determination of the precise taxonomic position of separate species. They will also have an important bearing on our understanding of the process of infection. In Encephalitozoon, spontaneous germination of spores still within the host seems a commonplace phenomenon. It had already been mentioned by Petri (Petri M. 1969: Acta Pathol. Microbiol. Scand. 204: 1–91) in E. cuniculi, and appeared to be quite striking in E. hellem as observed by Canning et al. (Canning E. U. et al. 1992: Europ. J. Protistol. 28: 226–237). There are numerous parallels in other microsporidia. In Nosema apis there have been claims of two types of spores (Fries I. 1992: Apidologie 23: 61–70), autoinfective and transmissive much alike the two types of oocysts in Cryptosporidium, and autoinfection is

Presented at the International workshop "Microsporidiosis and Cryptosporidiosis in ImmunoDeficient Patients", September 28 – October 1, 1993, České Budějovice, Czech Republic.
frequent e.g., in fish-infecting Glugea species. In Encephalitozoon infection of mammals and man, future studies will probably show whether this in-site, autoinfective germination is a feature associated with a certain stage of infection or possibly an atypical site, whether it is more proper to certain strains or species and to what extent it may depend on the type of the host cell response.

In epidemiology, the pivotal question is probably to reveal the source of human microsporidioses. Encephalitozoon cuniculi is, according to existing evidence, zoonotic, a conclusion falling in line with earlier work by Schaduck and coworkers (Schaduck J. A. et al. 1979: J. Parasitol. 65: 123–129) who infected mice, rats and Rhesus monkeys with rabbit strain of E. cuniculi, and who also found baboon cell cultures contaminated with this species (Schaduck J. A. et al. 1979: J. Parasitol. 65: 185–188). The results of DNA sequencing (Pie nia zek and coworkers, pers. comm.) also support this view. These facts do not preclude the possible existence of strains which may prove to be different entities, much alike in Pneumocystis carinii complex, where strains from rat and man constitute different species, and where Stringer et al. (Stringer S. L. et al. 1993: Abstracts, IX. Int. Congr. Protozool., July 25–31, 1993, Berlin, p. 123) claimed the existence in rat of even two species.

An animal source of the agent may also be suspected in rare findings of human infections in immunocompetent people due to species like Microsporidium ceylonensis or Nosema corneum, developing in an immunoprivileged site such as cornea. Cross-infections to test this possibility are feasible with the latter species, available in cell culture.

Enterocytozoon bieneusi seems to be species specific for humans, persisting in the population in light infections which are beneath the threshold of easy detection and thus escape attention. Such infections can probably be activated in individuals who become immunodeficient. The case of a self-limiting infection of a HIV-negative and immunocompetent man (Sandfort et al. 1993: Abstracts, IX. Int. Congr. Protozool., July 25–31, 1993, Berlin, p. 111) favours this assumption. The prevalence of E. bieneusi in AIDS patients might thus reflect the prevalence of low-level distribution of the agent in the "normal" population. In the lack of an animal model for enterocytozoonosis it may be difficult to explore the other possibility, that immunocompromised patients become highly susceptible to infection contracted from very few infected individuals in the "normal" population. The future may also reveal if there may be nosocomial infections, recently reported in cryptosporidiosis. They are easy to imagine especially in encephalitozoonosis, where in some cases respiratory acquisition of infection was postulated, as well as from cases of enterocytozoonosis disseminated to respiratory tract. Thus far, only enterocytozoonosis has been detected in AIDS patients in the tropics; most probably, future studies will also reveal infections with other species.

In the pathology and course of disease in human microsporidioses, comparison of observations with the wealth of data obtained in the research of animal infections with Encephalitozoon cuniculi may well further the understanding of how the infection proceeds. There are many questions in which the animal model may be of help. In man, one should know to what extent the individual microsporidian species are site specific. Such tissue specificity is obviously not very strict, as witnessed by species discovered in cornea and later reported from elsewhere in the body, or by enterocytozoonosis affecting nasal epithelium or bronchial tissue (Weber et al. 1992: Am. Rev. Respir. Dis. 146: 1603–1605) or causing cholangitis. Future research should also clarify, whether individual species prefer specific types of cells and, if they spread to other organs, which cells do they infect. There is a plethora of other questions. Enterocytozoon bieneusi stages are localized above the enterocyte nucleus - does such regularity exist also in other species? To what extent are the clinical symptoms of infections due to different species and strains of Encephalitozoon specific, and how do they overlap in their site preferences? Do the microsporidia infecting intestinal epithelium have some specific mechanism to delay the turnover of enterocytes?

Some of the interactions between the parasite and host cell can be studied in cell cultures; Enterocytozoon, however, still defies attempts at successful cultivation. Cell culture of human microsporidia is the most convenient way to obtain large amounts of parasite material for diagnostics and experimental work and chemotherapeutic trials, although in this respect it has not been fully exploited yet.

Diagnostic methods for detection of human microsporidioses have thus far been sufficiently elaborated and one can expect further development especially in two directions. One is the refinement and perfection of light microscopical (stains, fluorescence) and serological methods - especially to increase sensitivity and specificity of the latter. Much is expected from further development of DNA probes for rapid and safe detection of microsporidia in human material.

Treatment of human microsporidioses is still a sore point. Thus far, albendazole has been found effective towards Septata and Encephalitozoon. It has to be shown, however, to what extent the trophic stages and persisting spores are affected and eliminated. No obvious success has been achieved with Enterocytozoon. The treatment and epidemiology are two fields, at which the main thrust of the research will be directed, since these are the two areas crucial for checking the microsporidioses in man. Presently, there are many teams and laboratories working on these topics, in the USA, many European countries and Australia, and one can expect essential progress in the few coming years.

Received 1 November 1993

Accepted 17 November 1993

256