INTESTINAL CO-INFECTION BY *CYCLOSPORA* SP. AND *CRYPTOSPORIDIUM PARVUM* : FIRST REPORT IN AN AIDS PATIENT

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Summary :

Cyclospora is a recently described new human pathogenic coccidian causing intermittent diarrhoeal enteritis which may persist for weeks or months in immunocompetent subjects, particularly travellers visiting some tropical areas and countries, such as Nepal, the Caribbean, Peru and Mexico. More rarely this enteric pathogen affects immunocompromised humans, namely HIV-infected people or AIDS patients, with same clinical pictures recognized in normal hosts. We describe the first case of *Cyclospora* sp. and *Cryptosporidium parvum* associated diarrhoeal enteritis in an adult AIDS patient.

KEY WORDS : *Cyclospora* sp. *Cryptosporidium parvum*. AIDS.

MOTS CLES : *Cyclospora* sp. *Cryptosporidium parvum*. SIDA.

Résumé : CO-INFECTION INTESTINALE À *CYCLOSPORA* SP. ET *CRYPTOSPORIDIUM PARVUM* : PREMIERE OBSERVATION CHEZ UN SIDÉEN

Cyclospora est une coccidie pathogène de l'homme de description récente ; elle est responsable d'une diarrhée intermittente pouvant persister plusieurs semaines ou mois chez les sujets immunocompétents, particulièrement les voyageurs revenant de régions tropicales telles le Népal, les Antilles, le Pérou et le Mexique. Plus rarement, ce pathogène de l'intestin affecte les sujets immunodéprimés, à savoir les séropositifs à HIV ou les sidéens, induisant les mêmes signes cliniques que chez l'hôte normal. Nous décrivons le premier cas de diarrhée à *Cyclospora* sp. et *Cryptosporidium parvum* chez un adulte malade du SIDA.

INTRODUCTION

In 1986, Soave *et al.* reported the presence of coccidian-like bodies in the diarrhoeal stools of four American tourists who had spent some time in Haiti and in Mexico, and suggested that they might be a new enteropathogen. More recently, five AIDS patients and three international travellers with a similar diarrhoeal picture and organism were described (Hart *et al.*, 1990; Long *et al.*, 1990). Although the authors noted that the organisms resembled oocysts of coccidian protozoa, they concluded that they were photosynthetic, thylakoid algae-like structures (so-called "blue-green algae" or "cyanobacteria-like bodies"), ultrastructurally related to *Chlorella* spp. (Long *et al.*, 1990). Subsequent epidemiological studies showed that this pathogen is more widespread than previously thought, and it is prevalent in Asia, Latin America, and the Caribbean, particularly in Nepal, Peru and Mexico (Shlim *et al.*, 1991; Hoge *et al.*, 1993; Gascon *et al.*, 1993; Ortega *et al.*, 1993).

Two recent studies (Bendall *et al.*, 1993; Ortega *et al.*, 1993) have provided more evidence that the microorganism is a newly defined coccidian protozoan which had gone unrecognized in the human gut before the report of Soave *et al.* (1986), and classified it within the genus *Cyclospora*. Most reported cases have been in immunocompetent hosts, with "traveler's diarrhoea" (Shlim *et al.*, 1991; Hoge *et al.*, 1993; Gascon *et al.*, 1993; Ortega *et al.*, 1993). To date, only few cases have been reported in immunocompromised patients, specifically those with HIV infection (Hart *et al.*, 1990; Long *et al.*, 1990; Bendall *et al.*, 1993; Brandonisio *et al.*, 1993; Wurtz *et al.*, 1993).

We describe the first case of co-infection with *Cyclospora* sp. and *Cryptosporidium parvum* in an AIDS patient with diarrhoea.

CASE REPORT

The patient was a 33-yr old Italian male, shipyard worker with a long history of intravenous heroin abuse, dating back to before 1987. Between 1987 and 1989 the patient lived in Nepal. When he returned to Italy in July, 1989, he was admitted to the Infectious Diseases Unit of the Ravenna Hospital (Emilia Romagna region, central Italy) because of weight loss, profound weakness, persistent cough and malaise. He was found to be

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Fig. 1 - *Cyclospora* oocyst (arrow) and some others of *C. parvum* (arrowheads) are seen in stool sediment (Ziehl-Neelsen mod., $\times 1000$).

Fig.2 - Several immature *Cyclospora* oocysts in feces. Note the typical internal granulations and the thick wall (Nomarski interference contrast, $\times 1000$).

HIV-positive and to have bilateral cavitary pulmonary tuberculosis (TB), pityriasis of the hair, scabies and seborrheic dermatitis of the face. The patient appeared chronically ill and had lost 15 Kg; he was anemic (hemoglobin : 11 g/100 ml; RBC : 4,000,000/mm³) and had a CD4 count of 149/mm³, with a CD4/CD8 ratio of 0.16. Six months anti-TB therapy (rifampicin, isoniazide and ethambutol) and AZT (500 mg twice daily) resulted in resolution of the radiographic findings and an overall clinical improvement for the patient (CD4 > 400/mm³). The patient felt quite well until May, 1992, when he was re-admitted to the same hospital because of *P. carinii* pneumonia (PCP) and diarrhoea (5-6 bowel movements/day) of one month's duration. Stool specimens examined by two of us (M.S. and S.G.) with an acid-fast stain showed a mixed intestinal infection with both *Cyclospora* sp. and *C. parvum* oocysts (fig. 1). The CD4 count was 9/mm³.

Treatment with co-trimoxazole (120 mg/Kg/day for three weeks) for PCP and with spiramycin (9,000,000 IU/qd for one month) for cryptosporidiosis was started. The patient had a good clinical response both of pulmonary and intestinal symptoms. He had a reduction in diarrhoea frequency (1-2 bowel movements/day), despite persistence of the two pathogens in the stool and he was discharged. In September, 1992, after a brief admission to the Piacenza Hospital (Northern Italy), he was re-admitted to the Infectious Diseases Unit of the Ravenna Hospital because of CMV retinitis, persistent diarrhoea and mass lesions on a brain CT scan, probably due to *Toxoplasma gondii* infection. Empiric therapy with pyrimethamine (75 mg/day) and clindamycin (600 mg qd) was started, with progressive regression of symptoms and of the CNS lesions seen on CT scan.

A repeat stool examination was positive for *Cyclospora* sp. oocysts (fig. 2) but negative for *C. parvum*. An intestinal biopsy was not performed.

The patient died suddenly of a cardiac arrest on November 18, 1992. Autopsy was not permitted.

DISCUSSION

Chronic diarrhoea is a common problem in symptomatic HIV-infected subjects (Bartlett *et al.*, 1992). The spectrum of microorganisms potentially responsible for diarrhoea is widening; however, particularly in tropical areas, several cases are due to coccidian protozoa such as *Cryptosporidium parvum* and *Isospora belli* (Soave and Johnson, 1988).

Recently, in some cases of diarrhoeal enteritis in HIV positive patients (Hart *et al.*, 1990; Long *et al.*, 1990;

Bendall *et al.*, 1993; Brandonisio *et al.*, 1993; Wurtz *et al.*, 1993), the etiological agent has been identified as a new human coccidian of *Cyclospora* genus according to Ortega *et al.* (1993), and previously named "cyanobacterium-like" or "alga-like" by other authors (Long *et al.*, 1990 and 1991). To our knowledge, this is the first reported case of co-infection with *Cyclospora* sp. and *C. parvum* in an AIDS patient.

To date, the vast majority of human infection due to *Cyclospora* sp. has been reported in immunocompetent travellers and in foreigners residing in some tropical areas such as Asia (Nepal), the Caribbean and Latin America (Peru, Mexico) (Shlim *et al.*, 1991; Hoge *et al.*, 1993; Gascon *et al.*, 1993; Ortega *et al.*, 1993). An autochthonous epidemic outbreak in the USA among the medical and nursing staff working at an hospital of Chicago (WHO, 1991) and other sporadic cases elsewhere were also reported (Bendall *et al.*, 1993; Brandonisio *et al.*, 1993).

From the epidemiologic point of view, it is significant that our patient lived in Nepal for about two years. He complained abdominal symptoms only after several months from the return in Italy, when he was severely immunocompromised. This could mean for *Cyclospora*, as for other opportunistic AIDS-defining infections (Smith *et al.*, 1992), the re-activation of a latent infection due to a cell-mediated immunodeficiency.

Both in the immunocompetent and in the immunocompromised host, *Cyclospora* infection is usually associated with watery "intermittent" diarrhoea that tends to be self-limited, lasting weeks or months, even in AIDS patients.

In our patient it was impossible to evaluate temporary clinical remissions during *Cyclospora* infection, because of the presence of *C. parvum*. On the other hand, *Cyclospora* oocysts have been still recognized in patient stools five months after the first positive fecal identification.

It is likely that the treatment schedule with spiramycin and co-trimoxazole temporarily improved the diarrhoea due to these coccidia, in relation with the partial efficacy of spiramycin on *C. parvum* (Georgiev, 1993) and the preliminary data of the effectiveness of co-trimoxazole on *Cyclospora* both in immunocompetent (Madico *et al.*, 1993) and immunocompromised patients with AIDS (Wurtz *et al.*, 1993).

We also believe that the negative stool examination for *C. parvum* few days before the patient's death would be critically evaluated, in the light of the well-known irregular oocyst shedding and of the low sensitivity of the temporary and permanent staining methods (Weber *et al.*, 1991). Moreover, an intestinal

biopsy to confirm a complete parasitological cure was not performed.

In conclusion, clinicians and laboratory operators should be alerted that *Cyclospora* sp. is a new etiological agent of diarrhoeal enteritis both in immunocompetent and in immunodeficient humans. As in our patient, the possibility of a mixed infection with other intestinal pathogens, namely *C. parvum*, should also be considered: in these cases the careful microscopic evaluation of oocyst morphology in fresh and acid fast stained specimens (Long *et al.*, 1991) is an useful diagnostic method for differentiating these coccidia.

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