Intestinal helminths : a clue explaining the low incidence of inflammatory bowel diseases in Subsaharan Africa ? Potential benefits and hazards of helminth therapy

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Abstract

In their review, the authors state that the very low incidence and prevalence of IBD in sub-Saharan Africa cannot be explained by genetic factors since in Black populations of the USA and UK, the incidence of these diseases is approaching that of the white populations.

Beside helminths whose intestinal infestation is frequent in sub-Saharan Africa, other micro-organisms such as atypical mycobacteria, lactobacilli, etc, might have been reduced in Western population. This is a new variant of the Hygiene hypothesis. After Rook *et al.*, these micro-organisms were acting as adjuvants for induction of T regulatory cells which, associated with antigen-presenting cells secrete IL-10 and TGF-β, inhibiting the maturation of CD4 T cells to Th1 and Th2 effector cells, and consequently reducing the occurrence of Th1-mediated diseases like Crohn's disease and Th2-mediated diseases like ulcerative colitis.

The effects of intestinal helminths on host immunity have been studied in Ethiopian Jews emigrated to Israël. Thorough studies before and after deworming have demonstrated that chronic helminth infestation provokes a state of chronic immune activation with anergy, reversible after deworming.

Administration of ova of *Trichuris suis*, an helminth non pathogenic in man, has given encouraging results in the treatment o Crohn's disease and ulcerative colitis with a good safety record but long-term trials are needed since the potentially harmful effects of helminths on immunity. (Acta gastroenterol. belg., **2006**, 69, **418-422**).

Key words : Inflammatory bowel disease, Crohn's disease, ulcerative colitis, hygiene hypothesis, helminths, immunology, regulatory T lymphocytes.

Introduction

Following the Belgian IBD Immunology Meeting (Spa 2005), T. Moreels and P. Pelckmans (1) have reviewed the arguments explaining the lower incidence of Inflammatory Bowel Diseases (IBD) in developing countries versus developed countries by the *hygiene hypothesis*. As other authors, they have hypothesized that the high prevalence of chronic helminth infections in developing countries could reduce the immunological reactivity of the intestine towards its microflora, explaining the lower risk of eclosion of IBD. As confirmed by original studies of the authors, administration of helminths in different models of experimental colitis prevents or attenuates lesions mimicking IBD. Moreover, helminth therapy of Crohn's disease (CD) and ulcerative colitis (UC) has yielded encouraging results (2,3).

The aim of our review is to reevaluate the *hygiene hypothesis* by analyzing the impact of other factors than helminths on the pathophysiology of intestinal immunity, to report some immunological studies related to the effect of intestinal helminths on the host immunity in particular in migrating populations, and finally to discuss the potential benefits and hazards of helminth therapy.

Impact of other factors than helminths on the pathophysiology of intestinal immunity

In sub-Saharan Africa the very low incidence and prevalence of CD and UC cannot be explained only by genetic factors since the incidence of these diseases among Africans living since a long time in Western Countries is approaching that of white populations following studies reviewed by Loftus (4). There was indeed a similar incidence of IBD among African American children in Atlanta, Georgia (5). In Southern California, the prevalence of CD in the black population was about two thirds the prevalence in the white population (6). In Derby, UK, Fellows *et al.* (7) have demonstrated that the risk of CD was similar among whites and immigrant African-Carribean adults.

The role of environmental factors explaining these geographic differences was also suggested by the observation that over the last 50 years of the 20th century in Western countries, there was a concomitant decline of the incidence of prototypical infectious diseases (hepatitis A, measles, mumps, rheumatic fever, tuberculosis) and the rise of incidence of immune disorders such as IBD, asthma, multiple sclerosis, type 1 diabetes (8). The *hygiene hypothesis* was thus proposed with the possible roles of some classes of T-lymphocytes : Th1, Th2 and regulatory T cells (Treg) (8).

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A reinterpretation of the *hygiene hypothesis* has been proposed recently by Rook *et al.* (9,10). Instead of the unproved role of promoting domestic hygiene (detergents, etc), these authors have hypothesized the protective role of relatively harmless microorganisms : intestinal helminths, saprophytic mycobacteria and lactobacilli. These microorganisms could be transmitted in children, for example helminths in developing countries, others by contact with animals in farms, with pets, etc.

The possible protective role of intestinal helminths against IBD has been discussed in the preceding article (1). Although the prevalence of intestinal helminths in developing countries is very high, there are geographic variations. For example in Cameroon, in a survey of Ascariasis and trichuriasis among > 22.000 children of 512 schools Ratard *et al.* (11) found that the prevalence of helminths was high in two equatorial zones with different climates (50-85% for *A.lumbricoides* and 70-95% for *T. Trichura*) but was very low (< 5%) in the tropical zone.

The immunological mechanisms of prevention of IBD and other auto-immune diseases related to harbouring intestinal helminths are still hypothetical although some progress has been made recently in unraveling some pathways. The first hypothesis was a shift of immunological reactivity from the Th1 type, favouring Crohn's disease (CD), multiple sclerosis, diabetes type 1 toward a Th2 type under the influence of chronic helminth intestinal infestation. But ulcerative colitis (UC) and allergic diseases are related to an immunological reactivity of the Th2 type and could also be favorably influenced by intestinal helminths.

The role of regulatory T cells (Treg) has been proposed by Bach *et al.* (8). These T reg, a subset of CD4 + T cells have been discovered in 1997 by Groux *et al.* (12) in a model of experimental colitis in mice. In their model, these cells have suppressed antigen-specific immune responses and have downregulated the pathological immune responses *in vivo*. Many subsequent studies have confirmed the immunoregulatory role of Treg in experimental models of colitis. For example Veltkanp *et al.* (13) have shown that in transgenic mice for human CD3ε gene, bone marrow transplantation (BMT) of a pool containing regulatory CD4 + CD25 + cells reversed imbalances in the T cell pool and prevented colitis ; the latter could be induced by BMT of a pool containing CD4 + CD25-cells. Fantini *et al.* (14) demonstrated that Treg generated in cell culture in the presence of TGF-β suppressed Th1 mediated colitis induced in SCID mice after transfer of CD4 + CD62L + cells.

The different types of T reg originating in the thymus or in the periphery (15) have been described in a preceding article (16). Expression of the transcription factor FOXP3 is a marker more specific than CD25 (17).

Following the development of the knowledge concerning regulatory T cells (Treg), Rook *et al.* (9,10) have proposed an unifying hypothesis explaining the 20th century rise of Th1 mediated- and Th2 mediated-autoimmune diseases by a defective maturation of Treg and regulatory antigen presenting cells (Fig. 1). The microorganisms already mentioned : helminths, mycobacteria, lactobacilli, considered as "old friends", would have protected Homo sapiens during its evolution

by modulating the reaction of dendritic cells originating from the innate immune system against allergens, self and gut contents. Instead. of driving effector cells toward a Th1 or Th2 mode of aggressive immune responses like with pathogens, the dendridtic cells modulated by "old friends"would drive effector cells to the regulatory mode of reaction, through the secretion of the cytokines IL-10 and TGF-β which down-regulate T effector cells.

A study of Strauch *et al.* (18) supports the hypothesis of Rook *et al.* (9,10). In a model of colitis in severe combined immunodeficient (SCID) mice, transfer of CD4 + CD62L + lymphocytes from germ free donor mice provoked an earlier onset of colitis than after transfer of CD4 + CD62L + lymphocytes from conventionally housed animals. The higher percentage of CD4 + GITR + expressing lymphocytes and the production of IL-10 after transfer of the lymphocytes from conventionally housed mice suggested the presence of Treg cells, which were absent in germ free mice. The authors concluded that bacterial antigens are crucial for the generation and/or expansion of Treg cells.

To our knowledge, there is only one study on Treg in IBD

patients. Maul *et al.* (19) have shown that Treg cells increased in peripheral blood in patients in remission but decreased during active disease, whereas they were increased in acute diverticulitis. In inflamed IBD mucosa, there was an increased number of Treg in comparison with normal mucosa.

Immunological studies related to the effect of intestinal helminths on the host immunity, particularly in migrating populations

Infection with the filaria Onchocherca volvulus (Ov)

This infection affecting about 20 million people in tropical regions and considered as the major cause of blindness in these regions has two forms of presentation after Doetze *et al.* (20) : some subjects despite heavy exposure remain free of infection and are thus putatively immune (PI), others develop a generalized onchocerciasis (GEO). By studying the response of peripheral blood mononuclear cells (PBMC) to the Ov Antigen, these authors have demonstrated that the hyporesponsiveness to the helminth in GEO patients was not mediated by a Th1 to Th2 shift but by IL-10 and TGF-β, the two cytokines associated with a Th3 response (regulatoryT cells) as already mentioned. This hyporesponsiveness was not present in PI individuals. The

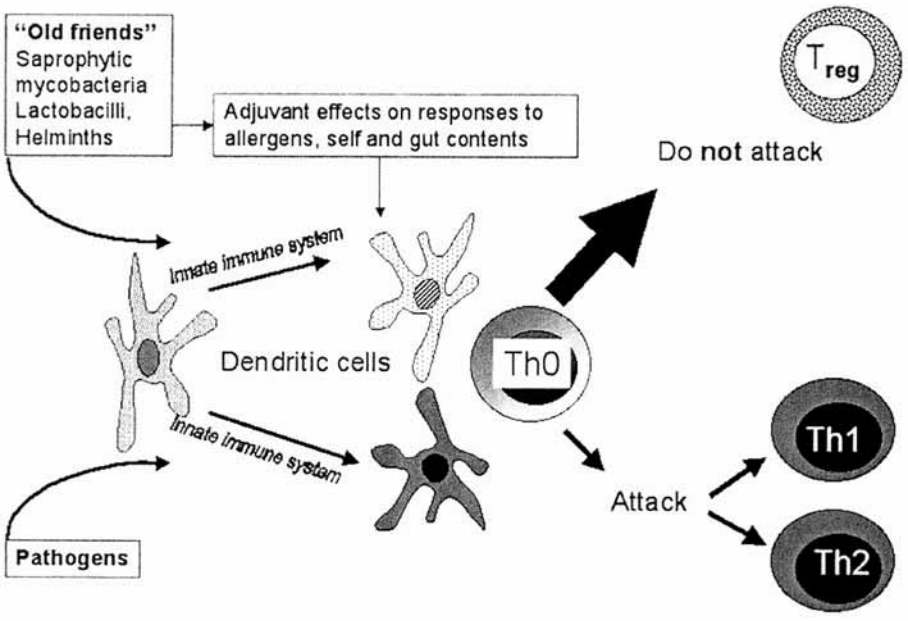

Fig. 1. — Actions of and harmless «old friends» and pathogens on the innate immune system with activation of antigen-presenting cells (Dendritic cells) which drive either regulatory T cells or effector cells (Th1 and Th2).
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authors could clone antigen-specific T regulatory cells from peripheral mononuclear cells of patrients with GEO.

Studies in Ethiopian Jews after emigration to Israel

A high prevalence of helminthic infections has been found in the population of Ethiopian Jews who emigrated to Israel in the eighties : one helminthic parasite in 80%, two in 40% and up to four in 3%. Borkow *et al.* (21) did an extensive immunological study in a group of Jews recently emigrated from Ethiopia ("New" Ethiopian Jews") who were compared to a group of Ethiopian Jews living in Israel since 3-10 years and who underwent likely a deworming ("Old" Ethiopian Jews), and to a group of Israeli-born individuals. The main results of the immunological investigations are summarized in Table 1.

Intestinal helminths were still found in 15% of the "old" Ethiopian Jews which explains significantly higher eosinophilia and IgE levels in this group as well in the group of of "new" Ethiopians compared to Israeli-born individuals. The CD4/CD8 ratio was reduced only in "new" Ethiopian Jews. There were significant differences of immunological parameters in the group of "new" Ethiopian Jews in comparison with the group of Israeli-born individuals without intestinal helminths : increased percentage of HLA-DR + / CD3 + cells, which indicated more activation of Tcells, and poor. transmembrane signal transduction. In "new"Ethiopian Jews infested by helminths, the percentage of positive Mantoux tests (intracutaneous PPD skin injections) was low and increased significantly 6 months after deworming.

In presence of these findings as well as those of other elaborated tests before and after deworming, the authors (21) have concluded that chronic intestinal helminth infestation causes persistent immune activation with anergy and therefore reduces to immunological defense toward infections and could reduce. the immunological response to vaccines.

Recommendations of the World Health Organization (WHO) about deworming

Following a meeting of the Partners for Parasite Control at the WHO in Geneva at the end of 2004, an editorial of the Lancet (22) summarized evidence-based. recommendations : Helminth control is crucial toward improving health in developing countries following several arguments given in Table 2. Moreover the drugs for mandatory deworming are cheap and effective : a single tablet of

albendazone or mebendazole (0.02 US $) + a single dose of praziquantel (0.20 US $) once or twice a year. Praziquantel is indicated owing the high prevalence of schistosomiasis.

Potential benefits and hazards of helminth therapy

The two important articles of Summers *et al.* (2,3) about *Trichuris suis (T. suis)* therapy of IBD, commented in the preceding article (1), have shown that a higher

Table 1. — **Main results of immunological evaluation in two groups of Ethiopian Jews compared to Israeli-born individuals, studied by Borkow *et al.*** (*J. Clin. Invest.*, 2000, **106** : 1053-60)

	Israeli-born Individuals (N31)	"New" ethiopian jews (N111)	"Old" ethiopian jews (N48)
Time in Israel		< 12 months	3-10 years
Intest. helminths	0%	> 80%	15%
Eosinophils (/µl)	127 +/- 8.4	748 +/- 58.6***	322 +/- 40.7**
IgE (IU/ml)	105 +/- 23.3	1650 +/- 125***	203 +/- 31,5*
CD4/CD8 RATIO	1.38 +/- 0.07	1.06 +/- 0.04**	1.38 +/- 0.06
%HLA-DR+/CD3+	4.9 +/- 0.4	10.3 +/- 0.75***	5.9 +/- 0.55
Transmembrane signal transduction	Normal	Poor	
Mantoux tests		More (+) responses 6 mo post-deworming (P < 0.001)	

Statistical differences between the Ethiopian groups and the Iraeli born individuals : *P < 0.05 **P < 0.01 ***P < 0.001.

Table 2. — **Recommendations of the World Health Orgnization about deworming*** (*Lancet 's Editorial*, 2004, **364** : 1993-4)

Helminth control* is crucial toward improving health in developing countries :
• improves children's nutrition, growth and intellectual development
• reduces children's vulnerability to other infectious diseases (malaria, etc)
• improves maternal survival (prevention of severe anaemia) and reduces perinatal deaths
• slows down the progression from HIV to AIDS

*A single tablet albendazole or mebendazole + a single dose of praziquantel once or twice a year (cheap and effective drugs).

rate of response was observed in Crohn's disease patients but it was an open-label study (2). In the randomized controlled trial of ulcerative colitis therapy (3), the rate of response was modest but significantly higher (43%) in the *T. suis* group than in the placebo group (16%). Other trials under way will confirm or invalidate these first encouraging results and the absence of adverse events.

The issue of long term safety of this still experimental therapy will also benefit from further trials. Since chronic helminth infection may alter immunity following the findings of the Israeli authors (21) and the warnings of the WHO already mentioned, untoward side effects might occur, as they occur with immunosuppressive and immunomodulatory therapy of IBD. In a letter to Gastroenterology, Hsu and Tseng (23) were concerned about the case-report of life-threatening colitis associated with *Campylobacter jejuni* and concomitant T. suis ova in feces (24). Summers *et al.* stated that it was unclear in this report (24) that the patient was infested by *T. suis* or *T. Trichiura.* Hsu and Tseng were also concerned by the fate of the ulcerative patients non-responding to *T. suis* in the absence of subsequent anti-helminth therapy. Summers *et al.* responded that subsequent eradication of *T. suis* was unnecessary since it is not a human parasite and it is spontaneously eliminated over a few weeks, even in the immunocompromized host.

Conclusions

The epidemiological research about the causes of the discrepancy of incidence and prevalence of IBD between countries of sub-Saharan Africa and Western countries led to interesting hypotheses about the role of helminths and perhaps other microorganisms in regulating the immunological homeostasis of the intestine and preventing Th1- and Th2-mediated diseases.

Consequently, the knowledge of the important role of regulatory T cells for immunoregulation has progressed (25). Treg therapy is promising and has already started in transplanted patients (26). Helminth therapy might be an indirect way to stimulate the production of Treg. Owing to numerous successful studies on the prevention and attenuation of experimental colitis by administration of intestinal helminths, trials on the effect of ova of *Trichuris suis* have started with encouraging results and a good safety record. However long- term studies are needed to confirm the efficacy and also the safety of this new therapy, since intestinal infestation by helminths does modify immunity.

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