

***Mycobacterium ulcerans* disease: role of age and gender in incidence and morbidity**

M. Debacker¹, J. Aguiar², C. Steunou², C. Zinsou^{1,2}, W. M. Meyers³, J. T. Scott¹, M. Dramaix⁴ and F. Portaels¹

¹ Institute of Tropical Medicine, Antwerp, Belgium

² Centre Sanitaire et Nutritionnel Gbemoten, Zagnanado, Benin

³ Armed Forces Institute of Pathology, Washington, DC, USA

⁴ Ecole de Santé Publique, Brussels, Belgium

Summary

During the 5-year period, 1997–2001, 1700 patients with a clinical diagnosis of *Mycobacterium ulcerans* disease [Buruli ulcer (BU)] were treated at the Centre Sanitaire et Nutritionnel Gbemoten, Zagnanado, Benin. The patients lived in the four regions of southern Benin: Atlantique, Mono, Oueme and Zou, with the largest number coming from the Zou Region where the centre is located. The median age of BU patients was 15 years (q1 = 7, q3 = 30). Lower limbs are involved 3.2 times more frequently than upper limbs in older patients and younger patients have the highest prevalence of multiple lesions. The latter are frequently associated with bone lesions. Specific detection rates for age and gender showed a distribution with maximum peaks in the 10–14 years group and among adults between 75 and 79 years. Over 59 years, males are more at risk of developing *M. ulcerans* disease than females. Children under 15 years represent the largest part of the BU disease burden and of the general population. The highest detection rates (per 100 000 population) were in the 75–79-year-old patients. The most likely explanation of this was reactivation of disease from a latent infection of *M. ulcerans*. Educational programmes should target especially these two groups of population at risk.

keywords *Mycobacterium ulcerans*, Buruli ulcer, Benin, age, gender, osteomyelitis

Introduction

Increasing numbers of Buruli ulcer (BU) patients have been reported during the last few years, especially in West Africa (World Health Organisation (WHO) 2000; Debacker *et al.* 2004). The infectious agent, *Mycobacterium ulcerans*, is an environmental mycobacterium, which produces a necrotizing toxin that diffuses and provokes tissue destruction far beyond the inoculation point. Different forms of the disease (nodules, oedema, plaques and ulcers) are directly related to the delay in seeking medical advice (Debacker *et al.* 2004). *Mycobacterium ulcerans* also causes osteomyelitis, leading sometimes to amputation (Lagarrigue *et al.* 2000; Portaels *et al.* 2003). Surgery, the recommended treatment (WHO 2001 a), often leads to long periods of hospitalization with serious socioeconomic (Asiedu & Etuaful 1998) and psychosocial implications (Stienstra *et al.* 2002; Aujoulat *et al.* 2003).

Although large number of patients have been reported, the epidemiology of BU remains obscure, even in endemic countries. In 1997, a first report was published on 867 BU

patients from the Centre Sanitaire et Nutritionnel Gbemoten (CSNG), Republic of Benin, for the period 1989–96 (Aguiar *et al.* 1997). Our study covers the patients admitted to the CSNG in the ensuing 5 years (1997–2001) during which a collaborative project was initiated to improve detection and control of BU. This study aims to describe BU in Benin, building on our previous work (Debacker *et al.* 2004), focusing on the influence of age and gender factors in BU patients.

Patients and methods

Data collection

This study is based on observations on all 1700 patients clinically diagnosed as BU and admitted to the reference centre for BU treatment, the CSNG, from 1997 through 2001, at Zagnanado in the Zou Region, Benin. Age, gender, origin, date of disease onset as reported by the patient, date of diagnosis, duration of hospitalization, clinical characteristics and evolution of the disease were recorded.

Selection of Buruli ulcer patients

The WHO clinical criteria for suspecting BU were used at the CSNG (WHO 2000). Thus, BU was suspected in patients with one or more of the following features:

- presence of a chronically developing lesion (several weeks or months), i.e. 'a wound that will not heal';
- typical nodular, indurated plaque or oedematous cutaneous lesion: one or more painless chronic ulcers with undermined edges, or a depressed scar;
- swelling over a painful joint, suggesting bone involvement.

The following ancillary criteria should be considered:

- no fever or regional lymphadenopathy;
- patient is under 15 years old;
- patient lives or travelled in an endemic area.

Between 1997 and 2001, the yearly admissions were, respectively, 299, 355, 403, 301 and 342 patients, giving a total of 1700 admissions. In 13 of these admissions, BU was in the initial clinical differential diagnosis for 13 patients who were later confirmed to have another disease (Debacker *et al.* 2004) and were excluded from the study. There were recurrences of BU after discharge from CSNG in 57 patients (3.3%). These were removed from the study as they constituted a particular group of patients with long hospitalization times and/or many episodes of the disease. Thus, a total of 70 patients were excluded, leaving 1630 patients for analysis. No patient was included more than once. A recurrent case is defined as a patient with a previous history of BU who presents with another lesion at the same or different site of the body within 1 year of apparently successful treatment (WHO 2000) and discharge from hospital.

Demographic data

Demographic data of the general population were based on the most recent national census (Ministère du Plan et de la Restructuration Economique 1992). Additional statistical information came from the Benin Demographic and Health Survey (1996). For the 5-year period, 1997–2001, demographic data were extrapolated from 1992 statistics assuming an annual 3.2% growth rate, corrected by projections of the National Institute of Statistics and Economic Analysis of Benin (1996).

Diagnostic procedures

Specimens of tissue and exudates from 906 patients were analysed to confirm the clinical diagnosis. One or more of

the following examination were performed: direct smear examination for acid-fast bacilli (AFB), culture, IS2404-PCR and histopathological examination (WHO 2001b). The remaining 724 patients were diagnosed clinically: all were typical of BU, and did not present reasonable differential diagnostic problems. All BU patients whose disease was confirmed by laboratory analyses were positive for at least one bacteriological test, or by histopathology evaluation.

Statistical analysis

Data were analysed by EPIINFO (Centers for Disease Control and Prevention, Atlanta, GA) and SPSS vrs 9.0 (SPSS, Chicago, MI, USA) for windows. Contingency tables were analysed by the Pearson chi-square test. Incidence rates (IR), relative risks (RR), incidence ratios (IR) and their confidence interval (CI) were calculated. Cases were excluded from each analysis when information was missing for a specific variable. A total of 1630 patients satisfied the aforementioned criteria for inclusion in the study as BU; however, age was not recorded for 25 patients and gender was missing for eight patients.

Results

Place of residence of patients

Most patients came from the Region of Zou (66.3%), where the CSNG is located, followed by the Region of Oueme (17.4%), Atlantique (9.5%) and Mono (6.0%) (Figure 1). Twelve (0.7%) were from neighbouring countries (Nigeria, Togo, Côte d'Ivoire and Ghana) (Debacker *et al.* 2004). The median age was 15 years ($q_1 = 7$ years, $q_3 = 33$ years), the age range 2 months to 83 years. The gender ratio (M/F) of the entire sample was 1.01.

Incidence rates in the Zou Region

Incidence rates were calculated only for the Zou Region because most patients (66.3%) came from this region. Figures 2 and 3 show the incidence rates of BU by age, gender and period of diagnosis. In males and females, a higher than expected percentage of BU patients over 59 years of age was observed. In this group, an increase in the age-specific detection rates of BU was found between 1997 and 2001. For the two other age groups, the incidence rates decreased during this period. Specific detection rates (/100 000) by age and gender for patients coming from the Zou Region from 1997 to 2001 are presented in Figure 4. The distribution peaked in the



Figure 1 Map of Benin with the four Buruli ulcer (BU) endemic regions: the Region of Zou (Z), the Region of Atlantique (A), the Region of Mono (M) and the Region of Oueme (O).

10–14-year-old group for patients under 45 years of age, and in those 75–79 years old in over 45 year olds. Patients over 59 years of age were 3.15 times more at risk of developing BU than 15–59 year olds (RR: 3.15, CI: 2.68–3.71, $P < 0.001$). The 0–14-year-old group had almost the same risk as the 15–59 year group of developing BU (RR: 1.19, CI: 1.07–1.32, $P = 0.001$). There was no difference in the risk for males and females to develop BU disease under age 59 years. Beyond the age of 59, males were 1.49 times more at risk than females (RR: 1.49, CI: 1.12–1.98, $P < 0.006$).

Localization of the lesions by age and gender

The site and frequency of lesions by age group are given in Table 1. There were no significant differences between the frequency of lesions on the right or left limbs (data not shown). Lower limbs were involved 3.2 times more often than upper limbs in older patients (15 years and oldest). Children (0–14 years) tended to develop the disease more often on the trunk, head and neck (IR: 2.65, CI: 2.07–3.40, $P < 0.001$) and upper limbs (IR: 1.19, CI: 1.01–1.41, $P = 0.041$). Lesions at other sites such as the genitalia were not frequent enough to establish trends. No lesions were observed on the palms or soles.

Males had more lesions on lower limbs than females (IR: 1.12, CI: 1.04–1.21, $P = 0.002$) while upper limbs were more frequently involved in females (IR: 1.26, CI: 1.07–1.49, $P = 0.006$). Under 15 years, males had more lesions on lower limbs than females (IR: 1.17, CI: 1.07–1.28, $P < 0.001$). Over 14 years, there are more females than males with lesions on the trunk, head and neck (IR: 1.82, CI: 1.14–2.91, $P = 0.011$).

Number of Buruli ulcer sites and age

The majority of patients (91.0%) had lesion(s) only at one body site (Table 2). Patients <30 years had the highest prevalence of lesions at multiple sites (10.7%). The difference is statistically significant compared with patients ≥ 30 years ($P < 0.001$). The relation between number of cutaneous lesions and bone involvement is shown in Table 3. The percentage of patients with bone lesions increased with the number of BU skin sites. Dissemination of BU in the skin seems to be directly related to the development of bone lesions.

Discussion

Aguiar *et al.* (1997) reported passive detection rates for 867 BU patients in southern Benin for the period 1992–96. This study supplements their data by analysis of BU patients seen at the same medical centre during the 5 successive years (1997–2001). Cases described here represent only those patients detected passively at the same highly experienced centre for BU treatment (CSNG). The CSNG is the most widely recognized reference centre for BU treatment and treats the largest number of BU patients in Benin. To our knowledge, the only other centre for the treatment of BU in Benin during the period of this study was the centre of Lalo. This centre, situated in the Mono Department, treated approximately 400 patients during the period 1997–2001, and no data on these patients are included in our study. Other centres were not treating BU

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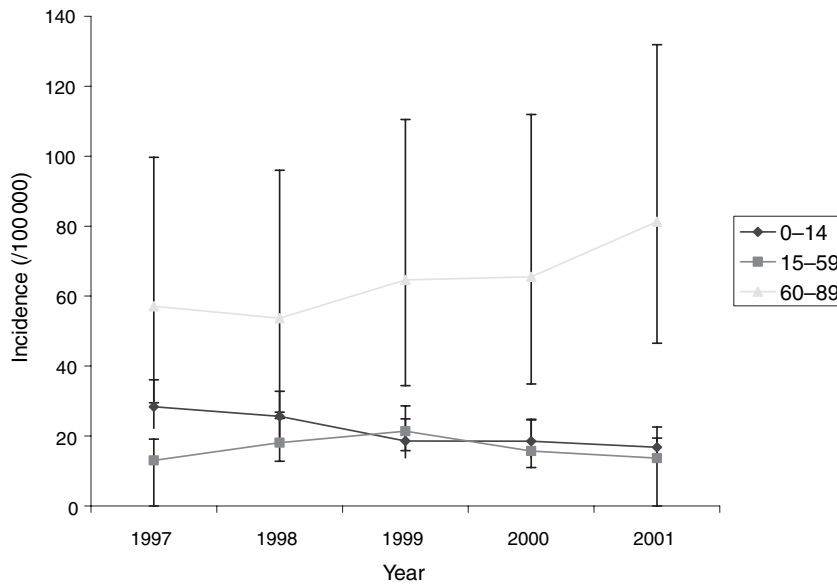


Figure 2 Age-specific detection rates of Buruli ulcer (BU) in Zou males by year. The vertical bars represent 95% confidence intervals.

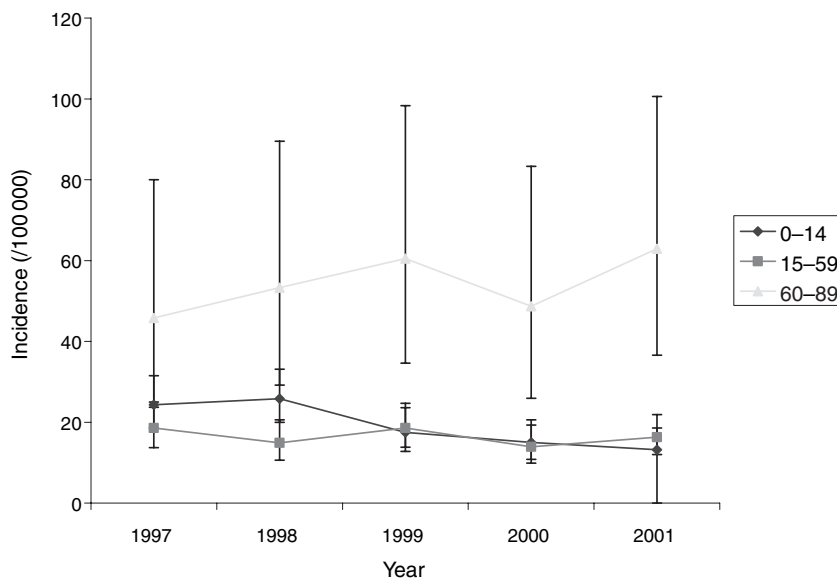


Figure 3 Age-specific detection rates of Buruli ulcer (BU) in Zou females by year. The vertical bars represent 95% confidence intervals.

during this period and referred some BU patients to the CSNG.

Data collected from 1997 to 2001 indicate that the CSNG receives patients mainly from the Zou Region, where the centre is situated. Patients seem to choose the CSNG for a variety of reasons including accessibility, financial concerns and cultural compatibility. Health-seeking behaviours may have influenced our results because

BU is often viewed as being associated with witchcraft, and patients often initially consult a traditional healer (Aujoulat *et al.* 2003). Financial burden is an important factor for BU involving primarily the cost of transportation and treatment. Because hospitalization for BU is frequently prolonged, the patient and the caregiver (usually a family member) may be away from home for long periods, making the purchase of food a decisive burden. Other

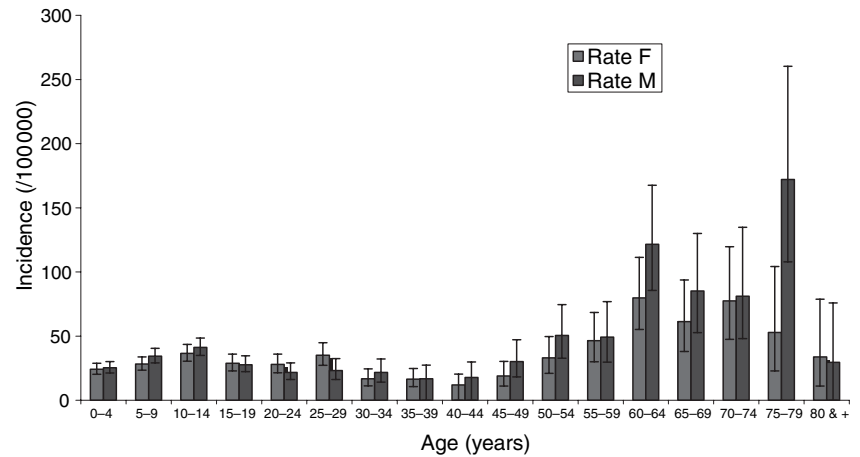
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Figure 4 Specific detection rates by age and gender in the Zou Region between 1997 and 2001.

Table 1 Localization of lesions by age and gender for all admissions

	Age groups (years)					
	0-14	15-29	30-44	45-59	60-74	75-89
Males						
Lower limbs	256 (53.4)	126 (68.1)	43 (76.8)	47 (72.3)	67 (88.2)	23 (92.0)
Upper limbs	111 (23.0)	43 (23.2)	11 (19.6)	13 (20.0)	7 (9.2)	2 (8.0)
Trunk	86 (18.0)	14 (7.6)	2 (3.6)	3 (4.6)	1 (1.3)	
Head/neck	20 (4.2)	2 (1.1)		1 (1.5)	1 (1.3)	
Genitalia	6 (1.3)			1 (1.5)		
Total	479	185	56	65	76	25
Females						
Lower limbs	206 (47.8)	119 (52.6)	43 (61.4)	55 (82.1)	62 (82.7)	11 (84.6)
Upper limbs	124 (28.8)	74 (32.7)	18 (25.7)	8 (11.9)	10 (13.3)	1 (7.7)
Trunk	73 (16.9)	28 (12.4)	8 (11.4)	4 (6.0)	2 (2.7)	1 (7.7)
Head/neck	28 (6.5)	5 (2.2)			1 (1.3)	
Genitalia			1 (1.4)			
Total	431	226	70	67	75	13

Table 2 Comparison of number of Buruli ulcer (BU) lesion sites and age for all admissions

Cutaneous sites with lesions	Age groups (%)						Total (%)
	0-14	15-29	30-44	45-60	60-74	75-89	
1 site	711 (89.3)	327 (89.1)	113 (93.4)	127 (96.9)	147 (96.7)	36 (94.7)	1461 (91.0)
2 sites	52 (10.7)	22 (10.9)	6 (6.6)	2 (3.1)	5 (3.2)	1 (5.3)	88 (5.5)
3 sites	15	10	2	2		1	30 (1.9)
4 sites	9	6					15 (0.9)
5 sites	9	2					11 (0.7)
Total	796	367	121	131	152	38	1605

Age of 25 patients was not recorded.

Table 3 Bone and cutaneous lesions in disseminated Buruli ulcer (BU)

Cutaneous sites with lesions	Patients with bone lesions (%)	Patients with only cutaneous lesions (%)	Total
1 site	142 (9.7)	1323 (90.3)	1465
2 sites	43 (48.3)	46 (51.7)	89
3 sites	14 (48.3)	15 (51.7)	29
4 sites	11 (78.6)	3 (21.4)	14
5+ sites	3 (37.5)	8 (62.5)	11

For 22 patients, the type of lesion was not recorded.

factors that may influence passive BU incidences either positively or negatively at the health centre (Debacker *et al.* 2004) are:

- the marked development of treatment facilities at the CSNG during the period covered by the study, and the fact that more BU patients had become aware of the effective therapy offered by the CSNG;
- the operation of rural public health training and publicity programmes: population surveys were conducted in 1999 for the future development of treatment centres in other regions, and all cases detected during these surveys were referred to the CSNG for treatment;
- significant cyclic environmental changes such as excessively dry or wet periods that differ from region to region;
- patients' fear of surgery and the ready access to practitioners: patients may initially prefer traditional therapists who do not perform surgery;
- the diminishing reservoir of untreated patients achieved by the programme;
- occasional reductions in contamination with *M. ulcerans* of endemic environmental sites: for example, we found that some endemic sites in the district of Ouinhi often became less positive for *M. ulcerans* DNA (F. Portaels, J. Aguiar, C. Steunou *et al.*, unpublished data).

Incidence rates were calculated only for the Zou Region because most patients came from this region. The specific incidence rate by age gives two peaks: a large one in adults (older than 45 years) and a smaller one among children and young adults (<45 years old). This finding differs from the usual perceptions in the field where young boys are considered most at risk of BU. The high rate among older people, especially the 75–79-year-old group, may be related to down-regulation of the immune status permitting reactivation of latent disease. In such a scenario, trauma may activate latent *M. ulcerans* infection (Meyers *et al.* 1974). The primary infection can go unnoticed, especially

if it is confined to a papule or a self-healing nodule. Younger people have closer contact with environmental sources of the aetiological agent and are usually more scantily attired. Although adults aged between 15 and 59 years are frequently exposed to wetlands through their daily activities, their specific detection rates were lower. One factor may be that young adults have a more effective protective immunity.

In the age group >59 years, Zou males are more at risk to develop BU than females; but there is no difference in risk between males and females in people under age 59. Factors that may promote the preponderance of men in this group include work-related activities and greater access to health care. In Ghana, Stienstra *et al.* (2002) investigated beliefs and perceptions of BU patients compared with a control group. Although they found that BU was a stigmatizing disease, there was no difference in the impact the disease had on men compared with women. Such studies have not been carried out in other countries.

In general, in the Zou Region, there is an increase in incidence rates of BU in males and females in >59 year olds. The elderly are more apt to follow traditional practices, for example, they refuse vaccinations and fear surgery. Such patients are often influenced by sorcery with unfortunate results (lesions do not heal or relapse). Adults traditionally are more prone to go to the traditional healers. However, compared with other studies we observed a higher incidence of BU in patients over 60. This could mean that a larger number of older patients than would be expected are now coming to the treatment centre, in contrast to the situation in previous published studies in other countries, where a low incidence of elderly patients is described (Smith 1970; Uganda Buruli Group 1971; Amofah *et al.* 2002). Perhaps our study reflects a change of attitude in this group of older patients. The larger number of infections in the elderly in 2001 compared with 2000 may be related to the BU publicity programme initiated in 2000 (M. Debacker, J. Aguiar, C. Steunou *et al.*, unpublished data). These patients are no longer engaged in agriculture, freeing them to go to hospital. However, other factors may be involved and require additional study.

In our study, the anatomic distribution of BU lesions by age and gender does not differ from other studies (Uganda Buruli Group 1971; Amofah *et al.* 2002). Lesions on lower limbs predominate for all age and gender groups. Lower extremities are most frequently in contact with the soil, water, plants and insects. Because of their smaller stature, the entire body of children is nearer to the soil than the upper body of adults. Under 15 years of age there is a preponderance of lesions on lower limbs in males (Barker 1972), and adult females develop more lesions than adult males on the head, neck and trunk (Uganda Buruli Group

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1971). The difference in site of lesion by age and gender probably relates to differences in work and recreational activities. For example, adult females carry materials of all kinds on their heads, especially foodstuff, water and wood more frequently than men. Thus, the head, neck and trunk are more at risk of contamination.

Most frequently the disease begins with a single lesion. Younger patients (<15 years) are more apt to develop multiple lesions with dissemination to distant sites. The percentage of bone lesions is high in disseminated forms. These are the most severe forms of the disease, often leading to amputation and other disabilities or even death. Why children are prone to develop disseminated lesions remains unknown but the cause is probably multifactorial; including, for example, immunity and mode of transmission. The percentage of multiple lesions in patients older than 30 years was low, perhaps because many of these patients died of complications associated with traditional treatment or complete lack of health care. Osteomyelitis can affect one or more bones and treatment of such lesions by traditional methods is likely to lead to infections and septicaemia. Combined infections of BU with other diseases such as schistosomiasis (Scott *et al.* 2004), HIV (Portaels *et al.* 2003) or sickle cell anaemia (Pszolla *et al.* 2003) may favour the dissemination of the disease and increase mortality rates.

Our study suggests that people of advanced age are most at risk for *M. ulcerans* disease, possibly as a result of reactivation of an infection, apparent or subclinical, acquired during childhood, the period when the largest number of individuals may develop the disease. Educational programmes should especially target children and the elderly. Publicity programmes should be organized for schools to promote the early detection of the disease. Teachers could be recruited and trained to detect and refer BU cases to the hospital. Special attention should be given to the motivation of patients with BU that involves multiple body sites. Such patients delay seeking medical advice, and as a result have longer hospital stays (Portaels *et al.* 2003). The social and economical impact of these patients is burdensome for their family and for the health care systems.

Our study of 1630 BU patients demonstrates differences in the frequency and severity of BU related to age and gender, which may be partly explained by immunological and sociocultural parameters. More studies on both aspects are urgently needed to assess their influence on *M. ulcerans* disease.

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Authors

M. Debacker, J. T. Scott and **F. Portaels** (corresponding author), Institute of Tropical Medicine, Nationalestraat 155, 2000 Antwerp, Belgium. Tel: +32 3 247 63 17; Fax: +32 3 247 63 33; E-mail: mdebacker@itg.be, janet.scott@worc.ox.ac.uk, portaels@itg.be

J. Aguiar, C. Steunou and **C. Zinsou**, Centre Sanitaire et Nutritionnel Gbemoten, Zagnanado, Benin. Tel: +229 52 02 93; Fax: +229 52 02 93; E-mail: ghemontin@ifrance.bj

W. M. Meyers, Armed Forces Institute of Pathology, Washington, DC 20306, USA. Tel: +1 202 782 18 73; Fax: +1 202 782 71 61; E-mail: wmeckmeyers@erols.com

M. Dramaix, Ecole de Santé Publique, route de Lennik 808, 1070 Brussels, Belgium. Tel: +32 2 555 40 72; Fax: +32 2 555 40 47; E-mail: miwilmet@ulb.ac.be