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Iatrogenic outbreak of *M. chelonae* skin abscesses

D. CAMARGO[1], C. SAAD[1], F. RUIZ[2], M. E. RAMIREZ[2], M. LINEROS[2], G. RODRIGUEZ[3], E. NAVARRO[2], B. PULIDO[2] AND L. C. OROZCO[1]*

[1] *Laboratorio de Patología, Instituto Nacional de Salud, AA 78091 CUAN, Sante Fé de Bogotá, D.C. Colombia, Sur América*
[2] *Departamento Administrativo de Salud del Atlántico, DASALUD, Edificio Lotería del Atlántico, Carrera 45 No. 44-12, Barranquilla, Colombia, Sur América*
[3] *Facultad de Medicina, Universidad Nacional, Bogota, Columbia, Sur América*

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SUMMARY

We describe an outbreak of skin lesions due to *Mycobacterium chelonae* subsp. *abscessus* associated with injections of lidocaine (lignocaine) given by a 'bioenergetic' (a practitioner of alternative medicine) in Colombia. The lidocaine carpules and the lesions of the patients yielded mycobacteria with identical biochemical characteristics.

Using the methodology of Sartwell and a case control design we examined the incubation period and assessed risk factors. Of 667 potentially exposed individuals, a total of 298 patients were interviewed, of whom 232 had skin lesions. The median incubation period was 30·5 days (range 15–59 days). Male sex (OR 2·85, 95% CI 1·26–6·51), increasing age (OR 1·25, 95% CI 1·03–1·53), subcutaneous injection route (OR 3·72, 95% CI 1·09–12·7) and number of injections (OR 1·01, 95% CI 1·00–1·03) were risk factors for disease.

To our knowledge, this is the largest reported outbreak of *M. chelonae* infection, the first in which the organism has been isolated from the putative vehicle of infection, and the first in which the incubation period could be determined.

INTRODUCTION

The nature and prevalence of disease due to environmental mycobacteria are determined by three factors (1): environmental factors, opportunities for spread of mycobacteria and susceptibility of the human population.

Mycobacterium chelonae has been isolated from freshwater such as rivers and lakes; it may be found in soil, house dust and water supplies where it is resistant to standard levels of chlorination [2, 3]. Contamination due to this mycobacterium commonly occurs in watery environments and after colonizing piped water supplies, may gain access to medical products if these are not properly handled and stored [3].

Mycobacterium chelonae is a non-photochromogenic, rapidly growing mycobacterium [4]. It has been included within the *M. fortuitum* complex and three subspecies have been described: *M. chelonei* subspecies: *chelonei*, *abscessus* and *M. chelonei-like* (unnamed) [5], but only two subspecies are recognized: *abscessus* and *chelonae*. Recently a proposal has been made to elevate the subspecies *abscessus* to species status [4].

* Author for reprints: Luis Carlos Orozco Vargas, AA 78091 CUAN, Santa Fé de Bogotá, D.C., Colombia, Sur América.

M. chelonae has been aetiologically implicated in several clinical syndromes which include not only organ-specific pathology but also disseminated disease [6]. Isolated cases are common [7–14] but outbreaks are not [15–22].

Several kinds of injuries have been reported to favour the disease produced by *M. chelonae*. These include vaccinations [15], injections with home-sterilized needles, gunshot wounds, farm machinery accidents [5] and surgery [5, 17, 23–25]. Use of non-disposable syringes or needles and multiple injections with the same needle, have also been found to be risk factors [10].

Disease can occur at any site where the skin is broken and may result in a wart-like skin lesion, soft tissue abscess that drains small amounts of watery fluid, or nodular lesions that despite their dark red appearance, are only minimally tender [5]. Extensive tissue destruction is also possible [26].

We describe an outbreak by *M. chelonae* due to contamination of lidocaine carpules. We established risk factors and measured the incubation period.

METHODS

In May 1993, DASALUD (Departamento Administrativo de Salud del Atlántico), informed the National Institute of Health (INS) in Santa Fé de Bogotá about several patients with chronic skin supurative lesions, apparently secondary to 'neural therapy' given by a 'bio-energetic' (a practitioner of alternative medicine).

To study the outbreak, professionals of INS and DASALUD interviewed the 'bioenergetic' and a clinical history and a physical examination was performed on the patients.

Laboratory methods

Samples of disinfectant and cleansing solutions, pieces of cleaning and sterilizing material, syringes, needles, cottons, injectable solution (lidocaine carpules) and swabs of environmental surfaces obtained from the bioenergetic's office, were cultured.

The samples cultured from the patients were purulent secretions or cloudy serous fluid from open lesions.

Bacteriological diagnosis was established by acid-fast staining with the Ziehl-Neelsen method and confirmed by culture in Ogawa-Kudoh (O.K.) medium [27–28]; mycobacterial strains were identified by conventional biochemical tests [4, 29, 30].

The biopsies from the lesions were processed with hematoxylin and eosin (HE) and Ziehl Neelsen stain [31].

Incubation period

The incubation period was established in patients who attended only once at the bioenergetic's office and who presented lesions at the time of the interview.

Sartwell's recomendations [32, 33] were followed in order to establish the incubation period and the dispersion factor.

Case-control study

To study risk factors, a case-control design was used. A definite case of mycobacterial soft-tissue disease was defined as a patient with lesions at the site of lidocaine injection, accompanied by a positive culture for *M. chelonae*. A probable case was defined as a patient with closed lesions, like nodules, from which no cultures were obtained but, who presented with symptoms after lidocaine injections, or patients with granulomatous reactions with acid-fast bacilli or a strong granulomatous reaction by histopathological analysis.

Patients who had received injections, but who did not have lesions at the time of the interview, were defined as controls.

A multivariate logistic regression analysis, comparing all definite and probable cases with controls was done. The model and its goodness of fit were evaluated following Hosmer and Lemeshow's recommendations [34–36].

The software Epi-Info 5.01b was used for data entry and STATA 3.1 for logistic regression and goodness of fit.

RESULTS

The information obtained from the 'bioenergetic', showed that he had attended 667 patients from January to April 1993, but only 298 patients could be interviewed.

Patients were residents of Barranquilla (44%), Santa Marta (37%), Cartagena (6%), Magangué

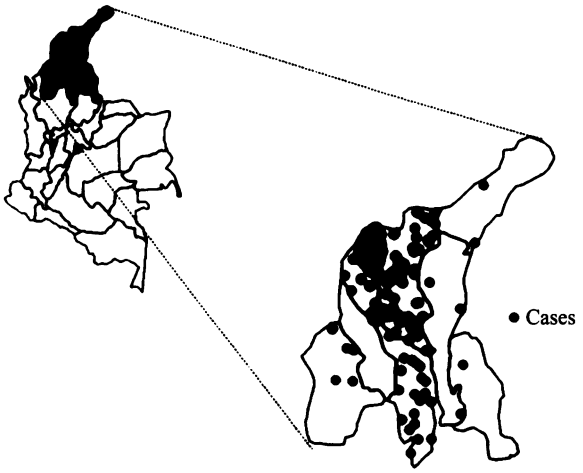

Fig. 1. Geographical distribution of cases. All cases are resident on or near the Atlantic coast of Colombia.

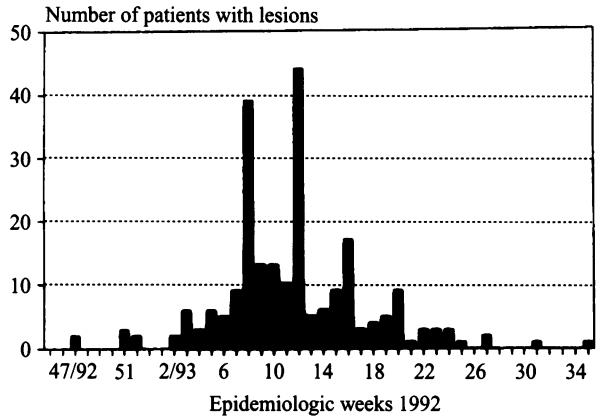

Fig. 2. Distribution of cases in time. Numbers of patients with lesions by week, between week 47, 1992 and week 35, 1993.

Table 1. *Characteristics of lesions by gender*

Variable	Male ($n = 61$)	Female ($n = 171$)
Site of lesions		
Abdomen	57*	139
Buttocks	9	41
Type		
Nodules	43	139
Abscesses	30	68
Fistulas	18	38
Symptoms		
Pain	40	122
Redness	45	140
Oedema	42	118
Fever	14	26

* Number of patients.

(6%) and other municipalities of the Atlantic Coast (6%) (Fig. 1).

The outbreak extended from November 1992 to August 1993, with peaks every fourth epidemiological week: 8th, 12th, 16th and 20th of 1993 (Fig. 2).

Table 2. *Biochemical characteristics of the strains isolated*

Tests	Results
Growth in	
Ogawa–Kudoh at 37 °C in less than 7 days	(+)
Lowenstein-Jensen in less than 7 days	(+)
MacConkey agar	(+)
Sauton agar	(+)
Picrate 0·2%	(+)
NaCl 5% on Lowenstein-Jensen medium	(+)
At	
45 °C	(−)
37 °C	(+)
22 °C	(+)
Pigment production	
In dark	(−)
Response to light	(−)
Enzymatic activity	
Nitrate reductase	(−)
Arylsulphatase after 3 days	(+)
Acid phosphatase	(+)
Catalase 68 °C	(+)
Catalase > 45 mm	(+)
Tween 80 hydrolase	(−)
Urease	(+)
Niacin production	(−)

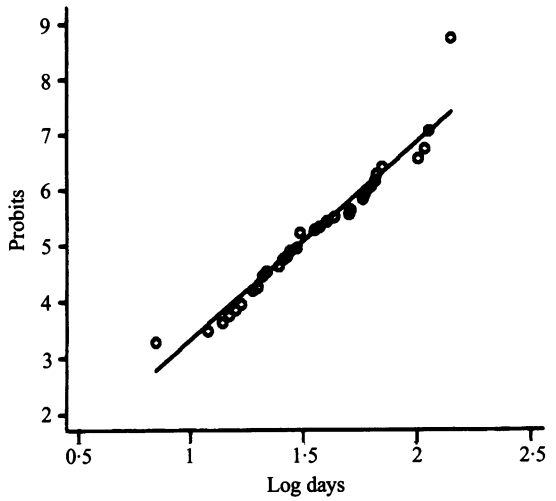

Fig. 3. Incubation period. The ordinate represents the cumulative percentages of cases, expressed in probits; the abscissa is the time scale in logarithms of days. The probit for 50% is equal to 5·21.

The patients, 229 women and 69 men, ranged in age from 4–83 years [median 40 years] and from 5–78 years [median 43 years], respectively. All of them had received about five diluted lidocaine injections at intervals of 20 days. The range of injections per

Table 3. *Relative risks obtained by simple and multiple logistic regression*

Factor	Unadjusted OR	95% CI	Adjusted OR	95% CI
Gender: (male)	2·58	[1·16–5·73]	2·85	[1·26–6·51]
Age: by 10 years	1·29	[1·06–1·56]	1·25	[1·03–1·53]
Injections: number	1·01	[1·00–1·03]	1·01	[1·00–1·03]
Route:				
Subcutaneous	5·38	[1·65–17·6]	3·72	[1·09–12·70]
Intramuscular	1·09	[0·64–1·90]		
Injections in:				
Abdomen	3·90	[1·49–10·30]		
Buttocks	0·90	[0·50–1·62]		
Arms	2·32	[0·29–18·90]		
Legs	1·29	[0·27–6·12]		
Hypertension	0·91	[0·39–2·13]		
Arthritis	1·20	[0·47–3·10]		
Diabetes	2·30	[0·29–18·90]		
Alcohol consumption	1·48*	[0·72–3·03]		

* Once a week.

patient was from 1–432 [median 13] for women and from 1–384 [median 9·0] for men. Many patients had been under bioenergetic treatment for 3–4 years. The body sites that had received more injections were the abdomen (94%) and the buttocks (66·4%). The most common injection route was subcutaneous (96·0%). Three percent of the patients had diabetes and 21·5% admitted drinking alcohol once a week.

A total of 232 patients had at least one lesion. In total we registered 655 lesions in all of them, with a median of 2 and a range from 1–9. The cutaneous lesions were nodules (78·4%), abscesses (42·2%) and fistulas (24·1%), all of them without regional lymphadenopathy (Table 1).

Before the beginning of this investigation, the patients had received from the bioenergetic and another physician a great variety of antibiotics and combinations for the treatment of the lesions; the most common were: amikacin, cephalosporins, penicillins and tetracyclines, but when the bioenergetic suspected a mycobacterial disease, monotherapy with clarithromycin, 1 gram per day was established. A total of 161 (69·4%) patients had been taken clarithromycin for periods of between 15 and 60 days, at the beginning of this investigation.

Laboratory results

A total of 69 samples from the same number of patients was cultured, of which 68 were positive for a rapidly growing mycobacterium identified as *M. chelonae* subsp. *abscessus*; 60 patients had received clarithromycin as monotherapy.

The macroscopic examination of lidocaine carpules showed deteriorated rubbers with several punctures and different liquid volumes, which suggested refilling, repeated use and sterilization. We isolated a strain from one of these lidocaine carpules, obtained from the bioenergetic's office, with the same biochemical characteristics as the strain isolated from the patients. The results of the biochemical tests are presented in Table 2.

The other samples obtained from the bioenergetic's office were negative for mycobacteria.

Histopathology

Histopathology was characterized by superficial and deep abscesses and granulomatous inflammation, involving the hypodermis; a discrete epidermic hyperplasia was observed with formation of fistulas which opened into the epidermis. The granulomas were made up of epithelioid cells, with a few Langhans cells. Several biopsies showed classical tuberculoid granulomata with central caseous or fibrinoid necrosis.

Lymphocytes were abundant and sometimes formed clusters; plasma cells were present in moderate number. Fite-Faraco stain was positive in 19/71 biopsies (26·8%), the bacilli being found in microabscesses or within small clear spaces with the

appearance of fatty vacuoles. Fibrosis and haemorrhage were always present, and secondary vasculitis of small vessels surrounded by neutrophils was common.

Incubation period

The analysis following Sartwell's recommendations showed a median incubation period of 30·5 days and a dispersion factor of 1·95, with a range between 15·6–59·5 days (Fig. 3).

Risk factors

The logistic regression analysis was done with 232 definite or probable cases and 66 controls.

The results for simple logistic regressions for the variables with a probable biological role as risk factor, are presented in Table 3.

The adjusted risks for the multiple logistic model includes: gender, age grouped by 10 year periods, subcutaneous route and number of injections (Table 3). We did not find interaction between risk factors.

The results of goodness of fit statistics were: H^*g 9·26 D.F. = 8 ($P = 0{\cdot}32$), Pearson χ^2 290·7, D 311·7 both with D.F. = 293 indicating that the model fits the data quite well.

The graphic analysis (Leverages, Pearson Chi-square and Delta β) for the goodness of fit showed an adjusted model without extreme or influential values (graphics available from the corresponding authors).

DISCUSSION

A few outbreaks due to *M. chelonae* have been described in the literature after injections of DPT (15), open heart and cardiac surgery [16, 19, 38], mammoplasty [17], dialysis [20–22] and in people who had undergone invasive procedures at a podiatry office [18].

In Colombia, since 1981, there have been reports of isolated cases with soft tissue lesions [14]. Two outbreaks have been recorded. The first was observed after yellow-fever vaccination in Bucaramanga (unpublished observations), and the other following intradermal desensitisation treatment with allergens in Medellin [39], but in these outbreaks the responsible strain was not isolated from the possible contaminated source.

Diagnosis of disease and establishing the nature of this infection is difficult when the entity is not suspected, because the primary agar plates and broth are examined for a couple of days and discarded [40]. In order to establish the diagnosis, appropriate culture techniques with prolonged incubation periods are necessary.

In most of the outbreaks caused by rapidly growing mycobacteria, the source and the nature of the responsible microorganism have not been established [1, 15, 20, 21, 24, 25, 41, 42] but, in general, water, aqueous solutions and the environment have been suggested as the source of contamination. Wallace discusses the importance of geographic location, seasonal factors, adequacy of air filtration and the presence of nonsterile water in the operating room [38].

America is the most frequent geographic location for outbreaks due to *M. chelonae* subsp *abscessus* [16–18, 20, 21, 24, 39, 41, 42]. Only two outbreaks have been reported in Europe, one in the Netherlands [15] and the other in Budapest [25].

The present outbreak is different from most of the others, due to the fact that it was possible to establish the source of infection. All patients had a prior history of penetrating injury at the site of lesion and we could isolate from the deteriorated carpules of lidocaine a mycobacterium with the same characteristics as the one from the patients. The evidence of refilling and repeated use of the carpules points to this procedure as the most likely source of contamination of the lidocaine carpules [3].

To our knowledge, this is the largest and the first outbreak of an infectious disease by *M. chelonae* linked to contaminated lidocaine injected by carpules. This instrument, used for many intradermal or subcutaneous injections, can result in multiple lesions per patient.

The median incubation period of 30·5 days with a length from 15·6–59·5 days and a dispersion factor of 1·95, suggest a great variation around the median [33]. Other studies have established periods between 3 days and 2 years [15, 17, 18, 20, 24, 25, 41, 42], but none of them followed Sartwell's methodology. Using data from one of the outbreaks [17], we could calculate a median incubation period of 34 days with a dispersion factor of 1·9, data very similar to ours.

It is difficult to explain the four peaks in Fig. 2, although it is probable that at least four batches of refilled lidocaine carpules could have been contaminated.

The evaluation of risk factors, showed an increment of risk with male gender, age, number of injections and subcutaneous route.

The male gender predominance in diseases such as tuberculosis and leprosy, and the decrease in immune response with ageing, are well known. The association between the number of injections and the increased risk of developing lesions, can be explained by the fact that the greater the number of injections, the larger the volume injected and higher the probability of mycobacteria being introduced.

With regard to the application route, the resistance of muscular tissue to the development of abscesses, when compared to subcutaneous tissue, is known.

Only a few authors have shown results about risk factors [17, 18, 21, 24, 25], but none of them found personal characteristics including gender, age, route or number of injections associated with mycobacterial disease.

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