MAJOR ARTICLE

High Rate of Acquisition but Short Duration of Carriage of Multidrug-Resistant Enterobacteriaceae After Travel to the Tropics

Etienne Ruppé,[1,2,3] Laurence Armand-Lefèvre,[1,2,3] Candice Estellat,[4,5,6,a] Paul-Henri Consigny,[7,a] Assiya El Mniai,[1] Yacine Boussadia,[4,5] Catherine Goujon,[7] Pascal Ralaimazava,[8] Pauline Campa,[9] Pierre-Marie Girard,[9] Benjamin Wyplosz,[10] Daniel Vittecoq,[10] Olivier Bouchaud,[11] Guillaume Le Loup,[12] Gilles Pialoux,[12] Marion Perrier,[1] Ingrid Wieder,[1] Nabila Moussa,[1] Marina Esposito-Farèse,[4,5] Isabelle Hoffmann,[4] Bruno Coignard,[13] Jean-Christophe Lucet,[2,3,14] Antoine Andremont,[1,2,3] and Sophie Matheron[2,3,8]

[1]Assistance Publique–Hôpitaux de Paris (AP-HP), Hôpital Bichat, Laboratoire de Bactériologie, [2]Institut national de la santé et de la recherche médicale (INSERM), Infection, Antimicrobiens, Modélisation, Evolution (IAME), Unité Mixte de Recherche (UMR) 1137, [3]Université Paris Diderot, IAME, UMR 1137, Sorbonne Paris Cité, [4]Département d'Epidémiologie et Recherche Clinique, AP-HP, Hôpital Bichat, URC Paris-Nord, [5]INSERM, CIC 1425-EC, UMR 1123, [6]Université Paris Diderot, UMR 1123, Sorbonne Paris Cité, [7]Institut Pasteur, Centre Médical, Centre d'Infectiologie Necker-Pasteur, [8]AP-HP, Hôpital Bichat, [9]AP-HP, Hôpital Saint-Antoine, Maladies Infectieuses et Tropicales, Paris, [10]AP-HP, Hôpital de Bicêtre, Maladies Infectieuses et Tropicales, Le Kremlin-Bicêtre, [11]AP-HP, Hôpital Avicenne, Maladies Infectieuses et Tropicales, Bobigny, [12]AP-HP, Hôpital Tenon, Maladies Infectieuses et Tropicales, Paris, [13]Institut de Veille Sanitaire, Saint Maurice, and [14]AP-HP, Hôpital Bichat, Unité d'Hygiène et de Lutte contre les Infections Nosocomiales, Paris, France

Background. Multidrug-resistant Enterobacteriaceae (MRE) are widespread in the community, especially in tropical regions. Travelers are at risk of acquiring MRE in these regions, but the precise extent of the problem is not known.

Methods. From February 2012 to April 2013, travelers attending 6 international vaccination centers in the Paris area prior to traveling to tropical regions were asked to provide a fecal sample before and after their trip. Those found to have acquired MRE were asked to send fecal samples 1, 2, 3, 6, and 12 months after their return, or until MRE was no longer detected. The fecal relative abundance of MRE among all Enterobacteriaceae was determined in each carrier.

Results. Among 824 participating travelers, 574 provided fecal samples before and after travel and were not MRE carriers before departure. Of these, 292 (50.9%) acquired an average of 1.8 MRE. Three travelers (0.5%) acquired carbapenemase-producing Enterobacteriaceae. The acquisition rate was higher in Asia (142/196 [72.4%]) than in sub-Saharan Africa (93/195 [47.7%]) or Latin America (57/183 [31.1%]). MRE acquisition was associated with the type of travel, diarrhea, and exposure to β-lactams during the travel. Three months after return, 4.7% of the travelers carried MRE. Carriage lasted longer in travelers returning from Asia and in travelers with a high relative abundance of MRE at return.

Conclusions. MRE acquisition is very frequent among travelers to tropical regions. Travel to these regions should be considered a risk factor of MRE carriage during the first 3 months after return, but not beyond.

Clinical Trials Registration. NCT01526187.

Keywords. antibiotics; carbapenemase; relative abundance; ESBL; importation.

Multidrug-resistant Enterobacteriaceae (MRE) producing extended-spectrum β-lactamases (ESBLs) and/or plasmid-encoded AmpC-type cephalosporinases (pAmpCs) were long confined to healthcare structures but have been spreading widely in the community over the last 2 decades [1]. The spread of MRE is a major public health issue, as a limited number of antibiotics remain active against these bacteria, and few new antibiotics are expected to reach the market in the foreseeable future [2]. The spread of MRE has been particularly intense in tropical regions, likely owing to poor hygiene and uncontrolled antibiotic usage [3]. Between 14.0% and 69.4% of travelers to high-prevalence countries are reported to acquire MRE, depending on the cohort and destination [4–11].

Received 23 January 2015; accepted 15 April 2015; electronically published 22 April 2015.

[a]C. E. and P.-H. C. contributed equally to this work.

Correspondence: Etienne Ruppé, PharmD, PhD, Laboratoire de Bactériologie, CHU Bichat-Claude Bernard, AP-HP, 46 rue Henri Huchard, 75018 Paris, France (etienne.ruppe@gmail.com).

Clinical Infectious Diseases® 2015;61(4):593–600

© The Author 2015. Published by Oxford University Press on behalf of the Infectious Diseases Society of America. All rights reserved. For Permissions, please e-mail: journals.permissions@oup.com.

DOI: 10.1093/cid/civ333

Downloaded from http://cid.oxfordjournals.org/ at Imperial College London Library on September 16, 2015

It is crucial to assess the dynamics of MRE acquisition and loss during and after travel to tropical regions. Indeed, if MRE importation from tropical regions was found to fuel a major reservoir, specific measures would be urgently required. The few available data suggest that most travelers clear MRE from their gut within a month following return [12], but that a minority become long-term carriers (>6 months after return) [4, 6, 7].

A better knowledge of MRE carriage after return from a tropical region might also improve patient management. A higher risk of MRE acquisition has been found among travelers returning from certain regions with high MRE endemicity (Southeast Asia [5–8, 11] and North Africa [5, 6]), and also among those having taken antibiotics [10] or having developed diarrhea [4, 10]. Yet no data are available on clinical or microbiological factors associated with the duration of MRE carriage, and it is therefore impossible to estimate the risk of MRE carriage at a given time after return.

We addressed these questions in the VOYAG-R study (ClinicalTrials.gov identifier NCT01526187), in which we determined the rate of MRE acquisition during travel to the tropics and monitored carriers for 1 year after their return.

METHODS

Population and Samples

From February 2012 to April 2013, all adults who planned to travel to tropical regions for between 3 days and 3 months and who attended 1 of 6 international vaccination centers in the Paris area were invited to participate in the study. The study was designed to include equal numbers of travelers to the 3 main tropical regions: sub-Saharan Africa, Latin America (including the Caribbean), and Asia. We included countries located in both tropical and subtropical regions. Travelers visiting >1 country were eligible provided they remained in the same region (eg, Vietnam and Cambodia in Asia). Only 1 volunteer per group of co-travelers was recruited. After receiving information on the study, volunteers were asked to complete a brief questionnaire before leaving the center and to provide a stool sample during the week preceding their departure. All stool samples were screened for MRE carriage. Only volunteers who had no detectable MRE fecal carriage before their departure were asked to send a further stool sample within a week after their return. Travelers who were carrying MRE after their return were asked to provide stool samples 1, 2, 3, 6, and 12 months later, or until MRE was no longer detected. Fresh stool samples were self-collected at the traveler's home and promptly shipped to the bacteriology laboratory of Bichat-Claude Bernard Hospital in Paris, France. Each sample was accompanied by a self-completed questionnaire recording (1) at inclusion: demographic data (age, sex), dietary habits (omnivorous, vegetarian), occupation, planned countries of visit, previous travel to the same region, dates of departure and return, prescribed malaria chemoprophylaxis, and the type and purpose of travel; and (2) after return: countries visited, occurrence of diarrhea and/or antibiotic use during the trip, use of the local healthcare system, and adherence to malaria prophylaxis. As doxycycline was exclusively used for malaria prophylaxis and not as an antibiotic, we considered it among antimalarial agents. Nevertheless, the impact of doxycycline and of other antimalarial agents on MRE acquisition rate was studied separately. During follow-up, travelers were asked to inform us if they took antibiotics (and, if so, why), were hospitalized, subsequently traveled to a tropical country, or experienced infectious events (particularly urinary tract infection [UTI]). Follow-up was terminated in case of further travel to a tropical region.

Detection of MRE

An MRE was defined as Enterobacteriaceae producing an ESBL, pAmpC, and/or carbapenemase. As the Enterobacteriaceae that produce these β-lactamases are resistant to most β-lactam antibiotics (which are key antibiotics for the treatment of infections caused by Enterobacteriaceae), we defined them as MRE [13]. Stool samples were cultured immediately on reception. Approximately 10 mg of stool was plated on ChromID ESBL agar (bioMérieux, Marcy-l'Etoile, France) and bi-valve ESBL agar (AES Chemunex, Ivry-sur-Seine, France). In parallel, an enrichment step was performed by diluting approximately 100 mg of stool in 10 mL of brain-heart infusion broth (BHI), 1 mL of which was diluted in 10 mL of BHI broth supplemented with 1.5 mg/L cefotaxime and incubated overnight before plating 100 µL on ChromID ESBL agar. Plates were incubated for 48 hours at 37°C in aerobic conditions. All colony-forming units with distinct aspects (size, color, shape) on each medium were further identified at the species level by mass spectrometry (MALDI Biotyper, Bruker Daltonics, Bremen, Germany) and tested for antibiotic susceptibility by the disk diffusion method as recommended by the French Society for Microbiology. Carbapenemase-producing Enterobacteriaceae (CPE) were detected by specific enrichment culture, as described elsewhere [14]. The relative abundance of MRE was determined as the ratio between the fecal concentrations of total MRE and total Enterobacteriaceae [15].

Characterization of Resistance Mechanisms

Total MRE DNA was extracted with the EZ1 DNA tissue kit on an EZ1 instrument (Qiagen, Courtaboeuf, France). First, ESBL-producing Enterobacteriaceae were tested for $bla_{\text{CTX-M}}$ from groups 1, 2, and 9 with real-time SYBR Green polymerase chain reaction (PCR). If negative, $bla_{\text{CTX-M-8}}$, $bla_{\text{CTX-M-25}}$, bla_{TEM}, bla_{SHV}, and bla_{VEB} were sought with specific PCR primers, as previously described [15]. pAmpC was sought by using specific primers for high-level cephalosporinase-producing Enterobacteriaceae [16]. Strains with reduced susceptibility to carbapenems were tested for bla_{KPC}, bla_{VIM}, bla_{IMP}, bla_{NDM}, and

Downloaded from http://cid.oxfordjournals.org/ at Imperial College London Library on September 16, 2015

bla_{OXA-48} [16]. Amplicons of carbapenemase-encoding genes, $bla_{TEM,}$ and bla_{SHV} were Sanger sequenced with the Big Dye terminator version 3 kit (Applied Biosystems, Courtaboeuf, France) for final identification.

Statistical Analysis

Sample size was calculated to obtain a precision of 5% for the estimated MRE acquisition rate in each region, with a type I error of 0.05. A total of 570 travelers were required (190 per region), assuming an MRE acquisition rate of 20% [4]. Baseline data and outcome measures were summarized using standard descriptive statistics for the total population and each region. All univariate analyses were adjusted for the region when it was required; otherwise, χ^2 tests were used. Independent risk factors for MRE acquisition were identified by logistic regression. Variables statistically significant at the 0.15 level were considered for the multivariate model. The final model was built with the criteria of noncollinearity and the lowest value of Akaike information criterion. Associations between covariates and the number of isolates per MRE-carrying subject (1 vs 2 or more) were identified by logistic regression. The duration of carriage was estimated from the dates of the first positive and the first negative stool sample for MRE ("event"). We used the actuarial (life table) method to represent time to event. Subjects lost to follow-up were censored at the last available time-point. Subjects who completed follow-up without an event were censored at the end of the study (12 months). The effects of covariates on the time to event were estimated using a Cox proportional hazards model. The rate of MRE acquisition was compared across regions by using the Kruskal–Wallis test. All statistical analyses were 2-tailed with a significance level of 5%, and were performed with R statistical software version 3.0.3.

Ethical Issues

The VOYAG-R study was approved by the Ile de France IV ethics committee on 14 November 2011. The study was observational and did not directly benefit the participants. French law requires that each participant sign a "nonrefusal" form. When MRE (including CPE) carriage was detected, the individual concerned was sent an information leaflet on MRE carriage, basic hygiene, and the need to mention this carriage when receiving medical care. Individuals with CPE carriage received a similar leaflet and were also contacted by the infection control practitioner of Bichat-Claude Bernard Hospital to ensure the information was correctly understood.

RESULTS

Description of the Travelers

A total of 824 subjects agreed to participate in the study, of whom 700 provided a stool sample before their departure. Eighty-one subjects (11.6%) were found to carry at least 1 MRE before departure and were thus excluded from the study (Supplementary Table 1). Travelers returning to Asia <1 year after a previous visit were more likely than other travelers to have MRE carriage before departure ($P = .01$).

A total of 574 subjects were finally included (Figure 1), of whom 195 (34.0%) traveled to sub-Saharan Africa, 183 (31.9%) to Latin America, and 196 (34.1%) to Asia (Supplementary Table 2). Mean age was 36 years (standard deviation [SD], 13 years), and the male to female sex ratio was 0.63 (222/352). Tourism was the main purpose of travel (479/574 [83.4%]); no medical tourism was reported. The median duration of travel was 20 days (interquartile range [IQR], 15–30 days). Only 10.4% (59/566) of subjects reported using antibiotics (other than doxycycline) during the trip. Diarrhea during the trip was reported by 228 of 560 (40.1%) travelers, 35 of whom (15.3%) took antibiotics.

MRE Acquisition

At least 1 MRE was acquired during travel by 292 of the 574 (50.9%) subjects. MRE acquisition rates ranged from 31.1% (57/183) in Latin America and 47.7% (93/195) in sub-Saharan Africa, to 72.4% (142/196) in Asia ($P < .001$). Among travelers to popular destinations (>10 study participants) who visited only 1 country, MRE acquisition was most frequent among those visiting Vietnam (13/14 [92.9%]), India (48/53 [90.6%]), Peru (22/26 [84.6%]), and Togo (9/12 [75.0%]) (Supplementary Figure 1, Supplementary Table 3). MRE carriage was rare after travel to French Guyana (2/42 [4.8%]) and Colombia (0/10 [0%]).

Variables Associated With MRE Acquisition

In univariate analysis (Table 1), factors associated with MRE acquisition were β-lactam use during travel (odds ratio [OR], 4.22; 95% confidence interval [CI], 1.47–12.08; $P = .007$), diarrhea during travel (OR, 1.89; 95% CI, 1.32–2.72; $P < .001$), and the type of travel (lower risk for all-inclusive resorts and higher risk for family visits, backpacking, and organized tours; $P = .03$). Regarding use of antibiotics (apart from doxycycline), fluoroquinolone exposure was not associated with a higher risk of MRE acquisition compared with other antibiotics (OR, 1.62; 95% CI, .28–9.41; $P = .59$). In multivariate analysis (Table 1), the region visited ($P < .001$), β-lactam use during travel (OR, 4.08; 95% CI, 1.39–11.97; $P = .011$), diarrhea during travel (OR, 1.90; 95% CI, 1.31–2.75; $P < .001$), and the type of travel (all-inclusive resorts vs others; $P = .033$) significantly influenced the risk of MRE acquisition.

MRE Characterization

On return, 526 MRE isolates were recovered from 292 subjects (mean, 1.8 distinct MRE per positive subject; median, 1.0 [IQR, 1–2]), of whom 135 subjects (46.2%) carried at least 2 MRE.

Downloaded from http://cid.oxfordjournals.org/ at Imperial College London Library on September 16, 2015

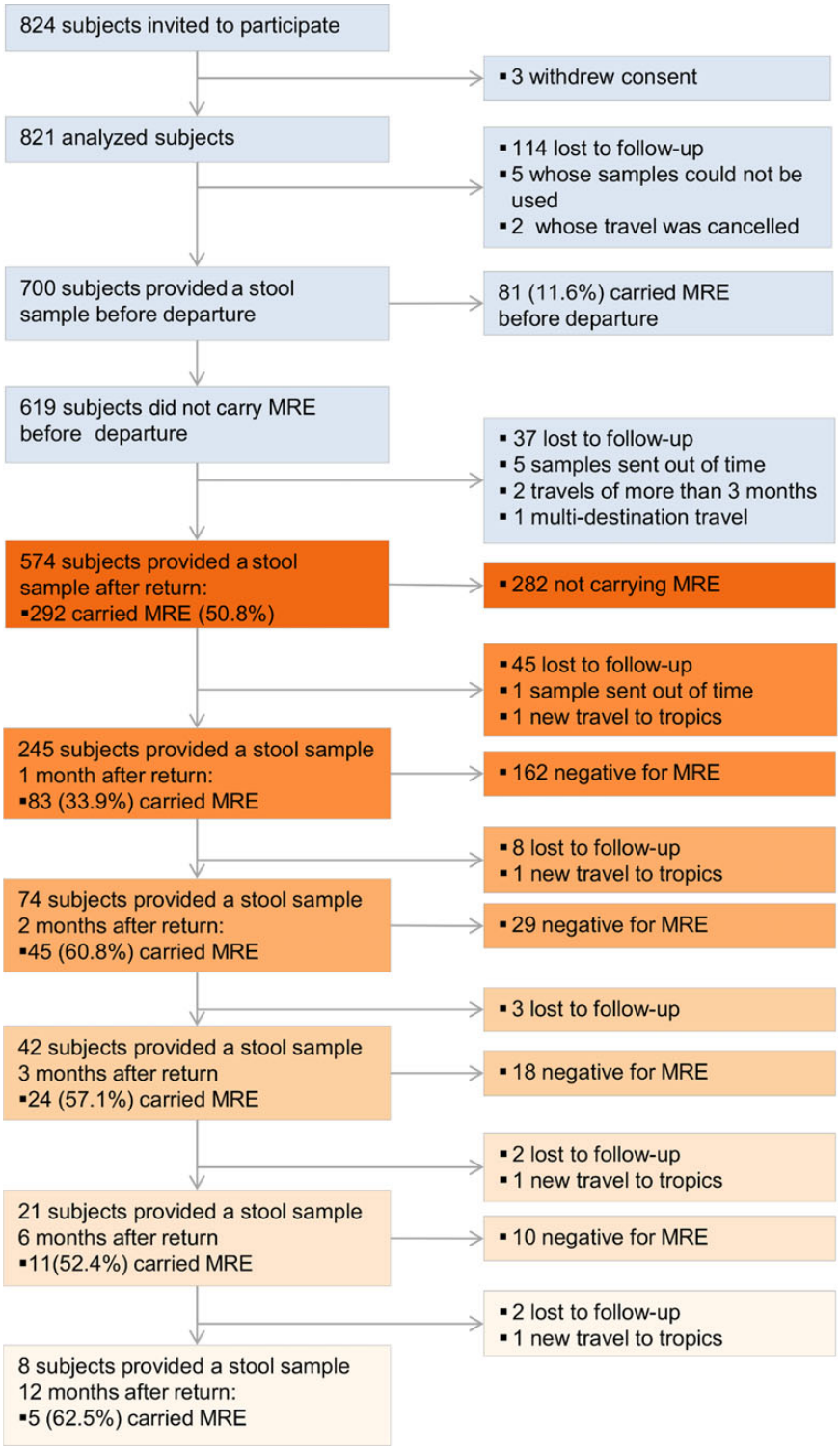


Figure 1. Flowchart of the study. The different colors refer to the time-line: boxes in blue refer to the time before the travel, while the shades of orange refer to the time after the travel. Abbreviation: MRE, multidrug-resistant Enterobacteriaceae.

Compared to travelers carrying only 1 MRE, these 135 subjects reported a higher frequency of diarrhea (56.3% vs 41.0%; $P = .01$) and antibiotic use during the trip (21.1% vs 9.7%; $P = .04$). Travelers to Asia were more likely than travelers to sub-Saharan Africa and Latin America to acquire >1 MRE (60.0%, 21.5%, and 18.5%, respectively, $P < .001$). One subject

Downloaded from http://cid.oxfordjournals.org/ at Imperial College London Library on September 16, 2015

Table 1. Univariate and Multivariate Analysis in Travelers With or Without Acquisition of Multidrug-Resistant Enterobacteriaceae

Variable	Total Travelers, No. (%)	Travelers Without MRE Acquisition, No. (%)	Travelers With MRE Acquisition, No. (%)	Univariate Analysis		Multivariate Analysis	
				Odds Ratio (95% CI)	*P* Value	Odds Ratio (95% CI)	*P* Value
Age, y							
18–34				1.00	.38		
34–49				0.82 (.54–1.25)			
49–64				0.67 (.39–1.13)			
≥65				1.31 (.46–3.69)			
Sex							
Total	574	281	292		.36		
Male (reference)	222 (38.7)	119 (42.2)	103 (35.3)	1.00			
Female	352 (61.3)	163 (57.8)	189 (64.7)	1.18 (.83–1.69)			
Occupation							
Total	573	282	291		.12		
Professionals/senior managers	220 (38.4)	103 (36.5)	117 (40.2)	1.00			
Laborers	39 (6.8)	22 (7.8)	17 (5.8)	0.73 (.35–1.50)			
Middle managers	186 (32.5)	105 (37.2)	81 (27.7)	0.66 (.43–1.00)			
Unemployed	128 (22.3)	52 (18.4)	76 (26.1)	1.09 (.68–1.75)			
Malaria chemoprophylaxis							
Total	563	275	288				
Yes	306 (54.4)	134 (48.7)	172 (59.7)	1.28 (.83–1.97)	.26		
Doxycycline							
Total	561	275	286				
Yes	75 (13.4)	37 (13.5)	38 (13.3)	0.94 (.56–1.60)	.83		
Other							
Total	560	275	285				
Yes	228 (40.7	97 (35.3)	131 (46.0)	1.24 (.84–1.82)	.28		
Antibiotic use during travel							
Total	574	282	292				
β-lactam	25 (4.4)	5 (1.8)	20 (6.8)	4.22 (1.47–12.08)	.01	4.08 (1.39–11.97)	.011
Fluoroquinolone	13 (2.3)	3 (1.1)	10 (3.4)	2.44 (.62–9.54)	.2		
Nifuroxazide	45 (7.8)	16 (5.7)	29 (9.9)	1.79 (.91–3.51)	.09		
Other	5 (0.9)	4 (1.4)	1 (0.3)	0.26 (.03–2.52)	.24		
Diarrhea during the travel							
Total	568	277	291		<.001		
Yes	228 (40.1)	88 (31.8)	140 (48.1)	1.89 (1.32–2.72)		1.90 (1.31–2.75)	<.001
Attendance to a healthcare facility during travel							
Total	567	277	290				
Yes	24 (4.2)	9 (3.2)	15 (5.1)	1.49 (.60–3.66)	.39		
Type of travel							
Total	574	282	292		.03		.033
All-inclusive resort	27 (4.7)	19 (6.7)	8 (2.7)	1.00		1.00	
Mix of all-inclusive resorts and organized tours	78 (13.6)	45 (16.0)	33 (11.3)	1.58 (.59–4.24)		1.23 (.45–3.36)	
Family	142 24.7	79 (28.0)	63 (21.6)	2.23 (.88–5.64)		1.95 (.76–4.98)	
Backpacking	200 (34.8)	77 (27.3)	123 (42.1)	2.96 (1.18–7.47)		2.42 (.95–6.15)	
Organized tour	127 (22.1)	62 (22.0)	65 (22.3)	3.07 (1.20–7.86)		2.74 (1.07–7.06)	
Visited region							
Latin America (reference)						1	<.001
Sub-Saharan Africa						2.21 (1.40–3.48)	
Asia						5.72 (3.55–9.24)	
Duration of travel, wk, median (IQR)	2.86 (2.14–4.29)	2.71 (1.86–4.00)	3.14 (2.29–4.43)	1.06 (.99–1.12)	.09		

Abbreviations: CI, confidence interval; IQR, interquartile range; MRE, multidrug-resistant Enterobacteriaceae.

Downloaded from http://cid.oxfordjournals.org/ at Imperial College London Library on September 16, 2015

returning from Peru carried 8 distinct MRE (7 producing an ESBL, 1 producing pAmpC).

Most of the identified MRE were *E. coli* (93.3%; Supplementary Data 1). Of note, 1 ESBL-producing non-Typhi *Salmonella enterica* was isolated from a woman who had backpacked in Laos. ESBL production was the main resistance mechanism (91.8%), followed by pAmpC production (8.6%) (Supplementary Data 1). Four isolates (0.8%) produced both ESBL and pAmpC. Three isolates (0.6%) from 3 travelers returning from India produced a carbapenemase (2 OXA-181, 1 NDM-1) [16]. The vast majority of ESBLs were of the CTX-M type (95.4%), among which CTX-M group 1 predominated (83.7% of all CTX-M, Supplementary Data 1). Eventually, an OXA-48–producing *Escherichia coli* (coproducing pAmpC) was recovered in a pretravel sample from 1 subject who recently returned from Asia.

Follow-up of Travelers Who Acquired an MRE

Among the 292 travelers who acquired an MRE during travel, 245 provided a stool sample 1 month after their return (Figure 1), which was MRE positive in 83 cases (33.9%) (Figure 1, Table 2). Forty-five of the 74 (60.8%) subjects who provided a stool sample 2 months after return were still MRE carriers. Likewise, 57.1% (24/42), 52.4% (11/21), and 62.5% (5/8) subjects still carried an MRE 3, 6, and 12 months after their return (Figure 1). Only 4.7% of the 515 travelers still followed were MRE carriers 3 months after their return (Table 2). The dynamics of MRE loss differed significantly according to the region visited: travelers returning from Asia carried MRE for longer than travelers returning from sub-Saharan Africa or Latin America ($P < .001$; Figure 2, Table 2). Univariate analysis identified no travel-related variables significantly associated with longer carriage. However, carriers of at least 1 multidrug-resistant *E. coli* had a lower risk of prolonged carriage than carriers of other multidrug-resistant species (hazard ratio, 0.58; 95% CI, .38–.90; $P = .01$). Eight travelers reported an episode of UTI after their return, but no microbiological data were available.

Quantification of MRE

On return, the mean relative abundance of MRE (MRE-RA) was −3.2 log (SD, 1.9 log). Travelers returning from Asia had a higher mean MRE-RA than travelers to sub-Saharan Africa and Latin America (−2.9 [SD, 1.9] log vs −3.6 [SD, 1.9] log and −3.6 [SD, 1.8] log, respectively; $P = .01$; Figure 3*A*). Interestingly, patients with a low MRE-RA had shorter carriage: the hazard ratio for a negative subsequent stool sample per 1-log unit decrease in MRE-RA was 1.18 (95% CI, 1.1–1.26; $P < .001$; Figure 3*B*).

DISCUSSION

We found that MRE acquisition during travel to tropical regions was frequent but that carriage was generally brief. Half the travelers in this study acquired at least 1 MRE (an average of 1.8 species per carrier), a rate twice as high as that reported elsewhere [4–10]. This higher rate may be attributed to 3 characteristics of our study: (1) only travelers to tropical destinations were included, and (2) fresh stools (instead of rectal swabs) were cultured on various media, with (3) an additional enrichment step. Still, we cannot exclude that travelers with very low MRE carriage before travel could have gone undetected, then found to be positive at return because they used antibiotics during the travel.

In 2013, nearly 10.6 million persons (connecting flights excluded) departed from a French airport to a tropical region (4.5 million to sub-Saharan Africa, 1.8 million to Latin America, and 4.3 million to Asia) [17]. Based on the rates of MRE acquisition found here, we estimate that, in 2013, at least 5.8 million travelers (2.1 million to sub-Saharan Africa, 0.6 million to Latin America, and 3.1 million to Asia) brought an MRE from tropical regions to France. Interestingly, 3 months after their return, >95% of all the travelers studied here were free of MRE. Thus, importation of MRE by travelers does not fuel a sustained reservoir of MRE in the community. This finding

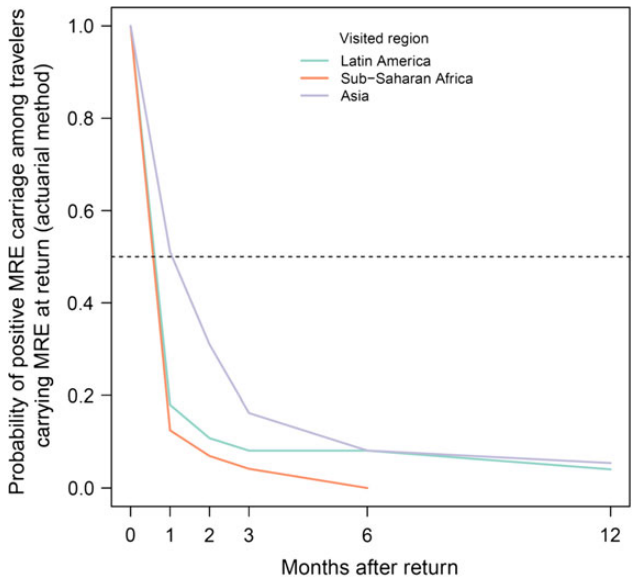
Table 2. Follow-up of Travelers After Their Return

Time After Return, mo	Proportion of MRE Carriers Among Carriers at Return (%)[a]				Proportion of MRE Carriers Among All Travelers (%)[a]			
	All	Sub-Saharan Africa	Latin America	Asia	All	Sub-Saharan Africa	Latin America	Asia
0	292/292 (100)	93/93 (100)	57/57 (100)	142/142 (100)	292/574 (50.9)	93/195 (47.7)	57/183 (31.1)	142/196 (72.4)
1	83/245 (33.9)	9/72 (12.5)	10/50 (20)	64/123 (52)	83/527 (15.7)	9/174 (5.2)	10/176 (5.7)	64/177 (36.2)
2	45/236 (19.1)	5/72 (6.9)	5/48 (10.4)	35/116 (30.2)	45/518 (8.7)	5/174 (2.9)	5/174 (2.9)	35/170 (20.6)
3	24/233 (10.3)	3/72 (4.2)	3/47 (6.4)	18/114 (15.8)	24/515 (4.7)	3/174 (1.7)	3/173 (1.7)	18/168 (10.7)
6	11/230 (4.8)	0/72 (0)	3/47 (6.4)	8/111 (7.2)	11/512 (2.1)	0/174 (0)	3/173 (1.7)	8/165 (4.8)
12	5/227 (2.2)	0/72 (0)	1/46 (2.2)	4/109 (3.7)	5/509 (1)	0/174 (0)	1/172 (0.6)	4/163 (2.5)

Abbreviation: MRE, multidrug-resistant Enterobacteriaceae.

[a] Travelers lost to follow-up, those whose stool sample was sent out of time as scheduled, and those who subsequently traveled to tropical regions were excluded at each time point.

Downloaded from http://cid.oxfordjournals.org/ at Imperial College London Library on September 16, 2015



Months after return	Likelihood of negative MRE carriage		
	Latin America	Sub-Saharan Africa	Asia
0-1	0.18	0.12	0.51
1-2	0.11	0.07	0.31
2-3	0.08	0.04	0.16
3-6	0.08	0	0.08
6-12	0.04	0	0.05

Figure 2. Survival curve representing multidrug-resistant Enterobacteriaceae (MRE) carriage rates among travelers during follow-up, according to the visited geographic area.

has major implications for physicians assessing the individual risk of MRE carriage among patients having recently traveled to a tropical region. During the first 3 months after return from a tropical region, MRE should be considered when prescribing empiric antimicrobial therapy, especially when the patient took antibiotics and/or developed diarrhea during the trip. Of note, 8 MRE carriers reported developing a UTI after their return. As UTI is more likely to be due to MRE in MRE carriers [15], our results suggest a need for routine antibiotic susceptibility testing in individuals who develop UTI shortly after returning from a tropical region.

We provide the first data on the relative abundance of MRE among all fecal Enterobacteriaceae during the first 12 months after return from the tropics. We found that carriage lasted longer in individuals with a high initial MRE-RA, suggesting that MRE-RA determination could help to predict the likely duration of carriage. We have previously shown that antibiotic exposure increases the MRE-RA [15], thereby possibly contributing to the persistence of MRE carriage. If this is confirmed, physicians should carefully consider the potential impact on the intestinal microbiota when prescribing antibiotics to subjects having recently returned from a tropical region.

The main factor associated with MRE acquisition was the region visited, travelers to Asia being at the highest risk. However, it is noteworthy that travelers to Peru and Togo were also at a high risk of MRE acquisition (84.6% and 75.0%, respectively). Other risk factors included diarrhea, which is a potential marker of exposure to contaminated food or water. Travel to all-inclusive resorts was associated with a lower risk of MRE acquisition. Exposure to β-lactam antibiotics increased the risk of acquisition, but this was not the case for doxycycline, an antibiotic used for malaria prophylaxis.

In conclusion, we show that MRE acquisition during travel to tropical regions is very frequent but relatively short-lived. The possible presence of MRE should thus be taken into account when prescribing empiric antibiotic therapy for infections that occur in travelers having returned from the tropics <3 months previously.

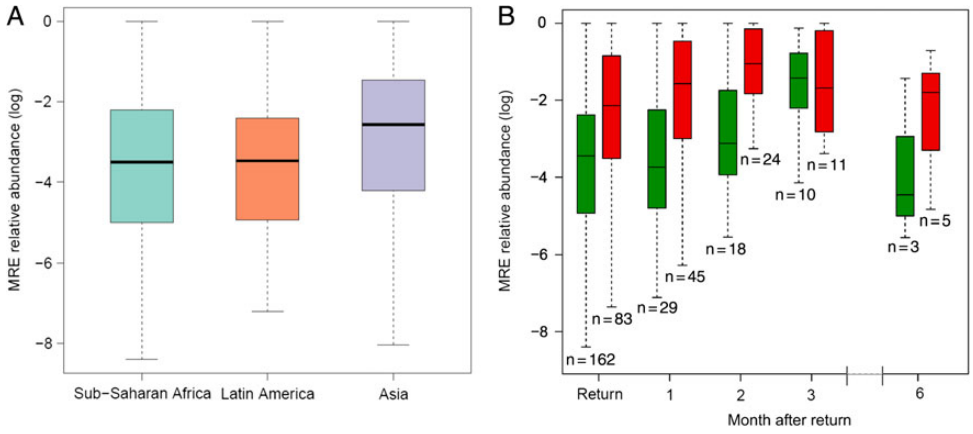


Figure 3. *A*, Fecal relative abundance of multidrug-resistant Enterobacteriaceae (MRE) (log) on return, according to the visited geographic area. *B*, Fecal relative abundance of MRE (log) among travelers during follow-up, with respect to the persistence or loss of MRE at the next time point. MRE carriers whose next sample was negative for MRE are shown in green; MRE carriers whose next sample was positive for MRE are shown in red.

Downloaded from http://cid.oxfordjournals.org/ at Imperial College London Library on September 16, 2015

Supplementary Data

Supplementary materials are available at *Clinical Infectious Diseases* online (http://cid.oxfordjournals.org). Supplementary materials consist of data provided by the author that are published to benefit the reader. The posted materials are not copyedited. The contents of all supplementary data are the sole responsibility of the authors. Questions or messages regarding errors should be addressed to the author.

Notes

Acknowledgments. The authors thank Amandine Fauquembergue and Mélanie Hamez for logistical support, research implementation and study monitoring, Cécile Kedzia and Fadela Amerali for help with research administration, Samira Laribi for developing the electronic case report form, and Marion Schneider for the data management. The authors also thank Dominique Monnet (European Centre for Disease Prevention and Control) for his continuous support and feedback, Mobidiag for their assistance in multidrug-resistant Enterobacteriaceae resistance typing, Raymond Ruimy for the implementation of CTX-M real-time polymerase chain reaction, and David Young for editorial assistance. We are grateful to all the travelers who agreed to participate in this study, and to the staff of the international vaccination centers.

Financial support. This work was supported by a grant from the French Ministry of Health (AOR 11101), and the sponsor was Assistance Publique–Hôpitaux de Paris.

Potential conflicts of interest. All authors: No potential conflicts of interest.

All authors have submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest. Conflicts that the editors consider relevant to the content of the manuscript have been disclosed.

References

1. Pitout JDD, Laupland KB. Extended-spectrum beta-lactamase-producing Enterobacteriaceae: an emerging public-health concern. Lancet Infect Dis **2008**; 8:159–66.
2. Boucher HW, Talbot GH, Benjamin DK, et al. 10×'20 progress—development of new drugs active against gram-negative bacilli: an update from the Infectious Diseases Society of America. Clin Infect Dis **2013**; 56:1685–94.
3. Woerther P-L, Burdet C, Chachaty E, Andremont A. Trends in human fecal carriage of extended-spectrum β-lactamases in the community: toward the globalization of CTX-M. Clin Microbiol Rev **2013**; 26: 744–58.
4. Tängdén T, Cars O, Melhus A, Löwdin E. Foreign travel is a major risk factor for colonization with *Escherichia coli* producing CTX-M-type extended-spectrum beta-lactamases: a prospective study with Swedish volunteers. Antimicrob Agents Chemother **2010**; 54: 3564–8.
5. Peirano G, Laupland KB, Gregson DB, Pitout JDD. Colonization of returning travelers with CTX-M-producing *Escherichia coli*. J Travel Med **2011**; 18:299–303.
6. Ostholm-Balkhed A, Tärnberg M, Nilsson M, et al. Travel-associated faecal colonization with ESBL-producing Enterobacteriaceae: incidence and risk factors. J Antimicrob Chemother **2013**; 68: 2144–53.
7. Paltansing S, Vlot JA, Kraakman MEM, et al. Extended-spectrum β-lactamase-producing Enterobacteriaceae among travelers from the Netherlands. Emerg Infect Dis **2013**; 19:1206–13.
8. Yaita K, Aoki K, Suzuki T, et al. Epidemiology of extended-spectrum β-lactamase producing *Escherichia coli* in the stools of returning Japanese travelers, and the risk factors for colonization. PLoS One **2014**; 9: e98000.
9. Weisenberg SA, Mediavilla JR, Chen L, et al. Extended spectrum beta-lactamase-producing Enterobacteriaceae in international travelers and non-travelers in New York City. PLoS One **2012**; 7: e45141.
10. Kennedy K, Collignon P. Colonisation with *Escherichia coli* resistant to 'critically important' antibiotics: a high risk for international travellers. Eur J Clin Microbiol Infect Dis **2010**; 29:1501–6.
11. Kuenzli E, Jaeger VK, Frei R, et al. High colonization rates of extended-spectrum beta-lactamase (ESBL)-producing *Escherichia coli* in Swiss travellers to South Asia—a prospective observational multicentre cohort study looking at epidemiology, microbiology and risk factors. BMC Infect Dis **2014**; 14:528.
12. Rogers BA, Kennedy KJ, Sidjabat HE, Jones M, Collignon P, Paterson DL. Prolonged carriage of resistant *E. coli* by returned travellers: clonality, risk factors and bacterial characteristics. Eur J Clin Microbiol Infect Dis **2012**; 31:2413–20.
13. Magiorakos A-P, Srinivasan A, Carey RB, et al. Multidrug-resistant, extensively drug-resistant and pandrug-resistant bacteria: an international expert proposal for interim standard definitions for acquired resistance. Clin Microbiol Infect **2012**; 18:268–81.
14. Ruppé E, Armand-Lefèvre L, Lolom I, et al. Development of a phenotypic method for detection of fecal carriage of OXA-48-producing Enterobacteriaceae after incidental detection from clinical specimen. J Clin Microbiol **2011**; 49:2761–2.
15. Ruppé E, Lixandru B, Cojocaru R, et al. Relative fecal abundance of extended-spectrum-β-lactamase-producing *Escherichia coli* strains and their occurrence in urinary tract infections in women. Antimicrob Agents Chemother **2013**; 57:4512–7.
16. Ruppé E, Armand-Lefèvre L, Estellat C, et al. Acquisition of carbapenemase-producing Enterobacteriaceae by healthy travellers to India, France, February 2012 to March 2013. Euro Surveill **2014**; 19.
17. Bulletin statistique trafic aerien commercial année 2013. Direction générale de l'aviation civile, **2014**. Available at: http://www.developpement-durable.gouv.fr/IMG/pdf/Bulletin_Stat_2013_20140527.pdf. Accessed 10 October 2014.
18. Ondov BD, Bergman NH, Phillippy AM. Interactive metagenomic visualization in a Web browser. BMC Bioinformatics **2011**; 12:385.

Downloaded from http://cid.oxfordjournals.org/ at Imperial College London Library on September 16, 2015