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Hepatitis E in a food handler – a rapid risk assessment to guide the public health response

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Objective: The Australian Capital Territory Health Directorate was notified of a food handler with hepatitis E virus (HEV) infection. To guide the public health response, a rapid risk assessment was undertaken to determine the risk of transmission of HEV from the infected food handler to restaurant patrons.

Method: The literature on HEV was reviewed and expert advice sought from clinical and public health specialists. This was supplemented by results of a site investigation and a case interview. The risk rating was determined to be the product of the likelihood of transmission and the consequence of the infection.

Results: The food handler was likely to have been infectious at the time he was working at the restaurant. He had handled high-risk foods, and the site inspection revealed potential opportunities for transmission. HEV is not common in Australia and it was assumed that the population was non-immune and hence susceptible to the disease. Therefore, there was a low but possible likelihood of transmission of HEV. If infected, HEV has the potential for major consequences in vulnerable populations especially among women who are pregnant. The overall level of risk was considered to be very high.

Discussion: The general public and health practitioners were alerted to enable early identification of symptoms and prompt disease management. There were no secondary cases of HEV associated with this event. In the absence of published guidelines and limited evidence, a risk assessment framework was a useful tool to inform public health decision-making.

In early 2011, the Communicable Diseases Control Section (CDC) of the Australian Capital Territory Health Directorate (ACT Health) in Canberra was notified of a food handler who was diagnosed with hepatitis E virus (HEV) infection. He had recently returned from an overseas trip to a country where there is known HEV activity and had presented to hospital with abdominal pain and jaundice. His liver function tests were abnormal (markedly elevated liver enzymes and high bilirubin) and serology showed a positive HEV IgM and a negative IgG. The clinical, epidemiological and laboratory features fit the case definition for HEV infection.[1] He was admitted to the hospital where he received symptomatic treatment. He worked at a restaurant and was involved in all aspects of food preparation.

The transmission of hepatitis A virus (HAV), a very similar disease, from food handlers to restaurant patrons has been demonstrated,[2,3] and national guidelines exist in Australia to guide the public health response.[4] However, there is less evidence and no guidelines available to guide public health decision-making when a food handler is infected with HEV. Although HAV and HEV share similarities, there are differences in disease characteristics, transmissibility and outcome that make the applicability of HAV guidelines less useful in this scenario.

In this paper, we describe and discuss a rapid risk assessment that was undertaken to guide the public health response for managing the HEV-infected food handler. The question we sought to answer was: what was the risk of transmission of HEV from the infected food handler to patrons at the restaurant? We describe the methods and results of our rapid risk assessment and the subsequent public health actions.

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RISK ASSESSMENT METHODOLOGY

To inform the risk assessment, published literature on HEV was reviewed. Additional information and expert advice was sought from specialists in the fields of public health, infectious diseases and gastroenterology. The results of a site investigation of the restaurant where the case worked, conducted by environmental health officers, and information obtained by interviewing the case also informed the risk assessment.

The risk assessment took into account the likelihood of transmission of the virus from the food handler to restaurant patrons and the consequence of the infection to determine the level of risk. The risk rating was the product of the likelihood of transmission and the consequence of the infection.

RESULTS

Likelihood of transmission

The first component of the risk assessment was to evaluate the likelihood of transmission and the potential for exposure of the restaurant patrons to the virus. The potential infectivity of the food handler, the transmissibility of the virus, the handling of high-risk foods, environmental factors that would facilitate spread of the disease and the susceptibility of the population were taken into account when assessing how likely it was that the patrons were exposed to the disease.

(1) Infectivity of the food handler

The infectious period of HEV is not known.[5] However, virus particles have been detected in stools 14 days after the onset of jaundice and approximately four weeks after the ingestion of contaminated food or water.[5] Given that the food handler worked at the restaurant up to four days before the onset of jaundice, it was very likely that he was infectious during the time that he worked there.

(2) Transmissibility of the virus

Transmission of HEV occurs predominantly via the fecal–oral route with contaminated water being the source of large epidemics.[6] Parenteral and perinatal routes have also been implicated.[6] Although person-to-person transmission is not thought to be common or to contribute significantly to morbidity in epidemics, it may occur.[7,8] Opportunities described for transmission of HEV from person to person were thought to include the use of common drinking, hand-washing and eating vessels.[8]

(3) Handling of high-risk foods

The food handler was responsible for the preparation of all foods at the restaurant including raw salads that were not further cooked before being served. The food handler stated that he used gloves when preparing these foods.

(4) Environmental assessment

Environmental health officers from ACT Health inspected the restaurant, and some deficiencies (the lack of a dedicated hand-washing facility) in the food preparation area were observed. This might have provided opportunities for transmission of the disease.

(5) Susceptibility of the population

The context of the risk assessment was the ACT population. Hepatitis E is an uncommon disease in Australia (notification rates 0.1–0.2 per 100 000).[9] In the ACT, from 2006 there had been only six cases of HEV notified, all overseas acquired. Similar to other notifiable diseases, it is possible that this is an underestimate due to asymptomatic cases or lack of testing; however, it was reasonable to assume that the underlying incidence is extremely low. Therefore, it was assumed that the ACT population was susceptible to HEV infection.

Consequence of infection

The next step in the risk assessment was to determine the potential consequences of infection with HEV if transmission did occur. Hepatitis E is an RNA virus, a major cause of waterborne hepatitis in tropical and subtropical countries and of sporadic disease in industrialised countries.[6] Typical signs and symptoms include jaundice, anorexia, hepatomegaly, abdominal pain, nausea and vomiting and fever.[6,7] Although the clinical course of HEV infection is similar to that of HAV

Figure 1. **Risk analysis matrix (adapted from the Australian Guidelines for the Prevention and Control of Infection in Healthcare)[11]**

LIKELIHOOD	CONSEQUENCES				
	Negligible	Minor	Moderate	Major	Extreme
Rare	Low	Low	Low	Medium	High
Unlikely	Low	Medium	Medium	High	Very high
Possible	Low	Medium	High	Very high ●	Very high
Likely	Medium	High	Very high	Very high	Extreme
Almost certain	Medium	Very high	Very high	Extreme	Extreme

Note: Black circle indicates the assessment of risk.

infection, HEV has been associated with greater severity (protracted coagulopathy and cholestasis) and higher mortality.[10]

HEV infection can range in severity from subclinical to fulminant disease. The disease is particularly severe for those with chronic liver disease and in pregnancy where the mortality rate can be 15–20%. Other complications during pregnancy include fetal death, premature delivery or death of the infant soon after birth.[6] There is no vaccine available in Australia or chemoprophylaxis for the prevention of the disease.

RISK CHARACTERIZATION AND MANAGEMENT

The likelihood of transmission and the consequence of the infection were assessed. It was highly likely that the food handler was infectious at the time he was working at the restaurant. He had handled high-risk (uncooked) food, and the site inspection had revealed potential opportunities for transmission of the disease. There is however limited evidence of the transmissibility of the virus, and although person-to-person transmission is not common, it may occur. The concern was that there could be transmission of the virus from the food handler to the restaurant patrons either via food or through plates or cutlery. Taking these factors into consideration, there was a low but possible likelihood of transmission. If transmission occurred however, the consequence of infection with HEV was considered to be major, especially in pregnant women and those with chronic liver disease. As there was no way to identify if there were high-risk patrons at the restaurant who had consumed meals made by the infected food handler, the overall level of risk was assessed to be very high (**Figure 1**).

The main limitation of this risk assessment was the limited literature and guidelines on transmission of hepatitis E. With the information and evidence available, and using a precautionary approach, the recommendation of the risk assessment was to provide advice to patrons who may have eaten at the restaurant during the time the food handler had worked there while potentially infectious. The advice was that there was a small risk of acquiring HEV infection, the symptoms to be aware of and the importance of seeking early medical treatment, especially those at high risk. Given that the restaurant had no booking lists and there was no way to identify those who were at higher risk, the public was alerted by a media release and a health alert that was placed on the ACT Health website. A communicable disease information hotline was available to deal with public inquiries. A letter was sent to all general practitioners and hospital emergency departments in the ACT to alert them of this event to enable the early identification and management of cases.

The decision to name publicly the restaurant was not made lightly and done only after the careful consideration of potential risks to the public. To mitigate adverse effects to the restaurant, a sensitive communication strategy was adhered to, involving restaurant management at all stages of the investigation and public health response. This ensured full cooperation by the restaurant management.

Enhanced surveillance of HEV notifications received by ACT Health after this incident did not find any secondary hepatitis E cases that were linked to the restaurant.

CONCLUSION

Conducting a rapid risk assessment in a novel situation where limited guidance was available was a useful way to ensure evidence-based decision-making and enabled a timely public health response. This has since been used within the Communicable Diseases Control section of ACT Health for other public health issues.[12] The World Health Organization has published guidelines for the rapid assessment of public health events,[13] and these are expected to be a useful framework for risk assessment to inform public health decision-making. A guideline for the public health management of hepatitis E would be a useful addition to the Australian Series of National Guidelines.[14]

Conflicts of interest

None declared.

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Investigating an outbreak of acute fever in Chuuk, Federated States of Micronesia

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Objective: In September 2012, there was an unexpected increase of acute febrile illness (AFI) in Chuuk State of the Federated States of Micronesia. At the same time, dengue outbreaks were occurring in two of the Federated States of Micronesia's other three states. The cause of AFI was suspected to be dengue; however, by the end of October, only one of 39 samples was positive for dengue. The objective of the investigation was to establish the cause of the outbreak.

Methods: A line list was created and data analysed by time, place, person and clinical features. Reported symptoms were compared with the published symptoms of several diagnoses and laboratory testing undertaken.

Results: Of the 168 suspected cases, 62% were less than 20 years of age and 60% were male. The clinical features of the cases were not typical for dengue but suggestive of respiratory illness. Nasopharyngeal swabs were subsequently collected and found to be positive for influenza. Public health measures were undertaken and the AFI returned to expected levels.

Discussion: Clinical diagnosis of acute febrile illness (AFI) can often be difficult and misleading. This can mean that opportunities for preventive measures early on in an outbreak are missed. In any outbreak, descriptive epidemiological analyses are valuable in helping to ascertain the cause of the outbreak.

The Federated States of Micronesia is an independent nation in free association with the United States of America, consisting of more than 600 islands extending across four states in the North Pacific Ocean: Pohnpei, Kosrae, Chuuk and Yap.[1] Chuuk State, made up of Chuuk Lagoon and a series of outer islands, has a population of approximately 53 000 people and the Federated States of Micronesia's highest population density.[1] Many of the outer islands are several hundred kilometres from Chuuk Lagoon, and Weno Island is the main island of Chuuk State (**Figure 1**). The hospital in Weno, the state's only hospital, has 150 beds and houses the state's only laboratory. Each of the other islands in the state has at least one dispensary; some have up to four. Each dispensary is run by a health assistant. Chuuk State Hospital has a dispensary coordinator who calls each dispensary health assistant on a weekly basis to monitor their activities and detect any unusual disease occurrences.

On 9 August 2012, an 18-year-old male from Weno Island presented to Chuuk State Hospital after experiencing three days of fever. A rapid diagnostic test for dengue was performed and was negative; however, the same sample was shipped to Brisbane for polymerase chain reaction (PCR) testing and was positive for dengue virus serotype-4 (DENV-4).

In mid-September, hospital doctors noted an increase in the number of cases of acute febrile illness (AFI). Due to the increase of and concern over concurrent dengue outbreaks in other Federated States of Micronesia states, clinicians commenced filling in patient encounter forms for suspected dengue cases using a case definition of: acute fever of at least 38 °C with two or more of the following: nausea, vomiting, severe headaches, orbital pain, joint pain, rash, haemorrhage, signs of leucopoenia.

In late September, a small number of cases of suspected dengue was reported in Satawan, an outer island several hundred kilometres south-east of Chuuk Lagoon. Then, on 3 October in Onoun, an island several hundred kilometres north-west of Chuuk Lagoon, a two-year-old boy who reported experiencing a runny

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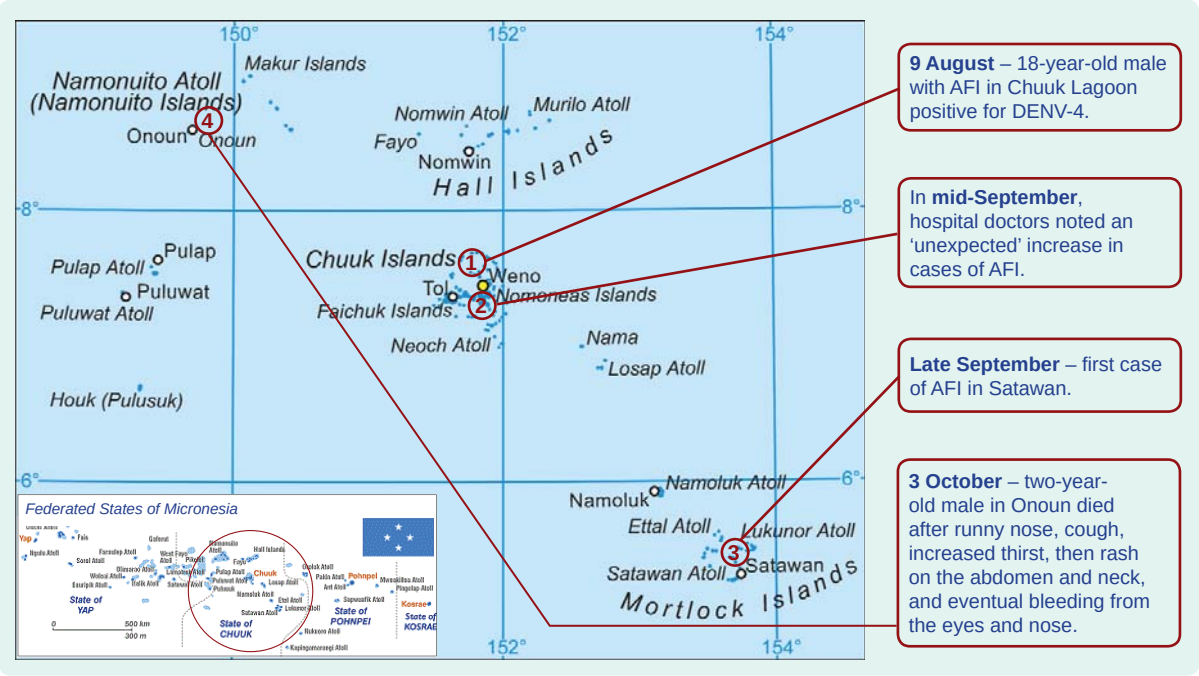
Figure 1. **Map of Chuuk State and sequence of events in acute fever outbreak, 2012**



AFI, acute febrile illness.

Source: Chuuk State map was reproduced from Wikimedia Commons, the free media repository (http://commons.wikimedia.org/wiki/File:Chuuk.png), and the inset map of the Federated States of Micronesia was reproduced from WHO Division of Pacific Technical Support (http://www.wpro.who.int/southpacific/pacelf/countries/fsm/en/).

nose, cough and increased thirst for two weeks, suddenly developed a rash on his abdomen that moved up to the neck area within minutes. The child started to bleed from the corner of his eyes and his nose and died soon after.

By 22 October, 137 cases of suspected dengue were reported with 39 blood samples sent to Brisbane for PCR confirmation; however, only the initial sample was positive for dengue. As there was uncertainty as to the cause of the outbreak, an investigation was undertaken in Chuuk State at the end of October 2012 to establish the cause of the acute febrile illness (AFI) outbreak and ensure an appropriate response.

METHODS

From 31 October to 2 November, an investigation was undertaken to establish the cause of the AFI outbreak. First, members of the EpiNet team, doctors, dispensary nurses, laboratory staff, the dispensary coordinator and a small number of patients were interviewed.

A line list of all suspected dengue cases was developed in Microsoft Excel using the Chuuk State Hospital emergency department register and the inpatient, outpatient and laboratory registers together with the dispensary coordinator's register. Clinical symptoms were included from patient medical records where possible. The line list had the following fields: line number; hospital/clinic name; data source/register; hospital number; first name; last name; age; sex; residence (village, island, island group); date of onset; date of presentation to hospital/clinic; attending clinician; symptoms (acute fever, headache, joint pain, muscle pain, bone pain, eye pain, nausea, vomiting, diarrhoea, weakness, rash, cough, runny nose, sore throat, other main symptoms); and laboratory results.

Analysis

Analyses were undertaken to examine and describe the outbreak in terms of time, place, person and clinical features. An epidemic curve by location was constructed using 'date of notification' on the *x*-axis because of insufficient data on date of onset. Attack rates and clinical features were compared across the three sites as was the distribution of cases by age and sex.

Attack rates were calculated using the Chuuk State estimated population for 2012.[2] There were 18 cases (17 from Onoun and one from Chuuk Lagoon) that had no gender status indicated in their records;

Table 1. **Age and sex distribution of acute febrile illness, Chuuk State, Federated States of Micronesia, 5 August to 4 November 2012 (*n* = 168)**

Age group	AFI cases			Population 2012*			Cases/1000 population		
	Total	Male	Female	Total	Male	Female	Total	Male	Female
	168	100	68	52 574	26 338	26 236	3.2	3.8	2.6
0–4	57	36	21	6388	3270	3118	8.9	11.0	6.7
5–9	17	8	9	6267	3200	3067	2.7	2.5	2.9
10–14	5	5	0	5929	3003	2926	0.8	1.7	0.0
15–19	25	16	9	5402	2709	2693	4.6	5.9	3.3
20–24	15	8	7	4590	2357	2233	3.3	3.4	3.1
25–29	4	2	2	3882	1941	1941	1.0	1.0	1.0
30–34	10	6	4	3215	1635	1580	3.1	3.7	2.5
35–39	6	4	2	3116	1546	1570	1.9	2.6	1.3
40–44	3	1	2	2921	1403	1518	1.0	0.7	1.3
45–49	3	2	1	2583	1256	1327	1.2	1.6	0.8
50–54	3	1	2	2394	1174	1220	1.3	0.9	1.6
55–59	6	2	4	2103	1060	1043	2.9	1.9	3.8
60–64	10	6	4	1578	783	795	6.3	7.6	5.0
65–69	1	1	0	882	415	467	1.1	2.4	0.0
70–74	1	1	0	549	252	297	1.8	3.9	0.0
75 and above	2	1	1	775	334	441	2.5	2.9	2.2

AFI, acute febrile illness.

* Source of population data: National Minimum Development Indicators.[10]

these were assumed to have a male-to-female ratio of 1:1, consistent with the ratio of cases with known sex in Onoun. There were also 19 cases with no age indicated (17 from Onoun and two from Chuuk Lagoon); these were assigned the same age distribution as the 149 cases for which age was known.

Information on clinical symptoms was extracted from the medical records for 55 (33%) of the 168 suspected cases. Reasons for not being able to extract clinical symptoms were a poor patient history or illegible clinician handwriting. The age and sex distribution of all cases was compared with these 55 cases. The distribution of symptoms was compared with other studies that had assessed symptom distribution in particular disease outbreaks.[3–6] Control measures were implemented.

Laboratory investigation

Diagnostic efforts included nasopharyngeal (NP) swabs for seven cases with a cough and/or a runny nose.These were tested by the reference laboratory in Hawaii using PCR. The reference laboratory was unable to grow the viruses or a sufficient titre for the haemagglutination-inhibition test for antigenic analysis.

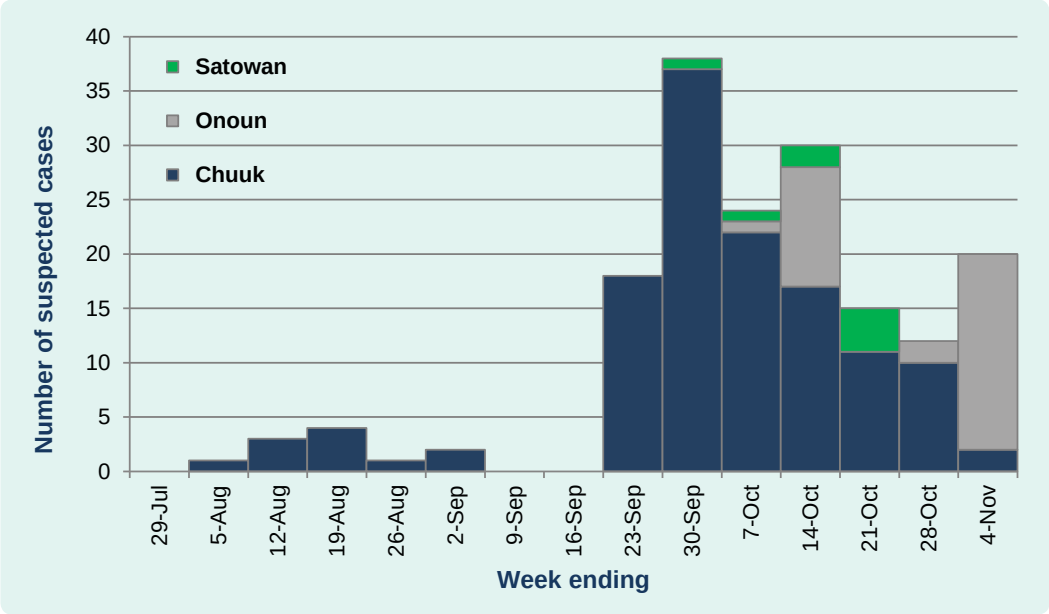
RESULTS

While the causative agent of the outbreak was unknown, the 18-year-old diagnosed with DENV-4 in August was interviewed. It was revealed he had no history of travel and lived adjacent to a construction company headquarters undertaking major road works. There were reportedly many potential mosquito breeding sites in and around these headquarters. The road construction workers residing at the headquarters were primarily Marshallese and Filipino. The Marshall Islands was experiencing a DENV-4 outbreak at the time, and dengue incidence peaks from July to September in the Philippines, although it was unclear whether DENV-4 was circulating at that time.[7] The 18-year-old recovered and was discharged from hospital on 13 August.

Cases

During the period 5 August to 4 November 2012, there were 168 cases of AFI reported in Chuuk State (**Table 1**). Of these, 62% were less than 20 years of age (34% less than five; 28% aged 5–19) and 60% were male (61% in Chuuk Lagoon, 53% in Onoun and 62% in Satawan). The highest attack rate was in the age group 0–4, followed by 60–64 and

Figure 2. **Reported cases of acute febrile illness per week by affected island, 5 August to 4 November 2012, Chuuk State, Federated States of Micronesia (*n*=168)**



15–19. The age group with the highest number of cases was 0–4, followed by 15–19, 5–9 and 20–24. A similar pattern by age was found by site, although in Onoun, the 60–64 age group had the third highest number of suspected cases.

Of all 168 cases, 128 (76%) were from Chuuk Lagoon, 32 (19%) were from Onoun, and eight (5%) from Satawan. Onoun had a substantially higher attack rate of suspected cases, given its relatively small population (53 suspected cases/1000 population compared to 3.2/1000 in Chuuk Lagoon and 2.7/1000 in Satawan).

Epidemic curve

There were three peaks in weekly totals (30 September, 14 October and 4 November). In Chuuk Lagoon, there was a steady decline from 30 September. Clinicians believed that this decline was a genuine decline in cases as opposed to less attention to case detection than in previous weeks. For the outer island of Onoun, the first suspected case fell ill on approximately 19 September, and the weekly number of cases peaked on 14 October and 4 November. This second peak was at the time of the investigation; case numbers declined the following week. For the outer island of Satawan, the first suspected case fell ill in late September, and the weekly number of cases peaked on 21 October and subsided after that (**Figure 2**).

Clinical features

The age and sex distribution of cases with symptom information (n = 55) was similar to the age and sex distribution in all cases (n = 168). In total, there were 195 symptoms reported in the records from these 55 cases (**Table 2**). Eleven cases reported one or two symptoms, 36 cases reported three or four symptoms, and eight cases reported five or more symptoms. The most commonly reported symptoms were acute fever (96%), cough (82%), headache (67%) and runny nose (44%). Rash was reported in only two of the 55 suspected cases, and eye pain and haemorrhage were reported once.

Twelve (7%) of the 168 suspected cases were hospitalized. Information on clinical symptoms was available from nine of these. Again, acute fever (100%), cough (78%) and headache (56%) were the most commonly reported symptoms. Joint pain was reported in three cases (33%). The Chuuk State Hospital doctors who were interviewed stated that the typical symptoms they observed in the suspected dengue cases were

Table 2. **Proportion of patients presenting with described symptoms extracted from medical charts by affected island, Chuuk State, Federated States of Micronesia, 5 August to 4 November 2012 (n = 55)**

	Total	Chuuk Lagoon	Onoun	Satawan
Population	53 000*	40 000	600	3000
Suspected cases	168	128	32	8
Cases with symptom data	55	20	30	5
Acute fever	96%	90%	100%	100%
Cough	82%	65%	90%	100%
Headache	67%	20%	93%	100%
Runny nose	44%	15%	70%	0%
Joint pain	13%	15%	7%	40%
Nausea	11%	10%	7%	40%
Vomiting	7%	10%	3%	20%
Dizziness	7%	20%	0%	0%
Muscle pain	5%	5%	7%	0%
Diarrhoea	4%	10%	0%	0%
Weakness	4%	5%	3%	0%
Rash	4%	0%	3%	20%
Dehydration	4%	0%	7%	0%
Eye pain	2%	0%	0%	20%
Signs of haemorrhage	2%	0%	3%	0%
Loss of appetite	2%	5%	0%	0%
Irritability	2%	5%	0%	0%
Numbness	2%	5%	0%	0%
Menorrhagia	2%	0%	0%	20%
Bone pain	0%	0%	0%	0%
Sore throat	0%	0%	0%	0%

* Source of population data: National Minimum Development Indicators.[10]

flu-like symptoms: very high fever, muscle pain, severe headache, nausea, vomiting and some eye pain and joint pain.

Determining the cause of the outbreak

The investigation revealed the average symptomatic presentation in the 55 cases was not typical for dengue[3] (**Table 3**). In the 55 cases, while fever and headache were common, there were very few cases with joint pain, eye pain, rash, nausea or vomiting and no cases with bone pain or body pain. Also, cough, which was present in 82% of the sample, is not a typical symptom of dengue. Rather the clinical presentation in the 55 cases was suggestive of a disease dominated by upper respiratory tract symptoms, so respiratory viruses, primarily influenza virus and corona virus, and bacterial infections such as *Mycoplasma pneumoniae* were considered. Measles virus infection seemed unlikely with the low frequency of rash[8] and 91% measles immunization coverage.[9]

Typical symptoms for people having influenza are fever (90–100%), cough (82–93%), runny nose (39–91%), muscle pain (18–94%), and headache (17–91%).[4–6] These were generally consistent with the symptomatic presentation of the 55 cases; the only exception was muscle pain, which was not reported, recorded or readable among the 55 cases. The influenza season in Chuuk is reported by programme staff to be August to December.

Laboratory investigation

Three of the seven samples were positive for influenza A(H3).

Control measures

Following the epidemiological investigation, public health measures for the prevention and control of respiratory diseases were undertaken. This included

Table 3. **Symptoms reported in acute febrile illness cases compared with dengue and influenza symptoms from other studies, Chuuk State, Federated States of Micronesia, 5 August to 4 November 2012 (n = 55)**

Cases with symptom data	Chuuk State	Dengue[3]	Influenza[4]	Influenza[5]	Influenza[6]
	55	3926	2470	200	153
Acute fever	96%	88%	90%*	90%	100.0%
Cough	82%	35%	93%	82%	89.5%
Headache	67%	78%	91%	17%	56.0%
Runny nose	44%	29%†	91%†	60%	39.0%
Joint pain	13%	64%	Not reported	Not reported	41.0%
Bone pain	0%		Not reported	Not reported	Not reported
Nausea	11%	53%	Not reported	5%	13.0%
Vomiting	7%		Not reported		6.0%
Muscle pain	5%	77%‡	94%	18%	56.0%
Diarrhoea	4%	30%	Not reported	2%	1.0%
Rash	4%	53%	Not reported	Not reported	Not reported
Eye pain	2%	63%	Not reported	Not reported	Not reported
Signs of haemorrhage	2%	24%	Not reported	Not reported	Not reported

* Feverishness

† Nasal congestion

‡ Body pain

triage of patients at health-care settings and ensuring all persons with symptoms of a respiratory infection adhere to respiratory hygiene, cough etiquette and hand hygiene. The incidence of AFI returned to normal levels by the end of November. It is difficult to assess how much these control measures contributed to this. Control measures would have had the greatest impact had they been introduced earlier in the outbreak.

DISCUSSION

This example demonstrated that outbreak investigations can be challenging, and unrelated cases, such as a laboratory-confirmed DENV-4 case or outbreaks in surrounding areas, may affect clinical diagnoses. The application of descriptive epidemiology to characterize outbreaks by clinical features, time, place and person may help prevent mischaracterizations. It also shows the potential for concurrent outbreaks with very different etiologies but relatively similar clinical manifestations that can add further to the challenge of finding the cause.

Limited on-island laboratory capacity for confirmatory testing and the need to utilize off-island reference laboratories increase costs and introduce delays in obtaining definitive diagnoses in many Pacific island settings. Descriptive epidemiology can guide appropriate laboratory testing and reduce the resource burden of shipping specimens for reference testing. Geographic remoteness, as was the case in the outer islands of Chuuk, can lead to delays in taking blood samples; these may often be taken after the viraemic period, and tests aiming to find pathogens (culture, antigen or PCR tests) may thus be falsely negative. Serological tests that rely on paired acute and convalescent specimens may be limited in this setting since very few patients return for a convalescent sample.

Investigation of respiratory disease outbreaks can be particularly challenging for a variety of reasons. The typical clinical presentation is often sufficiently indistinct and causes confusion as to the etiology. Further, good quality specimens and the correct biological testing are often difficult to obtain in a timely manner.[10,11] In Chuuk, it was also reported that there was some resistance from patients to having a nasopharyngeal swab performed.

While public health measures for the prevention and control of respiratory diseases were undertaken, the outbreak was largely over by the time these were implemented. Early alert and investigation of outbreaks is important in identifying the source of the outbreak and essential for control measures to be directed at the most effective time of the outbreak.[11] Also, as many pathogens can be found only a short time after the onset of illness,

patient consultation early in the course of the illness can help ensure the pathogen is still possible to detect in the clinical specimen.[11]

Despite the outbreak being almost over by the time of the investigation, there were several benefits to undertaking it. These include: identifying strengths and weaknesses of the surveillance and response system; making recommendations to prevent and contain future outbreaks; understanding disease patterns in the state, country and region for informing policy and practice; building capacity among the investigation teams; and demonstrating to the public that the outbreak is being taken seriously.[10]

With increasing domestic and international travel, respiratory disease outbreaks such as influenza can spread rapidly.[12] Since the SARS epidemic, researchers have found that air transportation is responsible for the global pattern of emerging diseases[13] and that every person is potentially no more than 24 hours away from being affected by any epidemic happening somewhere in the world. Outbreak surveillance and response is an essential process to limit the spread and impact of outbreaks. Global and regional partners need to continue to invest in building the capacity of low- and middle-income countries in the surveillance and response of outbreak-prone diseases.

Syndromic surveillance in the Pacific was established in 2010, whereby countries report syndromes using clinical signs and symptoms rather than laboratory confirmation. This is a very useful tool in resource-constrained and dispersed settings such as the Pacific. While the syndromic surveillance systems have improved across the region, there is still substantial work to do to improve the utilization, accuracy and confidence in these systems.[14] This will also assist countries to comply with the International Health Regulations (2005), which require countries to have the capacity to detect and notify public health events of international concern including infectious disease outbreaks.[15] This is even more important given the current extent of international travel.

Since the outbreak reported in this paper, Chuuk State has improved its syndromic surveillance system and now uses one integrated form that incorporates all four standard Pacific syndromic surveillance syndromes and their respective case definitions. These forms are reviewed twice a week to improve early detection of outbreaks.

Ascertaining the cause of this outbreak in a timely manner was greatly limited by the logistical barriers, particularly the remoteness of Chuuk, the limited on-island laboratory capacity and the cost and time required to obtain definitive diagnoses at reference laboratories. All of this meant the outbreak was largely over by the time the causative pathogen was identified. The low number of cases also limited the power of the investigation and the conclusions that can be drawn from it. Patients who had stronger or a greater number of symptoms may have been more likely to have them recorded and therefore the sample of 55 may not have been representative of the 168 cases in terms of symptomatic distribution. Also, some of the cases that presented with AFI in Chuuk may have been influenced by anxiety stemming from the concurrent dengue outbreaks in Yap and Kosrae states.

It is important to note that illegible clinician handwriting had a negative impact on the investigation. Conversely, those doctors whose writing was legible had an important influence on identifying the cause of the outbreak. A substantial amount of research on this issue has been undertaken in other areas of the world and has found that the problem of illegible handwriting among clinicians is common and widespread.[16–21] There is a need for research into approaches to solve this problem. Some have suggested electronic records will assist,[18] but this may not be feasible in the short term in many settings. Other solutions need to be explored.

CONCLUSION

Clinical diagnosis of AFI can often be difficult and misleading. This can mean that opportunities for preventive measures early on in an outbreak are missed. In any outbreak, health professionals should undertake prompt descriptive epidemiological analyses to help ascertain the cause of the outbreak. A line list using all available sources of information is a valuable tool in outbreak investigation, and analysis of the symptomatic presentation can greatly assist in finding the cause(s) of the outbreak. Laboratory systems to collect, ship where required and analyse specimens need to be prompt, well rehearsed and resourced to aid outbreak investigation.

Conflicts of interest

None declared.

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Seroprevalence survey of brucellosis among rural people in Mongolia

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Background: After the transition from socialism to a market economy in 1990, human brucellosis re-emerged in Mongolia. The aim of our study was to estimate a representative seroprevalence of *Brucella* spp. and to determine risk factors for brucellosis seropositivity among rural people.

Methods: A cross-sectional study with multistage random selection was conducted in eight provinces of Mongolia. Study participants were interviewed using a questionnaire to obtain their brucellosis history, current symptoms and likely risk factors. Blood samples were drawn to determine brucellosis seroprevalence.

Results: A total of 2856 randomly selected rural people aged four to 90 years were enrolled in the study. The seroprevalence of *Brucella* spp. was 11.1% (95% confidence interval [CI]: 10.0–12.1), ranging between 2.3% and 22.6% in the eight provinces; 39.2% (n = 609) of nomadic camps had at least one seropositive participant. Risk factors associated with brucellosis seropositivity were being older than 45 years (adjusted odds ratio [AOR] = 6.9, 95% CI = 5.1–8.7) and being a veterinarian (AOR = 2.8, 95% CI = 1.5–5.0).

Conclusion: Our study confirms that human brucellosis seroprevalence among rural people in Mongolia is high. Human brucellosis can be effectively controlled if high-coverage livestock mass vaccination is implemented with a coverage survey after the vaccinations to ensure completeness. This mass vaccination should be accompanied by public awareness and educational programmes.

Brucellosis is a zoonosis, and the infection is almost invariably transmitted by direct or indirect contact with infected animals or their products. It is an important human disease in many parts of the world, especially in the Mediterranean countries of Europe, North and East Africa, the Middle East, South and Central Asia and Central and South America.[1]

Brucellosis is caused by members of the *Brucella* genus. Transmisson of infection to humans occurs through breaks in the skin, following direct contact with tissues, blood, urine, vaginal discharges, aborted fetuses or placentas.[2] The most frequent symptoms of brucellosis are fever, chills or shaking, malaise, generalized aches and pains all over the body, joint and low back pain, headaches, anorexia, easy tiredness and general weakness.[3]

Mongolia has the second highest incidence of human brucellosis worldwide; another seven republics of the former Soviet Union are included in the 25 countries with the highest incidence. According to data from the National Statistical Office of Mongolia, a rapid increase in notified cases of brucellosis was observed between 1990 and 2000. The increase may have been the result of the evolution from a socialist state to a free market economy which led to the loss of rigorous livestock control.[4] During this period, changes to the health system precluded early recognition of the disease or interventions that considered the emerging trends in humans and animals.[5] In Mongolia, factors contributing to the incidence of brucellosis include traditional eating habits, standard hygiene measures, methods for processing milk and its products and rapid movement of animals.[3]

In 2011, a national brucellosis serosurvey was conducted that sampled 168 027 head of livestock from 11 528 nomadic camps (two to more than four herder families that share the same pasture and water source) of 337 districts of 21 provinces.[6] Twenty-one provinces, 57.3% of all districts and 8.0% of all nomadic camps

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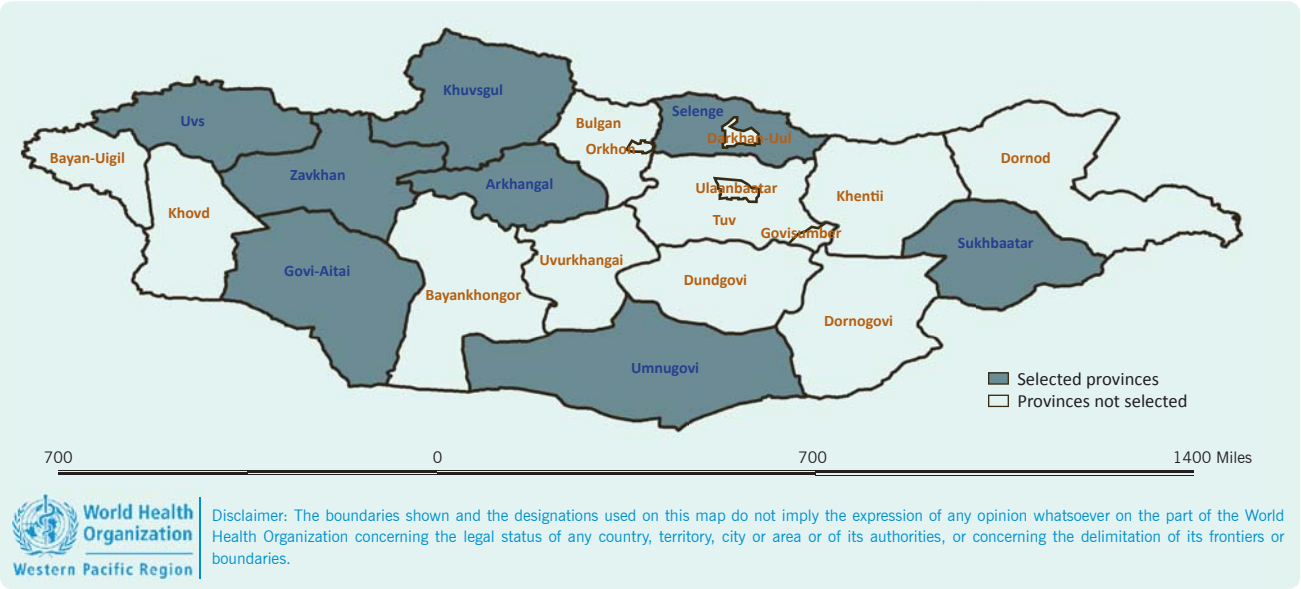
Figure 1. **Map of Mongolia by province highlighting provinces where the study was conducted**



had seropositive livestock including camels, cattle, sheep and goats. Livestock seroprevalence was found in 0.7% of camels, 1.8% of cattle, 0.7% of sheep and 0.5% of goats using parallel interpretations of Rose Bengal Tests (RBT), complement fixation tests and competitive-enzyme-linked immunoabsorbent assay (ELISA).[6]

The aim of our study was to estimate the seroprevalence of *Brucella* spp. and to determine risk factors for brucellosis seropositivity among rural people.

METHODS

Study design and population

Eight provinces were selected for the cross-sectional surveys. Between June and September 2010, surveys were conducted in Sukhbaatar and Zavkhan provinces, selected for convenience.[7] Between November 2011 and January 2012, the same surveys were conducted in a further six provinces: Arkhangai, Khuvsgul, Selenge, Uvs, Umnugovi and Govi-Altai (**Figure 1**). In each province, four districts were selected using simple randomization in Excel (the rand () command). Twenty nomadic camps and four to five individual participants were randomly selected based on the required sample size.

The cluster sample size calculation as described elsewhere[7] assumed a human brucellosis seroprevalence among Mongolian rural people of 20%.[8] In addition, the number of clusters and number of individuals per cluster was optimized according to the feasibility and the available budget.

The study was approved by the Ethics Committee of the Health Sciences University of Mongolia and the Ethics Committee of the Canton of Basel of Switzerland. All participants were informed about the study and what they could expect regarding diagnosis, reporting and treatment; all signed a consent form. A child younger than 16 years of age was included in the study with signed consent from of his/her parents.

Data collection

Study questionnaire

All study participants were interviewed using a questionnaire which included demographics, risk factors and clinical symptoms for brucellosis. The questionnaire was pre-tested during the 2010 study in Sukhbaatar and Zavkhan[7] and revised for the extended study to improve understanding of questions and to eliminate overly-sensitive questions.

Blood sample collection and handling

Venous blood was taken with 5 ml Vacutainer® tubes. The blood samples were centrifuged in 3000 rounds per minute for five minutes. Separated 1.5 ml tubes of serum were kept in a cool box and transported to the provincial laboratories for storage and cooling before shipment

Table 1. **Number of participants seropositive for *Brucella* spp.* by province, Mongolia, 2010 to 2012**

Province	Number of districts surveyed	Number of participants	Seropositives	% of positivity*	95% confidence interval
Khuvsugul	4	400	46	11.5	8.72–14.2
Umnugovi	4	400	49	12.3	9.64–14.9
Govi-Altai	4	398	30	7.5	4.17–10.8
Selenge	4	391	60	15.3	12.9–17.6
Arkhangai	4	400	9	2.3	0.45–9.15
Uvs	3	293	17	5.8	1.27–10.3
Sukhbaatar	4	318	72	22.6	20.5–24.6
Zavkhan	4	256	33	12.9	9.7–16.1
Total	31	2856	316	11.1	10.0–12.1

* Based on parallel interpretation of the RBT and ELISA test.

to the serological laboratory of the National Center for Communicable Diseases in Ulaanbaatar where they were tested for brucellosis.

Serological test

Sera were tested with the RBT for detection of antibodies to *Brucella abortus/melitensis* from Tulip Diagnostic Ltd (Bambolim, India). Positive sera were re-tested with the RBT using ½ to 1/32 serum dilutions,[9] and with enzyme immunoassay for the qualitative determination of IgG class antibodies against *Brucella* from the NovaTec Immundiagnostica GmbH (Dietzenbach – 63128 Germany). The ELISA test was performed according to manufacturer's instruction.

Data entry and statistical analysis

All data were double-entered in Access 2007, compared in Epi Info™ 3.5 to correct entry errors and analysed using STATA 10.1. Study participants who tested positive by either ELISA or RBT were considered seropositive for the statistical analysis.

To assess the association between risk factors and human brucellosis seropositivity we used Pearson χ^2 or Fisher's exact tests for explanatory variables such as demographics, behaviour-related risk factors and reported clinical symptoms. We also conducted univariate logistic regression using the binary serological outcome with the xtgee command and random effect on the nomadic camp level. A multivariate logistic regression model (with random effect at the nomadic camp) using backward stepwise selection and a removal level for covariates at $P = 0.10$ based on the likelihood-ratio test was then constructed. Variables with p values less than 0.05 in the univariate analysis were included in the multivariate model.

To determine the proportion of the general population seroconverting each year due to brucellosis exposure, the seroprevalence data were divided by the duration of seropositivity, assumed to be 10.9 years.[10] Using a conservative estimate of 20% of seroconversions representing true clinical cases (note that among all seropositives detected, 58.5% had at least two symptoms and 31.5% had at least three symptoms at time of interview), these proportions were multiplied by 0.3 and converted to rates per 100 000 for the general population.

RESULTS

There were 2856 study participants from 609 nomadic camps from 31 districts in the eight selected provinces between four and 90 years of age (median 38 years). This included 2260 (79.1%) herders, 142 (5.0%) students, 96 (3.4%) office workers, 70 (2.5%) workers, 37 (1.3%) retired people, 20 (0.7%) veterinarians, 18 (0.6%) entrepreneurs, 16 (0.6%) unemployed adults, 13 (0.5%) children under six years, and 184 (6.4%) other residents.

Seroprevalence

The seroprevalence of *Brucella* spp. among participants was 11.1% (95% CI: 10.0–12.1) ranging from 2.3% to 22.6% in the eight provinces (**Table 1**) and 4.1% to 43.8% in the 28 districts. Within nomadic camps, 39.2% (95% CI: 38.2–41.0) had at least one

Table 2. **Number of nomadic camps with members seropositive for *Brucella* spp., Mongolia, 2010 to 2012**

Province	Number of nomadic camps surveyed	Positive	% of positivity*	95% confidence interval
Arkhangai	79	7	8.9	1.89–15.9
Govi-Altai	80	28	35.0	32.0–37.9
Khuvsgul	82	35	42.7	40.2–45.1
Umnugovi	80	33	41.3	38.1–44.5
Uvs	58	13	22.4	17.5–27.2
Selenge	78	40	51.3	49.1–53.4
Sukhbaatar	83	56	67.5	65.9–69.0
Zavkhan	69	27	39.1	36.1–42.0
Total	**609**	**239**	**39.2**	**38.2–41.0**

* Based on parallel interpretation of the RBT and ELISA test

to four seropositive members (**Table 2**). This equated to an annual incidence of seroconversion of 1145 per 100 000 and an overall annual incidence of 229 clinical cases per 100 000.

Seroprevalence was higher in females than in males (11.2% compared with 10.9%, $P = 0.029$). By age group, the highest seroprevalence was found in those 45 years and above at 15.5% (95% CI: 13.9–17.0), with the lowest in the four to 10 year age group at 2.6% (95% CI: 1.5–20.4). All occupation categories included seropositive cases ranging between 2.8% and 30.0% (**Table 3**).

Analysis of risk factors for brucellosis

Risk factors associated with being seropositive in univariate analysis included: being 45 years old and above (odds ratio [OR] = 6.6, $P = 0.046$), being a veterinarian (OR = 3.5, $P = 0.016$), contact with aborted animal fetuses and placentas (OR = 1.35, $P = 0.016$) and consumption of undercooked liver (OR = 1.51, $P = 0.001$) (**Table 3**).

In the multivariate analysis, only two variables remained associated with being seropositive: being 45 years old and above (adjusted odds ratio [AOR] = 6.9, 95% CI: 5.1–8.7) and being a veterinarian (AOR = 2.8, 95% CI: 1.5–5.0). Among veterinarians who participated in the study, 72.7% assisted in livestock obstetric work, and 50% had direct contact with aborted animal fetuses and placentas. The risk factors for veterinarians was also much higher compared with other occupations ($P < 0.001$).

History of human brucellosis and clinical symptoms

Of the study participants, 2.7% ($n = 76$) reported receiving treatment for human brucellosis in the past; the median time since past brucellosis treatment was 14 years (Q1 = 3.3 and Q3 = 20 years). With the exception of testicular pain, there were significant differences between age groups in reporting clinical symptoms; the age groups of 20 to 44 years and 45 years and above reported more clinical symptoms for human brucellosis. Females also reported more headaches; joint, back and muscle pain; weakness and sleeping disturbances than males (**Table 4**).

Reported clinical symptoms at the time of the study were compared to the sero-status of participants. Overall, 165 of the 316 (52.2%) brucellosis seropositive participants and 1186 of the 2540 (46.7%) seronegative participants reported symptoms. Among all seropositives, 36.7% reported more than three symptoms; among the seronegatives, 23.1% reported more than three symptoms ($P < 0.001$). Headache; joint, back and muscle pain; night sweats and sleeping disturbances were significantly associated with brucellosis seropositivity (**Table 5**).

DISCUSSION

We report a seroprevalence of *Brucella* spp. among rural people of 11.1% (with a range between provinces from 2.3% to 22.6%) and an annual incidence of 229 per 100 000. The high incidence in the study likely reflects an increase in human brucellosis after

Table 3. **Univariate analysis of risk factors of brucellosis seropositivity* in Mongolia, 2010 to 2012**

Characteristic	Number of participants	Number seropositive (%)	OR (95% CI)	*p* value
Age group (years)				
4–9	39	1 (2.6)	1.0	–
10–14	69	4 (5.8)	2.3 (1.2–4.1)	0.440
15–19	96	3 (3.1)	1.2 (0.6–2.7)	0.864
20–44	1769	171 (9.7)	3.9 (1.2–7.6)	0.151
45 and above	883	137 (15.5)	6.6 (4.5–10.2)	0.046
Sex				
Males	1181	132 (11.2)	1.0	–
Females	1675	184 (10.9)	1.0 (0.9–1.2)	0.968
Occupation				
Herder	2260	263 (11.6)	1.3 (0.9–2.5)	0.087
Student	142	4 (3.0)	0.9 (0.3–2.5)	0.345
Office worker	96	7 (7.3)	0.7 (0.2–1.6)	0.267
Worker	70	7 (10.0)	0.9 (0.5–2.0)	0.733
Retired	37	7 (18.9)	2.0 (0.8–4.2)	0.112
Veterinarian	20	6 (30.0)	3.5 (1.6–7.9)	0.016
Entrepreneur	18	4 (22.2)	2.3 (1.0–4.6)	0.119
Unemployed	16	1 (6.3)	0.5 (0.3–1.3)	0.521
Children under six	13	1 (7.7)	0.7 (0.3–1.6)	0.708
Other	184	16 (8.7)	0.8 (0.4–1.7)	0.328
Risk factors				
Animal obstetric work	778	93 (11.9)	1.5 (0.9–2.5)	0.121
Contact with aborted animal fetuses and placentas	769	104 (13.5)	1.4 (1.0–2.1)	0.016
Consumption of raw milk	295	32 (10.8)	1.2 (0.7–1.8)	0.546
Consumption of raw liver	38	11 (28.9)	0.8 (0.5–1.2)	0.612
Consumption of undercooked liver	1067	146 (13.7)	1.5 (0.9–4.3)	0.001
Consumption of fresh animal blood	143	12 (8.4)	1.5 (1.0–1.7)	0.332

OR, odds ratio; CI, confidence interval.

* Based on parallel interpretation of RBT and ELISA

the transition in Mongolia from socialism to a market economy leading to livestock privatization and collapse of the veterinary sector.[4]

Although several earlier studies also estimated the seroprevalences of *Brucella* spp. in Mongolia among high-risk people including herders, veterinarians and raw animal processing technicians,[11–14] these differed from our study in time, study design and methodology and should not be compared. The result from our study was higher than the 0.1% to 10.1% reported among high-risk people in other countries,[10,15–21] which is not surprising as Mongolia is ranked second in the world for brucellosis incidence.[5] We also estimated a much higher incidence compared with that reported from notification data,[22] despite the fact that we have taken a conservative assumption that 20% of seropositive cases are clinical cases.

According to the multivariate analysis, adults aged 45 years and above and veterinarians had a higher risk for brucellosis. This age group plays an important role in livestock herding and birthing, and veterinarians have direct contact with animals and aborted materials when doing veterinary examinations. We also found seropositives in all age groups, including in young children (four to nine years), which may indicate ongoing exposure and transmission of brucellosis in rural Mongolia. These groups should be targeted with material about protection against brucellosis infection.

This study will serve as a baseline of the seroprevalence of *Brucella* spp. in rural people in Mongolia before the implementation of a nationwide livestock vaccination campaign; it also will be used for ongoing brucellosis surveillance. A decrease of human incidence and repeated sero-surveillance surveys

Table 4. **Reported clinical symptoms among study participants by age group and sex, Mongolia, 2010 to 2012 (N = 2856)**

Symptoms	n	Age group					*p* value*	Sex		*p* value*
		0–9	10–14	15–19	20–44	45 and above		Male	Female	
		%	%	%	%	%		%	%	
Fever	135	0.7	1.6	0.7	52.6	44.4	0.009	3.8	5.4	0.053
Headache	1268	0.3	0.7	2.0	57.9	39.1	< 0.001	34.3	51.8	< 0.001
Joint pain	1287	0.4	0.5	1.5	50.7	46.9	< 0.001	38.7	49.5	< 0.001
Back pain	1351	0.1	0.4	1.4	57.6	40.5	< 0.001	43.6	49.8	0.001
Muscle pain	590	0.5	1.0	1.0	46.4	51.1	< 0.001	14.9	24.7	< 0.001
Weakness	964	0.3	0.3	0.4	50.7	48.3	< 0.001	26.9	38.6	< 0.001
Night sweats	336	0.9	0.6	0.6	45.8	52.1	< 0.001	11.4	12.0	0.812
Sleeping disturbance	530	0.2	–	0.4	42.3	57.1	< 0.001	14.5	21.4	< 0.001
Weight loss	233	1.3	1.3	1.3	40.7	55.4	< 0.001	7.2	8.8	0.115
Miscarriage	31	–	–	–	90.3	9.7	0.015	–	100.0	< 0.001
Testicular pain	10	–	–	–	50.0	50.0	0.749	100.0	–	< 0.001

* Either derived from the χ^2 test or Fisher's exact test.

Table 5. **Reported clinical symptoms by sero-status among study participants, Mongolia, 2010 to 2012 (N = 2856)**

Clinical symptoms		Number of participants	Number seropositive (%)	*p* value
Fever	No	2721	301 (11.1)	
	Yes	135	15 (11.1)	0.561
Headache	No	1588	167 (10.5)	
	Yes	1268	149 (11.8)	< 0.001
Joint pain	No	1569	155 (9.9)	
	Yes	1287	161 (12.5)	0.014
Back pain	No	1505	151 (10.0)	
	Yes	1351	165 (12.2)	0.038
Muscle pain	No	2266	234 (10.3)	
	Yes	590	82 (13.9)	0.009
Weight loss	No	2623	287 (10.9)	
	Yes	233	29 (12.4)	0.379
Weakness	No	1892	194 (10.3)	
	Yes	964	122 (12.7)	0.058
Night sweats	No	2520	266 (10.6)	
	Yes	336	50 (14.9)	0.013
Sleeping disturbance	No	2326	242 (10.4)	
	Yes	530	74 (14.0)	0.010
Miscarriage	No	1644	182 (11.1)	
	Yes	31	2 (6.4)	0.713
Testicular pain	No	1171	131 (11.2)	
	Yes	10	1 (10.0)	0.620

in humans will indirectly assess the efficacy of the vaccination campaign in livestock.[23]

There were several limitations to the study. First, association between human and livestock seropositivity was not assessed in provinces (with the exception of Zavkhan and Sukhbaatar[7]). There also may have been temporal variations in risk factors for childhood brucellosis, interpretation of reported clinical symptoms for brucellosis based on seropositivity and pathogen exposure that were not captured by the cross-sectional study design.

CONCLUSION

Our study confirms that human brucellosis seroprevalence among rural people in Mongolia is high and that the incidence is much higher than the notification data suggests. As recommended by the Food and Agriculture Organization of the United Nations, the World Organization for Animal Health and the World Health Organization, mass livestock vaccination is required in Mongolia in the mobile livestock production system.

Safety measures to avoid brucellosis include wearing protective clothes such as gloves, using metal hooks to collect aborted fetuses and placentas for burial or burning, washing hands after handling livestock and completely cooking liver from small ruminants. This information should be included in educational materials to prevent as many as possible new cases, especially at the beginning of the mass vaccination campaign while strains still circulate. We have developed written and pictorial educational materials mainly for children. The literacy rate in Mongolia is extremely high and thus printed media are appropriate. In parallel, the surveillance, treatment and diagnostic capacities for human brucellosis must be increased in provinces and districts. Education and awareness programmes should be implemented particularly before the livestock birthing season.

Conflicts of interests

None declared.

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Seroprevalence survey of avian influenza A(H5N1) among live poultry market workers in northern Viet Nam, 2011

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Objective: Highly pathogenic avian influenza A(H5N1) is endemic in poultry in Viet Nam. The country has experienced the third highest number of human infections with influenza A(H5N1) in the world. A study in Hanoi in 2001, before the epizootic that was identified in 2003, found influenza A(H5N1) specific antibodies in 4% of poultry market workers (PMWs). We conducted a seroprevalence survey to determine the seroprevalence of antibodies to influenza A(H5N1) among PMWs in Hanoi, Thaibinh and Thanhhoa provinces.

Methods: We selected PMWs from five markets, interviewed them and collected blood samples. These were then tested using a horse haemagglutination inhibition assay and a microneutralization assay with all three clades of influenza A(H5N1) viruses that have circulated in Viet Nam since 2004.

Results: The overall seroprevalence was 6.1% (95% confidence interval: 4.6–8.3). The highest proportion (7.2%) was found in PMWs in Hanoi, and the majority of seropositive subjects (70.3%) were slaughterers or sellers of poultry.

Discussion: The continued circulation and evolution of influenza A(H5N1) requires comprehensive surveillance of both human and animal sites throughout the country with follow-up studies on PMWs to estimate the risk of avian–human transmission of influenza A(H5N1) in Viet Nam.

Highly pathogenic avian influenza A(H5N1) viruses re-emerged in south-eastern Asia in 2003, and these viruses continue to circulate widely among domestic poultry in the region.[1] Numerous outbreaks of influenza A(H5N1) viruses have occurred, with limited transmission to humans and as of yet unclear potential for sustained human-to-human transmission. However, the continuing evolution and genetic diversification of influenza A(H5N1) viruses is worrying since as few as four amino acid changes are necessary to render the viruses transmissible between ferrets, reinforcing the ongoing pandemic threat from these viruses.[2–4]

In Viet Nam, as of July 2014, there have been 127 human cases of influenza A(H5N1) infection with 63 deaths. Since the influenza A(H5N1) epizootic first began in Viet Nam in 2003, three main clades have circulated and been associated with human infections (clades 1, 2.3.4 and 2.3.2.1).[1,5] Contact with sick or dead poultry has been consistently identified as a risk factor for human influenza A(H5N1) infection, and live poultry markets have been shown to be important locations for amplifying influenza A(H5N1) virus transmission.[6,7] An antibody seroprevalence study conducted among 200 poultry market workers (PMWs) in Hanoi in 2001 detected antibodies against influenza A(H5N1) virus in 4% of subjects,[8] suggesting that there were human infections with influenza A(H5N1) before the first case was officially confirmed.[9] In addition, subclinical, asymptomatic or mildly symptomatic cases were reported during outbreak investigations.[9–11] Similarly, seroprevalence studies have been conducted in Thailand, Cambodia and Indonesia as part of comprehensive outbreak investigations to evaluate key clinical, epidemiological and serological aspects related to human influenza A(H5N1) infections.

To assess if exposure to influenza A(H5N1) viruses among PMWs has changed over this period, we conducted a seroprevalence study among PMWs in three provinces of northern Viet Nam in 2011.

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MATERIALS AND METHODS

Sample and protocol

A cross-sectional seroprevelance study was conducted among adult workers at five markets selling live poulty in the provinces of Hanoi, Thaibinh and Thanhhoa in northern Viet Nam. Sample size was estimated based on a reported seropositive rate of 4% among PMWs in Hanoi in 2001,[9] with a confidence level of 95% and 1.5% confidence interval (CI) ranging from 2.45% to 5.55%. To account for uncooperative participants and unqualified samples, a total of 600 samples were estimated for this study.

Live poultry markets were eligible if their regular number of poultry sellers exceeded 100 individuals and they were located in a large city with a history of laboratory-confirmed cases of human influenza A(H5N1) infection. With the support of local government, 11 poultry markets were nominated. Five markets from three provinces were then randomly selected. Individual participants were eligible if they were aged 18 years or older, currently a trader or slaughterer of live poultry (including waterfowl) and had worked for a minimum of six months in a live poultry market. We enrolled subjects, sampling market to market, until the required number of participants were recruited.

A questionnaire was used to collect information on demographic characteristics and potential occupational risk factors for exposures to influenza A(H5N1). The variables of age, gender, education history, medical history, province of occupation and poultry-related occupational risk exposures were collected. All participants were interviewed face to face. Data were entered into EpiData v3.1 and analysed using STATA v11. Frequencies were calculated with a 95% CI. Seroprevalence among workers was compared across the potential variables using the Pearson's chi-squared test or using Fisher Exact test if any observed value was less than five. Mean values were compared using a t-test to assess whether any differences observed were statistically significant at 95% CI.

All participants were asked to provide 5 ml of venous blood to determine serum antibody concentrations against influenza A(H5N1) viruses. Samples were transported by car in ice boxes at 4 °C to the National Influenza Center at the National Institute of Hygiene and Epidemiology (NIHE) in Hanoi within 24 to 48 hours of collection. Sera was used after centrifugation of whole blood at NIHE (2500 rpm for 15 minutes) and were aliquoted and stored at −70 °C until testing.

Serologic assays

The participants' sera were tested for antibodies to influenza A(H5N1) viruses by horse haemagglutination inhibition assay (HHI) and microneutralization assay (MN) at NIHE following the US Centers for Disease Control and Prevention protocols. All three clades of influenza A(H5N1) virus that have cirulated in Viet Nam since 2004 were used because the antigenic diversity of influenza A(H5N1) viruses may alter the sensitivity of assays to detect strain-specific antibodies. The influenza A(H5N1) viruses used were cultured from selected Vietnamese patients with positive reverse transcription polymerase chain reaction (RT–PCR) assays for clade 1 in 2005 (A/Viet Nam/HN30408/2005), clade 2.3.4 in 2007 (A/Viet Nam/HN31244/2007) and clade 2.3.2.1 in 2011 (A/Viet Nam/CM32/2011). Due to the unknown pathogenic potential of avian/human viruses, all tests involving live virus were conducted in animal-biosafety level 3 and biosafety level 3 laboratories.

The HHI assays were performed as described elsewhere[12] using reference antisera treated with receptor-destroying enzyme before testing. Serum samples were tested at a starting dilution of 1:10 using eight haemagglutinating units of virus and 1% volume/volume horse erythrocytes. All viruses were inactivated by 1% β propiolactone before use. HHI titres were read up to 60 minutes after the addition of erythrocytes and reported as the reciprocal of the highest serum dilution causing complete inhibition of agglutination. Results were accepted if negative sera and horse erythrocytes cell controls provided the correct non-agglutinated pattern. Samples that were negative by HHI assay in the lowest dilution (1:10) and samples resulting in HHI titres ≥40 were tested against the three influenza A(H5N1) clades by MN assay. MN assays were performed according to World Health Organization (WHO) protocols, using 100 x $TCID_{50}$ of live viruses as above.[12,13] Virus was incubated with twofold serial dilutions of sera starting at 1:10 and then incubated with MDCK cells (Madin-Darby canine kidney cells-American Type Culture Collection-ATCC, United States of America) overnight before virus quantitation by enzyme-linked immunosorbent assay (ELISA) to detect influenza nucleoprotein. The titre was

Table 1. **Characteristics of PMWs seropositive for influenza A(H5N1), northern Viet Nam, 2011 (n = 607)**

Characteristics	Number positive/total	%	95% CI	p value
Study areas				
Hanoi	22/305	7.2	4.3–10.1	
Thaibinh	9/169	5.3	1.9–8.7	0.49
Thanhhoa	6/133	4.5	0.9–8.0	
Age group				
0–24	1/31	3.2	0.0–9.8	
25–34	11/124	8.9	3.8–13.9	0.46
35–44	9/185	4.9	1.7–8.0	
45 and above	16/267	6.0	3.1–8.9	
Sex				
Male	13/214	6.1	2.8–9.3	0.99
Female	24/393	6.1	3.7–8.5	
Occupation				
Sellers and slaughterers	31/380	8.2	5.4–10.9	0.06
Others*	6/227	2.6	0.5–4.7	
Maximum education level attained				
High school, college or university	35/555	6.3	4.3–8.3	0.48
Primary/secondary school	2/52	3.9	0.0–9.3	
Medical history				
Chronic medical conditions	6/79	7.6	1.6–13.6	0.55
No chronic medical conditions reported	31/528	5.9	3.9–7.9	

CI, confidence interval.

* Breeders, transporters, veterinarians, drivers, feather collectors, cleaners, market managers.

reported as the reciprocal of the highest dilution that reduced infection by at least 50%.

The WHO seropositive criteria for an influenza A(H5N1)-confirmed case in single serum collected at day 14 or later of symptom onset is an HHI titre >160 and an MN titre >80.[14] However, this is not appropriate for seroprevalence studies as the participants are not suspected influenza A(H5N1) patients. In fact, the Consortium for the Standardization of Influenza Seroepidemiology has not yet devised positive criteria for influenza A(H5N1) assays.[7] For our study, a sample was considered seropositive for influenza A(H5N1) virus antibody if an HHI titre ≥80 and an MN titre ≥20 were obtained in duplicate HHI and MN tests with any influenza A(H5N1) clade. The clades of seropositive samples were classified based on the highest antibody titre by comparison between three clades.[13]

The study protocol was approved by the institutional review board of NIHE. Participation was voluntary and all subjects provided written informed consent to participate in the study.

RESULTS

Characteristics of study population

A serum sample was collected from 607 PMWs during four months (September to December 2011). Of those, 305 samples (50.3%) were from Hanoi, 169 samples (27.8%) from Thaibinh and 133 samples (21.9%) from Thanhhoa.The mean duration of working in a live poultry market was 7.7 years (95% CI: 7.1–8.2) and ranged from six months to 36 years.

The mean age of participants was 42.3 years (95% CI: 41.4–43.2) with a range of 18 to 74 years, and 214 (35.3%) were male. Almost two thirds (62.6%) reported that they were sellers or slaughterers of live poultry, with 37.4% being breeders, transporters and others (veterinarians, drivers, feather collectors, cleaners and market managers). A total of 79 (13.1%) participants reported having a chronic medical condition (hypertension, diabetes, hepatitis). There were 555 (91.4%) participants who had an education level to high-school standard or higher (**Table 1**).

Table 2. **Seropositive participants by influenza A(H5N1) clade and province, northern Viet Nam, 2011**

Study areas	Number positive/ total	Number seropositive				Proportion positive (%)
		Clade 1	Clade 2.3.4	Clade 2.3.2.1	Clades 2.3.4 and 2.3.2.1	
Hanoi	22/305	0	13	2	7	7.2
Thaibinh	9/169	0	6	0	3	5.3
Thanhhoa	6/133	0	5	0	1	4.5
Total	**37/607**	**0**	**24**	**2**	**11**	**6.1**

Seroprevalence of H5 antibodies

There were 37 participants seropositive for (21.9%), giving an overall seropositive rate of 6.1%; (95% CI: 4.6–8.3). Of the 37 seropositive samples, 24 were seropositive to clade 2.3.4, two were seropositive to clade 2.3.2.1 and 11 were seropositive to both (**Table 2**). By province, the proportion of seropositives was 7.2% (95% CI: 4.3–10.1) for Hanoi, 5.3% (95% CI: 1.9–8.7) for Thaibinh and 4.5% (95% CI: 0.9–8.0) for Thanhhoa; these differences were not statistically significant ($P = 0.49$) (**Table 1**).

By age, the highest proportion of seropositivity was in the 25 to 34 year old age group (8.9%, 95%CI: 3.8–13.9) although there was no statistically significant difference across the four age groups ($P = 0.46$). There was also no statistically significant difference by sex ($P = 0.99$), education level ($P = 0.48$) or chronic medical condition ($P = 0.55$) (**Table 1**). By occupation, the proportion seropositive was higher in slaughterers and sellers compared with all others but this was not significant (8.2% compared with 2.6%, $P = 0.06$) (**Table 1**).

DISCUSSION

Our study identified 37 (6.1%) PMWs seropositive for influenza A(H5N1) clade 2.3.4 and clade 2.3.2.1 viruses; these clades were predominant in northern Viet Nam from 2005 to 2013.[15–17] While clade 1 also circulated in Viet Nam from 2003 to mid-2005,[18] no PMWs seropositive for clade 1 was identified in our study. This was unexpected as some of the workers in our study would have been working when this clade was circulating. Our sample of PMWs were from Hanoi, Thaibinh and Thanhhoa where a large number of human influenza A(H5N1) infections have been reported since 2004.[6,18] Although studies conducted in 2004 in Viet Nam and Thailand did not identify contact with healthy poultry as a risk factor of influenza A(H5N1) exposure,[6] a study among PMWs in Hanoi in 2001 showed a 4% seroprevalence of influenza A(H5N1) clade A/Goose/Viet Nam/113/2001.[8] Circulation of influenza A(H5N1) in healthy ducks in Viet Nam[1] demonstrated the possible risk of influenza A(H5N1) infection among persons exposed to healthy poultry in Viet Nam.

In our study population, the working duration of PMWs was an average of 7.2 years (range from six months to 36 years). It is possible then that some of the workers in the study may not have been exposed to the clade 1 virus. That 11 participants were positive for both clade 2.3.4 and clade 2.3.2.1 may have been due to cross-reactivity, coinfections or reinfections since co-circulation of these clades was identified in 2010.[12]

We found no statistically significant difference in the proportion of seropositive PMWs between the three different geographical study areas, which suggests the working conditions or occupational habits of PMWs are similar in these areas. Our study found a higher seropositive rate among PMWs in Hanoi compared to a seroprevalence study conducted in 2001 (7.2% compared with 4.0%), which might be due to the spread of influenza A(H5N1) in Viet Nam occurring after late 2003.[15] We found a higher seropositive proportion in slaughterers and sellers compared with the other occupational groups; this is consistent with studies from China, Thailand and Bangladesh that showed that people who work directly with poultry are more likely to have been exposed to avian influenza A(H5N1).[9,15] A 2010 survey of live bird markets in Viet Nam found 3.3% of poultry samples tested were positive for influenza A(H5N1) by RT–PCR (personal communication: John Weaver, Viet Nam Avian and Human Influenza

Control and Preparedness Project). A survey of healthy ducks in northern Viet Nam in 2006 concluded that while influenza A(H5N1) viruses were not detected in any throat or cloacal swabs, serological analyses suggested that ducks were infected with influenza A(H5N1) viruses in the absence of any recognized outbreak.[11] That our study demonstrated seropositivity to influenza A(H5N1) viruses in all groups of poultry workers suggests that preventive measures such as personal protective equipment and vaccination for PMWs are needed.

Our criteria for seropositivity were modified from the WHO criteria for detection of influenza A(H5N1) infection by serological testing. The WHO criteria are for confirming suspected cases of influenza A(H5N1) and seems not to be practical or sensitive enough for serological surveillance studies since antibody titres against influenza A(H5N1) begin to decrease six to 12 months after exposure and may disappear.[19] We selected a seropositive cutoff titre of HHI ≥80 and MN titre ≥20 since MN's sensitivity is reduced (less than 80%) for adults[20] and all PMWs enrolled in our study were at least 18 years old. Previous comparisons have shown that horse red blood cells HHI assays have high reliability and good agreement with MN assays.[21] Using our cutoff titre might have resulted in an overestimate of the seroprevalence and these cutoffs should be tested with more seroprevalence studies in control populations where influenza A(H5N1) has not yet been detected. Further development of standard criteria for prevalence of antibodies against influenza A(H5N1) is needed for future studies.[7]

There may have been some misclassification in the reporting of occupational exposures in this study as PMWs may not have accurately recalled these exposures over a long period of time. It is also possible that participants may have had more than one poultry-related occupation during their exposure period. The results of our study, that 6.1% of PMWs in northern Viet Nam were seropositive for influenza A(H5N1), may not be representative of all PMWs in Viet Nam. Collecting further epidemiological information (using personal protective equipment; contact duration, influenza vaccination) may also be useful. The results of our study would be more convincing if control groups (vegetable, fruit and seafood sellers working in the same market with PMWs) were included or a follow-up study was conducted.

Despite these limitations, our study has shown that 6.1% of PMWs in northern Viet Nam are seropositive for influenza A(H5N1). The continued circulation and evolution of influenza A(H5N1) requires comprehensive surveillance of both human and animal sites throughout the country with follow-up studies on PMWs to estimate the risk of avian–human transmission of influenza A(H5N1) viruses in Viet Nam.

Conflicts of interest

None declared.

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Ongoing local transmission of dengue in Japan, August to September 2014

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In late August 2014, three autochthonous dengue cases were reported in Japan. Since then, as of 17 September 2014, a total of 131 autochthonous cases have been confirmed. While cases were reported from throughout Japan, the majority were linked to visiting a large park or its vicinity in Tokyo, and the serotype detected has been serotype 1. We report preliminary findings, along with the public health response activities, of the first documented autochthonous dengue outbreak in Japan in nearly 70 years.

Dengue is an acute, mosquito-borne febrile illness caused by a flavivirus found widely in the Asia-Pacific region, particularly in South-East Asia. While the most competent mosquito species for dengue virus transmission is believed to be *Aedes aegypti*, *Aedes albopictus* is also a competent vector present in much of Japan during the warmer months. Infection with dengue virus may cause fever, headache, muscle pain and/or rash but may also be mild or asymptomatic. While there is no specific treatment, with early and appropriate medical care, the likelihood of infections resulting in severe forms or death is rare.

In Japan, dengue has been a notifiable disease since April 1999. Physicians are required to report demographic, clinical and exposure history information of laboratory-confirmed cases to the local public health centre that are then reported to the Ministry of Health, Labour and Welfare (MHLW) and the National Institute of Infectious Diseases (NIID).[1]

In recent years, approximately 200 imported cases of dengue (those that had onset after returning to Japan following overseas travel) have been notified through national surveillance, the majority from South-East Asia.[2,3] While the last reported local transmission of dengue was during the 1940s,[4] given the growing number of imported cases and the recent emergence of dengue in areas such as the United States of America,[5] France[6] and Portugal,[7] the re-emergence of dengue in Japan had been a concern in recent years. A travel-associated German case was suspected to have been infected with dengue serotype 2 while in Japan in summer 2013,[8] and Japan's MHLW and NIID have since been preparing guidelines to manage response activities in case of local transmission.

On 27 and 28 August 2014, three autochthonous dengue cases, with no overseas travel, were reported from Tokyo and Saitama prefectures. All three reported that they had visited Yoyogi Park, a large (54 hectare) forested park in the centre of metropolitan Tokyo and were bitten by mosquitoes there; illness onset was on 18, 20 and 24 August. MHLW alerted local public health authorities of the autochthonous cases and called for vigilance and timely reporting by clinicians; awareness also increased among the general public via both official MHLW and unofficial media channels.

As of 17 September 2014, a total of 131 autochthonous cases have been reported; including cases that occurred before the initial three cases were detected (**Figure 1**). While cases were reported from 17/47 Japanese prefectures, the majority of cases (112/131 [85%]) were linked to Yoyogi Park or its vicinity. Of the 131 cases, 74 (56%) were male and 57 (44%) female. The median age was 26 years (range 4–77). Among both males and females, a large proportion were adolescents or young adults, with 42 (32%) in their 20s and 28 (21%) aged 10–19 years. Among children and the elderly, the distribution was skewed towards males: 4/5 children under 10 years of age were boys and 10/11 persons aged 60 years and over were men. At the time

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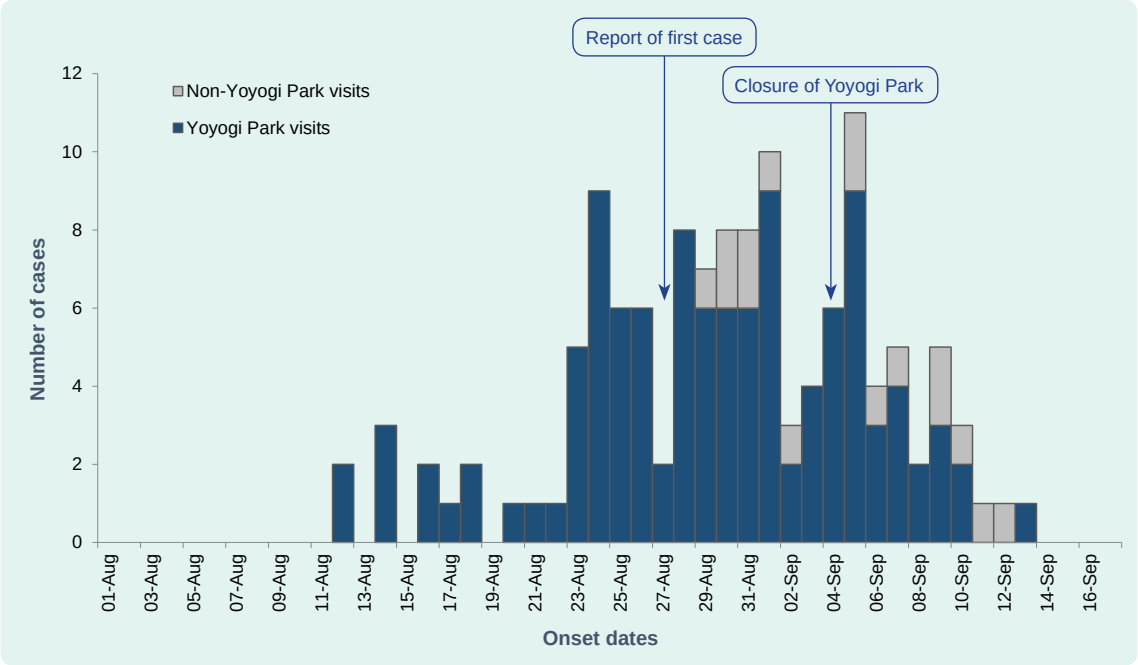
Figure 1. **Reported number of locally acquired laboratory-confirmed dengue cases in Japan by date of onset and suspected location of transmission, 1 August to 17 September 2014 (n = 128*)**



* Yoyogi Park visits, n = 112; non-Yoyogi Parks visits, n = 16.

of reporting, the majority had mild symptoms such as fever, myalgia and arthralgia. No fatalities have been reported. Sequence analysis of viral genomes presented high similarity with dengue serotype 1 viruses circulating in South-eastern Asia.[9]

Dengue virus has also been detected from mosquitoes collected in Yoyogi Park. After enhancing vector surveillance following the reporting of the initial cases, dengue virus was detected from mosquitoes captured on 3 September 2014 from 4/10 collection sites in the park.[10] This finding was a first since the Tokyo Metropolitan Government began testing mosquitoes captured from 16 collection sites in the Tokyo metropolitan area in 2004. Subsequently, dengue virus was also detected from a mosquito captured in Shinjuku Gyoen, a large park 2 km from Yoyogi Park, on 19 September; however, this park was already closed on 7 September.[11]

Local and national public health authorities have been responding to the ongoing domestic transmission. Risk communication messages have been disseminated via the Internet and traditional means (e.g. public notices at the affected sites, newspapers, television), focusing on personal protection (reducing exposed areas of skin when outdoors and applying insect repellent) and on elimination of mosquito breeding sites. MHLW is actively providing such information[12] and coordinating with local government counterparts in timely information collection. Updated guidelines for clinical management of dengue have also been disseminated. NIID, in coordination with partners, is providing technical support regarding virus testing and vector control, conducting risk assessments, epidemiological investigations and enhancing dissemination of information based on surveillance data. In addition, the Tokyo Metropolitan Government enhanced vector surveillance, increasing collection sites in Yoyogi Park,[10] and also temporarily closed the park on 4 September. Vector control by adulticide application was carried out at Yoyogi and several other parks in Tokyo; investigations indicated mosquito populations in most parks were lower after vector control activities (unpublished data).

There are important limitations, and it is unknown to what degree the magnitude of the current outbreak is due to enhanced awareness, surveillance, testing and/or reporting. It is also unknown when dengue emerged locally. While enhanced surveillance activities detected cases that occurred before the initial three cases, all cases detected so far have had onset since mid-August 2014, indicating that virus introduction may have taken place fairly recently. Regardless, the

confirmation of more than 100 domestically acquired dengue cases linked with a single urban park in Japan, within a period of a month, is remarkable. Another important potential limitation is ascertainment bias, such that clinically suspect cases who visited Yoyogi Park may have been more likely to be tested, as the initially reported cases were all linked to the park. Later, cases with no link began to be detected (**Figure 1**), indicating perhaps wider geographic circulation of the virus.

While the situation is still evolving, the detection of dengue virus-positive mosquitoes from Yoyogi Park has major public health, social and economic implications. Already, an Australian traveller who had visited Yoyogi Park has been reportedly confirmed with dengue infection in September 2014, following his return from Japan.[13] The unexpected magnitude and future concerns require ongoing monitoring, risk assessment and epidemiologic and entomologic studies. As *Aedes albopictus* is active only until November in most of Japan, notifications of autochthonous cases should soon start to decline. However, re-entry of the virus in the following year is possible via a dengue virus-infected mosquito(es) or person(s) who are viremic upon entry into the country. As the 2020 Summer Olympics are to be held in Tokyo, there are also significant international public health implications requiring effective surveillance and clear and routine communications of findings to regional and global partners and stakeholders.

Conflicts of interest

None declared.

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Detection of *Campylobacter* in human faecal samples in Fiji

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Introduction: Data on campylobacteriosis in developed countries are well documented; in contrast, few studies on campylobacteriosis have been conducted in developing countries. This study was undertaken to test for *Campylobacter* in human faecal samples sent to the two major pathology laboratories in Fiji.

Methods: A total of 408 diarrhoeal faecal samples were collected from the two major hospital pathology laboratories in Central Fiji (Suva) and Western Fiji (Lautoka) between December 2012 and February 2013 and from June to July 2013. Samples were analysed for the presence of *Campylobacter* using polymerase chain reaction (PCR) based methods.

Results: *Campylobacter* was detected in 241/408 (59.1%) of samples tested using PCR. Samples from children aged less than five accounted for 21.6% of positive cases.

Discussion: *Campylobacter* was detected in 59.1% of diarrhoeal samples collected from the two main laboratories in Fiji. A high proportion of children under five years with *Campylobacter* has been reported in other countries and could be due to parents being more likely to seek medical attention. Further studies are required to confirm the species of *Campylobacter* that are predominantly associated with gastroenteritis in Fiji.

Campylobacter spp. have been recognized as gastrointestinal pathogens in both developed and developing countries and are ubiquitous in food animals such as poultry, cattle, pigs, sheep, ostriches and shellfish and in pets such as cats and dogs.[1] Patients with campylobacteriosis present with symptoms similar to those seen in other enteric infections, and while symptoms are usually self-limiting, in severe cases symptoms may last for 5 to 7 days.[2]

Data on campylobacteriosis not exist in many developing countries due to the lack of surveillance programmes for *Campylobacter* infections. It has been reported that *Campylobacter* infections in developing countries are more frequently reported in children under five years of age and often regarded as a paediatric disease.[3] Based upon current studies, the rates of campylobacteriosis in the general population are an estimated 90 cases per 100 000 population in both developing and developed countries.[3]

Diarrhoeal disease is notifiable in Fiji; approximately 22 753 diarrhoeal cases and 281 cases of diarrhoea with blood were reported by the Ministry of Health in Fiji in 2011.[4] *Salmonella typhi* infections, classified as enteric fever, were detected in 404 of these cases;[4] however, no other pathogen-specific data were reported. This is despite studies showing widespread prevalence of gastrointestinal disease in developing countries.[5] The public health infrastructure is variable in the Pacific region. Many countries are geographically isolated and have limited economic resources;[6] therefore, specific communicable diseases including foodborne disease are neither notifiable nor monitored through laboratory-based surveillance systems.

In Fiji, when stool samples are collected, most pathology laboratories routinely screen for parasites, viruses and bacterial pathogens such as *Salmonella* and *Shigella* but not for *Campylobacter*.

This study was undertaken to test for *Campylobacter* in stool samples sent to the two major pathology laboratories in Fiji using polymerase chain reaction (PCR) methods.

MATERIALS AND METHODS

This study was approved by the Charles Sturt University Human Ethics Committee and the National Research Ethics Committee, Ministry of Health, Fiji.

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Sample collection

A total of 408 human faecal samples (stool samples) were collected from the Central and Western hospitals' pathology laboratories in Fiji from mid-December 2012 to the end of February 2013 and from June to July 2013. The samples were approximately equally divided between the two sites (Central [Suva] n = 208, Western [Lautoka] n = 200). Information on age and gender were extracted manually from the stool register held by the laboratories; due to time and access constraints, it was not possible to obtain this information for all samples.

Samples were collected in sterile containers, placed on ice and transported to the Fiji National University laboratory for PCR analysis. Samples were only made available to this study after completion of routine testing by the microbiology department in both laboratories: four to six hours after the sample was received in the laboratory in Suva and eight to 10 hours in Lautoka.

DNA extraction

Total DNA was extracted from the faecal samples using the Norgen DNA Stool Mini Kit (Norgen Biotek Corporation, Thorold, ON, Canada). An aliquot of the stool sample (200μL) was collected in 2ml Eppendorf tubes and heated at 70 °C for 10 minutes. The DNA was extracted using the manufacturer's protocol. The final elution volume of DNA was 100μl. Extracted DNA samples were stored at –20 °C before PCR analysis.

PCR detection of *Campylobacter*

PCR amplification was performed using previously described primers of a fragment specific for the *16SrRNA* gene, C412F,5′-GGATACACTTTTCGGAGC-3′ and C1288R,5′-CATTGTAGCACGTGTGTC-3′.[7] Each PCR reaction (25μl) contained GoTaq Green Master Mix 2x (Promega, Madison, WI, USA) that was used as the reaction buffer with 400μM dATP, 400μM dGTP, 400μM dCTP, 3mM $MgCl_2$ and 50 μM of each primer. DNA template (1μl) was used and subjected to 35 cycles of amplification in a thermal cycler (Eppendorf) with the following slightly modified conditions: denaturation at 94 °C for 5 minutes, annealing temperature for *16SrRNA,* 58 °C for 30 seconds, extension for 72 °C for 1 minute and further extension at 72 °C for 5 minutes.[7] The PCR amplicons were analysed by 1% agarose gel electrophoresis stained with ethidium bromide and visualized. All PCR assays were set up using DNA from *Campylobacter jejuni* NCTC 11351 as the positive control. No bacterial DNA was added to the negative control. Results were discarded if the negative control was positive. Species level identification was not undertaken on the *Campylobacter*-positive samples.

RESULTS

Of the 408 (Suva, n = 208; Lautoka, n = 200) stool samples analysed by PCR, 241 (59.1%) were positive for *Campylobacter*. A significantly higher proportion of PCR-positive samples were detected from the Central area (Suva, n = 141; 67.8%) than from the Western area (Lautoka, n = 100; 50%; $P < 0.008$).

Valid information was available on patient age for 229 samples and sex for 236 samples (132 males and 104 females). There was no significant difference regarding sex or age group distribution of cases between the Central and Western regions (P = 0.982 and P = 0.357, respectively). Although there were more PCR-confirmed *Campylobacter* infections in males compared to females (55.3% compared with 44.7%), this was not statistically significant (P = 0.125). The highest proportion of positive cases was observed in the 0–4 year age group at 21.6%, followed by a second peak (15%) in the 15–34 years age group (**Table 1**).

DISCUSSION

This study reports the detection of *Campylobacter* in faecal samples collected from patients with diarrhoea in Fiji from mid-December 2012 to February 2013 and in June and July 2013. In the study population, *Campylobacter* was detected in the majority (59%) of clinical samples. This finding is similar to a study undertaken in United States military personnel training in Thailand.[8]

The age distribution of PCR-confirmed cases of campylobacteriosis in this study was similar to the findings of other studies in developing countries, with a high proportion of cases in children aged less than five years.[5] One such example is a study carried out in Malawi where 21% of children under five years of

Table 1. ***Campylobacter*-positive specimens by age group and gender (n = 199)***

Age group (years)	Number tested	Total		Male		Female	
		Number positive	% positive	Number positive	% positive	Number positive	% positive
0–4	45	43	21.6	30	27.3	13	14.6
5–14	19	16	8.0	8	7.3	8	9.0
15–24	33	29	14.6	13	11.8	16	18.0
25–34	36	31	15.6	17	15.5	14	15.7
35–44	29	21	10.6	15	13.6	6	6.7
45–54	27	22	11.1	8	7.3	14	15.7
55–64	24	22	11.1	14	12.7	8	9.0
65–74	14	14	7.0	5	4.5	9	10.1
75 and above	2	1	0.5	0	0.0	1	1.1
Total		**199**	**100.0**	**110**	**55.3**	**89**	**44.7**

* Age and gender data were not available for 42 positive samples

age with diarrhoea had *Campylobacter* infections, and 14% of *Campylobacter* infections were detected in asymptomatic children.[9] Children may be more likely to be taken to hospitals by their parents when exhibiting symptoms of gastrointestinal disease, and consequently, faecal samples from these patients are frequently submitted to pathology laboratories for testing.

The sources of *Campylobacter* spp. infections in Fiji are not known. Poor food handling techniques and hygiene have been reported as the cause of diarrhoeal disease, and epidemiological studies also report cases of campylobacteriosis occurring through the consumption of raw milk[9] and undercooked or contaminated poultry meat.[10] All of these are possible risk factors in Fiji.

The diagnosis of *Campylobacter* infection is typically performed using culture techniques and in some cases PCR.[10] An important advantage of using PCR is that it can detect dead *Campylobacter* spp. in clinical samples, which may be important when poorly stored samples are analysed. Currently, in Fiji, pathology laboratories do not routinely test diarrhoeal samples for the presence of *Campylobacter*. To the best of the authors' knowledge, this is the first study to investigate the prevalence of *Campylobacter* spp. in diarrhoeal samples in Fiji.

The high rate of *Campylobacter* detected in this study requires further investigation; future studies should include healthy participants to determine the rate of *Campylobacter* carriage in the Fiji population. It would also be useful to determine the species of *Campylobacter* isolated in this population. Additionally, it would be valuable to determine if pathogens other than *Campylobacter* were present in the *Campylobacter*-positive samples from individuals with diarrhoea. Coinfection with *Salmonella typhi* or other pathogens were not investigated in this study.

In conclusion, the results of this study demonstrate that *Campylobacter* infections are common in Fiji, similar to many other developing and developed countries.

Conflicts of interest

None declared.

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Drug-resistant tuberculosis in the WHO Western Pacific Region

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Objective: To review the latest information about tuberculosis (TB) drug resistance and programmatic management of drug-resistant TB in the Western Pacific Region of the World Health Organization (WHO).

Methods: We analysed routine data reported by countries to WHO from 2007 to 2013, focusing on data from the following: surveillance and surveys of drug resistance, management of drug-resistant TB and financing related to multidrug-resistant TB (MDR-TB) management.

Results: In the Western Pacific Region, 4% (95% confidence interval [CI]: 3–6) of new and 22% (95% CI: 18–26) of previously treated TB cases were estimated to have MDR-TB; this means that in 2013, there were an estimated 71 000 (95% CI: 47 000–94 000) MDR-TB cases among notified pulmonary TB cases in this Region. The coverage of drug susceptibility testing (DST) among new and previously treated TB cases was 3% and 20%, respectively. In 2013, 11 153 cases were notified—16% of the estimated MDR-TB cases. Among the notified cases, 6926 or 62% were enrolled in treatment. Among all enrolled MDR-TB cases, 34% had second-line DST and of these, 13% were resistant to fluoroquinolones (FQ) and/or second-line injectable agents. The 2011 cohort of MDR-TB showed a 52% treatment success. Over the last five years, case notification and enrolment have increased more than five times, but the gap between notification and enrolment widened.

Discussion: The increasing trend in detection and enrolment of MDR-TB cases demonstrates readiness to scale up programmatic management of drug-resistant TB at the country level. However, considerable challenges remain.

Globally an estimated 1.6 million people develop tuberculosis (TB), and 110 000 die from this curable illness annually.[1] TB concentrates in vulnerable populations such as migrants, children, the elderly and the poor. The Western Pacific Region of the World Health Organization (WHO) has made substantial progress and reached the TB-related Millennium Development Goals and associated international targets in advance of the 2015 goal year: TB prevalence and mortality are below half of 1990 levels, and case detection and treatment success remain high.[1] Despite these gains, multidrug-resistant tuberculosis (MDR-TB) caused by *Mycobacterium tuberculosis* that is resistant to isoniazid and rifampicin (two of the first-line drugs used for treatment) poses a formidable challenge to controlling TB. MDR-TB cases can either be previously treated TB cases that develop resistance due to inadequate, incomplete or poor treatment quality (secondary drug resistance) or newly diagnosed TB cases infected with a drug-resistant TB strain (primary drug resistance). In 2013, among patients notified with pulmonary TB, a total of 300 000 (range: 230 000–380 000) people globally were estimated to have developed MDR-TB.[1] Among them, approximately one quarter occurred in countries of the WHO Western Pacific Region. In fact, every year, an estimated 71 000 MDR-TB cases are added to the Region's MDR-TB burden.[1] Most (more than 94%) of the MDR-TB cases in the Region live in three countries: China, the Philippines and Viet Nam.

MDR-TB is a public health challenge for several reasons: treatment duration is very long, up to two years, and complex due to severe side-effects of second-line drugs;[2,3] high management costs may contribute to the increased economic burden and catastrophic patient expenditures;[4] treatment outcome is poor;[1] and MDR-TB poses a huge burden on health systems in low- and middle-income countries where human resources are scarce and technical capacity is lacking to cope with the challenge.[5]

To address the challenge of drug resistance in the WHO Western Pacific Region, the *Regional strategy to stop tuberculosis in the Western Pacific 2011–2015*[6] declared scaling up the programmatic management of drug-resistant TB (PMDT) as one of its five objectives.

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This requires improvements in the following critical steps of the cascade of services: (1) step-wise increase of the proportion of TB cases who receive drug susceptibility testing (DST), (2) all diagnosed patients are promptly notified and enrolled in treatment, and (3) all enrolled patients complete their treatment with effective patient-centred support. In 2011, the Western Pacific regional Green Light Committee was established to assist Member States with a rational PMDT scale-up.

This article is the second in a series of regional TB reports after 'Epidemiology and control of tuberculosis in the Western Pacific Region: analysis of 2012 case notification data'.[7] In this current article, we reviewed the latest available data on TB drug resistance in the WHO Western Pacific Region and the status of its programmatic implementation. This analysis of MDR-TB case detection, notification, enrolment into treatment and outcome of treatment data will provide valuable information on programmatic progress and future direction.

METHODS

In this report, the data reported to WHO by countries and areas from 2007 to 2013 were used. In 2013, 32 of the 37 countries and areas of the WHO Western Pacific Region reported data to WHO, representing more than 99.9% of the total population. Data collection covers the following areas: TB case notification and treatment outcomes, diagnostic and treatment services, drug management, surveillance and surveys of drug-resistance, information on TB/HIV coinfection, infection control, engagement of all care providers and budgets and expenditures for TB control.

The estimated burden of drug-resistant TB by country was sourced from the global TB database,where the proportion of new and previously treated TB cases with MDR-TB from the latest available information from routine surveillance or a survey of drug resistance were used to estimate the total caseload of drug-resistant TB. The full description of these methods is available in the Global Tuberculosis Report 2014,[1] and the data sets can be downloaded from the WHO global TB database (www.who.int/tb/data).

This report describes the available data on TB drug resistance and progress in programmatic response for those countries and areas of the Region where at least one MDR-TB case was notified in 2013. Trends of MDR-TB rates over time were assessed for countries and areas with more than one survey or surveillance data points since 1996. A special focus was maintained on seven countries considered priority countries due to their high burden of TB: Cambodia, China, the Lao People's Democratic Republic, Mongolia, Papua New Guinea, the Philippines and Viet Nam.

In 2013, WHO published revised TB case definitions and introduced a case definition for TB cases resistant to rifampicin only (RR-TB).[8] A new rapid diagnostic technology, Xpert MTB/RIF assay (Cepheid, Sunnyvale, CA, USA), endorsed by WHO in 2010,[9] tests for RR-TB. RR-TB cases identified by the Xpert MTB/RIF test are recommended to start second-line TB treatment[10] and require similar management to MDR-TB cases. In this report, we included RR-TB cases with the confirmed MDR-TB cases.

Treatment outcomes are reported for the 2011 cohort, the most recent year for which there are data.

Analysis was conducted using the statistical package R (R Core Team, 2013, Vienna, Austria, www.R-project.org). For transparent and reproducible research,[11,12] we have published the programme code for generating the entire contents of this article using the R knitr package.

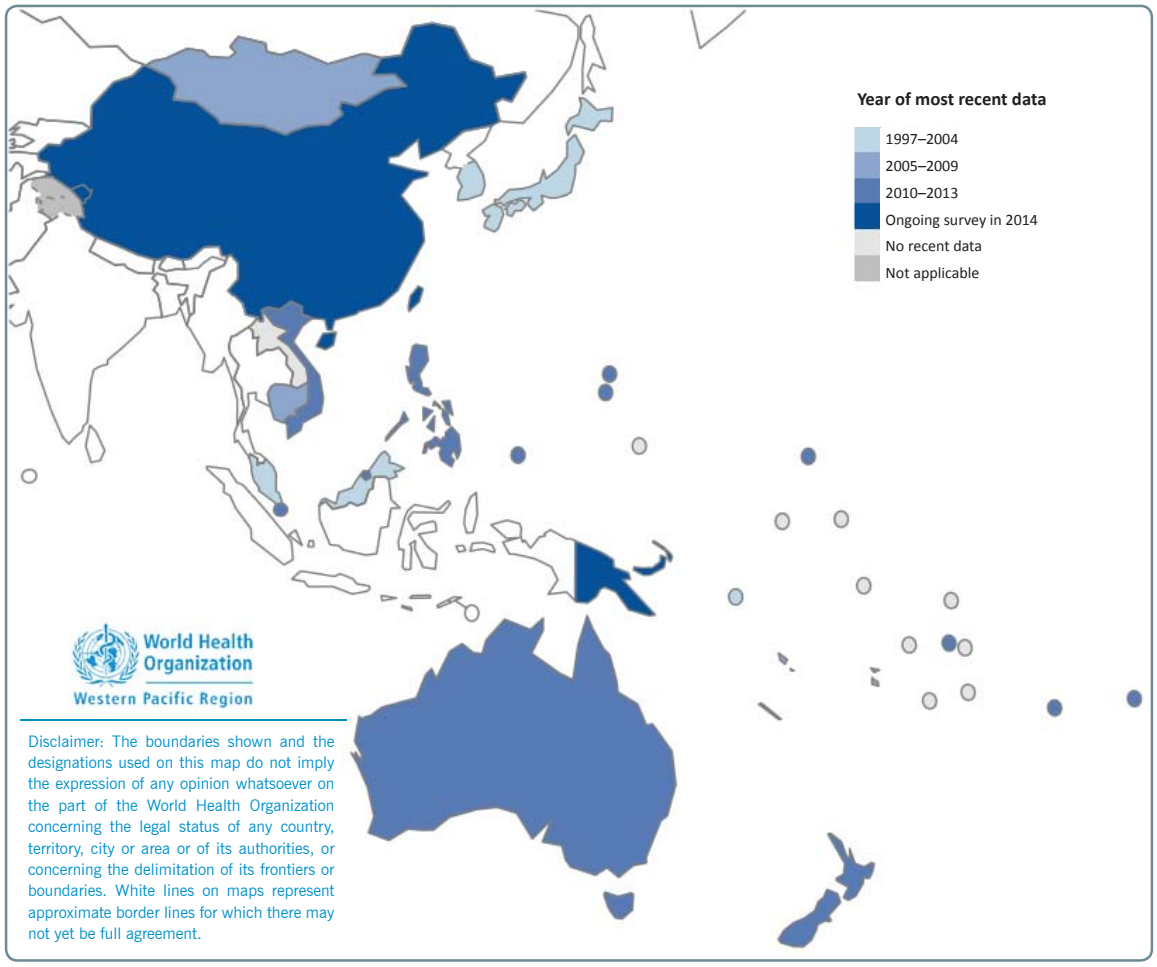
RESULTS

Coverage of drug resistance surveillance

Estimates of the MDR-TB burden depend on the availability of drug resistance data from either continuous surveillance or surveys. Drug resistance data were available from 25 of the 36 countries and areas of the WHO Western Pacific Region (**Figure 1**); 19 rely on routine surveillance and six rely on periodic surveys of representative samples of patients (Cambodia, China, Mongolia, Malaysia, the Philippines and Viet Nam).

National representative drug resistance survey (DRS) data are available from five out of seven priority countries of the Region (Cambodia, China, Mongolia, the Philippines and Viet Nam). Among them, Cambodia, Mongolia, the Philippines and Viet Nam have at least

Figure 1. **Year of most recent data on TB drug resistance by country, WHO Western Pacific Region, 1997–2014**



two national representative survey data points, and China is currently conducting its second national DRS. Malaysia's 1997 data originated from geographical areas that were not considered representative of the country as a whole. Two countries with outstanding DRS data are Papua New Guinea and the Lao People's Democratic Republic. Subnational DRS in Papua New Guinea is ongoing and results are expected to be available in 2015.

Estimated MDR-TB burden: cases and rates

Overall in the Region in 2013, 4% (95% confidenc interval [CI]: 3–6) of new and 22% (95% CI: 18–26) of previously treated TB cases were estimated to have MDR-TB: 71 000 in total, 53 000 (75%) among new cases and 18 000 (25%) among previously treated TB cases (**Table 1**). The proportion of new TB cases with MDR-TB ranged from 0% to 6%. China had the highest proportion at 6% (95% CI: 5–7) of MDR-TB among new cases. The proportion of previously treated TB cases with MDR-TB ranged from 0% to 34%. Mongolia had the highest estimated proportion at 34% (95% CI: 29–38) of MDR-TB among previously treated TB cases. Countries with more than 20% MDR-TB among previously treated TB cases were China (26%), Mongolia (34%), the Philippines (21%) and Viet Nam (23%). Although the highest proportion of MDR-TB was observed among previously treated TB cases, the absolute number of estimated MDR-TB cases is higher among new cases.

There was a positive correlation between the proportion of MDR-TB among new and previously treated TB cases (intercept 0.08%, coefficient 3.1%, F-statistics 3.84, $P = 0.123$; **Figure 2**), although this was not statistically significant. Most countries and areas fell within 95% confidence limits of the linear regression line except for Mongolia and the Philippines. Both had a higher proportion of MDR-TB among previously

Table 1. **Estimated number and proportion of MDR-TB cases among new and previously treated TB cases by selected country,* WHO Western Pacific Region**

Countries and areas	Data type	Year	MDR-TB among new cases		MDR-TB among previously treated TB cases		Total MDR-TB cases
			N (95% CI)	%	N (95% CI)	%	N (95% CI)
Australia	Surveillance	2013	16 (9–27)	2 (1–4)	1 (0–7)	4 (<1–21)	17 (8–26)
Brunei Darussalam	Surveillance	2013	1 (0–6)	<1 (<1–4)	0 (0–3)	0 (0–46)	1 (0–3)
Cambodia	Survey	2007	320 (160–580)	1 (<1–3)	180 (68–370)	11 (4–22)	510 (270–740)
China	Survey	2007	45 000 (35 000–55 000)	6 (5–7)	9200 (7800–11 000)	26 (22–30)	54 000 (48 000–61 000)
Hong Kong Special Administrative Region	Surveillance	2012	34 (21–52)	<1 (<1–1)	9 (3–20)	3 (<1–6)	43 (26–59)
Macao Special Administrative Region	Surveillance	2013	7 (2–16)	2 (<1–5)	4 (1–10)	12 (2–30)	11 (4–18)
Cook Islands	Surveillance	2013	0 (0–1)	0 (0–98)	0 (0–0)	0 (0–0)	0 (0–1)
Japan	Surveillance	2002	110 (63–160)	<1 (<1–1)	100 (72–130)	10 (7–13)	200 (150–260)
Lao People's Democratic Republic	Model†		160 (96–230)	5 (3–6)	65 (56–75)	24 (20–27)	220 (160–290)
Malaysia	Survey	1997	19 (0–120)	<1 (0–<1)	0 (0–340)	0 (0–17)	19 (0–57)
Marshall Islands	Surveillance	2013	2 (0–9)	1 (<1–7)	0 (0–5)	0 (0–71)	2 (0–5)
	New: Survey	2007					
Mongolia	Previously treated TB: Surveillance	2013	33 (16–59)	1 (<1–3)	210 (180–240)	34 (29–38)	240 (210–280)
New Zealand	Surveillance	2012	1 (0–5)	<1 (<1–3)	2 (0–5)	17 (2–48)	3 (0–6)
Palau	Surveillance	2013	0 (0–3)	0 (0–41)	0 (0–0)	0 (0–0)	0 (0–3)
Papua New Guinea	Model†		560 (340–800)	5 (3–6)	570 (480–650)	24 (20–27)	1100 (890–1400)
Philippines	Survey	2013	4400 (3100–6000)	2 (1–3)	4100 (3000–5500)	21 (16–29)	8500 (6900–10 000)
Republic of Korea	Surveillance	2004	780 (600–980)	3 (2–3)	1200 (850–1600)	14 (10–19)	1900 (1600–2300)
Singapore	Surveillance	2013	17 (8–30)	<1 (<1–2)	3 (0–12)	3 (<1–9)	20 (9–31)
Viet Nam	Survey	2012	3000 (1900–4100)	4 (3–5)	2100 (1500–2600)	23 (17–30)	5100 (4100–6100)
Western Pacific Region			**53 000 (31 000–75 000)**	**4 (3–6)**	**18 000 (15 000–21 000)**	**22 (18–26)**	**71 000 (47 000–94 000)**

Source: Global TB database.

CI, confidence interval; MDR-TB, multidrug-resistant tuberculosis.

* Countries that reported at least one MDR-TB case in 2013.

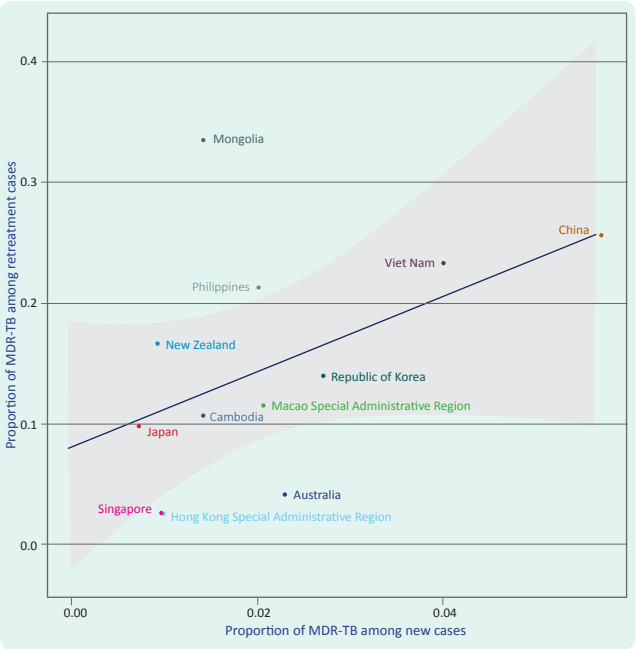
† Estimates of the proportion of new and previously treated TB cases that have MDR-TB were produced using modelling (including multiple imputation) that was based on data from countries that were considered to be similar in terms of TB epidemiology for which data do exist.

treated TB cases in relation to new cases. Australia, Hong Kong Special Administrative Region and Singapore, had a lower proportion of MDR-TB among previously treated TB cases relative to new cases.

Estimated MDR-TB burden over time

Among 25 countries with drug resistance data, 22 countries have more than one data point (direct measurement) either from continuous surveillance or from repeat surveys. Four scenarios are observed when comparing rates per 100 000 population of new TB cases with the estimated rate of MDR-TB among new TB cases from six countries in the Western Pacific Region (**Figure 3**). For Mongolia and Viet Nam, both the reported notification rate of new TB cases and estimated rate of MDR-TB increased over time (7% and 4% per year from 1999 to 2007 and 1997 to 2012, respectively). In Australia and the Philippines, TB notification rates increased while the rate of MDR-TB decreased (–4% and –10% per year from 2004 to 2012 and 2010 to 2012, respectively). In Hong Kong Special Administrative Region, both the TB notification rate and estimated rate of MDR-TB decreased (–8% for both from 1996 to 2012). In the Republic of Korea, TB case notification decreased, but the estimated rate of MDR-TB increased by 2% from 1996 to 2005. These findings need to be

Figure 2. **Correlation* between the estimated proportion of MDR-TB cases among new and previously treated TB cases respectively by selected country,† WHO Western Pacific Region, 2013**



MDR-TB, multidrug-resistant tuberculosis.

* Line represents the linear regression and the grey band represents 95% CI.

† Countries that reported their own MDR-TB estimates for new and previously treated cases in 2013.

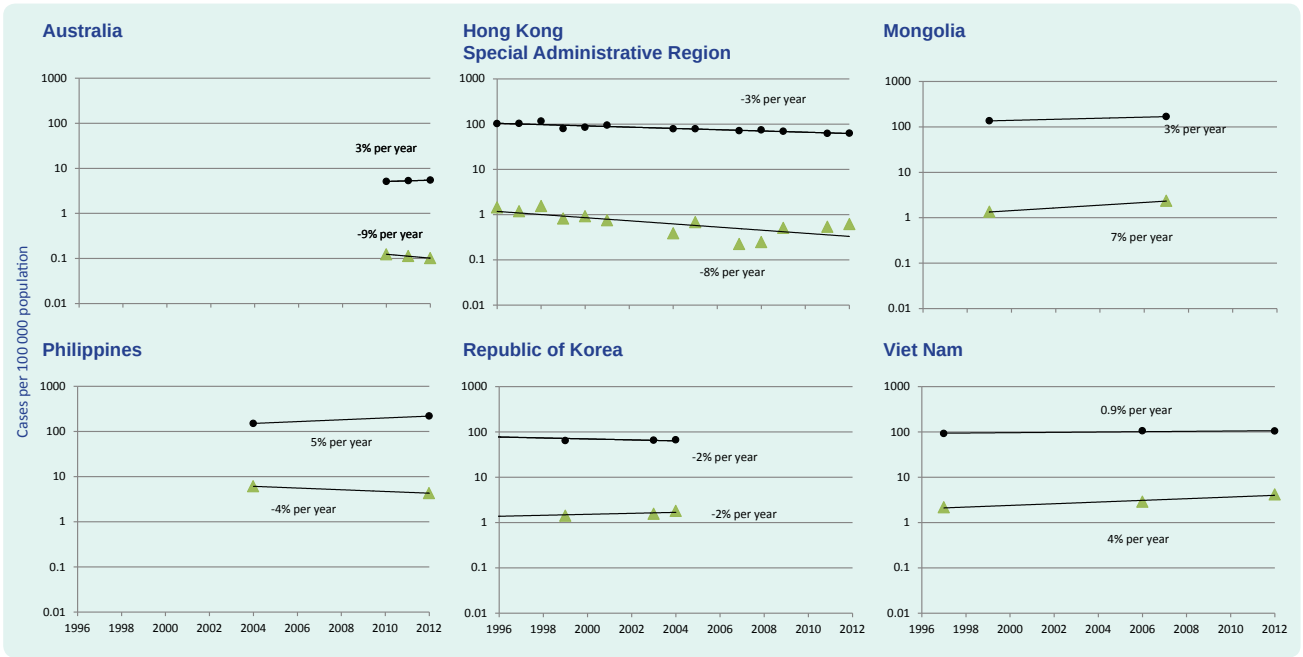
interpreted with caution as sufficient data points are not available for all of the countries to identify trends with confidence.

Drug susceptibility testing (DST)

DST data were reported from 18 countries in 2013. Only 3% of new bacteriologically confirmed TB cases and 20% of previously treated TB cases were tested for MDR-TB or RR-TB (**Table 2**). DST coverage for MDR-TB among new cases was less than 10% in China (3%), the Lao People's Democratic Republic (<1%), the Philippines (<1%), the Republic of Korea (4%) and Viet Nam (3%) and among previously treated TB cases. DST coverage remained below 50% in China (20%), Japan (43%), the Lao People's Democratic Republic (26%), Malaysia (9%), the Marshall Islands (43%), the Philippines (14%), the Republic of Korea (9%) and Viet Nam (45%).

Among cases tested in 2013, 8% were resistant to isoniazid only, 2% were resistant to rifampicin only and 11% were resistant to both (data not shown). The Xpert MTB/RIF test identified 17% of MDR-TB cases overall and was highest for Viet Nam at 81% and Papua New Guinea at 72% (**Table 2**).

Figure 3. **Rates per 100 000 population of new TB cases (black) and estimated MDR-TB cases among new TB cases (green) by selected countries, WHO Western Pacific Region, 1996–2012**



MDR-TB, multidrug-resistant tuberculosis.

Table 2. **Number and proportion of notified TB cases with DST results, confirmed MDR-TB and RR-TB cases and cases confirmed by Xpert, by new and previously treated TB cases and selected country,* WHO Western Pacific Region, 2013**

Countries and areas	Notified cases with DST results						MDR-TB and RR-TB* among cases with DST results						Cases confirmed by Xpert among MDR-TB and RR-TB†					
	New		Prev		Total		New		Prev		Total		New		Prev		Total	
	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%
Australia	570	82	25	76	604	83	13	2	3	12	17	3	0	–	0	–	0	–
Brunei Darussalam	146	92	6	100	152	93	1	<1	0	–	1	<1	0	–	0	–	0	–
Cambodia	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
China	20 080	3	7153	20	27 233	3	1612	8	2571	36	4183	15	244	15	246	10	490	12
Hong Kong Special Administrative Region	1919	55	198	56	2117	55	28	1	7	4	35	2	0	–	0	–	0	–
Macao Special Administrative Region	244	70	26	81	305	80	6	2	3	12	9	3	0	–	0	–	0	–
Cook Islands	1	100	0	–	1	100	0	0	0	–	0	–	0	–	0	–	0	–
Japan	7266	49	435	43	7		42	<1	22	5	64	<1	–	–	–	–	–	–
Lao People's Democratic Republic	11						0	0	7	10	7	9	0	–	0	–	0	–
Malaysia	2702	14	181	9	11 110	52	80	3	5	3	185	2	0	–	0	–	0	–
Marshall Islands	72	61	3	43	75	60	1	1	0	0	1	1	0	–	0	–	0	–
Mongolia	289	12	531	85	838	28	24	22	177	33	242	29	15	23	38	21	53	22
New Zealand	–	–	–	–	–	–	1	–	1	–	2	–	–	–	–	–	–	–
Palau	7						0	–	0	–	0	–	0	–	0	–	0	–
Papua New Guinea	–	–	–	–	93	<1	–	–	–	–	46	49	–	–	–	–	33	72
Philippines	25	<1	2631	14	2656	1	10	40	1349	51	1359	51	–	–	–	–	–	–
Republic of Korea	1249	4	726	9	1975	5	466	37	518	71	984	50	–	–	–	–	–	–
Singapore	1070	61	85	66	1155	62	14	1	4	5	18	2	0	–	0	–	0	–
Viet Nam	353	<1	3955	45	4531	5	40	11	997	25	1041	23	37	92	801	80	841	81
Western Pacific Region	**36 103**	**3**	**16 057**	**20**	**60 765**	**5**	**2379**	**7**	**5664**	**35**	**8195**	**13**	**296**	**12**	**1085**	**19**	**1417**	**17**

DST, drug susceptibility testing; MDR-TB, multidrug-resistant tuberculosis; Prev, previously treated tuberculosis; RR-TB, rifampicin-resistant tuberculosis.

* Countries that reported at least one MDR-TB case in 2013.

† Rifampicin-resistant only cases are included whether confirmed by DST or Xpert.

In countries with a high burden of MDR-TB, the DST coverage among previously treated TB cases increased steadily while the MDR-TB positivity rate remained high (**Figure 4**). In fact, there is a general paradoxical tendency for countries with a relatively higher DST coverage to detect a lower proportion of MDR-TB.

MDR-TB notifications and enrolment in treatment

There were 11 153 reported cases of MDR-TB and RR-TB in the Region in 2013, representing 16% of the 71 000 estimated MDR-TB cases among pulmonary TB patients. Of these, 2379 were reported among new TB cases, representing 4% of the estimated 53 000 MDR-TB cases among TB new cases; 5664 were reported among previously treated TB cases, representing 31% of the estimated 18 000 MDR-TB among previously treated TB cases. Mongolia, the Philippines and Viet Nam reported 84%, 33% and 47%, respectively, of their estimated MDR-TB among previously treated TB cases (**Table 3**).

Of those notified MDR-TB cases in 2013, only 62% (6926 of 11 153) were enrolled in treatment with second-line anti-TB drugs. For most countries, there has been a steady increase in enrolment in treatment over the years, especially since 2011; however, the gap between notified cases and enrolment in treatment for MDR-TB is widening in China, the Philippines and Viet Nam (**Table 4, Figure 5**).

Figure 4. **DST coverage among previously treated TB cases and MDR-TB and RR-TB positivity rate by year and selected countries, WHO Western Pacific Region, 2007–2013**

DST, drug susceptibility testing; MDR-TB, multidrug-resistant tuberculosis; RR-TB, rifampicin-resistant tuberculosis.

MDR-TB treatment outcomes

Treatment for the 2011 cohort were reported from 15 countries. Overall the proportion of MDR-TB cases who successfully completed treatment was 52%, with 21% lost to follow-up and 10% having died (**Figure 6**). Treatment success was 100% in Macao Special Administrative Region yet only 14% in Papua New Guinea.

In China, the treatment success rate was less than 50% between 2007 and 2011 with death and failure rates remaining high. Malaysia, and the Philippines showed a continuous decline of treatment success over the same time period; non-evaluation of cases is the main reason for Malaysia while loss to follow-up plays a major role in the decreasing success rate in the Philippines. Among the priority countries, Cambodia (86%) and Viet Nam (72%) had high treatment success for the 2011 cohort.

Second-line anti-TB DST and XDR-TB

Twelve countries reported testing data for the second-line anti-TB drugs in 2013, with 34% of enrolled MDR-TB cases having results for DST for the second-line anti-TB drugs. Combining data from all 12 countries, 12% of tested cases had resistance to fluoroquinolones (FQ), 8% to a second-line injectable and 13% to either a FQ or a second-line injectable agent or both. Extensively drug-resistant TB (XDR-TB) is a subset of MDR-TB that acquired additional resistance to FQ and one or more of the second-line injectable agents. In 2013, the total number of reported XDR-TB cases was

Table 3. **Number of estimated and notified MDR-TB and RR-TB cases, proportion notified among estimated MDR-TB and RR-TB cases and number and proportion of MDR-TB and RR-TB cases enrolled in treatment by selected country,* WHO Western Pacific Region, 2013†**

Countries and areas	Estimated			Notified			Percentage notified among estimated			Enrolled in treatment	
	New	Prev	Total	New	Prev	Total‡	New	Prev	Total		
	N (95% CI)	N (95% CI)	N (95% CI)				N (95% CI)	N (95% CI)	N (95% CI)	n	% among detected
Australia	16 (8–27)	1 (0–7)	17 (8–26)	13	3	24	81 (48–144)	300 (43–NA)	141 (92–300)	22	92
Brunei Darussalam	1 (0–6)	0 (0–3)	1 (0–3)	1	0	1	100 (17–NA)	– (0–NA)	100 (33–NA)	0	0
Cambodia	320 (270–580)	180 (68–370)	510 (270–740)	–	–	121	– (NA–NA)	– (NA–NA)	24 (16–45)	121	100
China	45 000 (48 000–55 000)	9200 (7800–11 000)	54 000 (48 000–61 000)	1612	2571	4183	4 (3–5)	28 (23–33)	8 (7–9)	2184	52
Hong Kong Special Administrative Region	34 (26–52)	9 (3–20)	43 (26–59)	28	7	35	82 (54–133)	78 (35–233)	81 (59–135)	22	63
Macao Special Administrative Region	7 (4–16)	4 (1–10)	11 (4–18)	6	3	10	86 (38–300)	75 (30–300)	91 (56–250)	8	80
Cook Islands	0 (0–1)	0 (0–0)	0 (0–1)	0	0	2	– (0–NA)	– (NA–NA)	NA (200–NA)	0	0
Japan	110 (150–160)	100 (72–130)	200 (150–560)	42	22	64	38 (26–67)	22 (17–31)	32 (25–43)	–	–
Lao People's Democratic Republic	160 (160–230)	65 (56–75)	220 (160–290)	0	7	7	0 (0–0)	11 (9–12)	3 (2–4)	4	57
Malaysia	19 (0–120)	0 (0–340)	19 (0–57)	80	5	277	421 (67–NA)	NA (1–NA)	1458 (486–NA)	49	18
Marshall Islands	2 (0–9)	0 (0–5)	2 (0–5)	1	0	1	50 (11–NA)	– (0–NA)	50 (20–NA)	1	100
Mongolia	33 (210–59)	210 (180–240)	240 (210–280)	64	177	257	194 (108–400)	84 (74–98)	107 (92–122)	192	75
New Zealand	1 (0–5)	2 (0–5)	3 (0–6)	1	1	3	100 (20–NA)	50 (20–NA)	100 (50–NA)	2	67
Palau	0 (0–3)	0 (0–0)	0 (0–3)	0	0	1	– (0–NA)	– (NA–NA)	NA (33–NA)	0	0
Papua New Guinea	560 (890–800)	570 (480–650)	1100 (890–1400)	–	–	119	– (NA–NA)	– (NA–NA)	11 (8–13)	145	122
Philippines	4400 (6900–6000)	4100 (3000–5500)	8500 (6900–10 000)	10	1349	3962	<1 (<1–<1)	33 (25–45)	47 (40–57)	2262	57
Republic of Korea	780 (1600–980)	1200 (850–1600)	1900 (1600–2300)	466	518	984	60 (48–78)	43 (32–61)	52 (43–62)	951	97
Singapore	17 (9–30)	3 (0–12)	20 (9–31)	14	4	18	82 (47–175)	133 (33–NA)	90 (58–200)	15	83
Viet Nam	3000 (4100–4100)	2100 (1500–2600)	5100 (4100–6100)	40	997	1204	1 (<1–2)	47 (38–66)	24 (20–29)	948	79
Western Pacific Region	**53 000 (47 000–75 000)**	**18 000 (15 000–21 000)**	**71 000 (47 000–94 000)**	**2379**	**5664**	**11 153**	**4 (3–8)**	**31 (27–38)**	**16 (12–24)**	**6926**	**62**

CI, confidence interval; MDR-TB, multidrug-resistant tuberculosis; NA, not applicable; Prev, previously treated tuberculosis; RR-TB, rifampicin-resistant tuberculosis.

* Countries that reported at least one MDR-TB case in 2013.

† All columns except estimates include RR-TB cases confirmed by Xpert only. Total MDR-TB cases detected included cases among extrapulmonary and from samples taken more than 2 weeks after start of treatment.

‡ Total column includes cases with treatment history unavailable.

107 (5% of tested MDR-TB cases) from six countries in the Region (17% of all countries and areas). The highest rate was reported from the Republic of Korea where 10% of all MDR-TB cases were XDR-TB.

Expenditure for MDR-TB management

Expenditure for MDR-TB management has significantly increased over the years (**Figure 7**). In 2013, a total of US $57.3 million was spent for MDR-TB management in the Region; this was 10.9% of the total national TB programme expenditure. Of the total funds reported, 18.4% was from domestic sources (government allocation) with the remaining 81.6% from external grants. Second-line drugs comprised 27.8% of the total cost for MDR-TB management.

DISCUSSION

The Western Pacific Region comprises countries with high MDR-TB caseloads, such as China, the Philippines, Viet Nam, and several Pacific island nations with very small, irregular caseloads. The Member States are also at different stages of PMDT implementation.

The first step for PMDT scale-up is to establish diagnostic capacity and increase the proportion of TB cases who receive DST. The Global Plan to Stop TB[13] set a target for DST at 100% of previously treated TB cases and 20% of new cases by 2015. Overall DST coverage remained low with the current coverage for new cases at 3% and previously treated TB cases at 20%, both far below the Global Plan to Stop TB target. The introduction

Table 4. **Number and proportion of MDR-TB and RR-TB cases resistant to second-line anti-TB drugs and XDR-TB, by selected country,* WHO Western Pacific Region, 2013**

Countries and areas	Second-line DST		Resistance to FQ		Resistance to second-line injectible		XDR-TB	
	N	% of enrolled	N	% of tested	N	% of tested	N	% of tested
Australia	12	55	2	17	1	8	0	–
Brunei Darussalam	–	–	–	–	–	–	–	–
Cambodia	–	–	–	–	–	–	–	–
China	–	–	–	–	–	–	–	–
Hong Kong Special Administrative Region	26	118	2	8	3	12	1	4
Macao Special Administrative Region	7	88	0	–	0	–	0	–
Cook Islands	0	–	0	–	0	–	0	–
Japan	–	–	–	–	–	–	–	–
Lao People's Democratic Republic	4	100	1	25	0	_	0	–
Malaysia	113	231	12	11	2	2	1	<1
Marshall Islands	–	–	–	–	–	–	–	–
Mongolia	113	59	6	5	15	13	5	4
New Zealand	2	100	0	–	0	–	0	–
Palau	0	–	0	–	0	–	0	–
Papua New Guinea	73	50	1	1	2	3	–	–
Philippines	927	41	59	6	38	4	5	<1
Republic of Korea	838	88	158	19	112	13	85	10
Singapore	12	80	0	–	0	–	0	–
Viet Nam	199	21	29	15	23	12	10	5
Western Pacific Region	**2326**	**34**	**270**	**12**	**156**	**8**	**107**	**5**

DST, drug susceptibility testing; FQ, fluoroquinolones; MDR-TR, multidrug-resistant tuberculosis; RR-TB, rifampicin-resistant tuberculosis; XDR-TB, extensively drug-resistant tuberculosis.

* Countries that reported at least one MDR-TB case in 2013.

of a rapid diagnostic tool has enabled a drastic increase in diagnostic capacity, as evidenced in Viet Nam, where 81% of notified MDR-TB cases were diagnosed by Xpert MTB/RIF in 2013. This analysis indicates that the expansion of DST coverage is still ongoing to cover the highest risk groups. Most countries starting PMDT focus on previously treated TB cases first, as diagnosing MDR-TB cases among new TB cases is a huge challenge in this Region. Although the proportion of MDR-TB among new TB cases is low, the absolute numbers are very high. It is a challenge to identify effective and efficient strategies to identify MDR-TB cases among new TB cases considering current financial and human resource capacity. To identify potential high-risk groups that can be targeted for selective DST, surveillance systems need to be expanded and strengthened.

XDR-TB is also a growing concern. Currently, second-line DST coverage is low at 34% as reported from 12 countries, and the true magnitude of XDR-TB burden is unknown. As PMDT is expanded with increased second-line DST capacity, more XDR-TB patients will likely be diagnosed. Programmes need to get ready to address XDR-TB with guidelines for the management of XDR-TB patients including policies for appropriate palliative care and infection control.

It is also of paramount importance that all TB cases diagnosed with MDR-TB are quickly put on treatment. Detection of MDR-TB cases is challenging and requires a lot of effort; if notified cases are not put on treatment, all these efforts will be in vain. It is alarming that overall in the Region in 2013, 38% of the notified cases were not put on treatment (48% in China, 43% in the Philippines and 21% in Viet Nam). As reported to the WHO Regional Office for the Western Pacific, reasons for this may be many-fold: some patients may be on the waiting list because of unavailability of treatment capacity including drugs, hospitalization and treatment support. Rapid scale-up of diagnostic capacity

Figure 5. **Number of MDR-TB and RR-TB cases notified and enrolled in treatment by year and selected country,* WHO Western Pacific Region, 2007–2013**

MDR-TB, multidrug-resistant tuberculosis; RR-TB, rifampicin-resistant tuberculosis.

* Countries that reported at least 10 MDR-TB cases in 2013.

especially with Xpert MTB/RIF may not be accompanied by an increased treatment capacity. Another reason may be loss to follow-up of confirmed cases that can occur due to non-accessibility of PMDT centres; non-coordination between diagnostic and treatment sites; and economic burden to patients, including direct and indirect cost. Instead, these patients return to their community and continue to spread their MDR-TB strain. Increasing diagnostic capacity must be aligned with drug and treatment provision, and political commitment to coordinated treatment capacity to match diagnostic capacity is essential. In addition, underlying causes for the initial high loss to follow-up need to be identified and addressed. The call for integrated patient-centred TB care and prevention, and the bold policies and supportive systems in the WHO End TB strategy,[14] need to be operationalized to address these issues. National TB programmes alone may not be able to address this misalignment between diagnostic capacity and treatment availability.

It is also imperative that all enrolled MDR-TB patients complete their treatment. As shown, treatment success rates in the Region are low at 52% for the 2011 cohort. This is similar to the global average of 48%.[1] High proportions of MDR-TB cases lost to follow-up or not evaluated (21% and 17%, respectively) are part of this low success rates and result in continual transmission of drug-resistant TB strains. These low success rates challenge the usefulness of PMDT; however, Cambodia

Figure 6. **Treatment outcomes of MDR-TB and RR-TB cases by year and selected country,* WHO Western Pacific Region, 2007–2011**

MDR-TB, multidrug-resistant tuberculosis; RR-TB, rifampicin-resistant tuberculosis.

* Countries that reported at least one MDR-TB case in 2013 and reported treatment outcomes for 2011.

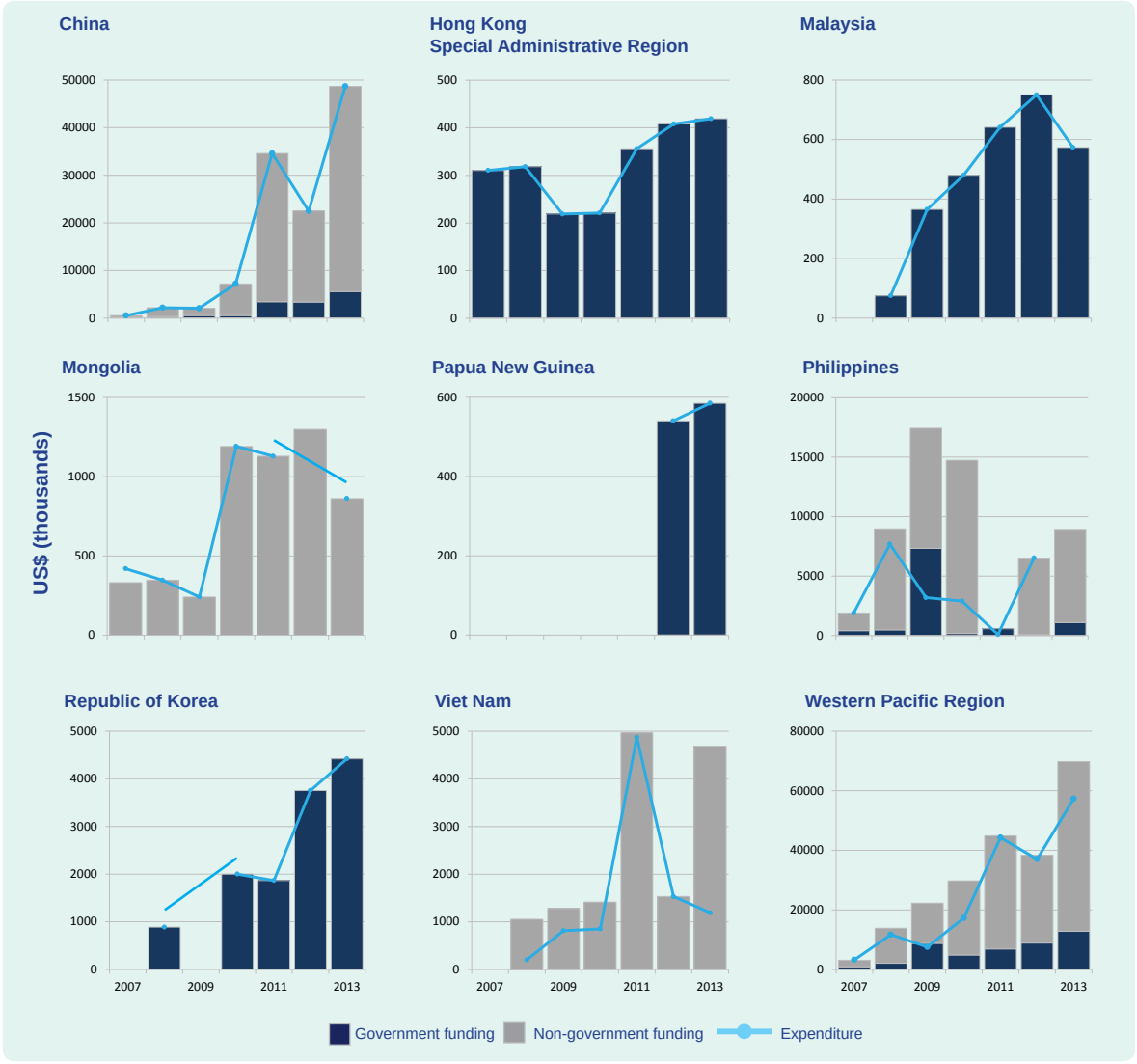
and Viet Nam showed treatment success rates above 70% over several years, showing that increasing treatment success for MDR-TB is possible.

The overall increase in notification and enrolment in treatment of MDR-TB cases demonstrates the readiness for PMDT scale-up at the country level. The results of this analysis showed that notification of TB cases is increasing; however, currently only 16% of the estimated MDR-TB among notified cases were reported under PMDT. Considerable challenges remain for scaling up PMDT with serious concerns regarding national political commitment and the long-term sustainability of donor-funded programmes that provide more than 80% of funding for MDR-TB.

The reader should note that the data reported in this analysis were not complete for all countries. As such, interpretation of the data should be made with caution.

MDR-TB is a man-made phenomenon, and unless the underlying cause is addressed, control efforts will not be successful. Prevention of MDR-TB including strengthening basic TB control needs to be at the centre of the strategy to address MDR-TB. Scaling-up of PMDT has to progressively improve all critical steps of the

Figure 7. **Expenditure on MDR-TB and RR-TB by funding source, year and selected country*, WHO Western Pacific Region, 2007–2013**



MDR-TB, multidrug-resistant tuberculosis; RR-TB, rifampicin-resistance tuberculosis.

* Countries that reported at least 10 MDR-TB cases in 2013.

cascade of services in a balanced manner. TB control has entered the most dynamic phase in decades with many opportunities. We have new diagnostic tools, new TB drugs and new strategies. It is therefore critical to invest both financial and technical resources to prevent MDR-TB and to scale up PMDT.

Conflict of interest

None declared.

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