

A community-based TB drug susceptibility study in Mimika District, Papua Province, Indonesia

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SUMMARY

SETTING: A district level tuberculosis (TB) control programme in Papua Province, Indonesia.

OBJECTIVE: To determine the nature and extent of drug-resistant TB in newly diagnosed sputum smear-positive patients.

METHODS: Sputum was collected from previously untreated smear-positive pulmonary TB patients diagnosed in the district over a 10-month period. Sputum specimens were processed and inoculated into a BACTEC MGIT960 tube. Isolates were identified by Ziehl-Neelsen staining, hybridisation with nucleic acid probes and biochemical investigations. Susceptibility testing was performed using the radiometric proportion method. Pyrazinamide testing was performed using the Wayne indirect method.

RESULTS: One hundred and seven patients had sputum

sent to a reference laboratory; 101 (94.4%) were culture-positive for *Mycobacterium tuberculosis*, with 87 (86.1%) fully sensitive to first-line anti-tuberculosis drugs. Two per cent were multidrug-resistant (MDR-TB) and 12 (11.9%) had other drug resistance. Each of the MDR-TB isolates was susceptible to amikacin, capreomycin, ciprofloxacin and para-aminosalicylic acid (PAS), but were resistant to rifabutin. One isolate was also resistant to ethionamide.

CONCLUSIONS: MDR-TB is present in Indonesia but is not a major problem for TB control in this district. Generalisability to other districts in Indonesia, particularly large urban areas, needs to be confirmed by future studies.

KEY WORDS: tuberculosis; Indonesia; multi-drug resistance; epidemiology; survey

MULTIDRUG RESISTANCE is a well recognised threat to tuberculosis (TB) control worldwide.¹ Since 1994, the World Health Organization (WHO), together with the International Union Against Tuberculosis and Lung Disease (IUATLD) has coordinated the Global Project on Anti-Tuberculosis Drug Resistance.^{2,3} The Project has concentrated on surveys of community-based samples of newly diagnosed, previously untreated sputum smear-positive patients.⁴ Using standard statistical sampling and laboratory methods and rigorous quality control, the Project has reported survey results from 72 geographical settings.⁴ All countries or regions surveyed have some drug-resistant cases, with rates of multidrug-resistant TB (MDR-TB, defined as resistance to at least isoniazid [H, INH] and rifampicin [R, RMP]) from 0% in several settings to 14.1% in Estonia (mean rate 1%).²

As with other global WHO initiatives in TB control, there has been a particular interest in drug resistance patterns in the 22 high-burden countries that account for 80% of the world's TB cases.⁵ Indonesia, with an annual TB incidence of 271 per 100 000 population, is estimated to have the third highest number of TB cases diagnosed each year, but there is little information about the extent of MDR-TB.⁶ In this paper, we present drug resistance data from a community-based study of newly diagnosed sputum smear-positive pulmonary TB (PTB) patients in eastern Indonesia.

SETTING

Indonesia, the fourth most populous country in the world (population 238 million), comprises 33 provinces consisting of 357 districts and 17 000 islands. A

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National TB Control Programme (NTP) has been operating in the country since 1996 and uses the WHO/IUATLD strategy known as DOTS.⁶ In 2002, DOTS coverage was 98%, 30% of the estimated number of sputum smear-positive patients were diagnosed and the cure rate was 86%.⁶

Mimika District, on the southern coast of Papua Province, has an estimated population of 131 000 (annual family planning census, unpublished data). A well functioning TB control programme has been operating in the district since 1997. The programme conforms to the NTP protocols and employs all the elements of the DOTS strategy. There is good coordination between the government, hospital and private health services, and a district register is maintained at the TB clinic attached to the Ministry of Health Primary Health Care Clinic in Timika, the main population centre for the district. TB control is supported by the Public Health and Malaria Control (PHMC) Department of the mining company located in the district. Staff employed by PHMC provide clinical services and are responsible for the enrolment, recording, default tracing and reporting of the NTP in the district. Quarterly TB control meetings are coordinated by the Ministry of Health District Health Official responsible for communicable disease control. There are three TB diagnostic laboratories in the district, and all service providers (hospitals, private practitioners, PHMC and government clinics) refer patients to the programme for treatment. The TB programme follows the guidelines of the NTP, with diagnosis by sputum microscopy using the Ziehl-Neelsen (ZN) method, a 6-month treatment regimen and directly observed treatment. The following medications are used for newly diagnosed sputum smear-positive PTB patients: RMP, INH, pyrazinamide (Z, PZA) and ethambutol (E, EMB) in the regimen 2RHZE/4RH.* In addition, streptomycin (S, SM) is used in the retreatment regimen (2SRHZE/RHZE/5RHE). In 2002, 222 patients with smear-positive PTB were commenced on treatment, with an 89% treatment success rate (Mimika District Ministry of Health, unpublished data).

METHOD

Selection of patients and specimen collection

This study formed part of a larger study on lung function in TB patients, which will be reported elsewhere. All adult new (previously untreated) sputum smear-positive PTB patients diagnosed in Timika district between 7 July 2003 and 5 May 2004 and notified to the district register were eligible for enrolment in the lung function study and thus eligible for the drug resistance survey. This method was consistent with the

100% sampling method in the WHO Guidelines for Drug Resistance Surveillance (DRS).⁴ Informed consent was obtained and a fresh sputum specimen was obtained from the patient prior to commencing TB treatment. Specimens were transported to the Institute of Medical and Veterinary Science (IMVS), Adelaide, South Australia for microscopy, culture, specific identification and drug susceptibility testing (DST). The IMVS Mycobacterium Reference Laboratory participates in external quality control programmes for DST through the WHO Global Supranational Reference Laboratory Quality Control Network and the Centers for Disease Control and Prevention, Atlanta, GA, USA. The IMVS Laboratory consistently achieves satisfactory performance in both programmes. Human immunodeficiency virus (HIV) testing is routine in the TB clinic for all TB patients using two tests (Determine™, Abbott Diagnostics, Jakarta, Indonesia and a confirmatory Western Blot). In this study there was 100% concordance between the two tests.

Reference laboratory methods

Sputum specimens were prepared using standard methods, and microscopy, culture and mycobacterial identification were performed as described in detail elsewhere.⁷

DST was performed using the radiometric BACTEC 460TB system (Becton Dickinson, Baltimore, MD, USA) using the recommended critical concentrations for the following antibiotics: INH (0.1 and 0.4 µg/ml), RMP (2.0 µg/ml), EMB (2.5 µg/ml), SM (2.0 µg/ml), amikacin (AMK, 1.0 µg/ml), capreomycin (CAP, 2.5 µg/ml), ciprofloxacin (CFX, 1.0 µg/ml), para-amino salicylic acid (PAS, 4.0 µg/ml), rifabutin (RBT, 1.0 µg/ml) and ethionamide (ETH, 1.25 µg/ml).^{8,9} The Wayne method of detecting pyrazinamidase activity was used to infer PZA susceptibility.¹⁰

Data analysis

All laboratory data were entered in MS Excel and patient details in MS Access (Microsoft Corp, Redmond, WA, USA). Data were imported to STATA™ (Stata Corp, College Station, Texas, USA) for analysis. Differences in continuous variables were examined using Student's *t*-tests and differences in proportions using Pearson's χ^2 test. All data were normally distributed except for patient's age, and for comparisons of this variable a log transformation was used to create an acceptable distribution. Ninety-five per cent confidence intervals (95%CI) were calculated using the exact binomial method. All tests were two-sided, and a threshold of $P = 0.05$ was used to define statistical significance.

Ethics

Ethical permission for this study was obtained from the Ethics Committees of the National Institute of

* Numbers before the letters indicate the duration in months of the phase of treatment.

Health Research and Development in Jakarta, Indonesia, and the Menzies School of Health Research in Darwin, Australia.

RESULTS

Patient selection and demographics

The method of selection of patients for this study is summarised in Table 1. There were 138 new sputum smear-positive TB patients diagnosed during the study period; 121 patients were registered with the district TB programme and therefore eligible for DRS. There were 109 patients enrolled in the lung function study, of whom 107 (88.4% of DRS eligible patients) provided a single sputum sample for culture, species identification and DST. Of the 107 samples, 101 (94.4%) successfully grew *Mycobacterium tuberculosis*. The majority of the patients (70.3%) were male, and the median age at time of diagnosis was 30.7 years (range 17.2–69.1 years). Patients were referred from a variety of private practitioners (3%), government clinics (44%), PHMC clinics (13%) and hospitals (40%). Three per cent of patients were identified as the result of contact tracing by the TB clinic. The ethnic groups of patients (50% Papuan) were representative of the total population of Mimika District (60% Papuan) and came from a broad distribution of residential addresses within the district. Five (4.4%) of the culture-positive patients were HIV-seropositive.

The DST results for first-line anti-tuberculosis drugs are shown in Table 2. Eighty-six per cent of the isolates were fully susceptible to first-line drugs, with only two MDR-TB isolates, and 12 others with resistance to either INH or SM or a combination of the two drugs. The two patients with MDR-TB were both Papuans but from different ethnic groups, were not epidemiologically linked and had different antibiograms suggesting two distinct isolates. Further DST was performed for the MDR-TB isolates. Both were susceptible to SM, PZA, AMK, CAP, CFX and PAS, and one was also susceptible to ETH and high INH. Both isolates were resistant to RBT. At the time of writing,

Table 1 Selection of patients for the drug resistance survey in Timika, July 2003–May 2004

	n (%)
Total smear-positive patients diagnosed	138 (100)
Not enrolled in TB study	
Refused	5 (3.6)
Clinician's decision*	5 (3.6)
Diagnosed at hospital†	17 (12.3)
Other‡	2 (1.4)
Enrolled in TB study	
No specimen collected	2 (1.5)
Specimen sent	107 (77.5)

* Too sick (n = 2); from very isolated area (n = 3).

† Diagnosed at community hospital but not enrolled in District TB Register.

‡ Early transfer out (n = 1); no reason given (n = 1).

Table 2 Drug resistance patterns against first-line TB medications used in the Indonesian TB control programme in Timika, Papua Province

	n (%)	95%CI
Total tested	101 (100.0)	
Fully susceptible	87 (86.1)	77.8–92.2
Any resistance	14 (13.9)	7.8–22.2
Monoresistance		
H	3 (3.0)	0.6–8.4
S	1 (1.0)	0.3–5.4
Other patterns		
HS	8 (7.9)	3.5–15.0
MDR		
HRE	2 (2.0)	0.2–7.0

TB = tuberculosis; CI = confidence interval; H = isoniazid; S = streptomycin; MDR = multidrug-resistant; R = rifampicin; E = ethambutol.

one patient had died and the other remained sputum smear-positive despite second-line anti-tuberculosis treatment. Among the patients with other drug resistance, one patient died during treatment (8.3%), one failed (8.3%) and three defaulted (25%). Of those who completed treatment, the cure rate was 87.5%.

DISCUSSION

This community-based study of drug resistance in the world's fourth most populous country has demonstrated that MDR-TB is present but at a low rate in this remote district with a relatively small population but well functioning TB control programme operating in a very challenging environment. This is an encouraging finding, but needs to be seen in the context of potential limitations.

The relatively low point estimate of MDR is similar to that found in neighbouring countries where data are available (see Table 3). No drug resistance survey data are available from Papua New Guinea or Brunei Darussalam, which also border Indonesia. This study is important in providing further information to inform both the global surveillance project and policy in Indonesia. The only previous published data on drug resistance are from a hospital-based study in the capital, Jakarta.¹⁵ That study showed an MDR-

Table 3 Drug resistance surveillance data for countries bordering on Indonesia

Country	Year of survey	Drug resistance		Ref
		Any %	MDR %	
Malaysia	1996–1997	4.8	0.1	11
Singapore	2001	5.0	0.9	2
Philippines	1997	14.6	1.5	12
Thailand	2001	14.8	0.9	2
Timor Leste	1999	17.2	0	13
Australia	2002	10.3	0.9	14

MDR = multidrug resistance.

TB prevalence in previously untreated patients of 4.6%, within the confidence interval (CI) of our estimated incidence in Timika.

The main limitations of this study are the relatively small sample size and the generalisability of the findings to other parts of Indonesia. While the 2% MDR-TB rate is reassuring, the upper limit of the 95%CI was 7%. It is not possible to compare the upper limit of the 95%CI for MDR with other countries in the region, because these are not included in the WHO reports.

Regarding generalisability, Mimika District has a very high population growth rate of 10–14%, with economically driven migration from other parts of Indonesia,¹⁶ and the non-Papuan study subjects came from a variety of Indonesian districts and ethnic backgrounds. This may mean that the results of this study could be more representative of the MDR-TB prevalence elsewhere in Indonesia than may be first suggested by the relatively remote location of Mimika District. It is also possible that the well functioning TB programme in Mimika could have attracted TB patients who were already sick to come to the district for treatment. Some patients diagnosed at the hospital who were not given the opportunity to participate in the present study, and others who refused to be part of the study or who were early defaulters and therefore not included in the study, may have been more likely to be drug-resistant cases, so the findings may be unduly conservative. However, in common with most WHO/IUATLD studies conducted elsewhere, this study has concentrated on patients who are registered with the NTP at district level.

It is usual for more than one district in a country to contribute to a drug resistance survey. This ideal was not possible for the present study, as it was logistically impossible to obtain specimens from other districts in Papua province. The findings of this study should therefore be viewed as preliminary and need confirmation from planned studies in more populous districts with a longer history of TB treatment in other parts of the country. Despite these limitations, this is an important first step to quantifying the prevalence of MDR-TB in provinces of Indonesia. Due to the large numbers of TB patients diagnosed each year in Indonesia, even a relatively low incidence of MDR-TB has major implications. If our estimate is generalisable to the whole country, then there could be between 5000 and 18 000 MDR-TB cases currently requiring treatment. Policy decisions and pilot treatment strategies need to be developed to cope with this potentially large number of patients.^{17,18}

CONCLUSION

It is feasible to perform drug resistance surveys in a remote area of Indonesia. Whilst it appears that MDR-TB is uncommon, this result will need to be confirmed in other districts of this large, diverse and populous

country before this result can be confirmed to be a true estimate for the whole country. In the meantime, the DOTS strategy needs to continue to be strengthened throughout Indonesia to prevent an escalation of drug resistance.

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R É S U M É

CONTEXTE : Un programme de lutte antituberculeuse au niveau du district dans la Province de Papouasie, Indonésie.

OBJECTIF : Déterminer la nature et l'importance de la résistance aux médicaments antituberculeux chez les patients à bacilloscopie positive récemment diagnostiqués.

MÉTHODES : Les expectorations ont été prélevées chez des patients atteints de tuberculose à bacilloscopie positive et sans traitement antituberculeux antérieur, diagnostiqués dans le district pendant une période de 10 mois. Les échantillons d'expectoration ont été traités et inoculés dans un tube BACTEC MGIT960 et sur des milieux de Löwenstein-Jensen. Les isolats ont été identifiés par la coloration de Ziehl-Neelsen, par l'hybridation au moyen de sondes à acides nucléiques et par des investigations biochimiques. Les tests de sensibilité ont été pratiqués par la méthode radiométrique des proportions. Pour le pyrazinamide, on a utilisé la méthode indirecte de Wayne.

RÉSULTATS : Le laboratoire de référence a reçu les expectorations de 107 patients. Chez 101 (94,4%), la culture a été positive pour *Mycobacterium tuberculosis*, dont 87 (86,1%) étaient totalement sensibles aux médicaments antituberculeux de première ligne. On a noté 2% de multirésistance (TB-MR) et 11,9% (12 patients) d'autres résistances aux médicaments. Chacun des isolats de TB-MR était sensible à l'amikacine, la capréomycine, la ciprofloxacine et l'acide para-amino-salicylique, mais était résistant à la rifabutine. Un isolat était également résistant à l'éthionamide.

CONCLUSIONS : La TB-MR existe en Indonésie mais n'est pas un problème majeur de lutte antituberculeuse dans ce district. Des études ultérieures devront démontrer dans quelle mesure ces résultats peuvent être généralisés vers d'autres districts d'Indonésie, en particulier les grandes zones urbaines.

R E S U M E N

MARCO DE REFERENCIA : Un programa de lucha contra la tuberculosis (TB) a nivel distrital en la Provincia de Papúa, Indonesia.

OBJETIVO : Determinar el tipo y difusión de la TB multidrogorresistente (TB-MDR) en los pacientes nuevos con baciloscopia positiva.

MÉTODOS : Se recogieron en el distrito, durante un periodo de 10 meses, muestras de esputo de los pacientes con TB pulmonar, baciloscopia positiva y sin antecedente de tratamiento antituberculoso. Las muestras se procesaron y se sembraron cultivos en tubos de BACTEC MGIT960 y en medio de Löwenstein-Jensen. Los microorganismos aislados se identificaron mediante coloración de Ziehl-Neelsen, hibridación con sondas de ácidos nucleicos y pruebas bioquímicas. Se practicaron también pruebas de sensibilidad a los medicamentos mediante el método radiométrico proporcional. Para la prueba de sensibilidad a la piracinamida se aplicó el método indirecto de Wayne.

RESULTADOS : Se enviaron al laboratorio de referencia muestras de esputo de 107 pacientes, y de estas 101 (94,4%) presentaron cultivos positivos para *Mycobacterium tuberculosis* y 87 (86,1%) fueron totalmente sensibles a los medicamentos antituberculosos de primera línea. Se observó MDR en 2% de los aislados de *M. tuberculosis* y el 11,9% (12) presentó otro tipo de farmacorresistencia. Todos los aislados con MDR fueron sensibles a amiacina, capreomicina, ciprofloxacina y ácido p-aminosalicílico, pero resistentes a rifabutina. Uno de los aislados fue también resistente a etionamida.

CONCLUSIONES : La TB-MDR está presente en Indonesia, pero no constituye un problema significativo para el control de la TB en este distrito. Esta afirmación no puede generalizarse a otros distritos de Indonesia, en particular a las grandes zonas urbanas sin llevar a cabo estudios complementarios.