Infection control, genetic assessment of drug resistance and drug susceptibility testing in the current management of multidrug/extensively-resistant tuberculosis (M/XDR-TB) in Europe: A tuberculosis network European Trialsgroup (TBNET) study

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ABSTRACT

Aim: Europe has the highest documented caseload and greatest increase in multidrug and extensively drug-resistant tuberculosis (M/XDR-TB) of all World Health Organization (WHO) regions. This survey examines how recommendations for M/XDR-TB management are being implemented.
Methods: TBNET is a pan-European clinical research collaboration for tuberculosis. An email survey of TBNET members collected data in relation to infection control, access to molecular tests and basic microbiology with drug sensitivity testing.
Results: 68/105 responses gave valid information and were from countries within the WHO European Region. Inpatient beds matched demand, but single rooms with negative pressure were only available in low incidence countries; ultraviolet decontamination was used in 5 sites, all with >10 patients with M/XDR-TB per year. Molecular tests for mutations associated with rifampicin resistance were widely available (88%), even in lower income and especially in high incidence countries. Molecular tests for other first line and second line drugs were less accessible (76 and 52% respectively). A third of physicians considered that drug susceptibility results were delayed by > 2 months.
Conclusion: Infection control for inpatients with M/XDR-TB remains a problem in high incidence countries. Rifampicin resistance is readily detected, but tests to plan regimens tailored to the drug susceptibilities of the strain of *Mycobacterium tuberculosis* are significantly delayed, allowing for further drug resistance to develop.

1. Introduction

The World Health Organization (WHO) 2015 report observed that the European Region had the highest caseload of documented patients and greatest increase in multidrug/extensively drug-resistant tuberculosis (M/XDR-TB) [1]. TBNET reported on the outcome of a review of the management of M/XDR-TB in 2010, noting that there were significant departures from the International Standards of Care for Tuberculosis and their European adaptation [2]. Most notably, there were deficiencies in recording patient outcomes, infection control [3], bacteriological analysis and laboratory support, as well as regimen selection and treatment duration. TBNET has also noted the problems of availability and cost of drugs in the management of M/XDR-TB [4].

In a consensus statement regarding the management of M/XDR-TB, it was identified that infection control measures should include a prompt diagnosis and isolation of patients in a well-ventilated single room with upper room ultraviolet germicidal irradiation [5]. Prompt diagnosis requires phenotypic drug susceptibility testing (DST) and genotypic tests for rifampicin resistance in those at risk of multidrug-resistant tuberculosis (MDR-TB), with further molecular testing for those with rifampicin resistance, especially for resistance to fluoroquinolones or injectable drugs. This survey aims to describe the current situation with regard to these basic recommendations.

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2. Methods

2.1. Study design

A cross-sectional survey.

2.2. Setting

Any hospital within the WHO European Region where physicians manage tuberculosis (TB).

2.3. Participants

The Tuberculosis Network European Trialsgroup (TBNET) has >650 members who are engaged in the management of tuberculosis. From October 2015 to January 2016, a standardized questionnaire was sent to TBNET members by e-mail to collect information about their management of patients with M/XDR-TB. Reminders were sent at weekly intervals after the first communication until more than 100 replies had been obtained. Members who were from the same hospital were considered to give a single answer and, where there were any differences in information, were contacted to confirm which answer was correct.

2.4. Variables

The questionnaire consisted of 7 identifiers, a confirmation of consent to participate in the study, 10 questions regarding inpatient and outpatient facilities, 19 questions regarding access to microbiology laboratories and 8 questions regarding participation in clinical research. The full questionnaire is available on request; included in the e-supplement are the questions relevant to this publication.

2.5. Bias

The title of the survey indicated an interest in MDR-TB. The responders were therefore less likely to reply if they had not seen patients with drug-resistant TB. Some replies were subjective and in particular all replies regarding the frequency of drug sensitivity testing results, which were received more than two months after the start of treatment, were rechecked by repeated email correspondence.

2.6. Statistical methods

Frequencies were compared using a chi-squared test and if a cell had <5, then Yates' correction [6] was employed, using the GraphPad free software (https://graphpad.com/quickcalcs/contingency1.cfm). High income (>$12,475 pa) and middle income ($1026-12,475) were based on the 2015 Gross National Income per capita from data collated by the World Bank website (http://data.worldbank.org/indicator/).

2.7. Ethics

The study did not require ethical approval after consultation with the Health Research Authority decision tool and Integrated Research Application System websites.

3. Results

3.1. Description of the sample

From 650 members, 105 replies were received. The data included 13 duplicates and 2 triplicates and replies without any information apart from the initial identification (Fig. 1). There were therefore 79 valid responses, of which 68 were from within the WHO European Region.

Twenty-four countries gave replies, of which 9 were from a single site, several of which were the major referral centres for that country (e.g. Belarus). The remainder had 2 or more site responders (Table 1; Fig. 2). Five countries had included all notified MDR-TB cases for 2015 and a further five covered >72% of all cases. If countries with a population <1 million and those with no MDR-TB cases per year were excluded, the significant omissions for the WHO Region were Bulgaria, Estonia, Hungary, Lithuania, Armenia, Azerbaijan and Georgia and the Central Asian republics Kazakhstan, Kyrgyzstan, Tajikistan, Turkmenistan and Uzbekistan. However, the survey covered countries having 96% of all MDR-TB cases within the WHO European Region, excluding the Central Asian republics. In terms of cases of MDR-TB, the survey participants had seen 15% of the recorded number for 2015 in the WHO region, excluding the Central Asian republics and 16% of those countries with a response.

Ten of the 68 responding sites had no MDR-TB cases in the period covered by the survey (Fig. 2). However, their responses indicate the preparedness for such cases. In one case, the responder had responsibility for MDR-TB, but had apparently not supervised their care (DC, Moldova). In other instances, another site in the same city could provide data, apart from Denmark, where there were few cases of MDR-TB.

In most cases, the responder was the head of department (57/68, 84%). The majority were pulmonary physicians (40/68, 59%), but there were significant contributions from infectious diseases physicians 20/68, 30%) and paediatricians (4/68, 6%). Specialist or university hospitals provided most responses (54/68, 80%), but some considered themselves district general hospitals even though they were the highest tier of health care for their area (e.g. Mathilde Jachym, at the national MDR-TB centre outside Paris). This is evidenced by the data in Table 1, noting that the percentage of the national figures for MDR-TB was generally high.

In order to assess consistency, replies were examined in detail. Repeated replies either gave the same information or only one reply contained complete information. The hospital with four responses differed according to the personal responsibilities of the individual physicians for inpatient facilities; the junior doctor's answers on this occasion were at variance with the other three physicians' and were therefore discarded. With regard to countries where more than one hospital replied (Table 1), inpatient, outpatient and local laboratory facilities differed, as would be expected, but regional and supra-regional laboratory access was consistent.

3.2. Facilities for treating M/XDR-TB

Five responding hospitals, located in Austria, Greece, Moldova and two in Spain had no inpatient facilities for treating patients with M/XDR-TB. The physicians indicated that inpatient facilities were accessed by referral to another specialist unit.

Table 2 shows the range of inpatient beds and the availability of different measures for infection control. Most notably, as the incidence of MDR-TB increases, the number of single rooms fails to follow accordingly (Fig. 3A and B). If there is any infection control in these hospitals managing large numbers of M/XDR-TB, this is through the occasional use of UV light irradiation, but single room isolation and negative pressure are rarely available.

Negative pressure was frequently used in multi-occupancy rooms in hospitals with more than 10 M/XDR-TB patients a year (Fig. 2C). Infectious disease physicians had greater access to negative pressure single rooms (15/19) compared to pulmonologists (16/34; χ^2 with Yates' correction $P = 0.049$).

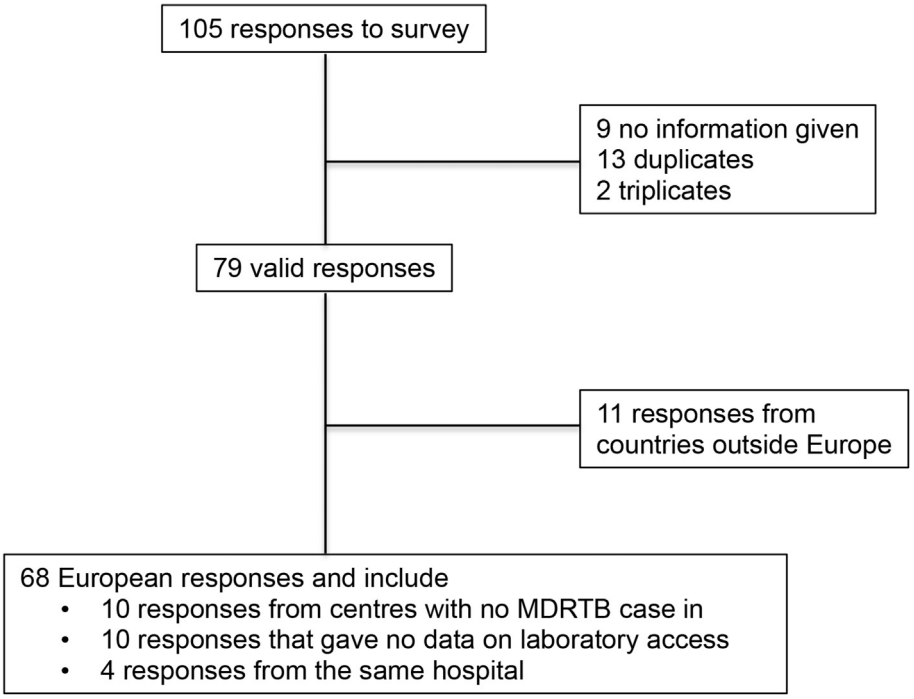

Fig. 1. Flow diagram for data.

Table 1
Epidemiological data of contributors and their institutions.

Countries covered by the survey				Countries not covered by the survey		
Country	No. of sites	MDR-TB cases in 2015[a] (% seen)	Population (millions)	Country	MDR-TB cases in 2015	Population (millions)
European Union at time of survey						
Austria	1	14 (0)	8.5	Bulgaria	80	7.1
Belgium	1	16 (75)	11	Croatia	0	4.2
Czechia	2	13 (100)	11	Cyprus	0	1.2
Denmark[b]	4	4 (100)	5.7	Estonia	47	1.3
France	2	63 (73)	64	Finland	10	5.5
Germany	4	170 (23)	81	Hungary	30	9.9
Greece	1	10 (10)	11	Ireland	2	4.3
Italy	6	110 (16)	60	Lithuania	270	2.9
Latvia	1	75 (80)	2	Luxembourg	0	<1
Netherlands	3	13 (100)	17	Malta	0	<1
Poland	1	60 (3)	39	Slovenia	0	2.1
Portugal	2	22 (14)	10			
Romania	1	670 (9)	20			
Slovakia	2	1 (100)	5.4			
Spain	8	25 (100)	46			
Sweden	4	27 (78)	9.8			
UK	7	58 (41)	65			
European Economic Area						
Norway	1	7 (72)	5.2	Iceland	0	<1
Switzerland	2	21 (29)	8.3	Liechtenstein	0	<1
WHO European Region, excluding Central Asian Republics						
Belarus	1	1800 (56)	9.5	Albania	8	2.9
Moldova	4	1700 (23)	4.1	Andorra	0	<1
Serbia	1	20 (15)	8.9	Armenia	150	3
Russia	3	42000 (1)	143	Azerbaijan	1400	9.8
Ukraine	2	12000 (3)	45	Bosnia & Herzegovina	6	3.8
				FYC Macedonia	6	2.1
				Georgia	550	4
				Israel	20	8.1
				Monaco	0	<1
				Montenegro	0	<1
				Montserrat	0	<1
				San Marino	0	<1
				Vatican	0	<1
TOTAL (%)	63	58,896 (16)	766.9		2579	73

[a] Data for MDR-TB cases and populations taken from the WHO 2016 report [1].
[b] Including Greenland. Central Asian Republics include Kazakhstan, Kyrgyzstan, Turkmenistan and Uzbekistan (total 12,560 MDR-TB; population 59.3 million).

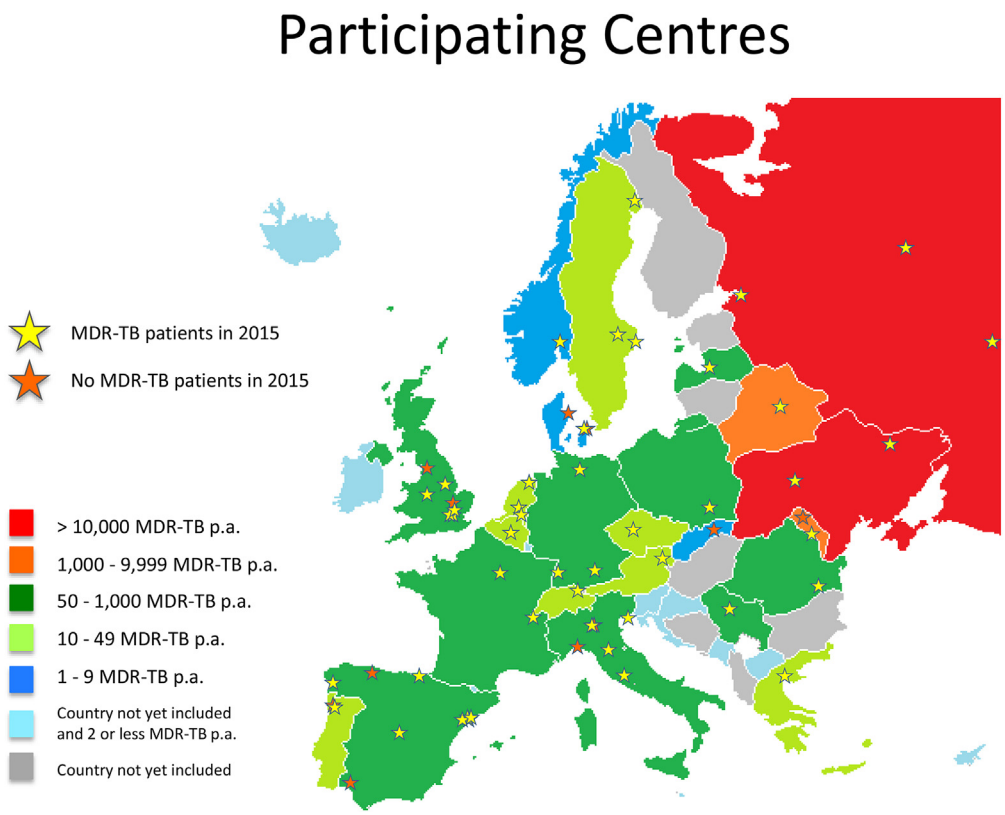

Fig. 2. Map of TBNET MDR-TB centres. Yellow stars indicate responding centres with MDR-TB in 2015; orange stars indicate responders with no MDR-TB in 2015.
Countries with responding centres are coloured by the number of MDR-TB cases in 2015 (data from World Health Organization [1]): red >10,000; orange 1000–9999; dark green 50-1000; light green 10–49; blue 1–9.
Countries without data and 2 or less MDR-TB cases are coloured light blue.
Grey areas indicate no responders.
Kosovo is indicated in white as WHO does not contain data for this area.

Table 2
Inpatients beds and infection control measures by hospital site.

MDR-TB per year	No. sites	Inpatient beds (mean ± SE)	Single rooms median (range)	Negative pressure median (range)	UV light median (range)
0	10	3 ± 1	4 (0–5)	1 (0–4)	0 (0–0)
1–4	20	5 ± 1	4 (1–24)	4 (0–13)	0 (0–8)
5–9	8	7 ± 2	8 (1–35)	3 (0–15)	0 (0–10)
10–20	11	19 ± 5	6 (1–18)	8 (0–26)	2 (0–12)
>20	11	48 ± 10	2 (0–20)	0 (0–20)	3 (0–20)

3.3. Molecular tests of drug resistance

The majority (52, 77%) stated they had access to tests for mutations in *rpoB* associated with rifampicin resistance at their hospital (10 had no access and 6 did not reply to this question; Table 3). The test was unavailable locally in all four hospitals in Denmark, 2/8 hospitals in Spain and 3/6 hospitals in Italy. However, PCR tests for rifampicin resistance were available at all regional laboratories (but not in some specialist TB hospitals, Fig. 4A).

The communication of rifampicin resistance to the physician occurred at a median of 2 days, whether performed locally or after referral to a regional or specialist unit (Fig. 3). Surprisingly, 42 (62%) also claimed to have access at their local hospital to PCR testing for first line drugs other than rifampicin; 21 without such access could obtain these from a regional reference laboratory (Fig. 4; 5 did not reply to these related questions).

3.4. Time to drug sensitivity results

Physicians were asked to estimate the percentage of drug susceptibility testing (DST) results, which were not available until after 2 months - the point at which a decision on the continuation phase drug regimen would normally be made. More than a third of physicians (24; 35%) from 10 countries could not remember any delay in the previous year. Each of the physicians who estimated a delay in DST of >10% was approached to confirm their estimates approximately 3 months after the initial response. The subsequent emails produced a downgrading in the percentage of samples with delays, largely through misinterpretation of the question. One responder indicated that a recent long delay had coloured the first response. However, the majority (93%) were consistent in their estimate. In the final analysis, a third (23; 33%), from 14 different countries, confirmed the delay (range 10–50% of all samples sent

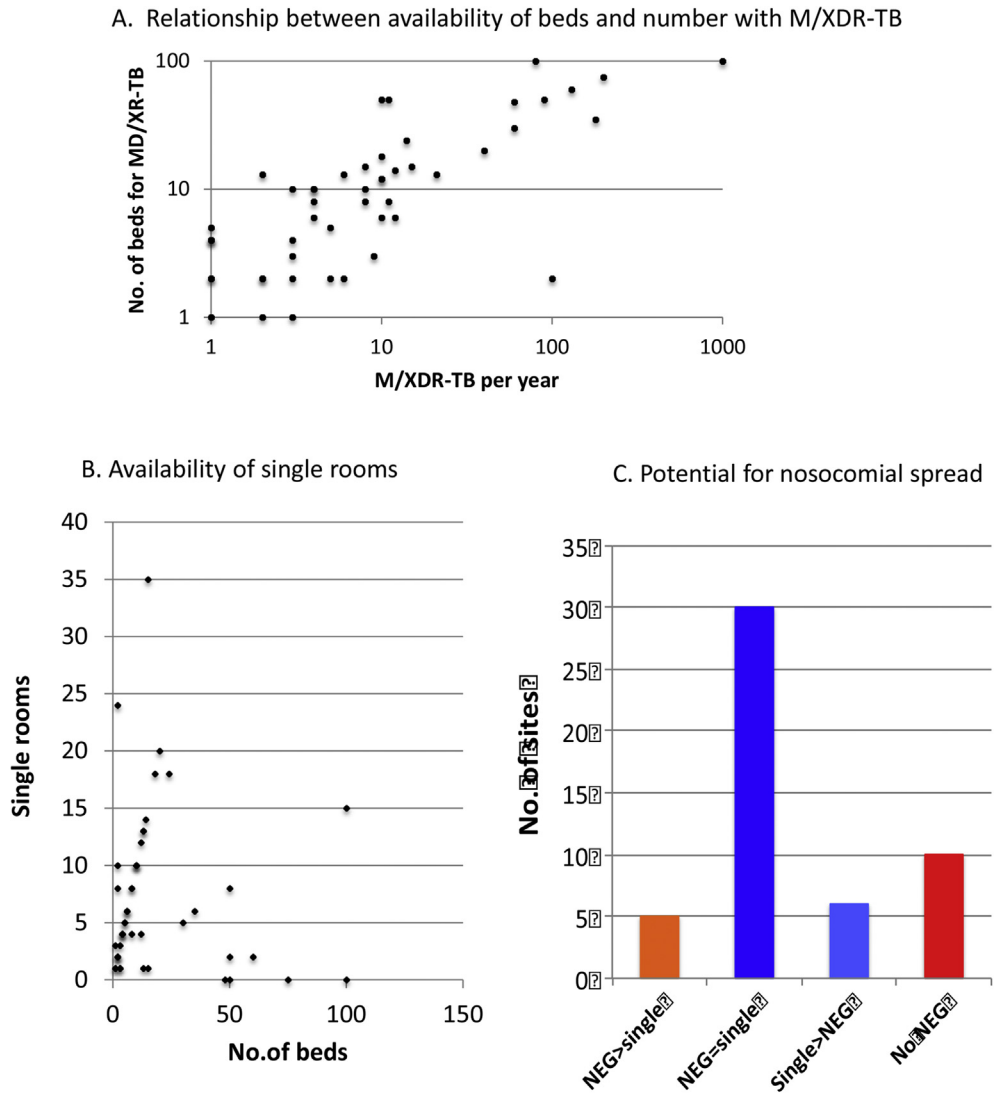

Fig. 3. Inpatient facilities for the treatment of M/XDR-TB in Europe.
C. The potential for nosocomial spread is suggested by the number of hospital sites where: there is a negative pressure room (NEG) that contains several occupants (NEG>single); the number of single rooms is greater than the number of negative pressure rooms (NEG); or where there are no negative pressure rooms.

Table 3
Availability of molecular testing.

	MDR-TB		No MDR-TB
	University hospital n = 34 (%)	Regional/specialist hospital n = 16 (%)	n = 9 (%)
rpoB test on site:	**31 (91)**[a]	**13 (81)**	**4 (45)**
Results within 1 day	14 (44)	6 (40)	1 (11)
Time to result: median (range), d	2 (1–21)	2 (1–14)	4 (1–15)
PCR for other first-line drugs:	**27 (79)**	**11 (69)**	**2 (22)**
Results within 1 day	3 (12)	2 (18)	0 (0)
Time to result: median (range), d	6 (1–30)	5 (1–30)	5 (2–7)
PCR for second-line drugs on sputum (+ve smear)	**19 (56)**	**7 (44)**	**1 (11)**
Results within 1 day	2 (11)	1 (14)	nk
Time to result: median (range), d	7 (1–60)	5 (1–30)	nk
Specific mutations available	13 (69)	6 (86)	4 (45)

[a] Percentages are accurate, missing data noted in text.

for DST, median 22.5%; mean 24.3 ± 2.9). However, 19/23 had in-hospital access to PCR tests for rifampicin resistance and two could obtain these on request to the regional centre.

3.5. High vs. low incidence MDR-TB and effect of average income on facilities

A high incidence of MDR-TB was associated with a universal

access to tests for *rpoB* mutations on site with a shorter time to result (Table 4). However, genotyping for mutations causing resistance in second-line drugs was rarer and the time to DST results longer.

The incidence of MDR-TB was higher in medium income countries. The difference was that Latvia and Romania are considered high-income countries but have a higher rate of MDR-TB, whereas Serbia is a medium-high income country but with a low incidence of MDR-TB. When these three countries were changed, the number of single rooms, with negative pressure ventilation and UV light sterilization were lower in low-income countries and genotyping for mutations in second-line drugs was less available.

4. Discussion

The survey demonstrates that single rooms for the management of M/XDR-TB are not readily available in the countries from which responses were obtained, especially in geographical areas where M/XDR-TB is common. Negative pressure rooms are rare and even ultraviolet light decontamination is unusual. The availability of molecular tests for rifampicin resistance is good but in a substantial proportion results are unnecessarily delayed. The use of molecular tests to determine a suitable regimen is available in the hospitals of approximately half of those responding to the survey. However, if M/XDR-TB is not considered, a third might not discover resistance before beginning the continuation phase of treatment.

4.1. Nosocomial transmission

Although patients with M/XDR-TB can be treated as outpatients, for many the diagnosis is made after a brief hospital admission during which they are at their most infectious, especially compared to the later stages in treatment. The availability of single rooms for the management of M/XDR-TB is a significant problem in preventing nosocomial transmission [7,8]. The risk for the development of M/XDR-TB during a hospital stay was >5% in patients who were admitted with non-M/XDR-TB in Moldova and in three out of four cases was most likely due to nosocomial transmission [8]. In Belarus, the high incidence of M/XDR-TB in patients who had previously had fully sensitive disease was in part attributed to nosocomial transmission [9]. Systematic reviews have suggested that ambulatory treatment for M/XDR-TB is as effective as hospital-based treatment, where the latter was defined as inpatient treatment for the intensive phase of treatment or until culture-conversion occurred [10,11]. Due to the complexities of the management, possible adverse effects and frequent co-morbidities, treatment of MDR-TB should preferably be initiated in a hospital experienced in the management of patients with MDR-TB. The transmission of M/XDR-TB during treatment is suggested by M/XDR-TB in hospital staff [12], but in ambulatory care most

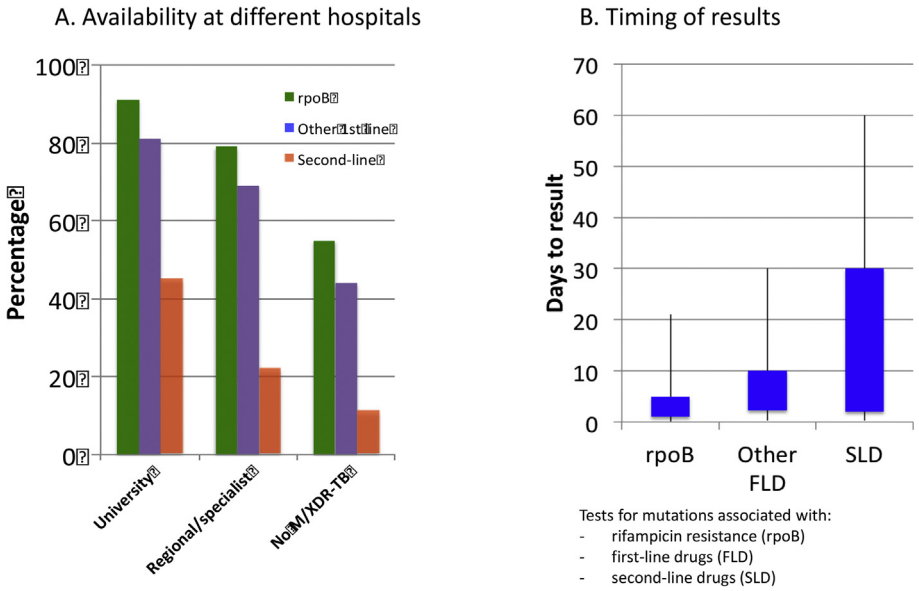

Fig. 4. Availability and timing of molecular tests of drug susceptibility of strains of *Mycobacterium tuberculosis*.

Table 4
Effect of MDR-TB incidence on available facilities.

	Incidence of MDR-TB		Per capita income	
	Low (0–0.46 per 100,000)	High (>4.8 per 100,000)	High >$12,475 p.a.	Medium ($1046-12,475)
Total MDR-TB patients/year	268	1900	423	1745
Inpatient beds for MDR-TB	397	500	474	423
Single, negative pressure rooms with UV light sterilization	65	61	83	43
rpoB mutation test on site (% of sites)	85	100	95	100
Time to rpoB test result median (range), days	2 (1–21)	1 (1–7)	2 (1–21)	1 (1–7)
Genotyping for second-line drug (SLD) resistance on sputum available (% of sites)	41	40	50	38
Time to SLD result Median (range), days	5 (1–60)	10 (1–14)	5 (1–60)	10 (1–10)

contacts would have already been exposed before the start of treatment. Prolonged hospital stays should be limited to those who are homeless or with difficult family situations, with adverse effects from treatment, significant co-morbidities or for end-of-life care [13]. A lower income per capita was associated with less access to suitable isolation rooms (Table 4). Natural ventilation and simple infection control measures can have an important role in reducing nosocomial transmission [14,15].

4.2. Molecular testing for drug resistance

The use of molecular testing for TB has expanded enormously since endorsement of this approach by WHO [16]. Despite the test time of 4 h, the median time to receiving the result was 2 days; a problem in connecting laboratories with physicians that has been well described but with longer delays than noted in this survey [17–19].

Molecular tests for other first line drugs and for second-line drugs were less commonly available and were usually employed on cultures rather than primary specimens (Table 2). Many mutations have been defined that can contribute to the decision regarding the use of the shorter treatment regimens (defining resistance to fluoroquinolones and injectable agents before starting treatment) or resolving potential problems regarding the choice of rifabutin in the presence of rifampicin resistance or aminoglycosides over polypeptide injectable agents [20]. This remains a costly exercise, which is therefore less commonly performed in lower income countries (Table 4). However, it seems likely that the decreasing cost of whole genome sequencing will permit a more comprehensive definition of drug resistance and a cost similar to that using gene probes for specific common mutations [21].

M/XDR-TB is frequently associated with resistance to other second-line drugs, especially in Eastern Europe [22]. Without access to genotypic testing for mutations in second line drugs, especially injectable agents and fluoroquinolones, the danger is that empirical regimens may have only one or two poorly effective drugs and further resistance arises. We would therefore encourage physicians to pursue a clear diagnosis of drug resistance once mutations associated with rifampicin have been identified and delay treatment if at all possible until a suitable regimen can be defined.

4.3. Delays, which might result in drug-resistance

A delay of more than 2 months from the time a clinical specimen has been received by the laboratory was noted in more than a third. There are few published estimates of the timing of DST results in relation to specimen collection. Indeed, at the turn of the century, empirical treatments were the rule rather than the exception [23]. Contamination of the specimen would be expected in perhaps 10% and these samples would then be re-processed. As with rapid molecular tests, it would appear that communication of results from the microbiology laboratory to the attending physician is a problem.

4.4. Limitations

By definition, responders to the survey would have included those with a particular interest in M/XDR-TB and who have a commitment to improving treatment through research. The availability of inpatient facilities, molecular testing and adequate local microbiological services may, therefore, have been overestimated. For 2015, the surveyed sites covered 16% of all MDR-TB recorded by WHO in their annual report (Table 1). However, in terms of access to regional and national laboratories, the countries of the respondents covered areas of the WHO European Region with 96% of all MDR-TB, excluding the Central Asian republics.

Estimates of delayed drug susceptibility results were subjective. A second email to confirm the figure, however, revealed only a single significant change. Consistency of the response over a 3-month period suggests that these were genuine perceptions. The possibility remains that some individuals consistently overestimate or underestimate problems with laboratories, depending on their working relationships. Objective audit would be the most sensible way to assess such delays.

Ten sites had no MDR-TB cases in the period of the survey. However, in these times of migration and population movements within Europe from countries with a high incidence of MDR-TB, their inclusion is a valuable indication of how well-prepared are European countries to deal with these patients.

4.5. Interpretation

The number of negative pressure rooms was greater than previously noted [3], but their absence in high incidence countries continues. By comparison with an earlier survey of a limited number of sites managing M/XDR-TB [2], access to molecular tests for tuberculosis have improved. There remains a problem with the early recognition of drug-resistance, except where, as recommended by WHO, molecular tests for rifampicin mutations are used routinely as an initial test in all patients suspected of having pulmonary TB.

4.6. Generalizability

Whilst most hospitals have a physician who can treat TB, the management of MDR-TB is usually localized to specialist centres. The response rate from TBNET members is therefore likely to reflect this increased specialization and overestimate rather than underestimate local access to isolation rooms and molecular tests of rifampicin resistance. Responders covered 15% of all MDR-TB reported within the WHO European Region, excluding the Central Asian republics.

The sample included countries within the EU, European Economic Area and some high incidence countries within Eastern Europe and covered countries whose total MDR-TB cases constitute 96% of all MDR-TB in the WHO European Region, excluding the Central Asian republics (Table 1). Thus, the survey represents the national facilities for access to other molecular tests for resistance and drug sensitivity testing most accurately.

4.7. Conclusions

Although the responders saw only 15% of all MDR-TB in the European WHO Region, the data indicate, even in these pro-active centres:

- There were too few isolation rooms in countries and hospitals with the most MDR-TB.
- The delay between a test result and its communication to the physician is many times that required for the test itself.
- A positive test for rifampicin resistance does not yet automatically lead to appropriate laboratory testing.

4.8. Recommendations

1. Inpatient facilities
   a. Those with possible pulmonary TB should have a rapid test for TB and rifampicin resistance.

   b. Outpatient management with home isolation and treatment is the best way to address the lack of inpatient facilities whenever possible, medical and social co-morbidities permitting.
   c. However, due to the complexities of the management, treatment of MDR-TB should preferably be initiated in a hospital experienced in the management of such patients.
   d. Isolation rooms for those with MDR-TB should be single occupancy and have negative pressure ventilation ± UV sterilization.
2. Rapid diagnosis of MDR-TB
   a. Near-patient tests to confirm the diagnosis of TB and the presence of rifampicin resistance should be widely available, especially in all hospitals with at least one case of MDR-TB within the last 5 years.
   b. A diagnosis of MDR-TB using near-patient tests should be made without requiring hospital admission.
   c. Once rifampicin resistance is found, DNA-based tests for resistance to other first-line and second-line drugs should be performed.
3. DST
   a. A positive genotypic test for rifampicin resistance should automatically mean that liquid cultures are set up to test susceptibility to other first-line and second-line drugs.
   b. MDR-TB should be prevented by ensuring that DST is available by the end of the initial phase of standard TB treatment.
4. Communication
   a. The physician, as well as the local microbiology department, should receive the results of TB diagnostic tests as soon as possible.

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References

[1] World Health Organization, Global Tuberculosis Report, WHO, Geneva, 2015, 2016.
[2] G.B. Migliori, G. Sotgiu, L. D'Ambrosio, R. Centis, C. Lange, G. Bothamley, D.M. Cirillo, S. de Lorenzo, G. Gue.nther, K. Kliiman, R. Muetterlein, V. Spinu, M. Villar, J.P. Zellweger, A. Sandgren, E. Huitric, D. Manissero, TB and MDR/XDR-TB in the EU and EEA/EFTA countries: managed or mismanaged, Eur. Respir. J. 39 (2012) 619–625.
[3] G. Sotgiu, L. D'Ambrosio, R. Centis, G. Bothamley, D.M. Cirillo, S. De Lorenzo, G. Guenther, K. Kliiman, R. Muertterlein, V. Spinu, M. Villar, J.P. Zellwegere, A. Sandgren, E. Huitric, C. Lange, D. Manissero, G.B. Migliori, TB and M/XDR-TB infection control in European TB reference centres: the Achilles' heel, Eur. Respir. J. 38 (2011) 1221–1223.
[4] G. Günther, G.B. Gomez, C. Lange, S. Rupert, van Leth, TBNET. Availability, price and affordability of anti-tuberculosis drugs in Europe: a TBNET survey, Eur. Respir. J. 45 (2015) 1081–1088.
[5] C. Lange, I. Abubakar, J.W. Alffenaar, G. Bothamley, J.A. Caminero, A.C. Carvalho, K.C. Chang, L. Codecasa, A. Correia, V. Crudu, P. Davies, M. Dedicoat, F. Drobniewski, R. Duarte, C. Ehlers, C. Erkens, D. Goletti, G. Günther, E. Ibraim, B. Kampmann, L. Kuksa, W. de Lange, F. van Leth, J. van Lunzen, A. Matteelli, D. Menzies, I. Monedero, E. Richter, S. Rüsch-Gerdes, A. Sandgren, A. Scardigli, A. Skrahina, E. Tortoli, G. Volchenkov, D. Wagner, M.J. van der Werf, B. Williams, W.W. Yew, J.P. Zellweger, D.M. Cirillo, TBNET. Management of patients with multidrug-resistant/extensively drug-resistant tuberculosis in Europe: a TBNET consensus statement, Eur. Respir. J. 44 (2014) 23–63.
[6] F. Yates, Contingency table involving small numbers and the chi-squared test, Suppl J Roy Statist Soc 1 (2) (1934) 217–235.
[7] N.R. Gandhi, D. Weissman, P. Moodley, M. Ramathal, I. Elson, B.N. Kreiswirth, B. Mathema, E. Shashkina, R. Rothenberg, A.P. Moll, G. Friedland, A.W. Sturm, N.S. Shah, Nosocomial transmission of extensively drug-resistant tuberculosis in a rurual hospital in South Africa, J. Infect. Dis. 207 (2013) 9–17.
[8] V. Crudu, M. Merker, C. Lange, E. Noroc, E. Romancenco, D. Chesov, G. Gunther, S. Niemann, Nosocomial transmission of multidrug-resistant tuberculosis, Int. J. Tuberc. Lung Dis. 19 (2015) 1520–1523.
[9] A. Skrahina, H. Hurevich, A. Zalutskaya, E. Sahalchyk, A. Astruko, S. Hoffner, V. Rusovich, A. Dadu, P. de Colombini, M. Dara, W. van Gemert, M. Zignol, Multidrug-resistant tuberculosis in Belarus: the size of the problem and associated risk factors, Bull. World Health Organ 91 (2013) 36–45.
[10] A. Bassili, C. Fitzpatrick, E. Qadeer, R. Fatima, K. Floyd, E. Jaramillo, Review article: a systematic review of the effectiveness of hospital- and ambulatory-based management of multidrug-resistant tuberculosis, Am. J. Trop. Med. Hyg. 89 (2013) 271–280.
[11] P. Weiss, W. Chen, V.J. Cook, C.J. Johnston, Treatment outcomes from community-based drug resistant tuberculosis treatment programs: a systematic review and meta-analysis, BMC Infect. Dis. 14 (2014) 333.
[12] N.R. Gandhi, D. Weissman, P. Moodley, M. Ramathal, I. Elson, B.N. Kreiswirth, B. Mathema, E. Shashkina, R. Rothenberg, A.P. Moll, G. Friedland, A.W. Sturm, N.S. Shah, Nosocmial transmission of extensively drug-resistant tuberculosis in a rural hospital in South Africa, J. Infect. Dis. 207 (2013) 9–17.
[13] World Health Organization, Companion Handbook to the WHO Guidelines for the Programmatic Management of Drug-resistant Tuberculosis, WHO, Geneva, 2014.
[14] A.R. Escombe, C.C. Oeser, R.H. Gilman, M. Navincopa, E. Ticona, W. Pan, C. Martinez, J. Chacaltana, R. Rodriguez, D.A.J. Moore, J.S. Friedland, C.A. Evans, Natural ventilation for the prevention of airborne contagion, PLoS Med. 4 (2) (2007) e68.
[15] E. Ticona, L. Huarato, D.E. Kirwan, M. Chumpitaz, C.V. Munayco, M. Maguina, M.A. Tovar, C.A. Evans, R. Escombe, R.H. Gilman, Impact of infection control measures to control an outbreak of multidrug-resistant tuberculosis in a human immunodeficiency virus ward, Peru, Am. J. Trop. Med. Hyg. 95 (6) (2016) 1247–1256.
[16] H. Albert, R.R. Nathavitharana, C. Isaacs, M. Pai, C. Denkinger, C. Boehme, Development, roll-out and impact of Xpert MTB/RIF for tuberculosis: what lessons have we learnt and how can we do better, Eur. Resp. J. 48 (2016) 1531–1532, https://doi.org/10.1183/13993003.00543-2016.
[17] E. Andre, C. Isaacs, D. Affolabi, et al., Connectivity of diagnostic technologies: improving surveillance and accelerating tuberculosis elimination, Int. J. Tuberc. Lung Dis. 20 (2016) 999–1003.
[18] J.Z. Metcalfe, S. Makumbirofa, B. Makamura, C. Sandy, W. Bara, P. Mason, P.C. Hopewell, Xpert MTB/RIF detection of rifampicin resistance and time to treatment initiation in Harare, Zimbabwe, Int. J. Tuberc. Lung Dis. 20 (2016) 882–889.
[19] M. Davids, K. Dheda, N. Pant Pai, D. Cogill, M. Pai, N. Engel, A survey on use of rapid tests and tuberculosis diagnostic practices by primary health care providers in South Africa: implications for the development of new point-of-care tests, PLoS One 10 (2015) e0141453.
[20] J. Dominguez, E.C. Boettger, D. Cirillo, et al., Clinical implications of molecular drug resistance testing for *Mycobacterium tuberculosis*: a TBNET/RESIST-TB consensus statement, Int. J. Tuberc. Lung Dis. 20 (2016) 24–42.
[21] L.J. Pankhurst, C. Del Ojo Elias, A.A. Votintseva, et al., Rapid, comprehensive, and affordable mycobacterial diagnosis with whole-genome sequencing: a prospective study, Lancet Respir. Med. 4 (2016) 49–58.
[22] G. Gunther, F. van Leth, S. Alexandru, et al., Multidrug-resistant tuberculosis in Europe, 2010-2011, Emerg. Infect. Dis. 21 (2015) 409–416.
[23] L.B. Heifets, G.A. Cengelosi, Drug susceptibility testing of *Mycobacterium tuberculosis*: a neglected problem at the turn of the century, Int. J. Tuberc. Lung Dis. 3 (1999) 564–581.