Frequency and distribution of drug-resistant tuberculosis gene mutations in Africa between 2010 and 2020

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Background. Tuberculosis (TB) has been a cause of millions of deaths worldwide for many years. According to the World Health Organization Global Tuberculosis Report from 2017 to 2019, Africa was one of the regions most affected by TB. There have been many studies of gene mutations accounting for drug resistance; however, research studying the distribution and the frequency of these mutations is still limited.
Objective. To describe the frequency and distribution of TB gene mutations throughout the African continent during the period 2010 - 2020, and to forecast the spread of drug-resistant TB in Africa.
Methods. Information from 113 articles on the PubMed database published in English from 2010 to 2020 was reviewed to identify gene mutations. The countries in which the mutations were reported were also recorded, to study the distribution. Finally, statistical data and graphs were established to track the mutations.
Results. There were 22 gene mutations reported from 25 countries throughout Africa. Among them, mutations of the genes *rpoB* (*f*=20.48%), *katG* (*f* =15.66%), *gyrA* (*f* =13.25%) and *inhA* (*f* = 7.23%) were most commonly reported from countries in all regions of Africa: *rpoB* (*n*=17), *katG* (*n*=13), *gyrA* (*n*=11) and *inhA* (*n*=16). Southern Africa had the greatest variety of TB mutated genes, while East Africa had the highest number of countries reporting mutations.
Conclusion. Many TB gene mutations were recorded throughout the African continent. Some countries reported unique mutated genes. This reflects the diversity of drug resistance. Many countries also reported the same mutated genes, which will help to forecast the spread of drug-resistant TB during the coming years. Gene mutation was related to multidrug-resistant TB. Further study is necessary to provide an accurate evaluation of multidrug-resistant TB in Africa.

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Tuberculosis (TB) is an emerging public health problem worldwide, and especially in Africa.[1,2] Treating and preventing TB have long been two key missions of modern medicine and public health. TB, which accounts for approximately 1.5 million deaths annually,[3] creates not only a medical burden worldwide but also a financial burden, with a medical cost of USD616 billion between 2010 and 2015.[4] Unlike the polio pandemic, which was eradicated in 2018, TB is still a pandemic worldwide, despite efforts to develop antibiotics. Multidrug-resistant TB (MDR-TB) plays an important role in the ineffectiveness of many antibiotics. Many studies have shown that MDR-TB and rifampicin mono-resistant TB (RFM-TB) worsened the treatment outcomes of TB.[5] Another factor that affects the treatment outcomes of MDR-TB is HIV co-infection.[6,7] With the development of cross-country transportation, the spread of MDR-TB is predictable. Since 2010, various mutations accounting for drug resistance have been identified in research from Africa, and there is a need to study the distribution of these mutations throughout the continent to evaluate the spread of TB mutations, and to estimate the TB burden in the future.

Methods

The keywords 'Africa' and 'multidrug-resistant *Mycobacterium tuberculosis*' and 'mutations' were applied to identify all related articles on the PubMed database. Information from 113 free full-text articles published in English between 2010 and 2020 was reviewed to identify gene mutations in MDR-TB. The gene mutations discussed in the present study were obtained by DNA sequencing or polymerase chain reaction (PCR). The frequency and territory of each mutation were extracted for statistical analysis. Since the aim of this research was to track the distribution of specific mutations throughout Africa from 2010 to 2020 according to statistical data, the main focus was on gene mutations reported in specific

countries from 2010 to 2020. Articles that did not report on gene mutations were excluded. Studies conducted outside Africa are not included in the statistical data. During the study process, articles from outside the PubMed database were also reviewed. The data-collecting process is briefly summarised in Fig. 1.

Results

There were 22 gene mutations reported from 25 countries throughout Africa (Table 1). Mutation of gene *rpoB* ranked the highest ($f = 20.48\%$), being reported in 17 (68%) countries. Mutations of genes *katG*, *gyrA* and *inhA*, respectively ranked in the subsequent positions according to report frequency. In the period 2010 - 2020 in Africa, mutations of the genes *rpoB*, *katG*, *gyrA* and *inhA* played dominant roles in drug-resistant strains of TB. Although mutations in *rpoB* and *katG* have been reported to play important roles in rifampicin and isoniazid resistance, respectively, there is evidence for specific TB strains that possess mutations in both *rpoB* and *katG*.[8-13] We also found studies showing that a *gyrA* mutation is able to cause fluoroquinolone resistance,[13-17] and an *inhA* mutation is also known to be related to bedaquiline resistance.[18,19] Because these antibiotic-resistant mutations are reported in all regions of Africa, one should be cautious treating TB in Africa using rifampicin, isoniazid, fluoroquinolone and bedaquiline in 2020.

Gene mutations in TB have been reported across a vast area of Africa. The data obtained also show evidence for the appearance of mutation genotypes throughout Africa. Among the 25 countries mentioned in this study, there are 4 countries from north Africa (Tunisia, Sudan, Morocco and Egypt), 9 countries from east Africa (Zimbabwe, Zambia, Uganda, Tanzania, Somalia, Ethiopia, Rwanda, Mozambique and Kenya), 4 countries from central Africa (Gabon, Congo, Cameroon and Angola), 3 countries from southern Africa (Botswana, eSwatini and South Africa (SA)), 5 countries from west Africa (Nigeria, Liberia, Guinea, Guinea-Bissau and Ghana). The east Africa group has the highest number of countries reporting gene mutations, while SA is dominant in terms of the diversity of gene mutations (Fig. 2).

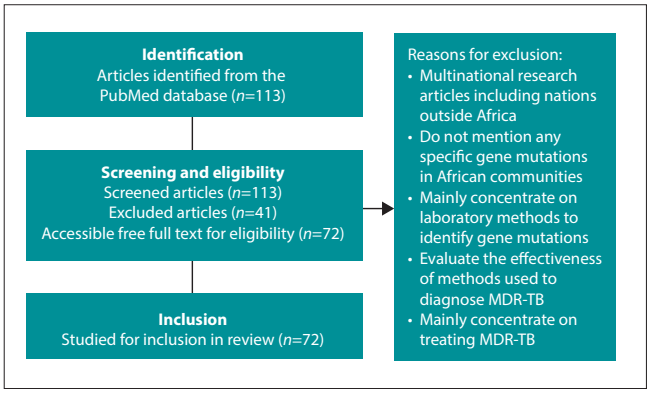

Fig. 1. Inclusion criteria.

Table 1. Frequency of specific gene mutations reported from 2010 to 2020 in Africa

Mutated gene	Country reported	Countries, *n*	Frequency (*f*), %
rpoB	Ethiopia, Zimbabwe, Kenya, Mozambique, Uganda, Rwanda, Tanzania, SA, Cameroon, Morocco, Guinea-Bissau, Sudan, Congo, Ghana, eSwatini, Angola, Gabon	17	20.48
katG	Zimbabwe, Kenya, Mozambique, Sudan, SA, Tanzania, Ethiopia, Cameroon, Morocco, Uganda, Congo, Guinea, Angola	13	15.66
gyrA	Mozambique, Morocco, Nigeria, SA, eSwatini, Somalia, Uganda, Botswana, Congo, Ghana, Tunisia	11	13.25
inhA	Sudan, SA, Ethiopia, Cameroon, Uganda, Angola	6	7.23
rrs	Kenya, Tanzania, SA, Botswana, Ghana	5	6.02
pncA	Tanzania, Tunisia, SA, Egypt, Zambia	5	6.02
gyrB	Tanzania, eSwatini, Somalia, Uganda, Congo	5	6.02
embB	SA, Tanzania, Ethiopia, Liberia	4	4.82
rpsL	Tanzania, SA, Congo	3	3.61
rpoC	SA, Uganda	2	2.41
sigA	SA	1	1.2
grcC1	SA	1	1.2
ubiA	SA	1	1.2
oxyR-ahpC	Tanzania	1	1.2
embC-embA	Tanzania	1	1.2
ethA/etaA	Uganda	1	1.2
GidB	SA	1	1.2
Eis	SA	1	1.2
tlyA	SA	1	1.2
ethA	Liberia	1	1.2
Gid	Liberia	1	1.2
mmpL3	SA	1	1.2
Total		83	100

SA = South Africa.

In the east African group, a study in Ethiopia revealed that, among infectious diseases, TB is ranked as the number-one killer among females (Table 2).[20,21]

Another study from Tanzania showed that the prevalence of smear-positive TB among women with a cough attending hospital was as high as 3.8%.[23] There is also a study reporting that Zimbabwe has among the highest estimated TB incidence per capita (603/100 000 population) in the world.[24]

Africa has always had a high burden of TB (Fig. 3).[25-27] In the case of SA, the country ranks among those with the highest MDR-TB burdens in the world, and the percentage of MDR-TB cases increased from 18.8% in 2011 to 23.9% in 2014.[28] Because almost all regions of Africa have now reported these gene mutations, screening TB patients using PCR or DNA sequencing has become necessary to choose the appropriate treatment, especially for MDR-TB patients.

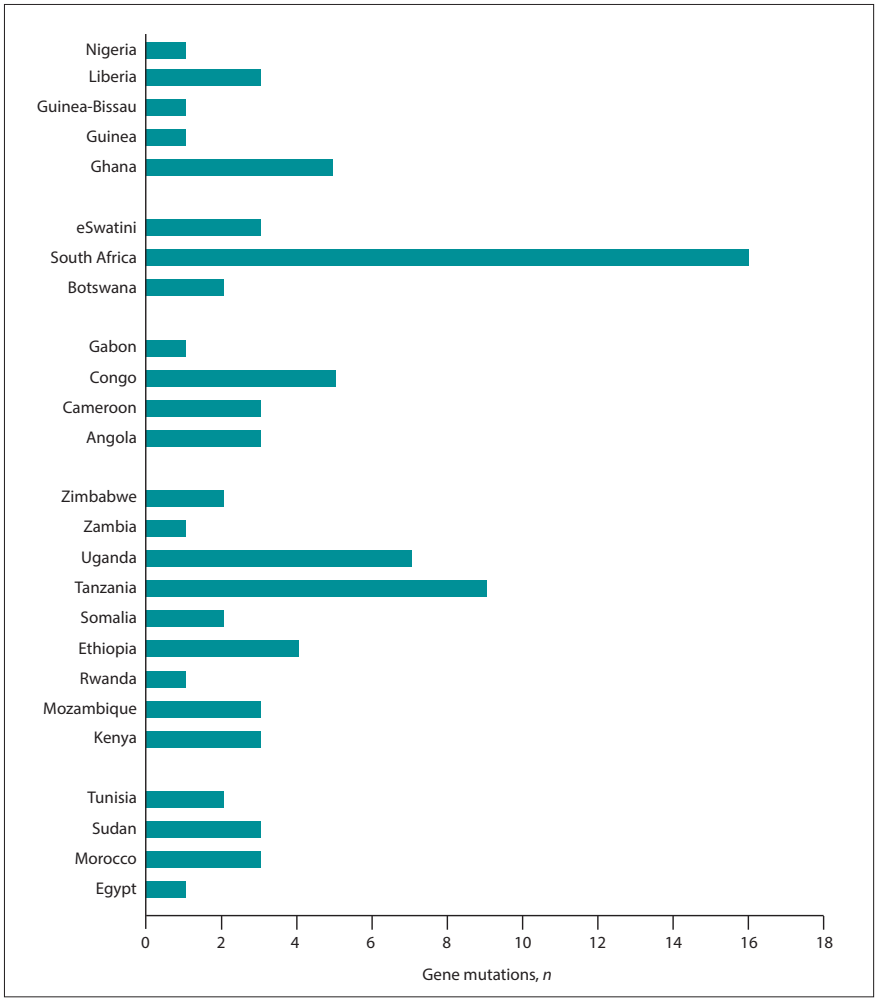

Fig. 2. Reported multidrug-resistant tuberculosis gene mutations in Africa, 2010 - 2020, by country.

Limitations

The database is limited as only 72 articles that were published from 2010 to 2020 were chosen, which is a short period for evaluating the TB situation. Information bias occurred because data from other countries were not reviewed. Secondly, using a limited number of countries to represent the whole African continent may result in bias. In the case of SA, for instance, its status as the country with the highest number of gene mutations in Africa is probably a result of the fact that the greatest amount of research has been conducted there.

Discussion

Various TB phenomena exist in different countries throughout Africa. Our study provides evidence that during the period 2010 - 2020, in Africa, mutations of the genes *rpoB*, *katG*, *gyrA* and *inhA* were the most common. Many other studies have shown that mutations in these genes are the main reason for the appearance of resistance to many antibiotics, including rifampicin, isoniazid, fluoroquinolone, bedaquiline and others.[11,13,14,29-34]

The distribution of gene mutations should also be noted. Table 1 shows that the most common gene mutations (mutations of *rpoB*, *katG*, *gyrA* and *inhA*) are reported in almost all regions of Africa, which provides

Table 2. Cause-specific mortality rate (per 1 000 person-years) among females in Kilite-Awlaelo health and demographic surveillance system, northern Ethiopia, 2010 - 2012[22,23]

		Total (105 793.9 person-years)		
Verbal autopsy (VA) diseases	VA code	*n* (*N*=398)	Rate	Rank
Infectious and parasitic disease	01	148	1.40	-
Tuberculosis	01.03	61	0.58	1
Acute lower respiratory infection	01.13	20	0.19	2
Intestinal infectious disease	01.01	17	0.16	3
Infectious disease, unspecified cause	01.99	16	0.15	4
Meningitis	01.11	14	0.13	5
HIV/AIDS	01.09	10	0.09	6
Malaria	01.10	8	0.08	7
Viral hepatitis	01.08	2	0.02	8

Table 3. Collaborative TB/HIV activities in 30 countries with high TB/HIV burden[26-28]

Year	WHO region	Total estimated TB cases, *n*	Estimated HIV-positive incident TB cases, *n*	Notified TB patients with known HIV status, *n* (%)
2016	Africa	1 303 483	764 000	1 062 438 (82)
	Global	6 624 523	1 030 000	3 595 018 (57)
2017	Africa	1 323 450	663 000	1 106 521 (86)
	Global	6 708 123	920 000	3 755 671 (60)
2018	Africa	1 402 743	615 000	1 175 391 (87)
	Global	7 253 116	862 000	4 4337 402 (64)

TB = tuberculosis; WHO = World Health Organization.

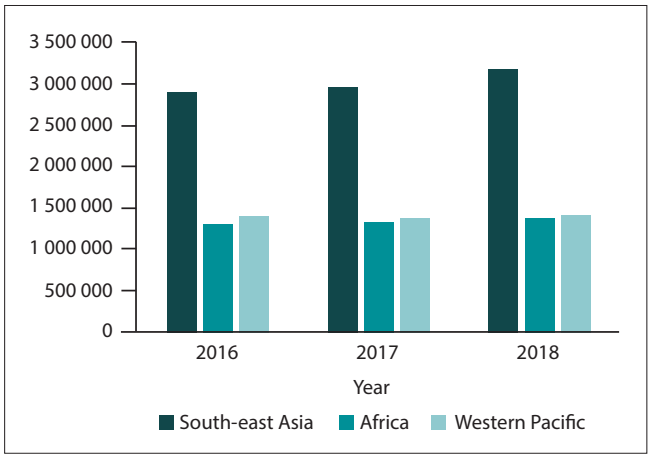

Fig. 3. Tuberculosis (TB) case numbers in Africa, south-east Asia and western Pacific (World Health Organization regions with the highest TB burdens), 2016 - 2018.[26-28]

evidence for the wide spread of rifampicin- and isoniazid-resistant TB throughout the entire continent. Our research also helps to clarify the spread of MDR-TB in Africa during the period 2010 - 2020. The research also shows the diversity of TB gene mutations in SA. According to the data we obtained, SA (16/22, 72.72%) and Tanzania (9/22, 40.1%) dominated in the number of TB gene mutations identified from 2010 to 2020. There were many mutations identified in SA that have not been reported in other countries in Africa. The high number of TB gene mutations in SA and Tanzania should be studied further.

Unlike the case of SA, the high number of TB gene mutations in Tanzania may be partly explained by reviewing antibiotic consumption in the country. According to the World Health Organization report on antibiotics consumption, 2016 - 2018, the defined daily dose per 1 000 inhabitants per day in Tanzania was 27.29, one of the highest antibiotic consumption rankings among countries that took part in the survey (ranked below Mongolia, Iran, Sudan and the Republic of Korea).[35] However, this explanation does not extend to the case of Sudan, where although the defined daily dose per 1 000 inhabitants per day was 35.29, only three types of gene mutation were identified in our study.

Another factor to which more attention should be paid is HIV co-infection. Africa is a region with a high prevalence of HIV, and the percentage of TB patients with HIV is also high compared with the TB mono-infection percentage (Table 3).

There is much research showing the relationship between HIV co-infection and the spread of MDR-TB. Because HIV co-infection affects the treatment outcomes of MDR-TB,[6,7,27] the only confirmed risk factors for MDR-TB are prior treatment for TB and refugee status.[36]

Conclusion

From 2010 to 2020, various mutations in the TB genome were recognised and confirmed across extensive areas of the African continent. In addition to well-known mutations that have been carefully studied for a long period, such as *rpoB*, *katG*, *gyrA*, etc., novel mutations with uncleared pathogenesis were also reported with smaller frequencies. Gene mutation or genotype modification play a key role in changing the phenotype of TB strains, and may strengthen the survivability of this bacteria in an antibiotic-containing environment. The confirmation of mutations, including novel mutations, raises awareness of the possibility of new antibiotic-resistant TB. From 2010 to 2020, east Africa was dominant in terms of the number of countries that reported mutations, while SA was significant for the diversity of confirmed mutated genes.

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