EJMA

DOI: 10.14744/ejma.2024.44127
EJMA 2024;4(4):184–190

Review

Balance Between Phenotyping and Genotyping Methods of N-Acetyltransferase 2 in Tunisia and Comparison with Other Countries: A Rapid Review

Yasmine Salem Mahjoubi, Daldoul Mouna, Mouna Ben Sassi, Ben Hammamia Syrine, El Jebari Hanene, Rim Charfi, Emna Gaies, Sameh Trabelsi

Department of Clinical Pharmacology, National center of Pharmacovigilance, Tunis, Tunisia

Abstract

Tunisia continues to experience tuberculosis at an intermediate level of endemicity. Isoniazid (INH) is a critical element in TB therapy. INH metabolism involves, mainly, the arylamine N-acetyltransferase 2 (NAT2) enzyme which is subject to genetic polymorphism.

The determination of NAT2 acetylation profiles is highly recommended for dose adjustment and predicting side effects in tuberculosis patients. Traditionally, acetylator status has been assessed through phenotyping tests. Recently, faster and simpler genetic methods have been proposed as an alternative for use in clinical practice.

This work provides a comprehensive review of main NAT2 phenotyping and genotyping results based on Tunisian experiences. In Tunisia, several studies have demonstrated the predominance of slow acetylator (SA) genotypes and phenotypes with the NAT2*5, being the most prelevant. When comparing acetylation phenotype with NAT2 genotype, a concordance value of 75% was observed.

The choice of method for determining NAT2 acetylation profile primarily depends on the laboratory's technical capabilities and expertise. Prospective clinical trials are essential to further evaluate the benefits of NAT2 genotyping specifically within the Tunisian population.

Keywords: Acetylation status, n-acetyltransferase, NAT2 gene, phenotyping, tunisian population, tuberculosis

Cite This Article: *Mahjoubi YS, Mouna D, Sassi MB, Syrine BH, Hanene EJ, Charfi R, et al. Balance Between Phenotyping and Genotyping Methods of N-Acetyltransferase 2 in Tunisia and Comparison with Other Countries: A Rapid Review. EJMA 2024;4(4):184–190.*

Tuberculosis (TB) caused by Mycobacterium tuberculosis, is a highly contagious airborne disease that primarily targets the lungs but has the potential to disseminate to other.[1] TB continues to pose a substantial global health threat, ranking as the second deadliest infectious disease following COVID-19.

As per the World Health Organization's data, in 2022, tuberculosis affected 10.6 million people globally, resulting in 1.3 million deaths.[2]

Tunisia continues to experience tuberculosis at an intermediate level of endemicity. In 2021, there were 2647 newly reported TB cases, resulting in an incidence rate of 22.46 new cases per 100,000 people.[3]

Isoniazid (INH) is a critical element in TB therapy for a long time, thanks to its strong bactericidal activity and minimal side effects.[4,5]

Address for correspondence: Yasmine Salem Mahjoubi, MD. Department of Clinical Pharmacology, National center of Pharmacovigilance, Tunis, Tunisia
Phone: 24161702 **E-mail:** m.yasmin7951@gmail.com

Submitted Date: November 22, 2024 **Revision Date:** November 22, 2024 **Accepted Date:** November 28, 2024 **Available Online Date:** December 23, 2024

INH is metabolized in the liver by the N-acetyltransferase2 (NAT2).[6]

This enzyme is subject to genetic polymorphism, and its activity is determined by a 2-allele autosomal gene located on chromosome 8.[6]

Based on NAT2 variability, individuals are categorized into three groups: rapid acetylators (RA), intermediate acetylators (IA), and slow acetylators (SA).

In fact, individuals who possess one or two high-activity alleles are classified as IA and RA respectively while SA carry two low-activity variants.[7]

To discern the acetylation profile, two distinct approaches emerge: genotyping and phenotyping, providing two complementary perspectives to characterize this enzymatic variability.

Significant inter-individual pharmacokinetic variability for INH has been observed, influenced by factors such as drug interactions, comorbidities, gastrointestinal abnormalities and genetic polymorphism. Therapeutic drug monitoring (TDM) is a method used to ensure adequate INH exposure by adjusting drug dose.[8]

INH treatment can lead to dose-dependent hepatotoxicity in SA and inadequate therapeutic response in RA.[6]

The determination of NAT2 acetylation profile has been highly recommended as a way to adjust doses and anticipate side effects in tuberculosis patients, ensuring maximum effectiveness and safety.[9,10]

Historically, distinguishing acetylator status relied on phenotyping tests which required administering a probe drug (caffeine, dapsone, and isoniazid).[11]

In more recent times, genetic techniques have emerged, allowing for the identification of distinct NAT2 alleles. Present choices for NAT2 genotyping encompass sequencing, multiple PCR assays, PCR along with hybridization probes and PCR coupled with restriction fragment length polymorphism analysis.[10]

The individualized therapeutic prescription based on genetic factors seems to be becoming a reality today, given the progress in understanding the functional consequences of NAT2 polymorphisms.

The choice between NAT2 phenotyping and genotyping methods has always sparked discussion.

This literature review aimed to achieve the following objectives:

- Provide an overview of the primary phenotyping and genotyping results related to NAT2 in Tunisia, drawing comparisons with findings from other populations.
- Determine the role of both phenotyping and genotyping approaches, exploring their interplay based on the Tunisian experience.
- Finally, recommend the preferred approach for application in both developing and developed countries.

Methods

We performed a comprehensive search across ScienceDirect, Google Scholar, PubMed and Cochrane Library utilizing the terms: "Acetylation profile, isoniazid, genotyping, phenotyping, N-acetyltransferase 2 enzyme, NAT2 gene, pharmacogenomics, correlation, Tunisian population." The search covered all publications available from the inception of each database up until February 29, 2024. Due to the limited number of relevant studies, we included all research that analyzed NAT2 variability in Tunisia for our review. Our review is designed to be narrative, providing a summary of the existing literature. The search process was carried out by two authors (YSM and MD), who also examined the references of the selected articles to identify additional relevant studies.

Results and Discussion

In total, we have identified 11 studies investigating NAT2 polymorphisms in Tunisia, comprising 4 studies focusing on phenotyping and 8 studies concentrating on genotyping.

Ragrading NAT2 phenotyping, in Tunisia, several investigations have explored the INH acetylation patterns within the adult population.

Almost all studies have demonstrated the predominance of a slow acetylation profile.

According to the study by Ben Rhouma M, conducted on 620 adult patients with pulmonary tuberculosis in 2009, 63.1% were slow acetylators (SA) and 36.9% were rapid acetylators (RA).12 These findings have been confirmed in 2021 by the study of Alshaikheid M that showed that SA was prevalent in around 58% of cases, with RA comprising the remaining 42%.[13]

However, Ben Tkhayat et al, revealed in a study involving 80 adult patients, a high frequency of RA phenotype found in 52.5% of cases (Table 1).[14]

The prevalence of the SA phenotype in Tunisia aligns with findings from previous studies in African and Asian Arab populations that reported an increased frequency of SA profiles[15,16] as well as in Europe.[17] In contrast, sub-Saharan African 18and Asian19 populations appear to exhibit a higher frequency of RA phenotypes (Table 1).

In clinical practice, the interest of identification of acetylation phenotype post-isoniazid administration is crucial for calculating the therapeutic range and determining the recommended dose.[6,11]

Table 1. Distribution of NAT2 acetylator status in Tunisian and other populations

Method	Year	Author	Geographical Origin	Slow	Intermediate	Rapid
Phenotyping	2021	ALsheikheid M et al	Middle coast of Tunisia	58	NS	42
	2017	Ben Tkhayat	North of Tunisia	47.5	NS	52.5
	2011	Jebabli et al	North of Tunisia	75	NS	25
	2009	Ben Rhouma et al	North of Tunisia	63.1	NS	36.9
	2004	Matar et al	Saudi Arabia	94.4	NS	5.6
	2002	Moussa et al	Morocco	61.8	NS	38.2
	2016	Toure et al	Senegal	44.3	NS	55.7
	1996	Kohno et al	Japan	13.1	NS	86.9
	1994	Carillo et al	Spain	65.4	NS	34.6
Genotyping	2017	Ben Fredj et al	Middle coast of Tunisia	59	38	3
	2016	Ouerhani et al	North of Tunisia	23.3	45	31.7
	2012	Ben Mahmoud et al	South of Tunisia	50	40.9	6.1
	2011	Ouerhani et al	North of Tunisia	31.1	45	23.9
	2009	Rouissi et al	North of Tunisia	28.8	39.2	32
	2009	Ouerhani et al	North of Tunisia	30.9	39.09	30
	2008	Khedhaier et al	Middle coast of Tunisia	49.7	44.3	5.9
	2019	Yuliwulandari et al	Indonesia	42	40	18
	2020	Mthiyane et al	South Africa	52.5	35.8	11.7
	2023	Sileshi et al	Ethiopia	74.2	22.4	3.3

NS: Not specified.

This approach is instrumental in tailoring the medication regimen to individual patient characteristics, thereby optimizing treatment outcomes.

In fact, the primary metabolic pathways of INH involve NAT2-catalyzed acetylation, forming acetylisoniazid (AcINH), and hydrolysis, which produces hydrazine (Hz) and isonicotinic acid (INA). AcINH can further hydrolyze to yield INA and acetylhydrazine (AcHz). It has been proposed that AcHz and Hz are responsible for INH hepatotoxicity.[20]

Clinical studies have consistently revealed a heightened risk of liver injury induced by INH, marked by increased plasma concentrations of INH and AcHz in slow acetylators compared to rapid acetylators. Additionally, NAT2 status influences hepatotoxicity in combination tuberculosis therapies involving INH and rifampicin. Rifampicin induces isoniazid hydrolases, intensifying Hz production, particularly in slow acetylators. Adjusting the INH dose in slow acetylators can reduce the incidence of hepatotoxicity.[21,22]

Having discussed the distribution of NAT2 phenotypes in Tunisia and underscored the significance of determining acetylation profiles for dose adjustments in clinical practice, now our focus turns to the emerging genotyping techniques.

The distribution of NAT2 polymorphism has been well studied in Tunisia and wordwide because variation in NAT2 activity has been not only linked to INH toxicity but also to the risk of variuos diseases and cancers due to its role in the metabolism of xenobiotics (Table 2).

This association has prompted an increased interest in understanding NAT2 genotypes, contributing to the elevated number of NAT2 genotyping-focused studies among our findings.

In Tunisia, NAT2 genotyping facilitated the identification of two rapid NAT2 allelic variants (NAT2*4 and NAT2*12) and four slow allelic variants (NAT2*5, NAT2*6, NAT2*7, and NAT2*14).[23]

Ouerhani et al have reported that the prevalence of NAT2 genotypes linked to slow, intermediate, and rapid phenitypes was approximated at 23.3%, 45%, and 31.7%, respectively, in the Tunisian.[24]

The incidence of SA phenotype in this study (North of Tunisia) was close to another study from the same geographical region 25 but appears to be less common compared to findings from similar research conducted elsewhere (on the middle coast26 and South 1 of Tunisia) (Table 2).

They have also found in the same study that the estimated frequencies of NAT2 alleles associated with SA, namely NAT2*5, NAT2*7, and NAT2*14, were approximately 50%, 3.75%, and 0.39%, respectively.[24]

The allelic frequencies of NAT2 variants were similar to thoses previously reported by Rouissi and Ben Fredj studies. However, the NAT2*14 was absent in these two latter studies.

Table 2. Distribution of NAT2 alleles in Tunisian and other populations

Year	Author	Geographical Origin	Slow				Rapid	
			NAT2*5	NAT2*7	NAT2*14	NAT2*6	NAT2*4	NAT2*12
2017	Ben Fredj et al	Midddle coast of Tunisia	44.4	3.5	0	30.3	16.2	5.6
2016	Ouerhani et al	North of tunisia	50	3.75	0.39	-	-	-
2011	Jebabli et al	North of Tunisia	34	1	3	20	24.5	13.5
2009	Rouissi et al	North of Tunisia	43.6	2.8	0	2	51.6	-
2023	Sileshi et al	Ethiopia	47.1	5.4	1.7	31.3	14.6	0
2023	Lopes et al	Brazil	36	-	-	18.2	20.4	-
2020	Thuli Mthiyane et al	South Africa	70.4				0.4	17.9
2019	Yuliwulandari et al	Indonesia	5	15	0	42	33	3
2014	Guaoua et al	Morroco	53	2	4	25	-	-
2011	Djordjevic et al	Serbia	38.2	-	-	26	24.4	-

Upon analyzing the distribution of NAT2 variants based on geographical regions, notable discrepancies in the frequency of SA emerged among the North, Central, and Southern parts of Tunisia. The highest prevalence was noted in populations residing in the North of Tunisia, reaching 60.1%.

Ethnic origin examination revealed no significant variation in the distribution of NAT2 phenotypes between Arab, Cosmopolitan, and Berber populations.[24]

When combining both factors, studies have shown that different ethnic origins from the same geographical origin appears to be genetically similar which indicates that genetic frequency is primarily influenced by geographical origin.[24]

Regarding the comparison of the distribution of NAT2 variants among the different geographical origins in Tunisia, almost all genotyping studies have been conducted in the northern part of the country, except for a single study by Ben Fredj et al. that was carried out in the middle coast of Tunisia. This particular study have shown a similar distribution of slow acetylator variants between the middle coast and the northern regions. In contrast, the NAT2*4 allele, associated with rapid acetylation, is more predominant in the North (51.6%) than in the middle coast (16.2%).[25,23]

This distribution of NAT2 alleles in our tunisian population are close to those reported in Morrocon,[27] African,[28] European[29] and American populations.[30] Contrastingly, NAT2*6 emerges as the predominant allele among Asian populations while NAT2*5 exhibits lower representation.[31]

In 2021, a Tunisian study have investigated the function of acetylator phenotype in adjusting INH dosage and proposed a population pharmacokinetic model. This method enabled the calculation of an individualized INH dose linked to achieving ideal therapeutic concentration 3 hours post-drug intake (C3). The study findings indicated that a daily dose of 450 mg in the RA/IA group and 225 mg in the SA group led to attaining a C3 of 1.5 mg/mL.[9]

These findings were close to a study in Germany which indicated that the INH dosage could be tailored to 150, 300 and 450mg/day, for SA, IA and RA groups, respectively to achieve comparable INH levels, assuming an average body weight (approximately 60 kg). (INH pharmacokinetics were not influenced by weight among Tunisian patients).[32]

However, these doses were found to be lower than those recommended by a Chinese study; dosages of around 300 500 and 800mg/day might be more suitable for slow, intermediate and rapid acetylators, respectively.[33]

Another study conducted in Mexico revealed that administering 225 mg of INH to patients weighing up to 50 kg and 300 mg to those weighing over 50 kg resulted in achieving a median 2 hours post-dose concentration (C2) of 6.78 mg/L in slow acetylators and 2.55 mg/L in rapid acetylators. (the recommended optimal C2 is 4.2±2.0 µg/ml).[34,35]

In 2012, a team of researchers from southern Tunisia conducted a study investigating the correlation between NAT2 gene variants and the occurrence of liver injury associated with INH, revealing a clear association between NAT2 genotype and the development of INH-induced hepatotoxicity.[1]

Specifically, patients with SA genotype exhibited a high incidence of serum aminotransferase elevation (78.60%) while RA genotype showed no abnormalities in liver function tests. Additionally, SA patients had threefold increase risk to experience liver enzyme elevation compared to IA patients.

In line with these findings, a Japanese study indicated that SA genotype patients were more prone to isoniazid induced hepatitis (83.3%), low incidence of aminotransferase elevations among IA genotype individuals (2.4%), and no instances in the RA genotype group.[36]

Finally, researchers from Northern Tunisia conducted a comparison between acetylation phenotype and NAT2 genotype.

The distribution of phenotypes within each genotype was examined in three patient groups classified based on their genotypes. The findings indicate that in group A, consisting of patients with genotypic slow acetylation, only one exhibited a phenotypically fast acetylation. In group B, encompassing heterozygotes with one wild-type and one substituted allele, 51.28% displayed a phenotypically fast acetylation, while 48.72% exhibited slow acetylation. Lastly, group C included five patients native allele (NAT2*4), with only one showing a phenotypically fast acetylation. The overall genotype-phenotype concordance rate for all NAT2 variations was 75%.[11]

In literature, there have been varied reports on the alignment of NAT2 genotype with acetylation phenotype.

While some studies have strongly correlated the NAT2 phenotype with the genotype beneath it, as demonstrated in several instances,[37-39] others have shown a lack of correlation or poor alignment between the NAT2 genotype and INH phenotype.[28,40]

Possible hypothesis have been suggested to justify the variability in the results of concordance between different populations.

Using NAT2 gene sequencing alongside caffeine and dapsone to determine acetylation phenotype, studies revealed discordance rates of 86% and 18% between the acetylator genotype and phenotype, respectively.[37]

In the Tunisian study by Jebabli et al, the acetylation phenotype was ascertained by calculating the acetylation index three hours after isoniazid administration, following the Vivien method. NAT2 genotyping was conducted using PCR amplification, leading to a moderate agreement between genotype and phenotype. The findings of this study indicate that the acetylation test has a high sensitivity of 98.2% in identifying slow acetylators. Moreover, a robust negative predictive value of 96% was noted.[11]

To conclude, the activity of an individual's NAT2 can be determined through phenotyping methods or predicted through genotyping methods.

Phenotyping and/or genotyping NAT2 could be useful either to alert to the need for close monitoring of liver function in slow acetylators or to recommend standard dosages for each acetylator type. However, broader clinical studies are still required to validate this approach.

Phenotypic determination of NAT2, on one hand, has drawbacks related to the technique itself, as its implementation is delicate. On the other hand, NAT2 activity can vary based on factors such as ethnic origin or concurrent medications.

These limitations justify the use of genotyping as multiple studies confirm that NAT2 activity polymorphism results from gene mutations with variable frequencies among ethnicities.[41]

However, lack of concordance between NAT2 phenotype and genotype is not uncommon, reflecting the respective limitations of both techniques and the still incomplete understanding of NAT2 enzyme.

The choice of the method used to determine the NAT2 acetylation status is mainly based on the technical capabilities available in the laboratory and its expertise in the field.

Research in pharmacogenetics not only holds medical significance but also economic value. Prospective knowledge of patients at risk of developing adverse effects should help better manage healthcare costs by avoiding the prescription of lengthy, costly, or potentially unnecessary treatments for a certain population.

While our understanding of the genetic mechanisms influencing inter-individual variations in NAT2 activity has significantly advanced over the past two decades, the practical application of this knowledge in clinical settings remains restricted. Worldwide, NAT2 genotyping is not yet routinely conducted.

In developed countries with low TB endemicity and a limited number of tuberculosis patients, there is minimal interest in widespread NAT2 genotyping across the population.

Conversely, in developing nations where tuberculosis is either moderately or highly prevalent, large-scale NAT2 genotyping could prove valuable for predicting rapid acetylators at risk of developing drug resistance and subsequent treatment failure and mimizing toxicity in slow acetylators.

However, the implementation of this approach is likely challenged by its feasibility in resource-limited countries.

Conclusion

In summary, our study revealed several key findings in the Tunisian population. Firstly, we found a significant predominance of the SA phenotypes. Secondly, we noted a moderate agreement between NAT2 phenotype and genotype. Lastly, across various regions of Tunisia, there were notable differences in NAT2 acetylation phenotypes, but similar genotype frequencies.

This study underscores the significance of striking a balance between phenotyping and genotyping approaches in investigating the NAT2 gene polymorphism in Tunisia. By integrating both methodologies, valuable insights into individual acetylator profile can be obtained, thereby facilitating the optimization of INH therapy.

Furthermore, prospective clinical trials are imperative to further assess the benefits of NAT2 genotyping specifically within the Tunisian population. This holistic approach not only enhances our understanding of NAT2 variability but also paves the way for tailored therapeutic interventions, ultimately improving patient outcomes.

Disclosures

Peer-review: Externally peer-reviewed.

Conflict of Interest: None declared.

Authorship Contributions: Concept – Y.S.M.; Design – Y.S.M., M.D.; Supervision – M.B.S.; Materials – Y.S.M., M.D.; Data collection and/or processing – Y.S.M., H.E.J., Analysis and/or interpretation – S.B.H., R.C., Literature search – Y.S.M., Writing – Y.S.M. Critical review – E.G., S.T.

References

1. Ben Mahmoud L, Ghozzi H, Kamoun A, Hakim A, Hachicha H, Hammami S; et al. Polymorphism of the N-Acetyltransferase 2 gene as a susceptibility risk factor for antituberculosis drug-induced hepatotoxicity in Tunisian patients with tuberculosis. Pathologie Biologie 2012;60(5):324–330.
2. WHO. Tuberculosis. Available at: https://www.who.int/news-room/fact-sheets/detail/tuberculosis. Accessed Feb 2, 2024.
3. Mansouri A, Bouguerra H, Belkahla M, Gara A. Epidemiological profile of tuberculosis in Tunisia. Eur J Public Health 2023;33: ckad160.636.
4. Heysell S, Mpgama S. Re-evaluating the role of isoniazid in treatment of pulmonary tuberculosis. Lancet Microbe 2020;1:E49-E50.
5. Li Y, Shi J, Song W, Shao Y, Zhu L, Chen C. A retrospective cohort study on the treatment outcomes and genotyping of isoniazid-resistant tuberculosis patients in Eastern China. J Glob Antimicrob Resist 2022;30:335-339.
6. Bouazzi OE. Pharmacological therapeutic monitoring of isoniazid. Eur Sci J [Article in French] 2020;16(3):401.
7. Kinzig-Schippers M; Tomalik-Scharte D, Jetter A, Scheidel B, Jakob V; Rodamer M, et al. Should we use n-acetyltransferase type 2 genotyping to personalize isoniazid doses? Antimicrob Agents Chemother 2005;49(5):1733–1738.
8. Akkerman OW, Dijkwel RDC, Kerstjens HAM, van der Werf TS, Srivastava S, Sturkenboom MGG, et al. Isoniazid and rifampicin exposure during treatment in drug-susceptible TB. Int J Tuberc Lung Dis 2023;27(10):772–777.
9. Fredj NB, Romdhane HB, Woillard JB, Chickaid M, Fadhel NB, Chadly Z, et al. Population pharmacokinetic model of isoniazid in patients with tuberculosis in Tunisia. Int J Infect Dis 2021;104:562–567.
10. Brans R, Laizane D, Khan A, Blömeke B. N-acetyltransferase 2 genotyping: An accurate and feasible approach for simultaneous detection of the most common NAT2 alleles. Clin Chem 2004;50(7):1264–1266.
11. Jebabli N, Gaies E, Kharrat M, Njima BB, Klouz A, Chaabouni H, et al. N-acetyltransferase 2 (NAT2) in Tunisian population: Correlation between acetylation phenotype and genotype: N-acetyltransferase 2 polymorphism. Pak J Sci Ind Res 2010;53(4):205–211.
12. Ben Rhouma Nadia M, Anissa Z, Jouda C, Basma O, Riadh D, Hedi OM, et al. Acetylator profile of a population of Tunisian tuberculosis patients: About 620 cases. Tunis Med [Article in French] 2005;83(7):385–389.
13. Alshaikheid M, Romdhane HB, Fredj NB, Fadhel NB, Aouam A, Chadli Z, et al. Isoniazid therapeutic drug monitoring in Tunisian patients with tuberculosis. J Clin Pharmacol 2021;61(7):972–978.
14. Ben Tkhayat A, Ben Saad S, Daghfous H, Tritar F. Acetylation phenotype in tuberculosis patients treated with the combination of fixed drugs. Rev Mal Respir [Article in French] 2017;34:A222.
15. Aït Moussa L, Khassouani CE, Hüe B, Jana M, Bégaud B, Soulaymani R. Determination of the acetylator phenotype in Moroccan tuberculosis patients using isoniazid as metabolic probe. Int J Clin Pharmacol Ther 2002;40(12):548-53.
16. Matar KM, Mayet AY, Ayoola EA, Bawazir SA, Al-Faleh FZ, Al-Wazzan A, et al. Isoniazid acetylation phenotyping in Saudi Arabs. J Clin Pharm Ther 2004;29(5):443–447.
17. Carrillo JA, Benítez J. Caffeine metabolism in a healthy Spanish population: N-acetylator phenotype and oxidation pathways. Clin Pharmacol Ther 1994;55(3):293-304.
18. Toure A, Cabral M, Niang A, Diop C, Garat A, Humbert L, et al. Prevention of isoniazid toxicity by NAT2 genotyping in Senegalese tuberculosis patients. Toxicol Rep 2016;3:826–831.
19. Kohno H, Kubo H, Takada A, Mori M, Arias TD. Isoniazid acetylation phenotyping in the Japanese: The molar metabolic ratio INH/AcINH. Am J Ther 1996;3(1):74–78.
20. Metushi I, Uetrecht J, Phillips E. Mechanism of isoniazid-induced hepatotoxicity: Then and now. Br J Clin Pharmacol 2016;81(6):1030–1036.
21. Wang P, Pradhan K, Zhong X, Ma X. Isoniazid metabolism and hepatotoxicity. Acta Pharm Sin B 2016;6(5):384–392.
22. Ramappa V, Aithal GP. Hepatotoxicity related to anti-tuberculosis drugs: Mechanisms and management. J Clin Exp Hepatol 2013;3(1):37–49.
23. Ben Fredj N, Gam R, Kerkni E, Chaabane A, Chadly Z, Boughattas N, et al. Risk factors of isoniazid-induced hepatotoxicity in Tunisian tuberculosis patients. Pharmacogenomics J 2017;17(4):372–377.
24. Ouerhani S, Ben Bahria I, Rouissi K, Cherni L. Distribution of xenobiotic metabolising enzyme genotypes in different Tunisian populations. Ann Hum Biol 2017;44(4):366–372.
25. Rouissi K, Ouerhani S, Marrakchi R, Ben Slama MR, Sfaxi M, Ayed M, et al. Combined effect of smoking and inherited

polymorphisms in arylamine N-acetyltransferase 2, glutathione S-transferases M1 and T1 on bladder cancer in a Tunisian population. Cancer Genet Cytogenet 2009;190(2):101–107.
26. Khedhaier A, Hassen E, Bouaouina N, Gabbouj S, Ahmed SB, Chouchane L. Implication of xenobiotic metabolizing enzyme gene (CYP2E1, CYP2C19, CYP2D6, mEH and NAT2) polymorphisms in breast carcinoma. BMC Cancer 2008;8:109.
27. Guaoua S, Ratbi I, Laarabi FZ, Elalaoui SC, Jaouad IC, Barkat A, et al. Distribution of allelic and genotypic frequencies of NAT2 and CYP2E1 variants in Moroccan population. BMC Genet 2014;15(1):156.
28. Mthiyane T, Millard J, Adamson J, Balakrishna Y, Connolly C, Owen A, et al. N-Acetyltransferase 2 genotypes among Zulu-speaking South Africans and isoniazid and N-Acetyl-Isoniazid pharmacokinetics during antituberculosis treatment. Antimicrob Agents Chemother 2020;24;64(4):e02376-19.
29. Djordjevic N, Carrillo JA, Ueda N, Gervasini G, Fukasawa T, Suda A, et al. N-Acetyltransferase-2 (NAT2) gene polymorphisms and enzyme activity in Serbs: Unprecedented high prevalence of rapid acetylators in a White population. J Clin Pharmacol 2011;51(7):994-1003.
30. Lopes MQP, Teixeira RLF, Cabello PH, Nery JAC, Sales AM, Nahn JR, et al. Human N-acetyltransferase 2 (NAT2) gene variability in Brazilian populations from different geographical areas. Front Pharmacol 2023;14:1278720.
31. Yuliwulandari R, Susilowati RW, Razari I, Viyati K, Umniyati H, Prayuni K. N-acetyltransferase 2 polymorphism and acetylation profiles in Buginese ethnics of Indonesia. Ann Hum Genet 2019;83(6):465-471.
32. Kinzig-Schippers M, Tomalik-Scharte D, Jetter A, Scheidel B, Jakob V, Rodamer M, et al. Should we use N-acetyltransferase type 2 genotyping to personalize isoniazid doses? Antimicrob Agents Chemother 2005;49(5):1733-8.
33. Jing W, Zong Z, Tang B, Wang J, Zhang T, Wen S, et al. Population pharmacokinetic analysis of isoniazid among pulmonary tuberculosis patients from China. Antimicrob Agents Chemother 2020;64(3):e01736-19.
34. Huerta-García AP, Medellín-Garibay SE, Ortiz-Álvarez A, Magaña-Aquino M, Rodríguez-Pinal CJ, Portales-Pérez DP, et al. Population pharmacokinetics of isoniazid and dose recommendations in Mexican patients with tuberculosis. Int J Clin Pharm 2020;42(4):1217–1226.
35. Bargaje M, Bharaswadkar S, Lohidasan S, Panda BK. Plasma drug concentrations of 4-drug fixed-dose combination regimen and its efficacy for treatment of pulmonary tuberculosis under National Tuberculosis Elimination Programme: A prospective pilot study. Indian J Tuberc 2022;69(3):311–319.
36. Hiratsuka M, Kishikawa Y, Takekuma Y, Matsuura M, Narahara K, Inoue T, et al. Genotyping of the N-acetyltransferase 2 polymorphism in the prediction of adverse drug reactions to isoniazid in Japanese patients. Drug Metab Pharmacokinet 2002;17(4):357–362.
37. Sileshi T, Telele NF, Burkley V, et al. Correlation of N-acetyltransferase 2 genotype and acetylation status with plasma isoniazid concentration and its metabolic ratio in Ethiopian tuberculosis patients. Sci Rep 2023;13:11438.
38. Zhao B, Seow A, Lee EJ, Lee HP. Correlation between acetylation phenotype and genotype in Chinese women. Eur J Clin Pharmacol 2000;56(9–10):689–692.
39. Zabost A, Brzezińska S, Kozińska M, Błachnio M, Jagodziński J, Zwolska Z, et al. Correlation of N-acetyltransferase 2 genotype with isoniazid acetylation in Polish tuberculosis patients. Biomed Res Int 2013:853602.
40. Birch Kristensen E, Yakimov V, Bjorn-Mortensen K, Soborg B, Koch A, Andersson M, et al. Study of correlation between the NAT2 phenotype and genotype status among Greenlandic Inuit. EXCLI J 2018;17:1043-1053.
41. Touré A. Etude du polymorphisme génétique de la N-Acétyltransférase de type 2 dans la population sénégalaise: prévention de la toxicité et de l'échec thérapeutique de l'isoniazide dans la prise en charge de la tuberculose. Médecine humaine et pathologie [Thesis]. Université du Droit et de la Santé.