

Original Article

Interferon gamma release assay post BCG among newborns and family members living in a crowded area in Jakarta, Indonesia

Francisca Srioetami Tanoerahardjo¹, Indri Rooslamati¹, Natalie Kipuw¹, Hadiyanto², Soegianto Ali², Edhyana Sahiratmadja³

¹ National Institute of Health Research and Development, Ministry of Health Republic of Indonesia

² School of Medicine and Health Sciences, Atma Jaya Catholic University of Indonesia, Jakarta, Indonesia

³ Faculty of Medicine, Universitas Padjadjaran, Bandung, Indonesia

Abstract

Introduction: BCG vaccine is a mandatory for newborn in Indonesia, an endemic tuberculosis (TB) country that ranks second worldwide. A close contact with untreated active pulmonary TB individuals in a crowded area may result in TB disease or otherwise develop a latent TB infection (LTBI), as shown by positive result of interferon gamma release assay (IGRA).

Objective: To explore LTBI among newborns and their family members living in a crowded area in Jakarta, Indonesia.

Methodology: A prospective analytical study was conducted among newborns between October 2016 and March 2017. IGRA was examined before BCG vaccination and after 12 weeks. In parallel, TB active case finding was performed among family members of the newborns.

Results: Out of 135 newborns, only 117 (86.7%) came for BCG vaccination. Of 346 family members screened, 8 (2.3%) were detected as untreated active pulmonary TB, confirmed by positive sputum and/or MTB culture. Family members living in the same house with active TB individuals ($p = 0.011$, OR 2.69) as well as being males ($p = 0.025$, OR 1.68) had a significant higher risk of having a positive IGRA.

Conclusions: Untreated pulmonary TB infection in a crowded area infects the surrounding neighbors, resulting in latent TB infection. An active program for detecting pulmonary TB cases and preventive measures need to be taken seriously to contain the potential spread of the infection.

Key words: IGRA; BCG; newborn; tuberculosis; Indonesia.

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Introduction

Tuberculosis (TB) is an airborne disease, caused by *Mycobacterium tuberculosis* (MTB) [1]. In a crowded area, an active TB patient is a potential source of MTB transmission that spreads the infection to the surrounding people by coughing and sneezing [2]. Moreover, young children living closely with MTB infected parents or family members are at high risk of contracting the disease. Furthermore, the immunity of newborns is still immature, making them more vulnerable, and resulting in a latent TB infection (LTBI) that may progress to active TB disease [3]. They may later serve as a reservoir for future transmission [4]. Interestingly, active pulmonary TB individuals are sometimes afraid to report to the health facilities due to lack of knowledge about the disease and stigma in the community and thus may further infect the community [5]. Therefore, education and awareness as well as an active TB case finding program is necessary.

Recently, interferon gamma release assays (IGRA) has been proposed to detect LTBI next to tuberculin

skin test (TST). The latent infection of MTB is defined by a positive result of IGRA or TST in the absence of active TB [3]. Early diagnosis and treatment of LTBI are therefore considered to be the most effective strategy for reducing the incidence of TB in the population. However, study in a high TB-burden setting comparing TST and IGRA in diagnosing LTBI has shown that neither was able to predict the active TB during the follow up [6]. Therefore, further exploration is required.

To prevent the disease, BCG vaccine is given to children in TB endemic countries, including Indonesia. The BCG vaccine is mandatory based on the National TB Program (NTP) in Indonesia, administered to newborns up to 12 weeks old. Interestingly, BCG efficacy studies have shown a range of 0 to 80% effectiveness [7]. Furthermore, despite the high coverage of BCG vaccination nationwide and the direct observed therapy system (DOTS) program, Indonesia still ranks as one of highest TB burden countries [8]. This study aimed to explore the IGRA test among

newborns living in a crowded area of Jakarta, Indonesia. In parallel, active case finding was conducted among family members living in the same house with the newborns.

Methodology

Study design

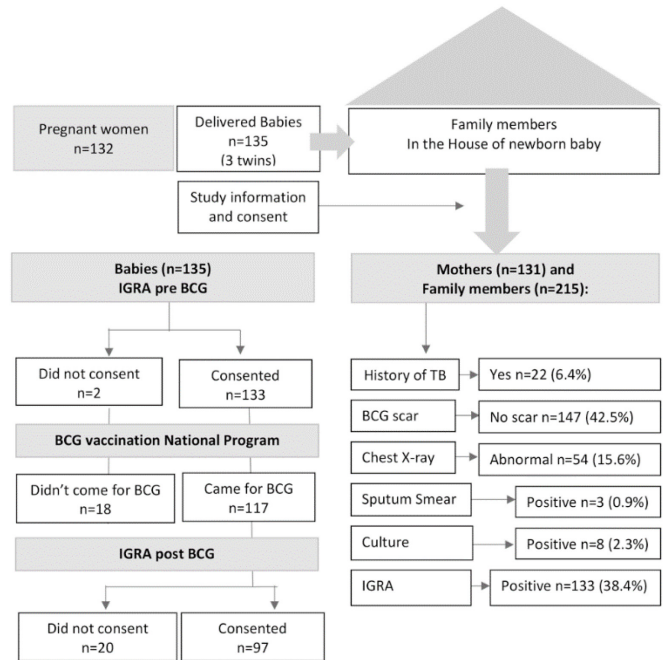
A prospective analytic study was conducted to explore the IGRA test among newborns living in a crowded district of Penjaringan and Tambora, Jakarta. The IGRA was performed before BCG vaccination and after 12 weeks. Furthermore, a cross-sectional analytic study was conducted to examine IGRA among the family members living in the same house of the newborns.

Study participants

Between October 2016 and March 2017, pregnant women (n = 132) who were at their last month of antenatal care were approached and informed about the study. They were further informed about the mandatory BCG vaccination for newborns up to 3 months old according to the National TB Program. Before the BCG vaccination, mothers were consented and peripheral blood was drawn using a wing needle into a heparinized tube for the IGRA test (QuantiFeron, NZ). With the mother's consent, the IGRA test was repeated after 12 weeks of BCG vaccination.

In a parallel study, during the home visit of the newborns, active TB case detection was directed to all individuals living in the same house with the babies, including history taking and chest X-ray. Individuals with abnormal chest X-ray were further examined for acid fast bacilli in their sputum and then cultured in Löwenstein–Jensen (LJ) medium. In addition, family members older than 15 years old were tested for IGRA, including the mothers of newborns. The research flow is shown in Figure 1.

Figure 1. Research Flow on Interferon Gamma Release Assay (IGRA) study among newborn babies before and after BCG vaccination and family members who lived in the same house in a crowded area in Jakarta, Indonesia.



Ethical Clearance

The study protocol was granted ethical clearance by the Ethical Committee of National Institute of Health Research and Development, Ministry of Health Republic of Indonesia no. LB.02.01/5.2/KE.397/2015.

Statistical Analyses

The frequency of the IGRA test before and after BCG vaccination was described in percentage. Furthermore, chi-square was adopted for the IGRA test among family members, living with or without active TB individuals and stratified by gender. A *p* value < 0.05 was considered statistically significant.

Table 1. Characteristic of the mothers of newborns from an overcrowded area who came for BCG vaccination of their newborns and those who did not come for vaccination.

	BCG vaccination		<i>p</i> value
	Vaccinated their newborn (N =117)	Did not show up for vaccination (N = 18)	
Age (years)			
median (min-max)	29 (17-40)	26 (18-33)	0.034*
Education^a (N)			
≤ 6 years education	31	-	0.193
6-9 years	40	8	
9-12 years	46	10	
Place of delivery (N)			
Hospital	46	3	0.063
Primary Health	71	15	

**p* was statistically significant (Chi-square); ^a Basic education ≤ 9 years vs. education > 9 years; BCG: Bacillus Calmette-Guerin.

Ethics approval and consent to participate

Ethical approval for this study was granted by the Ethical Committee of Ministry of Health Republic Indonesia LB.02.01/5.2/KE.397/2015.

Results

BCG vaccination among newborns

A total of 132 pregnant women that gave birth to 135 babies were identified, of whom 3 mothers were delivered of twins, consisting of boys 64 (47.4%) and girls 71 (52.63%) with a birth weight of 3050 gram (median; range 1700-4100 gram). However, only 117 of 135 (86.6%) newborns came for BCG vaccination (Figure 1). Some mothers (n = 18) refused to get their babies vaccinated.

At the time of BCG vaccination, the babies were weighed with median weight of 4,040 grams (range 2,350-6,100 grams) and were with median age of 39 days old (range 11-82 days). The characteristic of the mothers of newborns was analyzed based on BCG vaccination attendance (Table 1). The result showed that the older age mothers were significantly more likely to allow BCG vaccinations for the newborns than younger mothers ($p < 0.034$). Interestingly, the longer education of the mothers between mothers who came and did not come for the BCG vaccination showed no significant difference ($p = 0.193$). Furthermore, the location of delivery, either the hospital or the primary health care, was not significantly different ($p = 0.063$).

IGRA test result among newborn babies

From a total of 135 newborn babies, 133 had IGRA tests before BCG vaccination, however, only 97 babies came for IGRA after BCG. Furthermore, preBCG IGRA showed positive in 3 of 133 babies, however, these results turned to be negative 12 weeks after BCG,

Table 2. The IGRA test result among newborns living in an overcrowded area before and after BCG vaccination.

BCG vaccination		n (%)
Before	After	
No consent	-	2 (1.5)
Negative	n.d.	36 (26.7)
Negative	Negative	93 (68.9)
Negative	Positive	1 (0.7)
Positive	Negative	3 (2.2)

BCG: Bacillus Calmette-Guerin; IGRA: Interferon Gamma Release Assay; n.d.: not determined.

and they did not live with active TB family members. Interestingly, one baby who did not live with active TB family members turned out to be IGRA positive (Table 2).

IGRA results among family members

The active TB case finding showed that 2.3% (8 of 346) family members had active TB, confirmed by positive sputum examination and/or MTB culture. These individuals were not on TB therapy. Special attention was given to the 7 babies who had lived with active pulmonary TB parents or other family members (Table 3). However, 5 out of 7 babies were still IGRA negative after BCG; whereas others did not come for the IGRA test.

Furthermore, there was a significant statistical difference ($p = 0.011$) in the IGRA test result of family members living with active TB individuals compared to those without active TB. There was 2.69 times higher chance of positive IGRA in the individual living with active TB (95% CI, 1.22-5.94) (Table 4). Interestingly, there was a statistical difference in gender ($p = 0.025$); whereas males had a higher chance of having positive IGRA compared to females (OR 1.68; 95% CI, 1.07 – 2.64).

Table 3. The IGRA test of newborn babies who live with untreated pulmonary active TB family members.

ID no.	ID no. Baby	IGRA test		ID no. TB active	Relation to baby	IGRA Test	Other family members With age and IGRA result	IGRA Result in house	
		Before BCG	After BCG					Positive	Negative
123	123003	Negative	n.d.*	123001 123002	Father (39) Mother (33)	Positive Positive	-	2	-
444	444003	Negative	n.d.*	444005	Grand Ma (42)	Positive	Mother (17) (+)	2	-
121	121003	Negative	Negative	121006	Cousin (19)	Positive	Mother (36) (-), Aunt (40) (-)	1	2
142	142003	Negative	Negative	142001	Father (39)	Positive	Mother (36) (+), Aunt (36), (+), Grand Pa (75) (+), Grand Ma (73) (+), Uncle (32) (-), Sister (15) (-)	5	2
148	148003	Negative	Negative	148001	Father (24)	Positive	Grand Pa (64) (+), Grand Ma (45) (+), Mother (20) (-), Grand Pa (48) (-), Grand Ma (64) (-)	3	3
411	411003	Negative	Negative	411004	Grand Ma (52)	Positive	Father (36) (+), Grand Pa (55) (+), Mother (28) (-), Sister (5) (-)	3	2
117	117003	Negative	Negative	117004	Grand Ma (73)	Negative	Father (39) (+), Mother (32) (+)	2	1

n.d.*: not determined (did not come for BCG vaccination); BCG: Bacillus Calmette-Guerin; IGRA: Interferon Gamma Release Assay; TB: tuberculosis.

Discussion

Indonesia is a TB endemic country and the BCG vaccine has been prioritized to prevent TB disease. It is a mandatory according to the National TB Program, given to newborn babies up to 12 weeks. However, only 86.6% newborn babies were brought for BCG vaccination in our study. Despite the home visit from the cadre or health employee of the primary health care, some mothers refused to bring their baby for routine vaccination. Notably, older mothers were significantly more likely to visit the primary health care center for BCG vaccination as compared to young mothers ($p = 0.034$). In addition, a study in Malaysia has shown that several factors may play a role, including the assumption that vaccines have no effect, doubts about its contents, religious influence, and personal belief [9]. Therefore, education and awareness programs for pregnant women during their antenatal care need to be strengthened. Furthermore, the basic education of mothers and the birthplace have no significant associations with the willingness to allow vaccination for their children. The reason why mothers refuse vaccination for their babies needs further exploration.

As Indonesia ranks second in the global TB prevalence list, it is crucial to declare TB as a national health emergency, and an active TB case detection program plays an important role in mitigating this crisis [10]. This study has shown that active TB cases (2.3%) were identified and not all the individuals were on TB therapy (Table 3). TB is a chronic disease with clinical symptoms, which include coughing for over 3 weeks. The behavior of people suffering from chronic cough and their attitude towards a healthy lifestyle also appears to be a burden. Some of the reasons include stigma in the community, lack of awareness, and knowledge of TB [11]. Moreover, there is a spectrum of TB infection, ranging from latent to active TB. Individuals who are able to contain MTB infection are known as latent TB infection (LTBI) and may also harbor a low-grade, subclinical infection. These individuals are at a higher risk of reactivation [12]. The

reason behind infected individuals who do not visit health care providers need further clarification.

This study explored latent TB infection among newborns and family members living in the same house in a crowded area. Various studies have explored the usefulness of IGRA for the diagnosis of LTBI [13]. This study, particularly showed that family members living with active TB have 2.69 times higher chance of positive IGRA (95% CI, 1.22-5.94) compared to those without (Table 4). In addition, it is well known that the chance of being infected with MTB among individuals living in the same house is higher [1]. Also, positive IGRA among individuals who do not live in the same house with TB patients indicates that the infection is transmitted outside the house as well. The isolation of TB patients in a sanatorium or other institution in Indonesia is not possible, however, greater measures are required to reduce MTB transmission.

Furthermore, this study revealed that male has a higher chance of having positive IGRA compared to female (OR 1.68; CI 95% 1.07-2.64) (Table 4). This is due to their frequent outdoor activities and contact with others. Interestingly, male is more susceptible to TB disease as shown in a previous genetic study in Indonesia [14]. The variation in toll like receptor *TLR8* gene, located in chromosome X, has been related to the susceptibility to TB [14]. This finding suggests the role of host genetic factor in immune response differences. Moreover, since only half of the family members showed IGRA positivity, the immune response may play a role in the development of TB infection [12].

Children under 5 years old, especially those who live with active TB family members have a relatively higher risk to develop active TB. This depends on various factors, such as the proximity of contact with the index case, the MTB virulence, the environmental conditions of the house, which include solar radiation and air circulation as well as several other factors [4]. It is therefore important to detect LTBI cases and treat them early. Previously, TST was used to screen TB in adolescents, who have been in contact with TB patients

Table 4. The IGRA test result among family members living in the same house with active TB individuals and stratified according to gender.

	IGRA (+)	IGRA (-)	<i>p</i> value	OR (95%CI)
	N	N		
Family members				
With active TB	17	11	0.011*	2.69 (1.22 – 5.94)
Without active TB	116	202		
Gender				
Male	55	63	0.025*	1.68 (1.07 – 2.64)
Female	78	150		
TOTAL; N	133	213		
(%)	(32.6)	(67.4)		

TB: tuberculosis; IGRA: Interferon Gamma Release Assay; * *p* was statistically significant when $p < 0.05$ (Chi-square).

[15]. However, a study has shown that IGRA is quite sensitive in children younger than 15 years. Interestingly, the IGRA sensitivity is reduced compared to TST in children younger than 2 years [16]. Furthermore, IGRA seems to offer alternative LTBI detection, especially for babies living with parents who suffer from TB [17]. This study has shown that the newborns were still negative for IGRA 12 weeks after BCG vaccination, suggesting that BCG may stimulate immune response and play a protective role against TB disease [18].

Most importantly, the study traced active TB individuals with no treatment, therefore, an active case finding is needed in all family members living in the same house and using proper diagnostic tools for possible LTBI, followed by preventive therapy. Furthermore, education was given to the family members and active TB individuals who were also assigned for TB therapy. LTBI treatment requires a long period of therapy, for example, Isoniazid (INH) needs to be given for 6 or 9 months [4]. Interestingly, the population in Indonesia are predominantly intermediate acetylators [19]. Therefore, drug induced liver intoxication should be considered when giving positive IGRA individuals a long period of INH [20] and adherence as well as compliance, need to be well monitored. In addition, good education and awareness from the health care provider are required to ensure that positive IGRA individuals adhere to LTBI treatment for at least 6 months. This study further shows that 32.6% of the respondents are LTBI as indicted by IGRA positivity, as compared to Singapore where 12.7% of the respondents were LTBI [21]. This gives valuable information to the health authorities to set up preventive therapy using INH.

This study encountered several limitations, including the temporary address of some mothers, which was untraceable after delivery of the baby. As a result, some of them did not receive follow-up care. This issue needs to be raised to the authority to improve the population registration system. Also, education and awareness about TB and immunization need to be administered properly, to ensure that mothers have their babies vaccinated. Furthermore, a longer cohort study will provide more information on how the IGRA positivity in babies develops after exposure to active TB individuals. The data on babies and family members exposed to TB as well as others with IGRA positivity were given to the local health care providers for INH preventive therapy. Finally, good monitoring on this program requires the commitment of stakeholders.

Conclusions

Untreated active pulmonary TB individuals in crowded areas have been detected in this study (2.3%) and may infect the surrounding neighbors, resulting in latent TB infection or IGRA positive (32.6%). Family members living in the same house with active TB individuals as well as being males have a significant higher risk of having a positive IGRA. Since latent TB infection in the crowded area have a major impact on future MTB transmission, active pulmonary TB case finding program and preventive measures need to be taken to contain the potential spread of the infection. It is therefore, necessary to detect latent TB infection and to educate the community on how MTB is spreading.

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Authors' contributions

FST, IR and ES made substantial contributions to the conception and design of the study. NK, H and SA made contributions to the data collection. All authors were involved in data interpretation. FST and ES drafted and revised the manuscript. NK, IR, H, and SA critically revised the manuscript. The final version of the manuscript was approved by all authors, and all authors are accountable to the accuracy and integrity of all parts of the paper.

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Corresponding author

Francisca Srioetami Tanoerahardjo, MD, PhD
 National Institute of Health, Research and Development
 Ministry of Health Republic of Indonesia
 Jl. Percetakan Negara no. 23, Jakarta 10560, Indonesia
 Fax: +628112211615
 Phone: +628112211615
 Email: srioetami@gmail.com

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