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QuantiFERON-TB Gold In-Tube Reliability for Immigrants with Parasitic Infections, Boston

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SUMMARY

SETTING: Accurate testing and treatment for latent tuberculosis infection is necessary for tuberculosis elimination. Certain parasite infections are associated with increased tuberculin skin test positivity; species-specific effects on QuantiFERON-TB Gold In-Tube (QGIT) are not yet described.

OBJECTIVE: We sought to determine whether infection with helminths or protozoa affects QGIT results.

DESIGN: We retrospectively analyzed QGIT and parasite testing results for immigrants screened in Boston from 2012–2017. We also prospectively measured cytokines in QGIT supernatants for a subset (n=68) with 1) helminths, 2) *Blastocystis hominis*, 3) other protozoa, and 4) no parasites.

RESULTS: Of 527 immigrants screened, 141 (26.8%) had a positive QGIT and 229 (43.4%) had parasites detected: 27/527 (5.1%) helminths and 202/527 (38.3%) protozoa. Cytokine analysis revealed increased IL-10 concentrations with protozoa (p=0.04), and non-significantly higher Th2

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AUTHOR CONTRIBUTIONS

NH conceptualized the study, obtained funding, initiated and oversaw all study activities and helped draft and revise the manuscript; RE managed and helped conduct all study activities and data analysis, and wrote and revised the manuscript; MB, VS, and PS conducted the immunology studies, helped draft and critically revise the manuscript; PS also contributed to study concept and data analysis plan; LS, CK, JM, and PK consented patients, assisted with data collection and analysis and critical revision of manuscript; LW contributed to data analysis and manuscript revision; EB contributed to study design and conduct and critical manuscript revision.

concentrations with helminths compared to no parasites. No significant differences emerged in QGIT positivity or interferon-gamma concentrations in any group.

CONCLUSION: This study supports use of QGITs in parasite-endemic settings.

Keywords

Interferon-gamma release assays; *Mycobacterium tuberculosis*; tuberculosis diagnosis; cytokines; host-parasite interactions

INTRODUCTION

In 2015, the World Health Organization (WHO) released a blueprint for tuberculosis (TB) elimination worldwide¹. Pillars of this proposal included improving diagnosis and treatment of TB disease and latent tuberculosis infection (LTBI). Addressing the LTBI reservoir requires understanding whether comorbidities (e.g., diabetes mellitus, parasite infections) increase LTBI risk or progression from LTBI to TB. To evaluate such associations, we must assess the accuracy of diagnostic tests for LTBI in the presence of conditions that may affect host immune response and diagnostic test function.

Helminths are prevalent in 28/30 high TB-burden countries, and studies have suggested parasites affect tuberculin skin test (TST) positivity^{2–7}. Increased odds of TST positivity was observed with hookworm-infected individuals and decreased odds with non-invasive *Trichuris trichiura* and *Giardia lamblia*^{3,4}. Less is known about other protozoa such as *Blastocystis hominis*. The potential association between parasites and LTBI may reflect host immune response^{8–10}. Tissue-invasive helminths, including *Strongyloides stercoralis*, promote a T-helper 2 (Th2) response and suppress the Th1 response needed to control *Mycobacterium tuberculosis* (MTB) infection⁸. This cytokine response reverts after treatment for *S. stercoralis*^{9,11}. In mice, the lung Th2 response induced by invasive *Nippostrongylus brasiliensis* enhanced susceptibility to MTB by promoting MTB-permissive M2 macrophages¹⁰. Mice infected with non-invasive *Heligmosomoides polygyrus* had no such differential disease burden or regulatory cell responses.

Most low-incidence countries have shifted from TST towards interferon gamma release assays (IGRAs) such as QuantiFERON-TB Gold In-Tube (QGIT) for LTBI screening, especially in individuals with BCG exposure^{1,12}. Studies are needed to identify conditions associated with false negative IGRAs such as human immunodeficiency virus (HIV), diabetes mellitus, low body mass index (BMI), and older age^{13–15}. Strong induction of Th2 response and inhibition of Th1 in helminth-infected individuals could affect interferon gamma (IFN- γ) levels and IGRA reliability. The impact of parasites on IGRA testing remains unknown and is relevant in endemic countries and for immigrant screening. We sought to 1) investigate the epidemiologic association between QGIT results and parasite infections in a retrospective review and 2) in a prospective study characterize cytokine response to MTB with helminth and protozoal infections.

STUDY POPULATION AND METHODS

Epidemiologic Study

We retrospectively reviewed de-identified data for all refugees and asylum-seekers (henceforth collectively referred to as immigrants) seen at Boston Medical Center (BMC) and screened through Massachusetts Department of Public Health Refugee Health Assessment Program (RHAP) from October 2012-July 2017. Laboratory tests performed through RHAP include complete blood count (CBC) with differential and screening for HIV and LTBI. QGIT replaced TST in late 2012 and was used routinely starting January 2013. All immigrants were screened for parasites with stool antigen testing for *Cryptosporidium parvum* and *G. lamblia*; stool microscopy was performed before 2014, after which it was used selectively because of increased pre-departure parasite treatment^{16,17}. If prompted by suggestive signs or symptoms (e.g., eosinophilia), clinicians could order parasite serologic testing, performed preferentially at the National Institutes of Health (NIH) for filaria and the Centers for Disease Control and Prevention (CDC) for other helminths.

We collected demographic, medical, and laboratory data for eligible individuals through the electronic health record. Data included countries of birth and residence, days from arrival in the United States (U.S.) until first RHAP visit, documented pre-departure anti-helminthic therapy, BMI, and tobacco and alcohol use. We excluded individuals without QGIT or stool microscopy results.

Immunology Study

For inclusion in the immunology study, we prospectively recruited individuals from January 2016-November 2017 from primary care and specialty clinics at BMC that see many immigrants. Inclusion criteria were: 1) non-US born, 2) screened with QGIT 3) screened for parasitic infections with stool testing, and 4) language fluency in English or any of ten languages*. Individuals were excluded for 1) age <five years (as QGITs were not recommended), 2) failure to complete QGIT or parasite screening, or 3) QGIT collected >21 days prior (due to sample processing limitations). Stool ova and parasite microscopy was performed, if not already completed. Relevant results were reported to the patient's physician for treatment.

Measurement of Outcomes:

We classified QGIT results as positive (IFN- γ 0.35 IU/mL) or negative (<0.35 IU/mL), and as a continuous variable¹². The laboratory reports exact results to 10 IU/mL and higher values as >10 IU/mL (which were assigned a value of 11).

We defined eosinophilia as absolute eosinophil count ≥ 500 cells/ μ L and TB incidence by country of origin¹⁸. For pre-departure anti-helminthic treatment, missing values were imputed from CDC country-specific policy for refugees^{16,17}. We assumed individuals arriving in the US ≥ 12 weeks before their first visit were asylum seekers and not refugees, as RHAP visits occur within 12 weeks of immigration for refugees¹⁶. We assumed asylees

* Arabic, Burmese, Farsi, French, Haitian Creole, Mandarin, Portuguese, Somali, Spanish, and Vietnamese

were not empirically treated for helminthic infections as most asylum-seekers do not have pre-immigration health screening. We categorized BMI as underweight ($<18.5 \text{ kg/m}^2$ for adults, $<5^{\text{th}}$ percentile by CDC age- and sex-specific growth curves for children <18 years), normal weight ($18.5\text{--}25 \text{ kg/m}^2$ or $5^{\text{th}}\text{--}85^{\text{th}}$ percentile), and overweight ($>25 \text{ kg/m}^2$ or $>85^{\text{th}}$ percentile).

Cytokine Analysis:

Supernatants of QGIT samples from nil (control) and MTB antigen-stimulated tubes were stored at -80°C . Cytokines in the supernatants were measured using a multiplex cytokine panel (V plex pro-inflammatory panel 1 human kit) from Meso Scale Discovery (MSD) (Rockville, MD, USA). Plates were read on the MSD detector (MESO Quickplex SQ120). Calibration curve and analyte concentration calculations were performed using MSD DISCOVERY WORKBENCH® analysis software.

Statistical Analysis:

Data were analyzed using SAS 9.4 (SAS Institute Inc., Cary, NC, USA). We used the Student *t*-test or one-way analysis of variance (ANOVA) to describe continuous, normally distributed and χ^2 or Fisher's exact test for categorical normally distributed demographic characteristics, and Kruskal-Wallis or Wilcoxon Rank Sum for non-normally distributed variables and cytokine distributions given small sample sizes. For the epidemiology study, we employed multivariable logistic regression to examine the association between parasites and QGIT results, retaining variables causing change in effect size $>10\%$ for final model selection. For this model and immunology analyses, we compared those with: 1) helminths, 2) *B. hominis*, 3) other protozoa, and 4) no parasites. Those with indeterminate QGITs ($n=5$) or mixed *B. hominis* and other protozoa ($n=51$) were excluded from the model to isolate the effect on immune response. Given missing observations for alcohol and tobacco use, HIV, and pregnancy, we performed multiple imputation analysis using fully conditional specification creating 20 imputed datasets from all Table 1 variables to predict missing values. We separately performed multiple logistic regression on imputed data.

For the immunology study, to achieve 80% power to detect an effect size of 1.3 (large in the nomenclature of Cohen^{19,20}) for 2 group comparisons and 0.55 for 4 group comparisons, we aimed to include 10 individuals with positive and 10 with negative QGITs in each of the 4 groups noted above. The first 10 eligible participants were recruited into each group (where possible, without immunocompromising conditions, pregnancy, or pre-departure anti-parasitic therapy). We compared distributions of log-transformed concentrations of Th1 and Th2 cytokines. We examined nil tube Th2 cytokine concentrations for all samples and the difference between MTB antigen-stimulated and nil tube concentrations for QGIT-positive samples. For IFN- γ , we compared MTB antigen-stimulated minus nil tube concentrations among individuals with positive and negative QGITs. Finally, we compared continuous QGIT results among study groups combining individuals from epidemiology and immunology studies.

Ethics Statement

BMC and Rutgers New Jersey Medical School Institutional Review Boards (IRB) approved the protocol. All participants in the immunology study provided informed consent; the IRB waived consent for obtaining de-identified data for the epidemiology study.

RESULTS

Epidemiology Study

Of 1040 patients 5 years old screened through RHAP, 883 had QGITs performed; of those, 784 completed any stool testing, and 527 had stool microscopy and/or parasite serology (99 did not return stool samples, 257 only had *G. lamblia*/*C. parvum* antigen testing). The 527 analyzed individuals included 307 (58.2%) males; mean age was 30.2 years (range 5–86) (Table 1). Subjects emigrated most frequently from Haiti (n=118; 22.4%), Iraq (n=85; 16.1%) and Somalia (n=67; 12.7%); 29% were from the Americas, 26.6% from Sub-Saharan Africa, and 65.7% from a country with TB incidence 100/100,000 population. Thirty-five (6.6%) were underweight, 15 (2.8%) had diabetes mellitus, 9/485 (1.7%) tested for HIV had positive results, and none had immunocompromising conditions. Approximately half had pre-departure anti-helminthic treatment.

Stool testing revealed protozoa in 202/527 (38.3%) individuals and helminths in 3 (0.6%) (Table 2). Protozoa identified most frequently were *B. hominis* (n=126, 23.9%), *Endolimax nana* (n=66, 12.5%), and *G. lamblia* (n=29, 5.5%). Fifty individuals had helminth serologic testing, which revealed helminth infections in 24 (4.6% of study population): *S. stercoralis* in 15, *Schistosoma* spp in 13, and filaria in 2 (not mutually exclusive; Table 2).

Of 527 subjects, 141 (26.8%) had positive QGITs, which were observed more frequently in adults than children ($p<0.0001$), males ($p=0.03$), overweight/obese individuals (vs. normal/underweight, $p=0.002$), those from countries with higher TB-incidence ($p<0.0001$), those with HIV ($p=0.03$) or reporting alcohol ($p=0.008$) or tobacco use ($p=0.06$) (Table 1). Five subjects had indeterminate QGITs; one was underweight, one pregnant and one had HIV (Table 1).

The proportion of positive QGIT tests among those with protozoa ranged from 15% (3/20) with *G. lamblia* to 60% (3/5) of those with *E. hartmani* (Table 2). Positive QGITs occurred in 6/27 (22.2%) with helminths and 79/293 (27%) without parasites (Table 3).

In univariate and multivariate analyses of individuals with *B. hominis*, other protozoa, and helminth infections, no group had significantly increased odds of positive QGIT compared to individuals without parasites (Table 3). Only age, HIV, and country-specific TB-incidence remained significant predictors of QGIT positivity in multivariable analyses. Results did not substantially differ after multiple imputation analysis (Table 3).

Immunology study

We assessed 557 patients for inclusion in the immunology study. Of 386 eligible participants who had appropriate laboratory tests ordered, 265 (68.9%) consented to participate. Of these, 179 (67.5%) completed the necessary laboratory tests. Of the 49/179 (27.4%) with

positive QGITs, results were analyzed for 4 with helminths, 8 with *B. hominis*, 6 other protozoa, and 10 with no parasites; among QGIT-negatives, we analyzed ten in each of the four groups (Table 4).

We observed no significant differences in Th2 cytokines IL-4 or IL-13, but IL-10 concentrations were higher in protozoa and helminth groups compared to *B. hominis* and those without parasites ($p=0.04$) (Figure 1a-c). For QGIT-positive samples, we observed no significant differences in Th2 cytokine concentrations with MTB antigen stimulation (Figure 1d-f).

IFN- γ concentrations (in MTB antigen-stimulated minus nil tubes) were lower in QGIT-positive individuals with helminths, but the difference was not statistically significant (helminth mean concentration 1.32 pg/mL, versus 3.61 pg/mL in those without parasites; $p=0.20$ for all group comparison, $p=0.10$ for helminths vs. no parasites) (Figure 2a). IFN- γ concentrations did not differ among groups in QGIT-negative individuals ($p=0.23$) (Figure 2b).

Epidemiology and Immunology Studies

We found no significant differences in QGIT-measured IFN- γ concentrations among the four study groups in epidemiology and immunologic studies combined ($n=518$; some participants fit both inclusion criteria). Mean IFN- γ concentration was slightly lower in helminth-infected, QGIT-positive individuals ($p=0.58$) (Figure 3a). Among QGIT-negative individuals, we observed no difference in IFN- γ concentrations in the helminth group compared to other groups ($p=0.73$) (Figure 3b).

DISCUSSION

These findings suggest that QGIT results are valid in individuals with parasitic infections – a sizeable proportion of those tested in this immigrant population and worldwide^{5,7}. We found that, among immigrants from TB-endemic countries in the Boston RHAP program, parasites do not affect QGIT positivity, adjusting for covariates. Additionally, IFN- γ concentrations did not differ significantly after MTB antigen stimulation among those with helminths, *B. hominis*, other protozoa, or no parasites detected. These results suggest individuals with helminth or protozoal infections will not have false negative QGIT results from low IFN- γ concentrations.

As expected, Th2 cytokine concentrations were slightly elevated in individuals with helminths as was IL-10 for other protozoa. This is consistent with previous findings of increased Th2 cytokines from helminths^{8–11}. The cytokine bias however, does not appear to affect QGIT results.

We found that almost one-third of immigrants had positive QGITs and that older age, male sex, overweight/obese BMI, alcohol use, and HIV infection were univariate predictors of LTBI. Only age and HIV remained significant in multivariable analysis. These findings are consistent with the literature, but the association between BMI and LTBI has not been evaluated frequently²¹. Our findings mirror a recent meta-analysis showing that underweight

compared with overweight/obese BMI was associated with negative QGITs²². Malnutrition is a risk for TB disease^{23,24} but can also blunt cellular immune response, and has been associated with false negative IGRAs and TSTs^{14,25–27}. IGRAs offer the advantage over TST that if malnutrition altered the immune response, one would expect a poor mitogen response and indeterminate QGIT results, which we did not see, albeit with limited sample size. Additional studies would help determine whether low BMI alters QGIT reliability.

Our study's strengths lie in pairing epidemiologic and immunologic analyses that together support the lack of effect of parasitic infections on QGIT results. Furthermore, we recruited individuals with a variety of parasitic infections to compare effects on cytokine response. Our study had several limitations. First, decreases in refugee volume over 2017 and lower rates of helminth infection than predicted limited sample size in QGIT-positive parasite groups. This decreased power in two immunology study comparisons involving only QGIT-positives, and it is possible we failed to detect a true association. We addressed this by including both epidemiologic (n=471) and immunologic cohorts (n=68) in the analysis of quantitative QGIT response. All children recruited in the immunology study had parasites detected, and studies report differential cytokine responses with age^{28,29}; however, in sensitivity analyses, we did not see an association between age group and cytokine concentrations (data not shown), therefore it is unlikely that age confounded our results. Potential for under-detection of parasites existed as testing was limited to one stool test and additional testing per the patient's provider. Misclassification was likely minimal, as urinalysis and CBCs were performed; any clinical or laboratory evidence of helminth infection should have prompted further testing. As with any retrospective chart review, variables occasionally were not documented in the medical record (e.g., pre-departure anti-helminthic treatment). We incorporated CDC country-specific treatment guidelines and used multiple imputation analysis to address missing data. Finally, QGIT IFN- γ values >10 were not quantified, so we could not estimate the full distribution of QGIT results in the epidemiology study, but our immunology study utilized the full range of values and found similar results.

CONCLUSIONS

Our study of the association between parasite infections and QGIT testing in immigrants to the U.S. found no significant differences in QGIT results or IFN- γ concentrations among patients with helminths, *B. hominis*, or other protozoa compared with no parasites. This finding provides evidence that QGIT results can be trusted for individuals with parasite infections. Parasites are endemic in the majority of high TB burden countries, where it is important to identify TB contacts correctly, especially for contacts of MDR TB cases and young children^{5,7,30,31}. These data have relevance for contact tracing in high-burden countries and for screening immigrants to low burden settings.

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Figure 1:

Th2 cytokine concentrations in QGIT supernatants for immigrants with and without parasites, n = 68. 1A-1C. IL-4, IL-10, IL-13 concentrations in nil tube for QGIT-positive and QGIT-negative immigrants. 1D-1F. IL-4, IL-10, and IL-13 concentrations in MTB-antigen stimulated minus nil tube for QGIT-positive immigrants. Boxes indicate IQR, line within box denotes median, circles represent individual data points. Th2, T helper 2; QGIT, QuantiFERON-TB Gold-in-Tube; MTB, *Mycobacterium tuberculosis*; IQR, interquartile range.

Figure 2:

IFN- γ concentrations measured from QGIT supernatants for immigrants with and without parasite infections, n = 68, in MTB-stimulated minus nil tube (A) for QGIT-positive immigrants and (B) for QGIT-negative immigrants. Boxes indicate IQR, line within box denotes median, circles represent individual data points. IFN- γ , interferon gamma; QGIT, QuantiFERON-TB Gold-in-Tube; MTB, *Mycobacterium tuberculosis*; IQR, interquartile range.

Figure 3:

IFN- γ concentrations measured by commercial QGIT in MTB-stimulated minus nil tube for immigrants with and without parasite infections (epidemiology and immunologic groups combined), n = 518 in (A) QGIT-positive immigrants and (B) QGIT-negative immigrants. Horizontal line represents cutoff for positive QGIT (0.35 IU/mL) for reference. Boxes indicate IQR, line within box denotes median, circles represent individual data points. IFN- γ , interferon gamma; QGIT, QuantiFERON-TB Gold-in-Tube; MTB, *Mycobacterium tuberculosis*; IQR, interquartile range.

Table 1:

Demographic characteristics for epidemiology study of immigrants with completed QuantiFERON-TB Gold In-Tube and parasite screening, October 2012 - July 2017, Boston

Characteristic	QGIT positive n=141 (%)	QGIT negative n=381 (%)	QGIT indeterminate n=5 (%)	Total n=527 (%)	p-value [†]
Mean age, years (SD)	36.2 (13.9) [*]	28.0 (14.5)	25.8 (11.5)	30.2 (14.8)	<0.0001
Age					
Children (<18yo)	7 (5.0)	103 (27.0)	1 (20)	111 (21.1)	<0.0001
Adults (≥ 18yo)	134 (95.0)	278 (73.0)	4 (80)	416 (78.9)	
Time since arrival in US					
<12 weeks	126 (89.4)	353 (92.6)	4 (80)	483 (91.7)	0.22
12 weeks	15 (10.6)	28 (7.4)	1 (20)	44 (8.3)	
Sex					
Male	93 (66.0)	212 (55.6)	2 (40)	307 (58.2)	0.03
Female	48 (34.0)	169 (44.4)	3 (60)	220 (41.7)	
BMI (N=526)					
Underweight [‡]	6 (4.3)	28 (7.4)	1 (20)	35 (6.6)	0.002
Normal weight	55 (39.3)	204 (53.5)	2 (40)	261 (49.6)	
Overweight/Obese	79 (56.4)	149 (39.1)	2 (40)	230 (43.7)	
Alcohol use (N=451)					
Yes	18 (14.9)	22 (6.8)	0	40 (8.9)	0.008
No	103 (85.1)	303 (93.2)	5 (100)	411 (91.1)	
Tobacco use (N=490)					
Yes	17 (12.6)	25 (7.1)	0	42 (8.5)	0.06
No	118 (87.4)	325 (92.9)	5 (100)	448 (91.4)	
WHO region					
Africa	41 (29.1)	98 (25.7)	1 (20)	140 (26.6)	0.43
Americas	38 (27.0)	114 (29.9)	1 (20)	153 (29.0)	
Eastern Mediterranean	58 (41.1)	143 (37.5)	0	201 (38.1)	
Europe	0	7 (1.8)	0	7 (1.3)	
Southeast Asia	1 (0.7)	7 (1.8)	1 (20)	9 (1.7)	
Western Pacific	3 (2.1)	12 (3.2)	2 (40)	17 (3.2)	
Mean TB incidence, country of origin, cases per 100,000 (SD) [§]	195 (95)	154 (106)	176 (123)	165 (104)	<0.0001
TB incidence, country of origin [§]					
<100/100,000	27 (19.2)	152 (39.9)	2 (40)	181 (34.3)	<0.0001
100–200/100,000	56 (39.7)	123 (32.3)	1 (20)	180 (34.2)	
>200/100,000	58 (41.1)	106 (27.8)	2 (40)	166 (31.5)	
Pre-departure anti-helminthic treatment					
Yes	76 (53.9)	196 (51.4)	2 (40)	274 (52.0)	0.62
No	65 (46.1)	185 (48.6)	3 (60)	253 (48.0)	

Characteristic		QGIT positive n=141 (%)	QGIT negative n=381 (%)	QGIT indeterminate n=5 (%)	Total n=527 (%)	p-value [†]
HIV (N=485)						
	Yes	5 (4.0)	3 (0.8)	1 (33.3)	9 (1.9)	0.03
	No	120 (96.0)	354 (99.2)	2 (66.7)	476 (98.1)	
Pregnant (N=448) [¶]						
	Yes	1 (0.8)	7 (2.2)	1 (33.3)	9 (2.0)	0.45
	No	123 (99.2)	314 (97.8)	2 (66.7)	439 (98.0)	
Diabetes mellitus						
	Yes	7 (5.0)	8 (2.1)	0	15 (2.8)	0.13
	No	134 (95.0)	373 (97.9)	5 (100)	512 (97.2)	
Absolute eosinophil count (mean, [SD]), cells/ μ L (N=521)		185 (19)	199 (24)	180 (22)	195 (22)	0.56
Eosinophilia (N=521)						
	Yes (≥ 500 cells/ μ L)	10 (7.2)	34 (9.0)	1 (20)	45 (8.6)	0.51
	No (<500 cells/ μ L)	129 (92.8)	343 (91.0)	4 (80)	476 (91.4)	

* No. (%) unless otherwise indicated; for each variable with any missing data, total No. without missing data indicated.

[†] p-values indicate comparison between QGIT positive and QGIT negative individuals, using Student *t*-test for continuous, normally distributed variables (age), Kruskal-Wallis for continuous, non-normally distributed variables (TB incidence, absolute eosinophil count) and χ^2 or Fisher's Exact test for categorical variables.

[‡] For adults: underweight denotes BMI <18.5 kg/m², normal weight 18.5–25 kg/m², and overweight/obese >25 kg/m². For children, underweight denotes CDC BMI percentile $<5^{\text{th}}$ percentile, normal 5th–85th percentile, and overweight $>85^{\text{th}}$ percentile by age and sex.

[§] Denotes TB incidence by 2016 WHO statistics for primary country of residence prior to the United States, by cases per 100,000 population and divided into lower and higher categories of TB incidence (17).

[¶] By pregnancy test or diagnosis in medical record. Not pregnant indicates either negative pregnancy test or not a female of reproductive age (13–50 years). The 79 women of reproductive age without documented negative pregnancy tests were considered as having missing data.

Abbreviations: QGIT QuantiFERON-TB Gold In-Tube SD Standard Deviation BMI Body Mass Index WHO World Health Organization TB Tuberculosis HIV Human Immunodeficiency Virus

Table 2:

Parasite testing results for epidemiology study of immigrants with completed QuantiFERON-TB Gold In-Tube and parasite screening, October 2012 - July 2017, Boston

Parasites detected	All n=522 (%)	QGIT positive n=141 (27%)	QGIT negative* n=381 (73%)	p-value [†]
None	293 (56.1)	79 (27)	214 (73)	
Any Protozoa:	202 (38.7)	56 (27.7)	146 (72.3)	0.85
<i>Blastocystis hominis</i> [‡]	70 (13.4)	21 (30)	49 (70)	
<i>Dientamoeba</i>	1 (0.2)	0	1 (100)	
<i>Endolimax nana</i>	17 (3.3)	6 (35.3)	11 (64.7)	
<i>Entamoeba coli</i>	8 (1.5)	2 (25)	6 (75)	
<i>Entamoeba hartmanni</i>	5 (1.0)	3 (60)	2 (40)	
<i>Entamoeba histolytica/dispar</i>	4 (0.8)	2 (50)	2 (50)	
<i>Giardia lamblia</i>	20 (3.8)	3 (15)	17 (85)	
<i>Iodamoeba butschlii</i>	3 (0.6)	1 (33.3)	2 (66.7)	
Mixed Protozoa	74 (14.2)	18 (24.3)	56 (75.7)	
Any Helminth:	27 (5.2)	6 (22.2)	21 (77.8)	0.59
<i>Strongyloides stercoralis</i>	10 (1.9)	2 (20)	8 (80)	
<i>Schistosoma</i> spp.	9 (1.7)	3 (33.3)	6 (66.7)	
<i>Schistosoma</i> spp. & <i>S. stercoralis</i>	3 (0.6)	1 (33.3)	2 (66.7)	
<i>Hymenolepis nana</i>	2 (0.4)	0	2 (100)	
Hookworm	1 (0.2)	0	1 (100)	
<i>Schistosoma</i> spp. & <i>S. stercoralis</i> & Filaria	1 (0.2)	0	1 (100)	
<i>S. stercoralis</i> & Filaria	1 (0.2)	0	1 (100)	

* Individuals with indeterminate QGITs not shown in this table (n=5), as all individuals with an indeterminate result had no parasites detected.

[†] p-values reflect χ^2 test comparing QGIT positivity for any protozoa detected compared with no parasites detected and for any helminths detected compared with no parasites detected.

[‡] Counts of protozoa denote indicated organism alone was isolated; mixed infections are included in 'Mixed Protozoa' category.

Abbreviations: QGIT QuantiFERON-TB Gold In-Tube

Table 3:

Odds of positive QuantiFERON-TB Gold-in-Tube by parasite group, epidemiologic study

Characteristic	Crude OR	95% CI	aOR [†]	95% CI	aOR [‡]	95% CI
Parasites Detected						
None (n=293)	Ref	Ref	Ref	Ref	Ref	Ref
<i>B. hominis</i> (n=70)	1.16	(0.66, 2.06)	1.12	(0.49, 2.56)	1.19	(0.65, 2.18)
Other Protozoa (n=81)	1.28	(0.75, 2.18)	1.44	(0.70, 2.98)	1.46	(0.82, 2.60)
Helminth (n=27)	0.77	(0.30, 1.99)	0.47	(0.10, 2.19)	0.61	(0.23, 1.64)
Age						
Children (<18yo)	Ref	Ref	Ref	Ref	Ref	Ref
Adults (≥ 18yo)	7.25	(3.08, 17.0)	8.44	(2.81, 25.4)	9.19	(3.80, 22.2)
Time since arrival to US						
<12 weeks	Ref	Ref				
≥ 12 weeks	1.26	(0.63, 2.53)				
Sex						
Male	Ref	Ref				
Female	0.63	(0.41, 0.96)				
BMI						
Underweight	Ref	Ref				
Normal Weight	1.94	(0.92, 4.08)				
Overweight/Obese	3.59	(1.73, 7.44)				
Alcohol use						
Yes	2.67	(1.32, 5.38)	1.43	(0.60, 3.40)		
No	Ref	Ref	Ref	Ref		
Tobacco use						
Yes	1.94	(1.00, 3.77)	2	(0.79, 5.04)		
No	Ref	Ref	Ref	Ref		
Mean TB incidence, country of origin, cases per 100,000	1.004	(1.002, 1.006)	1.006	(1.003, 1.009)	1.01	(1.003, 1.007)
Pre-departure anti-helminthic treatment						
Yes	1.11	(0.74, 1.66)				
No	Ref	Ref				
HIV						
Yes	4.69	(1.10, 19.9)	10.24	(1.01, 104)		
No	Ref	Ref	Ref	Ref		
Pregnant						
Yes	0.4	(0.05, 3.37)	0.4	(0.04, 3.58)		
No	Ref	Ref	Ref	Ref		
Diabetes Mellitus						
Yes	2.66	(0.91, 7.72)				
No	Ref	Ref				
Absolute Eosinophil Count (cells/hpf)	0.68	(0.26, 1.80)	0.3	(0.04, 2.10)		

* Total No. for this table is 471 (Those with mixed *B. hominis* and other protozoa detected [n=51] or indeterminate QGIT results [n=5] were excluded from multivariable analysis).

[†] OR of positive compared to negative QGIT adjusted for age (≥ 18 vs. <18), alcohol use, tobacco use, tuberculosis incidence of country of origin, absolute eosinophil count, pregnancy, and HIV.

[‡] Using multiple imputation analysis to account for all missing data, OR of positive vs. negative QGIT adjusted for age (≥ 18 vs. <18) and tuberculosis incidence of country of origin.

Abbreviations: QGIT QuantiFERON-TB Gold In-Tube aOR Adjusted Odds Ratio CI Confidence Interval

Table 4:

Demographic characteristics for immigrants included in immunologic analysis, January 2016 - November 2017, Boston

Characteristic	All n=68 (%) [*]	No parasites n=20 (%)	<i>B. hominis</i> n=18 (%)	Other protozoa n=16 (%)	Helminth n=14 (%)	p-value [†]
Mean age, years (SD)	29.6 (13.6)	31.4 (8.7)	24.8 (15.0)	25.1 (9.5)	38.4 (17.5)	0.01
Age						
Children (<18yo)	13 (19.1)	0 (0)	7 (38.9)	4 (25)	2 (14.3)	0.009
Adults (≥ 18yo)	55 (80.9)	20 (100)	11 (61.1)	12 (75)	12 (85.7)	
Time since arrival to US						
<12 weeks	48 (70.6)	9 (45)	17 (94.4)	14 (87.5)	8 (57.1)	0.002
≥ 12 weeks	20 (29.4)	11 (55)	1 (5.6)	2 (12.5)	6 (42.9)	
Sex						
Male	39 (57.4)	11 (55)	9 (50)	9 (56.2)	10 (71.4)	0.66
Female	29 (42.6)	9 (45)	9 (50)	7 (43.8)	4 (28.6)	
BMI (N=67)						
Underweight [‡]	3 (4.5)	0	1 (5.6)	1 (6.2)	1 (7.7)	0.85
Normal weight	28 (41.8)	9 (45)	7 (38.9)	8 (50)	4 (30.8)	
Overweight/Obese	36 (53.7)	11 (55)	10 (55.6)	7 (43.8)	8 (61.5)	
Alcohol use (N=64)						
Yes	9 (14.1)	3 (15)	3 (17.6)	1 (7.1)	2 (15.4)	0.89
No	55 (85.9)	17 (85)	14 (82.4)	13 (92.9)	11 (84.6)	
Tobacco use (N=66)						
Yes	5 (7.6)	2 (10)	1 (5.6)	1 (6.7)	1 (7.7)	0.96
No	61 (92.4)	18 (90)	17 (94.4)	14 (93.3)	12 (92.3)	
WHO region						
Africa	20 (29.4)	3 (15)	5 (27.8)	6 (37.5)	6 (42.9)	0.32
Americas	35 (51.5)	14 (70)	7 (38.9)	8 (50)	6 (42.9)	
Eastern Mediterranean	13 (19.1)	3 (15)	6 (33.3)	2 (12.5)	2 (14.3)	
Europe	0	0	0	0	0	
Southeast Asia	0	0	0	0	0	
Western Pacific	0	0	0	0	0	
Mean TB incidence, country of origin per 100,000 (SD) [§]	157 (79)	185 (65)	120 (91)	184 (72)	135 (71)	0.09
TB incidence, country of origin, cases per 100,000 [§]						
<100/100,000	17 (25)	2 (10)	8 (44.4)	2 (12.5)	5 (35.7)	0.19
100–200/100,000	33 (48.5)	12 (60)	6 (33.3)	10 (62.5)	5 (35.7)	
>200/100,000	18 (26.5)	6 (30)	4 (22.2)	4 (25)	4 (28.6)	
Pre-departure anti-helminthic treatment						
Yes	14 (20.6)	0	5 (27.8)	6 (37.5)	3 (21.4)	0.01
No	54 (79.4)	20 (100)	13 (72.2)	10 (62.5)	11 (78.6)	

Characteristic	All n=68 (%) [*]	No parasites n=20 (%)	<i>B. hominis</i> n=18 (%)	Other protozoa n=16 (%)	Helminth n=14 (%)	p-value [†]
HIV (N=61)						
Yes	3 (4.9)	0	1 (5.9)	1 (6.2)	1 (11.1)	0.47
No	58 (95.1)	19 (100)	16 (94.1)	15 (93.8)	8 (88.9)	
Pregnant [‡] (N=63)						
Yes	2 (3.2)	2 (11.1)	0	0	0	0.24
No	61 (96.8)	16 (88.9)	17 (100)	16 (100)	12 (100)	
Eosinophilia (N=63)						
Yes (≥ 500 cells/μL)	12 (19)	1 (5.3)	1 (5.6)	2 (13.3)	8 (72.7)	<0.0001
No (<500 cells/μL)	51 (81)	18 (94.7)	17 (94.4)	13 (86.7)	3 (27.3)	
Diabetes mellitus						
Yes	0	0	0	0	0	--
No	68 (100)	20 (100)	18 (100)	16 (100)	14 (100)	

^{*}No. (%) unless otherwise indicated; for each variable with any missing data, total No. without missing data noted.

[†]p-values indicate comparison between QGIT positive and QGIT negative individuals, using ANOVA for continuous, normally distributed variables (age), Kruskal-Wallis for continuous, non-normally distributed variables (TB incidence, absolute eosinophil count) and χ^2 test for categorical variables.

[‡]For adults: underweight denotes BMI <18.5 kg/m², normal weight 18.5–25 kg/m², and overweight/obese >25 kg/m². For children, underweight denotes CDC BMI percentile <5th percentile, normal 5th–85th percentile, and overweight >85th percentile by age and sex.

[§]Denotes TB incidence by 2016 WHO statistics for primary country of residence prior to the United States, by cases per 100,000 population and divided into lower and higher categories of TB incidence (17).

[¶]By pregnancy test or diagnosis in medical record. Not pregnant indicates either negative pregnancy test or not a female of reproductive age (13–50 years). Women of reproductive age without documented negative pregnancy tests were considered as having missing data.

Abbreviations: QGIT QuantiFERON-TB Gold In-Tube SD Standard Deviation BMI Body Mass Index WHO World Health Organization TB Tuberculosis HIV Human Immunodeficiency Virus