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Latent *tuberculosis* co-infection is associated with heightened levels of humoral, cytokine and acute phase responses in seropositive SARS-CoV-2 infection

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SUMMARY

Objectives: Latent Tuberculosis infection (LTBI) is postulated to modulate immune responses and alter disease severity in SARS-CoV-2 co-infection. However, no data exist on the effect of LTBI on the immune responses in SARS-CoV-2 co-infected individuals.

Methods: We examined the SARS-CoV-2 specific antibody responses, plasma cytokines, chemokines, acute phase proteins and growth factor levels in LTBI positive and negative individuals with SARS-CoV-2 infection.

Results: Our results demonstrated that individuals with LTBI (LTBI+) and seropositive for SARS-CoV-2 infection were associated with elevated SARS-CoV-2 specific IgM, IgG and IgA antibodies, as well as enhanced neutralization activity compared to those negative for LTBI (LTBI-) individuals. Our results also demonstrate that LTBI+ individuals exhibited significantly higher plasma levels of IFNγ, IL-2, TNFα, IL-1α, IL-1β, IL-6, IL-12, IL-15, IL-17, IL-3, GM-CSF, IL-10, IL-25, IL-33, CCL3 and CXCL10 compared to LTBI- individuals. Finally, our results show that LTBI+ individuals exhibit significantly higher levels of C-reactive protein, alpha-2 macroglobulin, VEGF and TGFα compared to LTBI- individuals.

Conclusions: Thus, our data clearly demonstrates that LTBI+ individuals seropositive for SARS-CoV-2 infection exhibit heightened levels of humoral, cytokine and acute phase responses compared to LTBI- individuals. Thus, LTBI is associated with modulation of antibody and cytokine responses as well as systemic inflammation in individuals seropositive for SARS-CoV-2 infection.

Introduction

Latent Tuberculosis infection (LTBI) is known to infect about a quarter of the world's population, while SARS-CoV-2 has thus far infected over 130 million people worldwide.[1, 2] Although both LTBI and SARS-CoV-2 are co-prevalent in many parts of the world, there is a paucity of information about the effect of co-infection on the host immune responses. Tuberculosis (TB) has been postulated to play a role in the development of SARS-CoV-2 infection and exacerbation of COVID-19 disease in small studies conducted in China and India.[3] It has been shown that COVID-19 can occur before, after or simultaneously with the diagnosis of TB and that mortality is likely to be increased in elderly individuals with these co-infections.[4] A recent study showed that indeterminate result in Interferon Gamma Release Assays was associated with severe lymphocytopenia in COVID-19 patients.[5] In addition, LTBI was not shown to affect the ability to in vitro respond to SARS-CoV-2.[6] Various papers have suggested that in areas of high LTBI prevalence, the profound lymphopenia induced by SARS-CoV-2 and use of steroids as a treatment for COVID-19 could (a) predispose patients to TB reactivation as a consequence of a transient suppression of cellular immunity and/or (b) increase the risk of progressive primary TB infection.[7–11]

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The pathogenesis of COVID-19 is driven by immune responses, excessive inflammation and increased coagulation.[12–14] COVID-19 can manifest as an asymptomatic, mild, moderate or severe disease.[15,16] Both LTBI and asymptomatic SARS-CoV-2 infections are associated with enhanced cytokine, chemokine, acute phase protein and growth factor responses.[17] Moreover, antibody responses to the spike protein of SARS-CoV-2 and enhanced neutralizing antibody responses are integral hallmarks of SARS-CoV-2 infection.[18] Therefore, to elucidate the interaction between LTBI and seropositive SARS-CoV-2 infection, we examined binding and neutralizing antibody responses to SARS-CoV-2 as well as the systemic levels of cytokines, chemokines, acute phase proteins and growth factors in seropositive SARS-CoV-2 individuals with or without LTBI.

Material and methods

Study population

Consenting individuals with age range among 60 - 80 years of age, living in hotspots for SARS-CoV-2 infection in Chennai, India were recruited between July 2020 and September 2020. Those who were diagnosed with tuberculosis (TB) in the previous 6-months or were currently on anti-TB treatment were not included in the study. The study participants were negative for HIV or malignancy. SARS-CoV-2 seropositivity and prior infection was diagnosed by IgG positivity for spike protein. All the seropositive individuals were asymptomatic and did not report any signs or symptoms of COVID-19 and did not seek any treatment. LTBI was determined on the basis of positivity for QuantiFERON TB Gold in tube (QGIT) test, with no symptoms or signs suggestive of active TB. Individuals with SARS-CoV-2 acute infection (as determined by RT-PCR) were excluded from the study. Individuals with no history of previous TB and normal chest radiographs were included. QGIT was done based on the manufacturer's recommendations (Qiagen). The study groups are defined as those who are positive for both SARS-CoV-2 and LTBI (hereafter LTBI+) and those who are SARS-CoV-2 positive but negative for LTBI (hereafter as LTBI-).

Measurement of SARS-CoV-2 IgA, IgM and IgG

The SARS-CoV-2 serology was measured by an iFLASH 1800 chemiluminescent immunoassay from Shenzhen YHLO Biotech which measures IgM and IgG assays against both SARS-CoV-2 S- and *N*-proteins. The tests were performed following the manufacturer's protocol (Shenzhen YHLO Biotech Co., Ltd.) The results were determined by chemiluminescent reaction as relative light units (RLUs). IgM and IgG concentrations were obtained using the iFlash 1800 assay, and $\geq$ 10 AU/ml were defined as positive and $<$ 10.00 AU/mL was considered as non-reactive. Nucleocapsid specific IgA levels were detected using COVID-19 Human IgA ELISA kit (RayBiotech) based on the manufacturer's protocol. The assay was validated by the positive control mean optical density (PC:OD450) greater than 0.5 and negative control mean less than 0.3.

Measurement of circulating neutralizing antibodies

Plasma samples were used to measure the circulating neutralizing antibodies levels using SARS-CoV-2 Surrogate Virus Neutralization Test Kit according to manufacturer's (GenScript) instructions. The cut-off value for SARS-CoV-2 neutralizing antibody detection, according to the manufacturer, the SARS-CoV-2 Surrogate Virus Neutralization more than or equal to 30% was considered as positive and $<$30% was considered as non-reactive.[19]

Multiplex assays

Systemic plasma levels of acute phase proteins, cytokines and chemokines were estimated using Luminex Magpix Multiplex Assay system (Bio-Rad, Hercules, CA). Milliplex MAP Human CVD Panel Acute Phase magnetic bead panel was used to measure the acute phase proteins and Luminex Human Magnetic Assay kit 45 Plex (R & D systems) was used to measure the cytokines and chemokine levels. The minimum detection levels for acute phase proteins was as follows: alpha-2 macroglobulin ($\alpha-2$-M), 0.49 ng/mL; C-reactive protein (CRP), 0.05 ng/mL; haptoglobin, 0.06 ng/mL; and Serum Amyloid A-1 (SAA-1) 0.06 ng/mL; The lowest detection limits for cytokines were as follows: IFNγ, 5.7 pg/mL; IL-2, 3.6 pg/mL; TNFα, 12.4 pg/mL; IL-1α, 10.6 pg/mL; IL-1β, 3.5 pg/mL; IFNα, 3.9 pg/mL; IFNβ 3.25 pg/mL; IL-6, 9.0 pg/mL; IL-12, 18.5 pg/mL; IL-15, 2.5 pg/mL; IL-17, 9 pg/mL; IL-3, 17 pg/mL; IL-7, 3.5 pg/mL; G-CSF, 8.4 pg/mL; GM-CSF, 18.4 pg/mL; IL-4, 1.1 pg/mL; IL-5, 6.2 pg/mL; IL-13, 31.8 pg/mL; IL-10, 32.2 pg/mL; IL-25, 18.4 pg/mL; IL-33, 13.8 pg/mL; IL-1Ra, 11.7 pg/mL. The lowest detection limits for chemokines were as follows: CCL2, 5.9 pg/mL; CCL3, 5.1 pg/mL; CCL4, 103.8 pg/mL; CCL5, 297 pg/mL; CCL11, 21.6 pg/mL; CCL19, 3.9 pg/mL; CCL20, 2.4 pg/mL; CXCL1, 19.1 pg/mL; CXCL2, 21.1 pg/mL; CXCL8, 1.4 pg/mL; CXCL10, 2.6 pg/mL and CX3CL1, 188 pg/mL. The lowest detection limits for growth factors was as follows: VEGF, 5.9 pg/mL; EGF, 8.6 pg/mL; FGF-2, 8.7 pg/mL; PDGF-AA, 5.2 pg/mL; PDGF-BB, 7.31 pg/mL; TGFα, 8.6 pg/mL; Flt-3 L, 22.9 pg/mL; Granzyme B (GZB), 4.9 pg/mL; PDL-1, 69.3 pg/mL; TRAIL, 22.5 pg/mL.

Statistical analyze

We used the linear regression curve fit model for the consideration of the values below or above the detection limit and further we normalized the data by performing the normality test prior to statistical analysis. Central tendency were measured using Geometric means. Nonparametric Mann-Whitney U test was used to compare the LTBI+ versus LTBI- to identify the statistical significant differences. Multiple comparisons were corrected using the Holm's correction. Data analyses were done using GraphPad PRISM version 9 (GraphPad Software, Inc., San Diego, CA, USA). Principle Component Analysis (PCA) was applied to distinguishing possibly significant trends of cytokines, chemokines, acute phase proteins and growth factors, which are dependable for any of the clustering/separation between LTBI+ versus LTBI- groups. JMP14 software was used to plot Principle Component Analysis (PCA).

Ethics statement

The study was approved by the Ethics Committees of NIRT (NIRT-INo:2020010). Informed written consent was received from all study individuals. The study is part of the clinical study entitled, Study to evaluate the effectiveness of the BCG vaccine in reducing morbidity and mortality in elderly individuals in COVID-19 hotspots in India (NCT04475302).

Results

Study population characteristics

The study population demographics and clinical characteristics are shown in Table I. There was no significant difference in age or sex or clinical characteristics between the study groups.

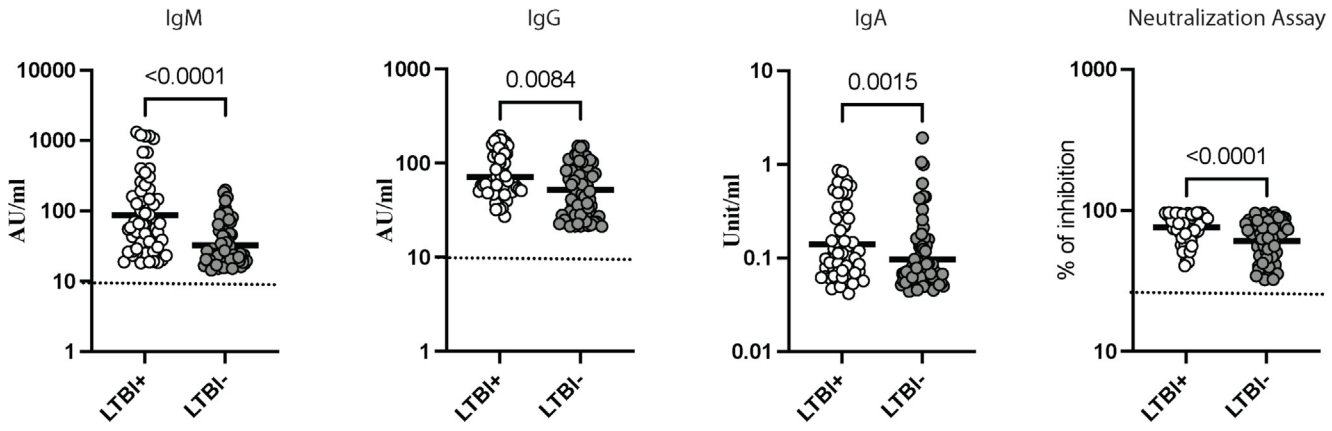

Fig. 1. LTBI with seropositive SARS-CoV-2 infection is associated with heightened levels of IgA, IgM, IgG and neutralizing antibodies The plasma levels of SARS-CoV-2 spike protein specific IgM and IgG, N protein specific IgA and neutralizing antibodies were measured in LTBI+ ($n = 61$) and LTBI- ($n = 72$) individuals. The data are represented as scatter plots with each circle representing a single individual. p values were calculated using the non-parametric Mann Whitney *U* test and with Holms correction for multiple comparisons.

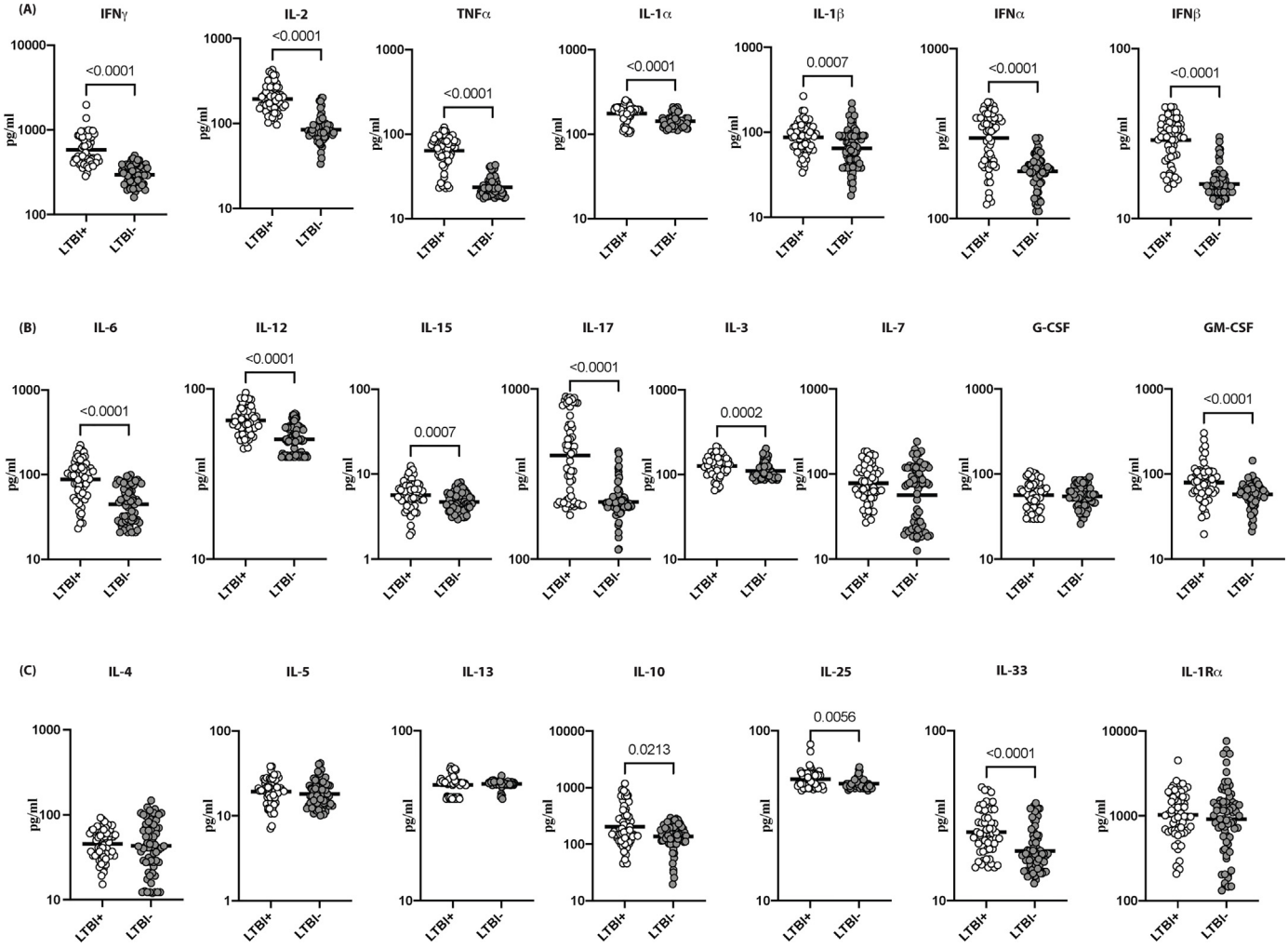

Fig. 2. LTBI with seropositive SARS-CoV-2 infection is associated with heightened systemic levels of pro and anti-inflammatory cytokines (A) The plasma levels of Type 1 cytokines, IL-1 family and IFNα and β cytokines were measured in LTBI+ ($n = 61$) and LTBI- ($n = 72$) individuals with seropositive SARS-CoV-2 infection. (B) The plasma levels of inflammatory cytokines, IL-17 cytokines were measured in LTBI+ ($n = 61$) and LTBI- ($n = 72$) individuals seropositive SARS-CoV-2 infection. (C) The plasma levels of Type 2 cytokines, and other anti-inflammatory cytokines were measured in LTBI+ ($n = 61$) and LTBI- ($n = 72$) individuals seropositive SARS-CoV-2 infection. The data are represented as scatter plots with each circle representing a single individual. *P* values were calculated using the non-parametric Mann Whitney *U* test and with Holms correction for multiple comparisons.

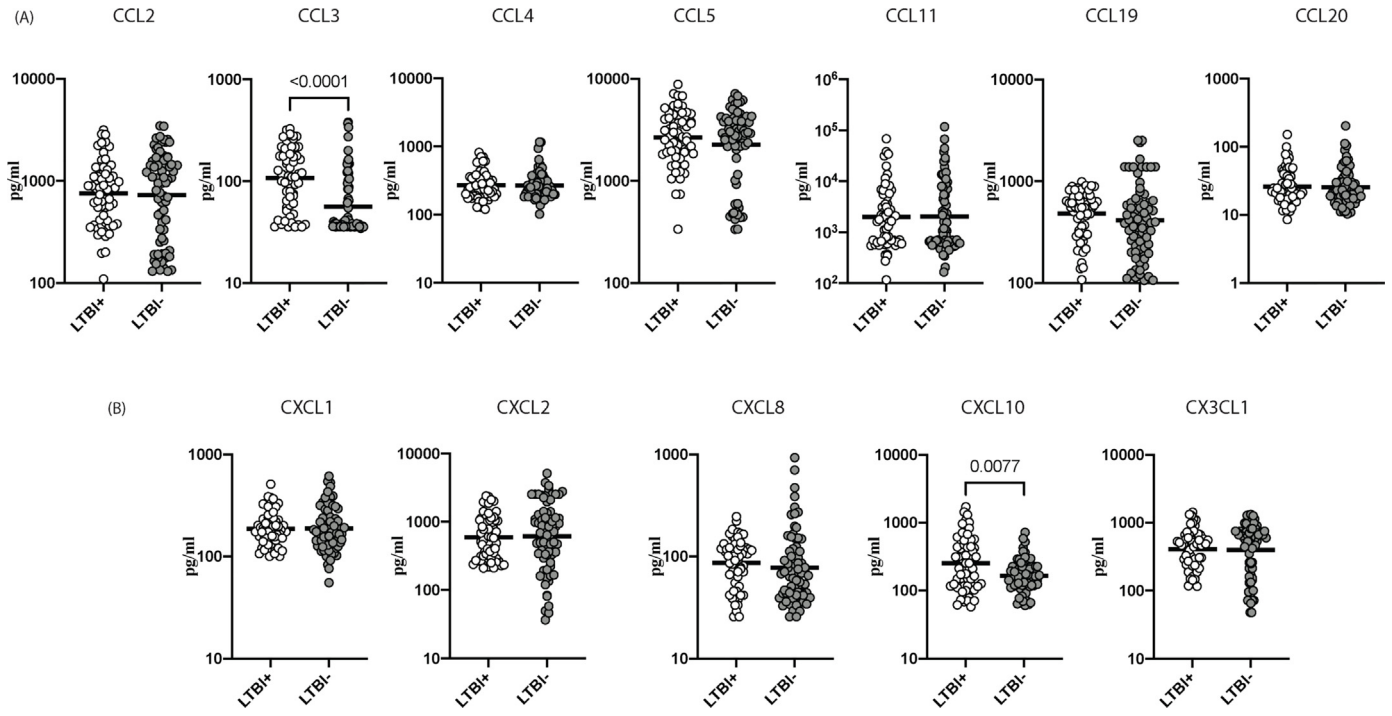

Fig. 3. LTBI with seropositive SARS-CoV-2 infection is associated with heightened systemic levels of CCL3 and CXCL10 chemokines (A) The plasma levels of CC chemokines were measured in LTBI+ (n = 61) and LTBI- (n = 72) individuals seropositive SARS-CoV-2 infection. (B) The plasma levels of CXC chemokines were measured in LTBI+ (n = 61) and LTBI- (n = 72) individuals seropositive SARS-CoV-2 infection. The data are represented as scatter plots with each circle representing a single individual. *P* values were calculated using the non-parametric Mann Whitney *U* test and with Holms correction for multiple comparisons.

LTBI with seropositive SARS-CoV-2 infection is associated with heightened levels of IgM, IgG and IgA binding antibodies as well as neutralization capacity

To estimate the impact of LTBI on humoral immunity in SARS-CoV-2 infection, we compared the plasma levels of SARS-CoV-2 specific IgM, IgG, IgA and neutralizing antibodies in seropositive SARS-CoV-2 infected individuals with or without LTBI. As illustrated in Fig. 1, LTBI+ individuals showed increased plasma levels of IgM (GM of 87.35 AU/ml in LTBI+ versus 32.30 AU/ml in LTBI-), IgG (GM of 71.04 AU/ml in LTBI+ versus 52.20 AU/ml in LTBI-), IgA (GM of 0.1406 Units/ml in LTBI+ versus 0.0967 Units/ml in LTBI-) and enhanced neutralizing antibody capacity (GM of 75.95% in LTBI+ versus 60.83% in LTBI-) in comparison to LTBI- individuals. Further, we wanted to determine the relationship between antibody levels and the IFNγ levels (QGIT values). As shown in S.Fig. 1 there was no significant correlation between the antibody levels and the IFNγ values. Thus, LTBI with seropositive SARS-CoV-2 infection is linked with enhanced humoral immune responses.

LTBI with seropositive SARS-CoV-2 infection is linked with elevated systemic levels of pro and anti-inflammatory cytokines

To estimate the impact of LTBI on cytokine responses in seropositive SARS-CoV-2 infection, we compared the plasma levels of pro and anti-inflammatory cytokines in seropositive SARS-CoV-2 infected individuals with or without LTBI.

As illustrated in Fig. 2A and Supplementary Table.1, LTBI+ individuals exhibited increased plasma levels of IFNγ, IL-2, TNFα, IL-1α and IL-1β in comparison to LTBI- individuals.

As illustrated in Fig. 2B and Supplementary Table.1, LTBI+ individuals exhibited increased plasma levels of IL-6, IL-12, IL-15, IL-17, IL-3 and GM-CSF in comparison to LTBI- individuals.

Finally, as illustrated in Fig. 2C and Supplementary Table.1, LTBI+ individuals exhibited increased plasma levels of IL-10, IL-25 and IL-33 in comparison to LTBI- individuals. Thus, LTBI with seropositive SARS-CoV-2 infection is linked with heightened systemic levels of pro and anti-inflammatory cytokines.

Table.1
Demographics of the study population.

	LTBI+	LTBI-
Subjects Enrolled	n = 61	n = 72
Age (Median)	64 (61–78)	63 (60–77)
Gender (M/F)	30 /31	45/32
Height (Median)	157 cm	153 cm
Weight (Median)	61 Kg	62 Kg
Pulse rate (Median)	90	92
Systolic Blood Pressure (Median)	135	140
Diastolic Blood Pressure (Median)	90	85
Saturation of Peripheral Oxygen (SpO2)% (Median)	98	97
QGIT	Positive	Negative
SARS-CoV-2 specific IgM	Positive	Positive
SARS-CoV-2 specific IgG	Positive	Positive

QGIT, QuantiFERON-TB® Gold In-Tube.

LTBI with seropositive SARS-CoV-2 infection is linked with heightened systemic levels of CCL3 and CXCL10 chemokines

To determine the influence of LTBI on chemokines in seropositive SARS-CoV-2 infection, we compared the plasma levels of CC and CXC chemokines in seropositive SARS-CoV-2 infected individuals with or without LTBI. As shown in Fig. 3A, LTBI+ individuals showed increased plasma levels of CCL3 (GM of 107.4 pg/ml in LTBI+ versus 56.20 pg/ml in LTBI-) and as illustrated in Fig. 3B, CXCL10 (GM of 253.5 pg/ml in LTBI+ versus 164.2 pg/ml in LTBI-) in comparison to LTBI- individuals. Thus, LTBI with seropositive SARS-CoV-2 infection is associated with heightened systemic levels of CCL3 and CXCL10 chemokines.

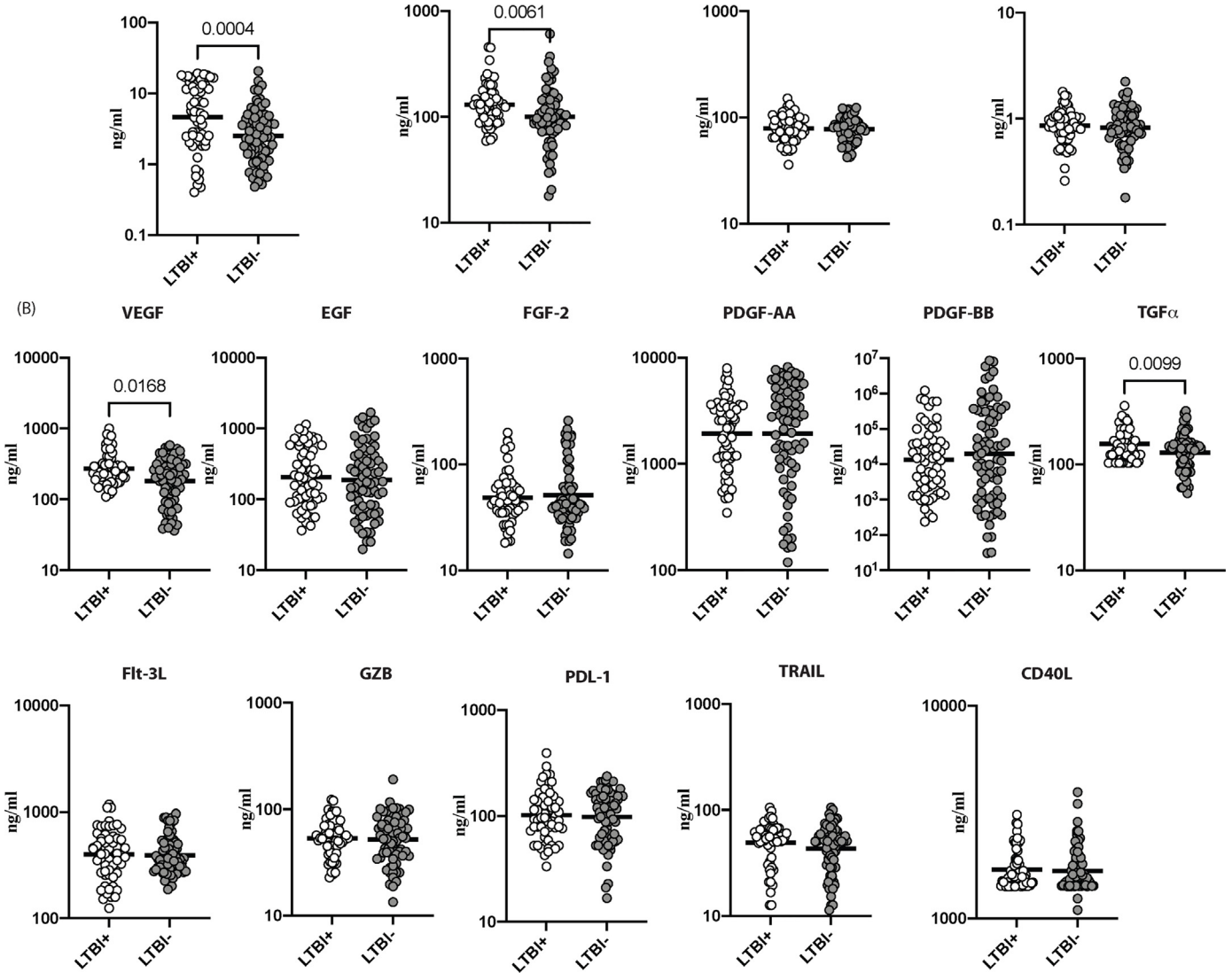

Fig. 4. LTBI with seropositive SARS-CoV-2 infection is associated with heightened systemic levels of acute phase proteins and growth factors The plasma levels of acute phase proteins were measured in LTBI+ ($n = 61$) and LTBI- ($n = 72$) individuals seropositive SARS-CoV-2 infection. (B) The plasma levels of growth factors were measured in LTBI+ ($n = 61$) and LTBI- ($n = 72$) individuals seropositive SARS-CoV-2 infection. The data are represented as scatter plots with each circle representing a single individual. *P* values were calculated using the non-parametric Mann Whitney *U* test and with Holms correction for multiple comparisons.

LTBI with seropositive SARS-CoV-2 infection is associated with heightened systemic levels of acute phase proteins and growth factors

To determine the influence of LTBI on systemic levels of acute phase proteins in seropositive SARS-CoV-2 infection, we compared the plasma levels of these markers in seropositive SARS-CoV-2 infected individuals with or without LTBI. As illustrated in Fig. 4A, LTBI+ individuals showed increased plasma levels of CRP (GM of 2.31 ng/ml in LTBI+ versus 1.3 ng/ml in LTBI-) and $\alpha-2$-M (GM of 130.9 ng/ml in LTBI+ versus 100.8 ng/ml in LTBI-) in comparison to LTBI- individuals.

Next, we wanted to determine the influence of LTBI on systemic levels of growth factors in seropositive SARS-CoV-2 infection, we compared the plasma levels of these markers in seropositive SARS-CoV-2 infected individuals with or without LTBI. As illustrated in Fig. 4B, LTBI+ individuals showed increased plasma levels of VEGF (GM of 270.7 ng/ml in LTBI+ versus 180.3 ng/ml in LTBI-) and TGFα (GM of 15.64 ng/ml in LTBI+ versus 12.90 ng/ml in LTBI-) in comparison to LTBI- individuals. Thus, LTBI with seropositive SARS-CoV-2 infection is associated with heightened systemic levels of acute phase proteins and growth factors.

Principle component analyses divulges patterns in cytokines, chemokines, acute phase proteins and growth factors

Principal components analyze (PCA) was used to visualize differences between the groups created on the entire data set. To visualize the clustering pattern of cytokines between SARS-CoV-2 positive with or without LTBI and LTBI+ denoted as red circle and LTBI- denoted as blue circle individuals, we performed PCA analysis with the cytokines which are statistically different (IFNγ, IL-2, TNFα, IL1α, IL1β, IFNα, IFNβ, IL-6, IL-12, IL-15, IL-17, IL-3, GM-CSF, IL-4, IL-10, IL-25 and IL-33) between LTBI+ and LTBI- individuals. After excluding the factors with commonalities as low as 0.3, Wwe assessed PCA-1 (IFNγ, IL-2, TNFα, IL1α, IL1β, IFNα, IFNβ, IL-6, IL-12, IL-15) and PCA-2 IL-17, IL-3, GM-CSF, IL-4, IL-10, IL-25 and IL-33). As illustrated in Fig 5A, PCA analysis showed that cytokines clusters differs between LTBI+ and LTBI- individuals with seropositive SARS-CoV-2 infection. The score plot of the first two components revealed 33.8% and 9.18% of overall variance, respectively.

Next we performed PCA analyze for the chemokines, acute phase proteins and growth factors which are statistically different. After excluding the factors with commonalities as low as 0.3,we as-

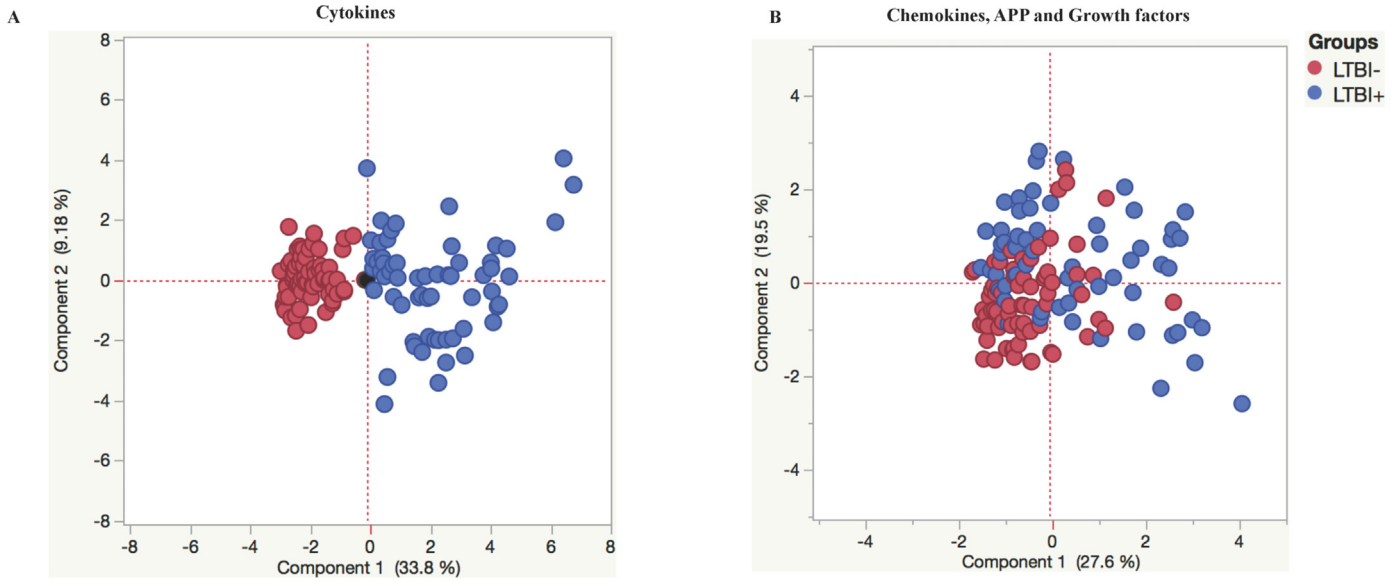

Fig. 5. Principle component analysis (PCA) divulges patterns in cytokines, chemokines, acute phase proteins and growth factors Principal component analysis (PCA) was done to exhibit the dissemination of data from the mixture of two groups LTBI+ (blue circles) and LTBI- (red circles). The PCA indicates the two principal components of variation. (A) PCA analysis was done with cytokines, between LTBI+ and LTBI- participants with seropositive SARS-CoV-2 infection. (B) PCA analysis of chemokines, acute phase proteins and growth factors between LTBI+ and LTBI- participants with seropositive SARS-CoV-2 infection (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.).

sessed PCA-1 (CCL3, CXCL-10) and PCA-2 as (CRP, α-2-M, VEGF and TGFα. As illustrated in Fig 5B, PCA analysis of chemokines (CCL3, CXCL10), acute phase proteins (CRP and α-2-M) and growth factors (VEGF and TGFα) showed two different clusters between LTBI+ and LTBI- individuals. The score plot of the first two components revealed 27.6% and 19.5% of overall variance, respectively. Thus, PCA analysis revealed the overall effect of cytokines, chemokines, acute phase proteins and growth factors of LTBI+ and LTBI- individuals with seropositive SARS-CoV-2.

Discussion

Since both LTBI and asymptomatic SARS-CoV-2 infection are highly prevalent in South India, it provided us the opportunity to examine the impact of LTBI on the humoral and innate immune responses in these co-infected asymptomatic individuals. We first examined the effect of LTBI on SARS-CoV-2 specific IgM, IgG and IgA binding antibodies as well as SARS-CoV-2 induced neutralizing antibodies. Our data clearly demonstrates elevated levels of binding antibodies and enhanced neutralizing antibody capacity in LTBI/SARS-CoV-2 co-infected individuals. Previous studies have shown that SARS-CoV-2 antibodies are induced in asymptomatic individuals but at a lower magnitude.[20] This has been taken as evidence for a weak adaptive humoral immunity in these individuals. Our data demonstrate that the neutralizing antibody capacity of asymptomatic individuals is not dampened but rather enhanced in the presence of LTBI co-infection. A meta-analysis of 23,320 individuals revealed that SARS-CoV-2 co-infection increased the risk of mortality in TB by a factor of 2.1 (Relative risk, 2.10, Confidence interval, 1.75–2.51).[21] Whether LTBI or active TB influences the clinical manifestations, disease severity or immune responses in COVID-19 is not well understood. Asymptomatic infection constitutes a variable but large proportion of SARS-CoV-2 infection and asymptomatic individuals are often considered to be a major source of transmission.[22] The enhanced capacity to produce both binding and neutralizing antibodies by LTBI individuals suggests that LTBI might offer a protective role against development of severe disease, although this remains to be formally proven.

The early onset of protective cytokines responses, which includes Type 1 cytokines such as IFNγ, TNFα and IL-2 as well as Type 17 cytokines such as IL-17 and Type I IFNs such as IFNα and IFNβ appears to be the key event in protection against infection as well as severe disease.[23] Asymptomatic patients appear protected from this manifestation perhaps due to their ability to mount protective cytokine responses early.[24] Our study clearly demonstrates that coexistent LTBI is associated with heightened plasma levels of a variety of protective cytokines, including IFNγ, IL-2, TNFa, IL-1a, IL-1β, IFNα, IFNβ, IL-6, IL-12, IL-17 and GM-CSF. In addition, LTBI is also associated with a concomitant increase in plasma levels of Type 2 and regulatory cytokines such as IL-4, IL-10, IL-25 and IL-33. Whether the increase in these cytokines is a compensatory response to the enhanced levels of pro-inflammatory cytokines remains to be determined. Of interest, Type 1 IFN levels were also elevated in LTBI/SARS-CoV-2 infected individuals. Thus, the capacity to produce protective cytokines appears to be enhanced in LTBI individuals, which again might be a potential mechanism to combat infection and morbidity.

Protection against COVID-19 is also dependent on the increased and early production of chemokines that exhibit the capacity to attract innate and adaptive immune cells to the lung.[25] Our data reveal that CCL3 and CXCL10 levels were significantly higher in LTBI/SARS-CoV-2 co-infected individuals suggesting that LTBI is associated with enhanced chemokine responses in addition the cytokine responses. Thus, the occurrence of elevated cytokine and chemokine levels might predispose asymptomatic individuals to protection against symptomatic disease in the presence of LTBI.

Heightened levels of acute phase proteins are a major hallmark of COVID-19.[26] CRP is both a diagnostic and prognostic marker of morbidity and mortality in COVID-19.[27] Other acute phase reactants such as α-2-M, SAP and Hp are less well studiedin SARS-CoV-2 infection. Our data on co-infected individuals suggests that LTBI is associated with elevated levels of CRP and $\alpha-$2-M. Finally, LTBI is also associated with enhanced levels of two other biomarkers of disease pathogenesis in COVID-19 – VEGF and TGFα.[28] Our study also reveals that cytokines for the most part (and chemokines, acute phase proteins and growth factors to a lesser extent) are significantly associated with LTBI positivity.

Thus, our study clearly demonstrates an important influence of LTBI on the humoral and innate immune responses in SARS-CoV-2 infection. Our study offers evidence for a positive protective effect of LTBI on both innate and adaptive immune responses in asymptomatic SARS-CoV-2 individuals. The enhanced binding and neutralizing antibody levels combined with enhanced levels of protective cytokines and chemokines could potentially contribute to decreased susceptibility to morbidity and mortality in COVID-19. Our study is limited by examination of LTBI in only the asymptomatic groups of individuals. Further examination of the influence of LTBI on COVID-19 patients with mild, moderate and severe disease should shed more insight on the interaction between these two major infections. Our study also examines only association and not causation. Nevertheless, our data provide a plausible mechanistic explanation for a positive effect of LTBI on SARS-CoV-2- infection and calls for more clinical and basic research studies on this interaction.

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CRediT authorship contribution statement

Designed the study (S.B., C.P); conducted experiments (A.R., N.P.K., A.N., N.S., S.M., R,M.R, V.V); acquired data (A.R., N.P.K.); analyzed data (A.R., N.P.K.); contributed reagents and also revised subsequent drafts of the manuscript (S.B. C.P.); responsible for the enrolment of the participants and also contributed to acquisition and interpretation of clinical data (C.P., K.K., S.B.M., T.C.A.); wrote the manuscript (A.R. and S.B). All authors read and approved the final manuscript.

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Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.jinf.2021.07.029.

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