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REVIEW – OPEN ACCESS

COVID-19 Disease and Interferon-γ: Has it a Protective Impact on Mortality?

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ABSTRACT

The complex coincidence of several immunopathological, socio-cultural, and health infrastructure factors may affect the COVID-19 related mortality among different populations. The impact of the age on disease progression has been confirmed in several studies. Recently limited ecological and clinical studies have sparked controversy among researchers about the protective impact of the non-specific effect of routinely used Bacille Calmette-Guerin (BCG), Hepatitis A virus (HAV), and influenza (Flu) vaccines or their natural infections against COVID-19. In the present study, variables, including BCG vaccination coverage, HAV prevalence, and population age distributions, from 59 countries were analyzed to examine their potential association with COVID-19 infection and related mortality rate. Concerning COVID-19 cases/million population (1MP) and mortality, there are significant differences between countries with and without BCG vaccination programs (p-value <0.001). A significant negative correlation between both BCG coverage and HAV prevalence with COVID-19 related mortality was also found (r (59)=-0.4, p-value <0.05), (r (59) =-0.3, p-value <0.01). Based on the results of the present study, previous ecological analyses and available epidemiological evidence, along with knowledge of the immune response to BCG, HAV and influenza vaccination, as well as COVID-19 infection progression, the current study suggest a hypothesis that IFN-γ induced immune response which could be triggered by BCG, HAV, and flu vaccination or natural infections may have a protective effect against COVID-19 related mortality.

Keywords: COVID-19, Interferon γ, mortality, BCG, HAV

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INTRODUCTION

In late 2019, a novel coronavirus infection later named the COVID-19 epidemic suddenly hit Wuhan, China, and quickly spread all around the world, leading to a global pandemic. High transmissibility among population and fatalities rate in specific high-risk groups made this novel infection a high impact health threat (1). Approximately 70% of the identified cases are between the ages of 30–69 years. More than 80% of patients who have died are over 60 years old and more than 75% of cases with death end had an underlying medical condition, such as Cardiovascular disease (CHF), diabetes, chronic respiratory disease, hypertension and cancer (2, 3). However, the impact of the disease is different among countries in different regions. The mortality varies from less than 0.05% to approximately 15.5% (4–6). Surprisingly, the mortality rate reported from countries with the fragile health systems is meaningfully low. Although differences in cultural norms, mitigation efforts, and health infrastructure highly impact on morbidity and mortality, several immunopathological factors also may contribute to making differences in pandemic features in various regions (4, 5).

The impact of the age on disease progression has been confirmed in several studies (7–11). Recently, limited ecological studies have sparked controversy among researchers about the protective impact of the non-specific effect of routinely used BCG vaccines against COVID-19 (4, 5). A similar claim has been made about the protective impact of hepatitis A and influenza vaccines (12, 13). However, several trials are running to investigate the potential influence of BCG vaccination and Anti-HAV presence on COVID-19 morbidity and mortality. The current review analyzes the available data to discuss the claims and offers an alternative hypothesis to clarify the issue.

MATERIALS and METHODS

In line with the study's aim, 59 countries from all over the world were included in this study. The vaccination schedules of countries were analyzed. This study makes use of the BCG World Atlas, a compendium of BCG vaccination policies in over 180 countries compiled by McGill University (14). Based on the presence or absence of BCG vaccination in the routine neonate vaccine schedules, countries were classified into two groups. Confirmed numbers of the COVID-19 cases and deaths until the middle of May 2020 were obtained from the Real-time Statistics Project Worldometers (6). Also, the HAV prevalence levels in different regions were obtained from

the centers for disease control and prevention (CDC) data sources (15). Annual flu death per 100,000 population data was also retrieved from the global map, which is pulled country by country from world health organization (WHO) (16) (Appendix 1).

Data analysis was performed using IBM SPSS 24 statistical program. Pearson correlation was used to analyze the correlation between COVID-19 cases/1 MP, and related death rate with BCG vaccination coverage variable. For the comparison of the COVID-19 cases/1 MP and mortality rate differences between countries that applied the BCG vaccination, and those who did not, an independent T-test was applied. For the assessment of the potential correlation between HAV prevalence level and Covid-19 mortality, nonparametric correlations (spearman rank correlations) were applied.

RESULTS

The countries that have the BCG vaccination schedule, on average, cover 92% of the total area of the country. The COVID-19 case/1MP and mortality rates showed significant differences (P-value <0.001) between countries that have a universal BCG vaccination program and the countries that have not. The mean of case/1MP and death rate among the countries that have not a BCG vaccination program were 2129.5 cases/1MP and 7.2% respectively. In contrast, the mean among countries with BCG vaccine schedule was significantly lower and was 680.54/1 MP and 3.6%, respectively.

A significant negative correlation between BCG coverage and COVID-19 mortality rate was found (r (59) =-0.4, P-value <0.05). The mean of the cumulative relative frequency of population over 60 years old in countries with and without universal BCG immunization was 25% and 15%, respectively. Also, a significant correlation between the cumulative relative frequency of the population over 60 years old and the mortality of COVID-19 was detected (r (59)=0.4, P-value <0.01). In the same way, there is a weak negative correlation between HAV prevalence level and death rate of Covid-19 (r (59)=-0.3, P-value <0.01), which means with the increase of HAV prevalence, the death rate of COVID-19 decreases.

DISCUSSION

In the current study, results demonstrate a significant difference between countries with and without a universal BCG immunization program concerning the COVID-19 case number and related mortality. Besides, a negative correlation between BCG vaccine coverage and mortality supports the idea that increasing population immunity level using BCG, leads to decreasing COVID-19 related mortality. Previous similar ecological studies obviously confirm these differences.

An ecological study conducted by Miller et al. (4) for the first time suggested that differences in BCG vaccination policies and practices may partially explain different mortality rates from COVID-19 between countries with and without universal BCG vaccination programs. Some researchers reported on a possible association between BCG vaccination and protection against severe disease and fatal outcome from SARS-CoV-2 infection.

Gursel et al. (5) later expanded the hypothesis that countries with continuing BCG immunization programs would pass the pandemic slightly less severe than those that did not have or have stopped their national BCG vaccination programs. Cases/MP and COVID-19-associated deaths/million in population with universal BCG vaccination were significantly lower than those that did not have/ceased their BCG vaccination programs (p-value <0.0001)

Ozdemir et al. (17) also reported similar results from their ecological analysis. Besides, the mean case and deaths per population ratio are also significantly higher in Northern hemisphere regions, comparing to the regions located in the Southern hemisphere (p-value <0.05). The mean case/death per population ratio among northern hemisphere regions was also significantly lower in countries with current national BCG vaccination programs comparing to non-applied countries.

Significant variations in COVID-19 cases between countries that have high and low tuberculosis incidence were shown by Madan et al. (18) where also a high BCG vaccination coverage associated with a lower incidence of COVID-19 was reported in their study.

BCG Vaccination in the first month of birth effectively protects infants and young children against life-threatening disseminated forms of TB, including TB meningitis and miliary TB (16, 19, 20). In addition, non-specific immunological effects of the BCG vaccine contribute to regulating the immune response and decreased susceptibility against subsequent infections caused by other pathogens, especially acute respiratory tract infections, through the induction of innate immune memory termed trained immunity, and heterologous lymphocyte activation, which leads to increased cytokine production, macrophages activity, T-cell responses, and antibody titers (21–23).

The findings of this research are in line with previous ecological studies. The results of these studies, along with the supportive epidemiological evidence, propose the presence of potential TB/BCG related protective mechanisms against COVID-19. Evidences from epidemiological studies relatively support the hypothesis. In South Africa, a relative impact of BCG vaccination on respiratory tract infections in adolescents by 73% reduction was shown (24). In Guinea-Bissau, a high mortality record, vaccination with BCG led to a 38% reduction of neonatal mortality from various diseases generally. In addition, the BCG vaccine causes yellow fever vaccine viremia reduction in 71% of volunteers, a virus with a similar genomic structure (25). Besides, BCG vaccination was previously used (once a month for three consecutive months) to produce a significant reduction in the prevalence of upper respiratory tract infections in elderly people (26). On the other hand, this study revealed a weak negative correlation between HAV prevalence level and mortality rate of COVID-19 (r (59) =0.4, P-value <0.01). This result is in line with the study of Sarialioglu et al.'s (12) study where the rate of COVID-19 among hemodialysis patients was investigated. As a result of this study, they found that the rate of COVID-19 infection among their patients was very low. Since 94.7% of patients were shown to be HAV antibody-positive, in the subsequent analysis, they suggested that the existence of Anti-HAV may take a protective role against COVID-19. Besides, Sarialioglu et al. (27), in another study, also showed a significant increase in COVID-19 mortality among countries with high HAV susceptibility. It makes it

clear that a higher prevalence of seropositivity of Anti-HAV either acquired by vaccination or natural infection may lead to lower mortality among COVID-19 infected individuals.

Current study data show that flu, another virus with similar quality of immune response with SARS CoV-2, mainly hit the southern hemisphere countries. Salem et al. (28) claimed that the quality and quantity of the immune performance that is shaped by the history of infections and vaccination against flu may minimize the severity of COVID-19 and contribute to explaining the differences in infection severity and susceptibility in different regions. This hypothesis is supported by the evidence of immunological cross-reactivity between flu and coronavirus due to the similarity in their structures, and subsequently, similarity in the quality of immunity toward both viruses (29–31). In addition to the cross-reactivity effect, the anti-flu immune responses can induce bystander immunity, which may trigger an immune response against other viral infections (13).

The impact of the population age distribution factor on the mortality rate is also considered in this work. Several studies obviously approved high age as a risk factor for severe COVID-19 and related mortality (7–11). Although ecological analysis alone provides a hypothesis that non-specific effects of BCG, HAV, and Flu vaccines may lead to a protective impact on COVID-19, several confounder factors, such as the demographic structure of countries, may interfere and challenge these conclusions. However, the demographic structure of the population alone cannot explain significant differences in the pandemic mortality rate between countries.

It is believed that several immunopathological factors may affect COVID-19 mortality among different populations. A rapid and well-coordinated innate immune response with the high collaboration of cytokines as the first line of defense plays an important role in immunopathology during viral infection. However, excessive immune responses, "cytokine storm", have been detected in critical patients with COVID-19, which lead to acute respiratory distress syndrome (ARDS) and multiple organ failure, which ends in death within a short time (32).

A cytokine storm syndrome is characterized by an increase in IL-6,IL-10,TNF-α,IL-2,MCP, IL-7, IP-10, granulocyte-colony stimulating factor (G-CSF), CXCL10, MCP-1, and macrophage inflammatory protein 1 alpha (MIP1A) (33, 34). Besides, lymphopenia (in CD4+ and CD8+ T cells) and decreased IFN-γ expression in CD4+ T cells showed an association with severe COVID-19 in several studies (33, 35). However, in some cases, increase in IFN-γ levels in the peripheral blood were detected in the severe cases compared to those in the mild cases (36), findings in children which were mainly affected by a mild form of the disease show increased IL-6, IL-10, and IFN-γ (37). It seems that IFN-γ mainly decreases in the severe form of COVID-19.

IFN-γ produced by CD4 T cells is a critical mediator in response to BCG, Hepatitis A and influenza vaccine/natural infection. Research shows BCG and HAV vaccination induces a high level of IFN-γ (38–43). Moreover, IFN-γ production is critical for viral clearance and the development of adaptive immune responses (44, 45). Interferon-induced transmembrane proteins (IFITMs) and tripartite motif-containing proteins (TRIMs) also contribute to the anti-viral immunity response. IFITMs play a role in protection against some viruses such as influenza-A, Flaviviruses, HIV-1, Ebola virus and coronavirus through restrict viral entry into the host cells (43).

Vaccination against BCG and high endemicity of viral infections, such as hepatitis A and influenza, which subsequently leads to a higher level of IFN-γ among the population of countries with lower COVID-19 mortality, may contribute to explain the mortality differences between countries. However, epidemiological studies alone cannot conclusively support the hypothesis, and lack of clinical evidences is the major limitation of this study. Further investigations are required to support this hypothesis.

CONCLUSION

IFN-γ induced immune response pathways induced by BCG, HAV, and flu vaccination and natural infection may trigger a protective effect against COVID-19 disease and mortality.

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Appendix 1. Data summary table

Countries	Income	Region	Current BCG	BCG coverage	COVID-19 cases/1MP	COVID-19 death/1MP	COVID-19 death rate	Cumulative relative frequency of population >60 years old	HAV prevalence level	Annual flu death/100 kp
USA	High	North America	No	*	3589.0	207	5.77	22.7	Very low	10.59
Canada	High	North America	No	*	1576.0	98	6.22	24.6	Very low	9.66
Italy	High	Europe	No	*	3485.0	478	13.72	29.8	Very low	4.89
Notherlands	High	Europe	No	*	2368.0	295	12.46	26.5	Very low	17.35
Belgium	High	Europe	No	*	4306.0	677	15.72	25.4	Very low	18.69
Newzealand	High	Pacific	No	*	308.0	4	1.30	22.1	Very low	8.39
Australia	High	Pacific	No	*	268.0	4	1.49	21.7	Very low	5.74
Norway	High	Europe	No	*	1447.0	39	2.70	23.1	Very low	18.33
Sweden	High	Europe	No	*	2210.0	265	11.99	25.9	Very low	8.04
Finland	High	Europe	No	*	948.0	42	4.43	28.9	Very low	3.77
Denmark	High	Europe	No	*	1644.0	84	5.11	25.9	Very low	19.42
Germany	High	Europe	No	*	1977.0	82	4.15	28.1	Very low	9.86
Czech	High	Europe	No	*	727.0	23	3.16	26.1	Very low	15.85
Slovakia	High	Europe	No	*	258.0	4	1.55	23.2	Very low	25.27
Austria	High	Europe	No	*	1732.0	66	3.81	23.8	Very low	5.70
Switzerland	High	Europe	No	*	3455.0	204	5.90	25.0	Very low	6.37
UK	High	Europe	No	*	2749.0	419	15.24	24.3	Very low	23.43
France	High	Europe	No	*	2584.0	381	14.74	26.7	Very low	10.01
Spain	High	Europe	No	*	5285.0	540	10.22	26.3	Very low	9.43
Ecuador	Upper middle	South America	No	*	1674.0	89	5.32	11.0	Intermediate	43.54
Ireland	High	Europe	Yes	94	4355.0	264	6.06	19.5	Very low	16.42
Portugal	High	Europe	Yes	95	2479.0	102	4.11	29.4	Very low	25.25
Poland	High	Europe	Yes	94	362.0	18	4.97	25.9	Low	16.11
Hungary	High	Europe	Yes	100	314.0	36	11.46	26.8	Low	6.46
Estonia	High	Europe	Yes	90	1282.0	41	3.20	26.7	Low	8.64
Latvia	High	Europe	Yes	96	475.0	8	1.68	27.5	Low	7.54
Rassia	Upper middle	Europe	Yes	95	923.0	9	0.98	22.3	Low	17.89
Belarus	Upper middle	Europe	Yes	98	1768.0	10	0.57	22.6	Low	10.04
Romania	Upper middle	Europe	Yes	90	684.0	41	5.99	25.8	Low	20.77
Bulgaria	Upper middle	Europe	Yes	98	235.0	11	4.68	28.3	Low	16.78
Ukraine	Upper middle	Europe	Yes	45	282.0	7	2.48	23.6	Low	8.64
Macedonia	Upper middle	Europe	Yes	95	725.0	40	5.52	20.7	Low	4.60
Turkey	Upper middle	Europe	Yes	94	1495.0	40	2.68	12.9	Intermediate	13.84

Appendix 1 (cont.). Data summary table

Countries	Income	Region	Current BCG	BCG coverage	COVID-19 cases/1MP	COVID-19 death/1MP	COVID-19 death rate	Cumulative relative frequency of population >60 years old	HAV prevalence level	Annual flu death/100 kp
Iran	Upper middle	Middle East Asia	Yes	100	1160.0	74	6.38	10.3	Intermediate	17.23
Saudi Arabia	High	Middle East Asia	Yes	98	776.0	5	0.64	5.8	Intermediate	44.89
Afghanistan	Low	South Asia	Yes	46	69.0	2	2.90	4.2	high	97.78
Pakistan	Lower middle	South Asia	Yes	95	91.0	2	2.20	6.6	high	62.83
India	Lower middle	South Asia	Yes	99	31.0	1	3.23	10.0	high	47.80
Bangeladesh	Lower middle	South Asia	Yes	99	57.0	1	1.75	7.8	Intermediate	63.12
Kazakhstan	Upper middle	Central Asia	Yes	95	211.0	1	0.47	12.0	Intermediate	21.16
Mongolia	Lower middle	Central Asia	Yes	99	12	0	0.00	7.2	Intermediate	29.63
Uzbekistan	Lower middle	Central Asia	Yes	98	65.0	0.3	0.46	8.2	Intermediate	27.94
Tajikistan	Lower middle	Central Asia	Yes	98	13.0	0.2	1.54	5.6	Intermediate	47.83
China	Upper middle	East Asia	Yes	95	58.0	3	5.17	17.2	Low	15.10
Japan	High	East Asia	Yes	98	118.0	4	3.39	34.4	Very low	32.07
S. Korea	High	East Asia	Yes	85	211.0	5	2.37	23.1	Very low	16.34
Malaysia	Upper middle	East Asia	Yes	95	195.0	3	1.54	11.0	Low	61.07
İndonesia	Lower middle	East Asia	Yes	92	41.0	3	7.32	10.0	Low	43.16
Tahiland	Upper middle	East Asia	Yes	99	43.0	0.8	1.86	19.0	Low	60.40
Egype	Lower middle	North Africa	Yes	95	63.0	4	6.35	8.2	Intermediate	21.06
Algeria	Upper middle	North Africa	Yes	95	102.0	11	10.78	10.0	Intermediate	29.19
South Africa	Upper middle	Sub-Saharan Africa	Yes	91	114.0	2	1.75	8.5	High	59.33
Cameron	Lower middle	Sub-Saharan Africa	Yes	95	78.0	2	2.56	4.2	High	181.62
Mexico	Upper middle	North America	Yes	95	182.0	17	9.34	11.2	Intermediate	19.92
Brazil	Upper middle	south America	Yes	87	479.0	33	6.89	14.1	Intermediate	43.59
Argantine	Upper middle	south America	Yes	95	106.0	5	4.72	15.8	Intermediate	40.06
UAE	High	Middle East Asia	Yes	95	1432.0	13	0.91	3.0	Intermediate	16.87
Qatar	High	Middle East Asia	Yes	95	5398.0	4	0.07	3.5	Intermediate	8.22
Iraq	Upper middle	Middle East Asia	Yes	95	57.0	2	3.51	5.0	Intermediate	24.43

All 59 countries which were included in the study, the vaccination schedules of countries, the confirmed numbers of COVID-19 cases and deaths until the middle of May 2020, the HAV prevalence levels, annual flu death per 100.000 population, and relative frequency of population higher than 60 years olds in each country, were summarized in the supplementary table