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Short Communication

Absence of SARS-CoV-2 antibodies in pre-pandemic plasma from children and adults in Vietnam ☆,☆☆

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ABSTRACT

We tested pre-pandemic (2015—2019) plasma samples from 148 Vietnamese children and 100 Vietnamese adults at high risk of zoonotic infections for antibodies against SARS-CoV-2 nucleocapsid and spike proteins. None was positive. The data thus demonstrated no evidence of prior serological cross-reactivity with SARS-CoV-2 that might explain the low numbers of COVID-19 in Vietnam. No pre-existing cross-reactivity might explain Vietnam success of COVID-19 control.

Introduction

SARS-CoV-2 emerged in late 2019 and is the cause of the ongoing COVID-19 pandemic. Yet, according to the World Health Organization, as of the end of February 2021, countries in the Western Pacific region, including Vietnam, have reported only a small fraction of the global COVID-19 cases (who.int). In Vietnam, this was helped by early preparedness and proactive responses encompassing timely border closure, physical distancing (including mask-wearing in public), contact tracing and testing, coupled with the isolation of infected cases and their direct contacts. However, another hypothesis is that there may be pre-existing immunity among the population in the region through exposure to SARS-CoV-2 related viruses. Knowledge of this might help explain why the burden posed by SARS-CoV-2 and the incidence of COVID-19 cases vary significantly across the world. It may also further shed light on the natural course of the infection.

Materials and Methods

A total of 148 young Vietnamese children with hand, foot and mouth disease (Nhan et al., 2020) and 100 Vietnamese adults were included for analysis of antibodies responses against SARS-CoV-2. The latter group was in close contact with domestic and/or wild animals and was thus at high risk for zoonotic infections, as detailed elsewhere (Nguyen et al. 2020). Of the 148 children, 8 children had reverse transcriptase-polymerase chain reaction (RT-PCR) confirmed evidence of current infection with either human coronavirus NL63 (HCoV-NL63) (n=2) or human coronavirus OC43 (HCoV-OC43) (n=6), and one adult had HCoV-OC43 detected in an earlier sample by RT-PCR. We used plasma samples collected at baseline and 2 years later for the adult cohort because they were part of a longitudinal cohort (Nguyen et al. 2020). We used

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Table 1
Demographics and animal contacts of the study participants

Variables	Children, N=148	Adults, N=100
Age[#]	18 (1 - 153)	44 (21 – 72)
Female/male	89/59 (1.5)	32/68 (0.47)
Occupation, n (%)		
Farmers	NA	42 (42%)
Slaughters	NA	32 (32%)
Animal health workers	NA	26 (26%)
Geographic locations of study participants		
HCMC (Ho Chi Minh City)	62 (41.89%)	0
Other provinces[*]	86 (58.11%)	100 (100%)[Ψ]
Collection period	June, 2015 – July, 2019	March, 2013 – September, 2016
Activity/event at risk of zoonotic infection		
Raising domestic animals	NA	63 (63%)
Raising wild animals	NA	16 (16%)
Bitten by animals	NA	19 (19%)
Bloody injuries[**]	NA	42 (42%)

Note to Table 1: [#]in months, median (range) for children and in years, median (range) for adults
[Ψ] 50 from Dong Thap province and 50 from Dak Lak province Domestic animals include dogs, chicken, pigs, cats, duck, muscovy duck, pigeons, cattle, geese, goat, rabbits, buffalo and turkeys.Wild animals include wild pigs, deers, porcupines and monkeys.
[*] Other provinces include An Giang (8), Ba Ria Vung Tau (2), Bac Lieu (1), Ben Tre (4), Binh Duong (4), Binh Phuoc (2), Binh Thuan (1), Ca Mau (1), Can Tho (3), Dong Nai (3), Dong Thap (2), Kien Giang (2), Lam Dong (1), Long An (15), Quang Ngai (1), Soc Trang (1), Tay Ninh (13), Tien Giang (18), Tra Vinh (3) and Vinh Long (1). The geographic locations of these provinces can be found in Figure 1.
[**] Bloody injuries while working with animals.

admission plasma from each participant and convalescent plasma samples from 2 human coronavirus positive children for the children. Thus, in total, we included 350 plasma samples in the analysis. After collection, all plasma samples were divided into small aliquots and stored at ≤20 °C until analysis. We extracted information about demographics, occupation and animal contact from the metadata of the aforementioned original studies.

We measured antibodies against 2 main immunogens (the nucleocapsid (N) and spike (S) proteins) of SARS-CoV-2 using 2 well-validated sensitive and specific serological assays, namely Elecsys Anti–SARS–CoV–2 assay (Roche, Germany) (Ainsworth et al. 2020) and SARS-CoV-2 Surrogate Virus Neutralization Test (sVNT) (GenScript, USA) (Tan et al. 2020). The former is an electrochemiluminescence immunoassay using recombinant N protein for qualitative detection of pan immunoglobulin (Ig) (including IgG) against SARS-CoV-2 in 20 uL of plasma samples. The latter is a surrogate assay for measuring S protein receptor-binding domain (RBD)-targeting neutralizing antibodies (RBD-targeting NAbs) in 10 uL of plasma samples (Tan et al. 2020). The Elecsys assay had the sensitivity of 99.6% when validated on samples collected at ≥30 days post symptom onset (Ainsworth et al. 2020), while the sVNT had the sensitivity of 98.9% when validated on samples collected at ≥14 days post symptom onset (Tan et al. 2020). These 2 assays detected SARS-CoV-2 antibodies in plasma samples from 11 of 11 Vietnamese patients with PCR-confirmed SARS-CoV-2 infection collected 2–3 weeks after diagnosis (data not shown).

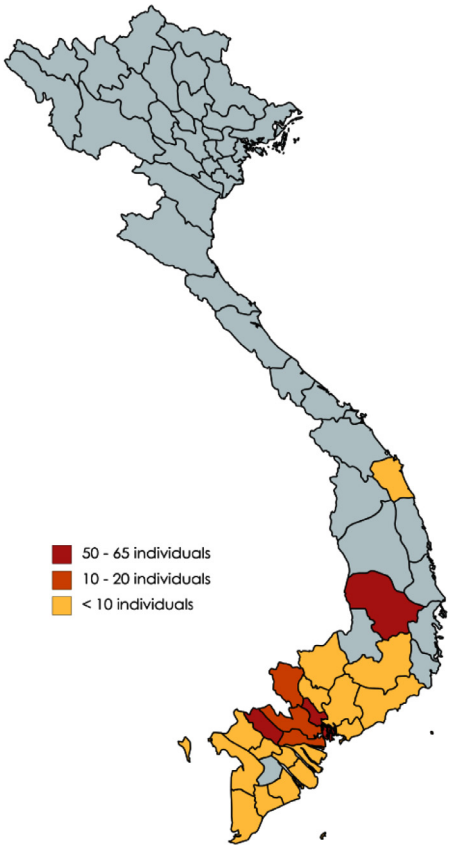

Figure 1. Map showing the geographic distributions of the study participants. (Inset map; https://mapchart.net).

Results

The 248 study participants came from various geographic locations in Southern Vietnam (Table 1 and Figure 1). They were all enrolled in the clinical studies between March 2013 and July 2019 (before the COVID-19 pandemic). Of the adult participants, farmers were predominant (n=42), followed by animal slaughterers (n=32) and animal health workers (n=26). The 148 children all had hand foot and mouth disease, and included 89 females and 59 males; the median age was 18 months.

None of the 350 plasma samples tested had detectable antibodies against N protein SARS-CoV-2. Additionally, RBD-targeting NAbs were not detected in 240 available plasma samples from the 100 adults and 38 children (including 10 plasma samples collected from 8 children positive for HCoV-NL63 or HCoV-OC43).

Discussion

Using in-house immunofluorescence assays, a recent study demonstrated that antibodies against S or N proteins were detected in 19% (n=105) and 14.1% (n=99) pre-pandemic plasma samples collected in Tanzania and Zambia, respectively (Tso et al., 2020). Cross-reactivity was strongly correlated with the presence of pre-existing antibodies against HCoV-NL63. Most recently, SARS-CoV-2 cross-reactive antibodies, especially neutralizing antibodies, were also detected in pre-pandemic plasma from SARS-CoV-2 uninfected individuals in the UK (Ng et al., 2020).

Human coronaviruses cause the common cold worldwide, with seroprevalence increasing with age (Dijkman et al., 2008). Thus, it is likely that in addition to the 9 individuals with RT-PCR evidence of human coronavirus infection, a proportion of the study participants were also exposed to these coronaviruses some time in the past. Therefore, previous exposure to known human coronaviruses alone might not determine the observed cross-reactivity in pre-pandemic plasma. Other possible contributing factors include the difference in assay performance and/or the heterogeneities between the study populations, which merits further research.

Cellular and humoral immunities are 2 major components of host responses. The former was not explored in the present study due to the unavailability of peripheral blood mononuclear cells. Of note, SARS-CoV-2-reactive T-cells were detected in 20% to 50% of blood collected from unexposed individuals from various geographic locations (Germany, Singapore, the Netherlands, the United States and the United Kingdom) (Grifoni et al. 2020; Le Bert et al. 2020; Mateus et al. 2020). How these pre-existing immunities correlate with protection remains unknown.

In summary, antibodies against SARS-CoV-2 N or S proteins were not detected in 350 pre-pandemic Vietnamese plasma samples. Future studies should look at pre-existing B-cell and T-cell memory in pre-pandemic samples, which might further shed light on the pathogenesis of the infection.

Conflict of Interest

None

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Ethical Approval

The institutional review board of collaborating hospitals in Vietnam and the Oxford Tropical Research Ethics Committee approved the clinical study.

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References

Ainsworth M, Andersson M, Auckland K, Baillie JK, Barnes E, Beer S, et al. Performance characteristics of five immunoassays for SARS-CoV-2: a head-to-head benchmark comparison. The Lancet Infectious Diseases 2020;20:1390–400. doi:10.1016/s1473-3099(20)30634-4.

Dijkman R, Jebbink MF, El Idrissi NB, Pyrc K, Muller MA, Kuijpers TW, et al. Human coronavirus NL63 and 229E seroconversion in children. J Clin Microbiol 2008;46:2368–73. doi:10.1128/JCM.00533-08.

Grifoni A, Weiskopf D, Ramirez SI, Mateus J, Dan JM, Moderbacher CR, et al. Targets of T Cell Responses to SARS-CoV-2 Coronavirus in Humans with COVID-19. Disease and Unexposed Individuals Cell 2020;181 1489-1501 e1415 doi. doi:10.1016/j.cell.2020.05.015.

Le Bert N, Tan AT, Kunasegaran K, Tham CYL, Hafezi M, Chia A, et al. SARS-CoV-2-specific T cell immunity in cases of COVID-19 and SARS, and uninfected controls. Nature 2020;584:457–62. doi:10.1038/s41586-020-2550-z.

Mateus J, Grifoni A, Tarke A, Sidney J, Ramirez SI, Dan JM, et al. Selective and cross-reactive SARS-CoV-2 T cell epitopes in unexposed humans. Science 2020;370:89–94. doi:10.1126/science.abd3871.

Ng KW, Faulkner N, Cornish GH, Rosa A, Harvey R, Hussain S, et al. Preexisting and de novo humoral immunity to SARS-CoV-2 in humans. Science 2020;370:1339–43. doi:10.1126/science.abe1107.

Nguyen TTK, Ngo TT, Tran PM, Pham TTT, Vu HTT, Nguyen NTH, et al. Respiratory viruses in individuals with a high frequency of animal exposure in southern and highland Vietnam. J Med Virol 2020;92:971–81. doi:10.1002/jmv.25640.

Nhan LNT, Khanh TH, Hong NTT, Van HMT, Nhu LNT, Ny NTH, et al. Clinical, etiological and epidemiological investigations of hand, foot and mouth disease in southern Vietnam during 2015 - 2018. PLoS Negl Trop Dis 2020;14. doi:10.1371/journal.pntd.0008544.

Tan CW, Chia WN, Qin X, Liu P, Chen MI, Tiu C, et al. A SARS-CoV-2 surrogate virus neutralization test based on antibody-mediated blockage of ACE2-spike protein-protein interaction. Nat Biotechnol 2020;38:1073–8. doi:10.1038/s41587-020-0631-z.

Tso FY, Lidenge SJ, Pena PB, Clegg AA, Ngowi JR, Mwaiselage J, et al. High prevalence of pre-existing serological cross-reactivity against SARS-CoV-2 in sub-Sahara. Africa Int J Infect Dis 2020. doi:10.1016/j.ijid.2020.10.104.