Infectious Diseases Diagnosis & Treatment
Katawa G, et al. Infect Dis Diag Treat 7: 242.
www.doi.org/ 10.29011/2577- 1515.100242
www.gavinpublishers.com

OPEN ACCESS

GAVIN PUBLISHERS

Research Article

Kinetics of Cytokines and Anti-SARS-CoV-2 Spike Glycoprotein S1 IgM and IgG1 in COVID-19 Infected Patients in Togo

Gnatoulma Katawa[1*], Wemboo Halatoko[2], Adjaho K. Koba[2], Christèle Nguepou Tchopba[1], Edlom P. Tchadie[1], Hombamane C. Simfele[1,2], Oukoe M. Amessoudji[1], Tomabu Adjobimey[3,4], Manuel Ritter[3], Ameyo M. Dorkenoo[5,6], Malewe Kolou[5]

[1]Unité de Recherche en Immunologie et Immunomodulation (UR2IM)/Laboratoire de Microbiologie et de Contrôle de Qualité des Denrées Alimentaires (LAMICODA)/Ecole Supérieure des Techniques Biologiques et Alimentaires (ESTBA), Université de Lomé, Lomé, Togo

[2]Institut National d'Hygiène, Lomé, Togo

[3]Institute for Medical Microbiology, Immunology and Parasitology (IMMIP), University Hospital Bonn (UKB), Bonn, Germany

[4]Département de Biochimie et de Biologie Cellulaire, Faculté des Sciences et Techniques (FAST), Université d'Abomey-Calavi, Abomey-Calavi, Benin

[5]Faculté des Sciences de la Santé (FSS), Université de Lomé, Lomé, Togo

[6]Division des Laboratoires, Ministère de la Santé et de l'Hygiène Publique, Lomé, Togo

***Corresponding author:** KATAWA Gnatoulma, Ecole Supérieure des Techniques Biologiques et Alimentaires (ESTBA)/ Université de Lomé, BP1515, Lomé, Togo.

Citation: Katawa G, Halatoko W, Koba AK, Tchopba CN, Tchadie EP, et al. (2023) Kinetics of Cytokines and Anti-SARS-CoV-2 Spike Glycoprotein S1 IgM and IgG1 in COVID-19 Infected Patients in Togo. Infect Dis Diag Treat 7: 242. DOI: 10.29011/2577-1515.100242

Received Date: 13 November 2023; **Accepted Date:** 21 November 2023; **Published Date:** 22 December 2023

Abstract

Background: COVID-19, caused by, had spread throughout the world. Its clinical manifestations ranged from asymptomatic to severe forms. The immunological markers that determine these forms are poorly investigated in Togo. This study aimed to investigate the dynamics of antibodies and cytokines in COVID-19 infected patients in Togo. **Methodology**: Sera from symptomatic (n = 40) and asymptomatic (n = 40) patients, collected at Day0, Day3 and Day10 post admission, were analyzed for SARS-CoV-2 specific IgM and IgG1 antibodies and IL-1β, IL-6, TNFα, IL-5, IL-10, IL-17A and IFNγ by sandwich ELISA. **Results:** In response to SARS-CoV-2 infection, IgM was expressed first followed by IgG1 in symptomatic patients. However, in asymptomatic patients, these antibodies were expressed simultaneously from the beginning of the infection with higher titers of IgG1. Except of IL-1β that increased from the admission to D10 all other cytokines were elevated at the admission and decreased to D10. Serum IL-6 levels were three times higher in symptomatic compared to asymptomatic patients. **Conclusion:** COVID-19 asymptomatic patients were characterized by high IgG1 antibodies titers whereas COVID-19 symptomatic patients, by high IL-6 levels.

Keywords: COVID-19; Antibodies; Cytokines; IgM; IgG; Togo

Introduction

In December 2019, a new coronavirus emerged in China and caused an acute respiratory illness known as coronavirus disease 2019 (COVID-19) [1]. The virus was identified as a beta-coronavirus related to the Severe Acute Respiratory Syndrome Coronavirus (SARS-CoV) and was therefore named SARS-CoV-2 [2]. In less than two decades, this virus is the third known coronavirus to cross the species barrier and cause severe respiratory infections in humans after SARS-CoV in 2003 and Middle East Severe Acute Respiratory Syndrome Coronavirus (MERS-CoV) in 2012; but with an unprecedented spread compared to the previous two viruses.

Due to the rapid increase in cases and uncontrolled spread worldwide, the World Health Organization declared SARS-CoV-2 a pandemic on March 11, 2020 [3]. By then, the virus had infected more than 118,000 people in 113 countries, with 4292 deaths [4]. In Togo, the first case was reported on March 6, 2020 and as of November 21, 2022; there were 39323 confirmed cases at COVID-19 with 290 deaths [5].

Although early reports described mainly patients with severe pneumonia [6], the spectrum of the disease is broad, with more than 80% of those infected showing moderate, mild or no symptoms [7].

Early studies involved hospitalized patients with severe or critical illness. In these patients, peak viral load in the upper respiratory tract occurs during the second week after the onset of symptoms, whereas viral clearance is achieved after 10 days in more than 90% of patients with mild disease [8]. Elevated cytokine levels, including interleukine 6 (IL-6) and IL-10 levels, increased C-reactive protein (CRP), and T-cell lymphopenia signal worsening disease [9]. This inflammatory response disorder is thought to result from an initial alteration in interferon production, which thus reduced early viral control. Early studies in China suggested that anti-SARS-CoV-2 antibody titers are higher in patients with a more severe form of the disease [10]. In order to better understand the trends in antibody and cytokine levels, other studies were conducted in patients at different stages of the disease (mild, severe, critical) [11-13].

Knowing that genetic diversity, environment and race may influence the kinetics of antibodies and cytokines while little is known about this aspect in Togo, this study aimed to investigate the dynamics of antibodies immunoglobulin M (IgM) and immunoglobulin G (IgG) and cytokines in COVID-19 infected patients in Togo.

Materials and methods

Type, period and area of study

This was a retrospective analytical study that focused on samples collected from June 3 to August 31, 2021. The study was conducted in the Autonomous District of "Grand Lomé" which recorded the most cases of COVID-19 (20404 out of 26291), i.e. 78% [14].

Sera were obtained from adults (age ≥18years) male and female volunteers SARS-CoV-2 infected and living in the "Grand Lomé" Autonomous District.

Antibodies and cytokines assays were performed at the "Unité de Recherche en Immunologie et Immunomodulation (UR2IM)/Laboratoire de Microbiologie et de Contrôle de Qualité des Denrées Alimentaires (LAMICODA)".

Study population

The study population consisted of 80 patients positive for SARS-CoV-2 by real time PCR, including 40 patients with severe symptoms requiring hospitalization at the Lomé-Commune care center (reference center for COVID-19 patients follow up in Togo) and 40 patients with no severe symptoms that were followed up at the IBIS Hotel in Lomé.

Sample size

The sample size (n = 66) was calculated using Schwartz formula $n = Z^2 P (1-P) / d^2$ where Z, the accepted risk error is 1.96; d, the precision, is 0.05; P, the prevalence of SARS-CoV-2 positive tests. As of December 04, 2021; out of 584,028 tests performed in Togo; 26,291 cases were confirmed SARS-CoV-2 PCR positive with a prevalence of 4.51% [14].

Samples

Sera were collected from symptomatic and asymptomatic patients at D0 (at admission), D3 (day 3 of admission) and D10 (day 10 of admission) and stored at - 20°C at the "Division des Laboratoires" biobank.

Anti-SARS-CoV-2 IgM and IgG1 antibodies assay

IgM and IgG1 antibodies to SARS-CoV-2 were determined in asymptomatic and symptomatic COVID-19 patient's sera by indirect Enzyme-Linked Immunosorbent Assay (ELISA).

In brief, 96 wells ELISA plates were coated with 50µl per well of "Recombinant human coronavirus SARS-CoV-2 Spike Glycoprotein S1" at a concentration of 25ng/ml diluted in "coating buffer 1x PBS" and then incubated at 4°C overnight.

On one hand sera were pre-diluted 1:101 in IgG/RF absorbent solution and incubated for 10 min at room temperature. Coated ELISA plate content was discarded and the plate was washed 5 times and dried. Then the "IgG/RF absorbent" was distributed 50µl to wells A1 and A2 as negative controls. Also, 50µl of each pre-diluted sample were added to the samples corresponding wells and the plate was incubated for 30mn at room temperature. After washing the plate 5 times and drying, 50µl of peroxidase-labeled anti-human IgM conjugate was dispensed in each well and the plate was incubated at room temperature for another 30mn.

On other hand, for anti- SARS-CoV-2 IgG1 antibodies, sera were pre-diluted 1:101 in "1X ELISA/ELISPOT Diluant" and distributed in samples corresponding wells after washing 5 times and drying the coated plate, followed by incubation for 30mn. "1X ELISA/ELISPOT Diluant" was used as negative control in wells in A1 and A2 wells. The "anti-human IgG1-Biotin" was diluted to 1/3000th with "1X ELISA/ELISPOT Diluant", content of ELISA plate was discarded and the plate was washed 5 times and dried. Thereafter, 50µl of diluted anti-human IgG1-Biotin antibody was spiked per well and the plate was incubated for 30mn at room temperature. Following steps consisted in the addition of 50µl per well of "Avidin-HRP 1X" after washing and drying the plate and the incubation of the plate for 30mn at room temperature.

At the end of peroxidase-labeled anti-human IgM conjugate and Avidin-HRP reactions, contents of ELISA plates wells were discarded and plates were washed 7 times and dried. 50µl of "1X TMB substrate solution" was added to each well and plates were incubated for 5 minutes at room temperature in dark. The reaction was stopped with 50µl of 2N sulfuric acid per well and plates were read on Huma-Reader HS plate reader (Human, Wiesbaden, Germany) at a double wavelength of 450nm - 620nm.

Cytokines assay

Levels of cytokines IL-1β, TNFα, IL-6, IFNγ, IL-5, IL-10 and IL-17A were measured in sera by sandwich ELISA using cytokines ELISA kits (Thermo Fisher Scientific, Bender MedSystems GmBH, Vienna, Austria) according to the manufacturer's instructions. Cytokines concentrations were measured at 450 nm on HumaReader HS plate reader (Human Diagnostics Worldwide, Wiesbaden, France).

Statistical analysis

Demographic data were analyzed using R software version 4.0.5. and immunological data were analyzed using GraphPad PRISM software version 5.02 for Windows (GraphPad Software, San Diego California USA). The Wilcoxon-Mann-Whitney, Fisher's exact and chi-square tests were used to compare variables between the two groups of patients. A p-value less than 0.05 was considered significant.

Results

Socio-demographic and clinical characteristics

A total of 80 adults aged between 18 and 76 years were included in the study. The median age of asymptomatic patients was 32 (26-37) years and 56 (46-64) years for symptomatic patients. Most of asymptomatic patients aged between 29 and 41years old while, symptomatic patients aged between 56 and 76 years old. Male were more reprensted in asymptomatic group and female, in symptomatic group. Regarding the occupation, 60% of asymptomatics were employees whereas 50% of symptomatics were self-employed. The togolese in couple were more represented in both group of patients. Principal symptoms recorded in symptomatic patients were cough (60%), fever (57%) and tiredness (48%) and high blood pressure (45%), diabetes (25%) were the most frequent comorbidities found in them (Table I).

Variable	Asymptomatic, n = 40	Symptomatic, n= 40	p-value [b]
Median age (Year)	32 (26 - 37)	56 (46 - 64)	**<0.001***
Age groups (Year)			**<0.001***
[18-23]	13 (32.5) [a]	4 (10.0)	
[29-41]	18 (45.0)	3 (7.5)	
[41-56]	8 (20.0)	13 (32.5)	
[56-76]	1 (2.5)	20 (50.0)	
Gender			**<0.001***

Variable	Asymptomatic, n = 40	Symptomatic, n= 40	p-value [b]
Female	4 (10.0)	25 (62.5)	
Male	36 (90.0)	15 (37.5)	
Occupation			**<0.001***
Student	7 (17.5)	1 (2.50)	
Self employed	7 (17.5)	20 (50.0)	
Housewife	1 (2.5)	8 (20.0)	
Retired	1 (2.5)	4 (10.0)	
Employee	24 (60.0)	7 (17.5)	
Nationality			0.13
Togolese	31 (77.5)	36 (90.0)	
Other	9 (22.5)	4 (10.0)	
Marital status			0.11
In couple	21 (52.5)	28 (70.0)	
Not in couple	19 (47.5)	12 (30.0)	
Education level			**0.005***
Not in school	1 (2.5)	5 (12.5)	
Primary	1 (2.5)	9 (22.5)	
Secondary	24 (60.0)	13 (32.5)	
Higher	14 (35.0)	13 (32.5)	
Symptoms			
Fever	13 (32)	23 (57)	**0.025***
Myalgia	0 (0)	4 (10)	
Rhinorhea	6 (15)	4 (10)	
Headache	5 (12)	11 (28)	
Tiredness	10 (25)	19 (48)	**0.036***
Cough	8 (20)	24 (60)	**<0.001***
Comorbidities			
HBP	NA	18 (45)	
Diabetes	NA	10 (25)	
Sinusitis	NA	1 (2.5)	
Obesity	NA	2 (5)	
Asthma	NA	1 (2.5)	
Prostate	NA	1 (2.5)	

Variable	Asymptomatic, n = 40	Symptomatic, n= 40	p-value [b]
Sickle cell disease	NA	2 (5)	
Kidney failure	NA	1 (2.5)	
Blindness	NA	1 (2.5)	
Pleurisy	NA	1 (2.5)	
Hemorrhoid	NA	0 (0)	
HBV	NA	0 (0)	
HIV	NA	0 (0)	

[a]: n (%); [b]: Wilcoxon-Mann-Whitney test, Independence chi-square test, Fisher exact test; *: p-value Significant; HBP: High Blood Pressure; HBV: Hepatitis B Virus; HIV: Human Immunodeficiency Virus.

Table 1: Socio-demographic and clinical characteristics.

IgM and IgG1 antibodies kinetics

In asymptomatic patients, IgM antibodies raised at D0 were expressed rapidly, peaking at D3 and gradually decreasing until D10. In symptomatic patients, IgM antibodies elevated at D0 were progressively expressed to reach higher levels at D10 (Figure 1A).

In asymptomatic patients, IgG1 antibodies were elevated at D0 and varied very little until D10. In symptomatic patients, IgG1 were progressively expressed and reached higher levels at D10 (Figure 1B).

We observed that IgG1 was less produced in symptomatics compared to asymptomatics at D0 and D3. At D10, IgG1 had quite the same levels in both groups of patients. In addition, IgM did not vary among both groups (Figures 1C, 1D).

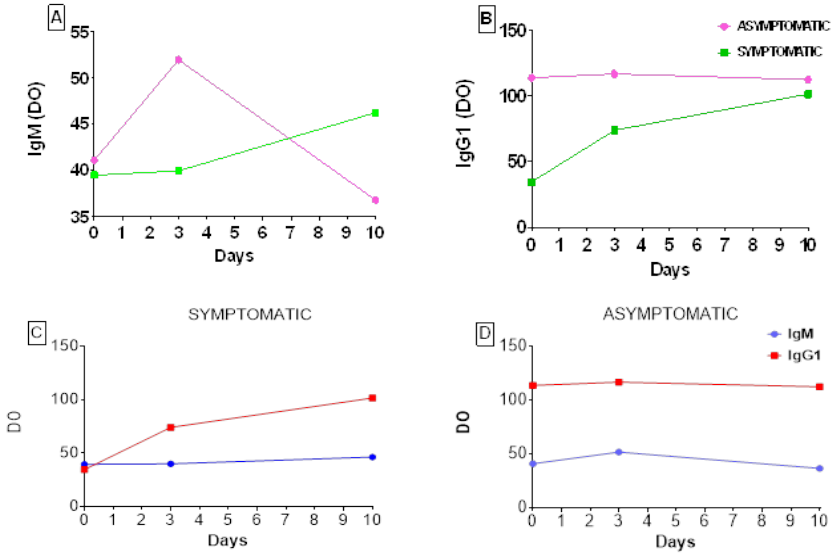

Figure 1: Kinetics of anti-SARS-CoV-2 antibodies. (A) represents the kinetics of IgM antibodies and (B) IgG antibodies, in the sera of asymptomatic patients in pink (n=20) and symptomatic patients in green (n=20). The mean DO (ELISA) obtained at D0, D3 and D10 was used to establish the antibody kinetics. (C) represents the kinetics of the IgM/IgG1 ratio of symptomatic patients (D) represents the kinetics of the IgM/IgG1 ratio of symptomatic patients.

Kinetics of innate cytokines

IL-1β cytokine levels were showed to increase progressively and were similar in both symptomatic and asymptomatic patients (Figure 2A). In contrast TNFα levels decreased in both groups rapidly from D0 to D3 and remained constant until D10. However, they were higher in asymptomatic patients than in symptomatic patients at D0 (Figure 2B). Regarding IL-6 cytokine, its levels were higher in symptomatics than asymptomatics at D0, then we observed a decrease in both groups from D0 to D3 (Figure 2C).

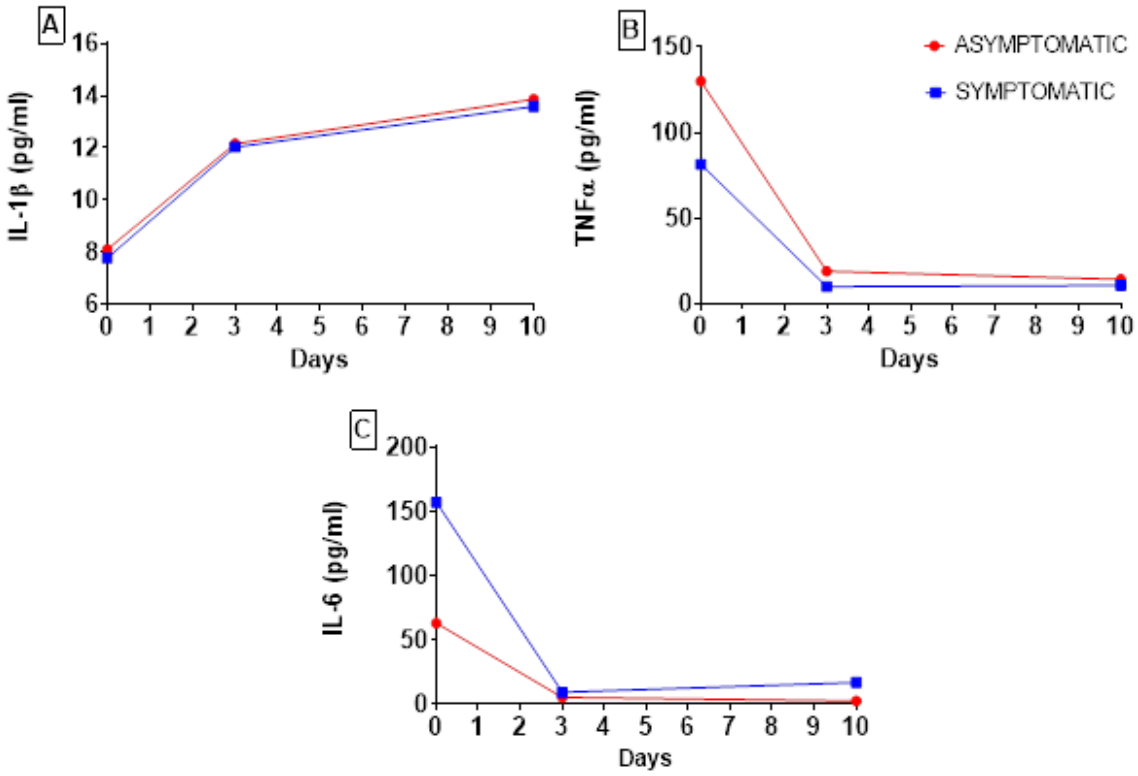

Figure 2: Kinetics of type I cytokines. (A) represents the kinetics of the innate immunity cytokines IL-1β (B); TNFα and (C) IL-6 in the sera of asymptomatic patients in red (n=40) and symptomatic patients in blue (n=40). The mean of the concentrations in pg/ml, obtained at D0, D3 and D10 were used to establish the kinetics of the cytokines.

Kinetics of adaptive cytokines

We observed a decrease in IL-5, IL-10 and IFNγ production from D0 to D3 in both symptomatic and asymptomatic patients (Figures 3A, 3B, 3D). IL-5 was more expressed at D0 in asymptomatic patients and augmented in symptomatic patients from D3 to D10 (Figure 3A). IL-10 levels remained high in symptomatics whereas IL-17A remained elevated in asymptomatics. This latter cytokine increased from D0 to D3 and decreased from D3 to D10 in both groups (Figure 3C).

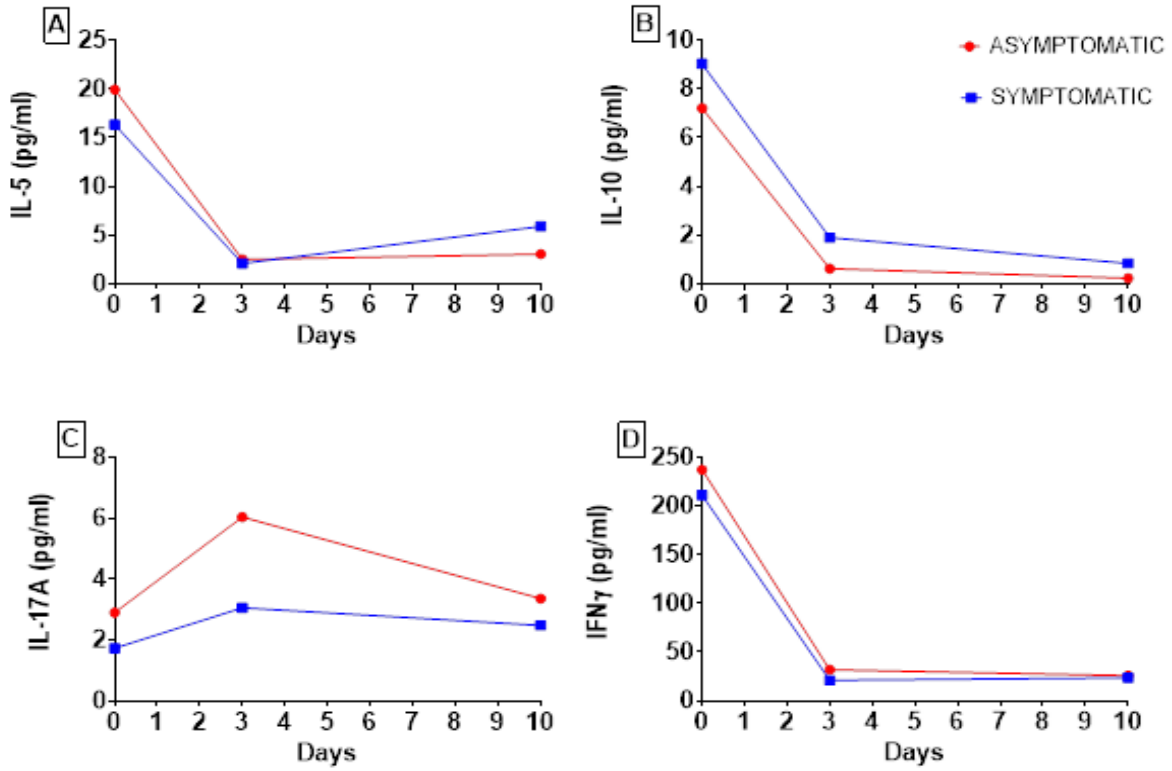

Figure 3: Type II cytokine kinetics. (A) represents the kinetics of the adaptive immunity cytokines IL-5; (B) IL-10; (C) IL-17A and (D) IFNγ, in the sera of asymptomatic patients in red (n=40) and symptomatic patients in blue (n=40). The mean pg/ml concentrations obtained at D0, D3 and D10 were used to establish cytokine kinetics.

Discussion

This study provides important insights into the COVID-19 pandemic and immunological data in SARS-CoV-2 infection in Togo. We had a limitation in considering COVID-19 patients admission day as D0 and could not then determine when they were infected.

Among symptomatic patients, the most important symptoms reported in COVID-19 in our study were cough, fever and tiredness and this corroborate with those reported by Adil *et al*, (2021) [15]. Indeed, a study on epidemiological and clinical characteristics of the first wave of the pandemic showed that the most frequently reported symptoms were headache (66.4%), fever (63.5%), and cough (59.9%) [16].

With regard to age groups, all age groups were susceptible to SARS-CoV-2 infection in our study. However, the elderly and those living with comorbidities such as HBP and diabetes were more likely to develop the severe form of the disease. Some studies have also shown that men were twice as likely to die from COVID-19, and patients over 65 years of age with comorbidities such as diabetes, HBP and cardiovascular disease were factors associated with death from COVID-19 [17,18].

Our data revealed that housewives, pensioners and self-employed people were at greater risk of developing the symptomatic form of the disease. This could be explained not only by the fact that they were more exposed but also by their advanced age. Indeed, in the present study, age was a risk factor for the disease.

In response to SARS-CoV-2 infection, the body produced specific IgM and IgG1 antibodies to SARS-CoV-2 in both asymptomatic and symptomatic subjects. However, in symptomatic patients, our data showed that SARS-CoV-2 induced a classical viral response pattern, where IgM is the first isotype to appear, followed by IgG1. In contrast, in asymptomatic patients, the kinetics suggest simultaneous production of IgM and IgG1 from the onset of infection with higher titers for IgG1. This last result can have several explanations: either patients have been in contact

with SARS-CoV-2 several times and have developed antibodies or they have already been vaccinated against SARS-CoV-2.

Several types of seroconversions have been reported, such as synchronous IgG and IgM seroconversion, IgM seroconversion earlier than IgG and IgM seroconversion later than IgG [19,20]. In a study analyzing specific IgM and IgG by enzyme-linked immunosorbent assay, several patients were found to be seropositive for IgG than for IgM on day 0 and day 5 of hospital admission, with some patients showing earlier IgG than IgM seroconversion [21].

In the present study, innate cytokines were expressed in patients with elevated levels upon admission to the care center. In particular, TNFα was higher in asymptomatic patients. The cytokine IL-1β increased progressively and similarly in both groups of patients during the follow-up time. As COVID-19 is an inflammatory disease, it is characterised by cytokine storms associated with disease severity [22]. Thus, macrophage-related type I cytokines, in particular IL-6 and TNF-α, were significantly increased in the majority of severe cases. It has been also shown that the interaction between SARS-CoV Spike protein and murine macrophages could induce the release of IL-6 and TNFα cytokines through the activation of NF-κB [23]. This would explain the expression of these cytokines at the beginning of SARS-CoV-2 infection found in both patients' groups in our study. Indeed, innate immunity is the first line of host defence against viral infection and experimental infection in SARS-CoV-infected mouse models has provided evidence that innate immunity is important for viral clearance [23]. Moreover, IL-6 plays a central role in the innate and acquired immune response, and is a predominant inducer of the acute phase response to infection.

Comparing the level of IL-6 production at D0 with that of anti- SARS-CoV Spike protein IgM antibodies, we find that IL-6 would dictate the expression of SARS-CoV Spike protein IgM in symptomatic patients at the beginning of the infection. Indeed, IL-6 plays an important role in the subsequent development of acquired immunity against incoming pathogens by stimulating antibodies production by B cells [24]. This would explain the decrease in IL-6 and the increase in anti-S IgM over time in our study.

Symptomatic patients maintained their inflammatory environment from D0 to D10, as demonstrated by IL-6 expression which remained significant even though it decreased over time. The hypothesis of a regulatory mechanism was verified by investigating the expression of adaptive cytokines, notably IL-5, IL-10, IL17-A and IFNγ.

In the present study, IL-10 was expressed with low levels in both asymptomatic and symptomatic patients. In contrast to our data, serum IL-10 levels were significantly higher in COVID-19 patients in the intensive care unit than in non-intensive care patients in Wuhan [7]. In addition, a meta-analysis of 1242 non-severe and 915 severe COVID-19 patients from 18 clinical studies identified IL-6 and IL-10 as covariates accurately predicting disease severity [25]. The low level of IL-10 observed in our study would then explain the low lethality rate of COVID-19 recorded in Togo.

The level of cytokine IL-5 was low in both groups of patients in our study. It was observed that IL-5 levels were not significant between control subjects, severe cases and mild cases with COVID-19 (52). We did not include healthy controls which would allow us to assess the differences in concentrations observed between the two groups.

The mean level of IL-17A was increased in asymptomatic patients compared to symptomatic cases. Ghazavi et al found the same with a significant difference [26]. Indeed, Th17 cells are the main source of IL-17 production which stimulates the secretion of IL-8, which is a potent chemoattractant of neutrophils involved in conditions such as COVID-19 [27,28].

Th1 cells, NK cells and $CD8^+$ T cells are the main sources of IFN-γ [29]. The increase in IFNγ production early in the infection indicates a Th1 cell response in COVID-19 [30]. Developing the Th1 response to eliminate the virus is part of the immune system's strategies [31], and an IFNγ response may lead to better outcomes in patients with COVID-19. The decrease in IFNγ levels over time in our study is thought to be due to the treatment given to patients during follow-up.

Conclusion

The present study highlights the dynamic nature of immune response, which can escalate and then decline. The aim of this study was to investigate the dynamics of antibodies and cytokines in symptomatic and asymptomatic COVID-19 patients in Togo.

On the one hand, the present study showed that in symptomatic patients, the immune system expressed IgM antibodies first, followed by IgG1 in response to SARS-CoV-2 infection. In contrast, in asymptomatic patients, expression was simultaneous from the onset of infection with higher titers for IgG1. This would explain the non-severity of the disease in some infected subjects. On the other hand, high IL-6 production was noted in symptomatic patients, a pro-inflammatory cytokine that is the predominant inducer of the response in the acute phase of the infection. The present study adds to the theory that IL-6 is a predictor of disease severity in patients with COVID-19.

Acknowledgements

We thank the "Division des Laboratoires" for providing us with their biobank for the sample collection.

We also thank Dr Essoham Ataba from "Programme National de Lutte Contre le Paludisme, Ministère de la Santé de l'Hygiène

Publique et de l'Accès Universel Aux Soins" for the samples collection.

Ethical considerations

This study received ethical approval, under registration number 024/2022/CBRS of June 23, 2022 from the Bioethics Committee for Health Research of the Ministry in charge of Health in TOGO.

Conflict of interest

The authors declare that they have no conflict of interest.

Author contributions

G.K., E.P.T, A.K., realised the study design; A.D., performed the sample collection and clinical data, A.T., M.R., provide the protein file assay; G.K., M.R., C.N., M.O.A, did the experimental design; H.C.S., C.N. wrote the original draft; H.C.S., E.P.T performed statistical analysis, MK approved the manuscript.

References

1. Zhou P, Yang XL, Wang XG, Hu B, Zhang L, et al., (2020) A pneumonia outbreak associated with a new coronavirus of probable bat origin. Nature 579:270-273.
2. Christèle Tchopba Nguepou EA, Gnatoulma Katawa, Banfitebiyi Gambogou, Manuel Ritter, Simplice D, et al., (2020) COVID-19: Epidemiology, Pathogenesis and Immununological Basis. Al-Nahrain Journal of Science. 4:1-12.
3. WHO (2020) Allocution liminaire du Directeur général de l'OMS lors du point presse sur la COVID-19 - 11 mars 2020.
4. WHO (2023) Coronavirus disease (COVID-19) pandemic 2023.
5. OMS (2020) Epidémie à Coronavirus, COVID-19, le Togo déclare un premier cas confirmé 2020.
6. Zhou F, Yu T, Du R, Fan G, Liu Y, et al., (2020) Clinical course and risk factors for mortality of adult inpatients with COVID-19 in Wuhan, China: a retrospective cohort study. Lancet (London, England) 395:1054-1062.
7. Huang C, Wang Y, Li X, Ren L, Zhao J, et al., (2020) Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. Lancet 395:497-506.
8. Vetter P, Vu DL, L'Huillier AG, Schibler M, Kaiser L, et al., (2020) Clinical features of covid-19. BMJ 369:m1470.
9. Han H, Ma Q, Li C, Liu R, Zhao L, et al., (2020) Profiling serum cytokines in COVID-19 patients reveals IL-6 and IL-10 are disease severity predictors. Emerg Microbes Infect 9:1123-1130.
10. Huang C, Huang L, Wang Y, Li X, Ren L, et al., (2021) 6-month consequences of COVID-19 in patients discharged from hospital: a cohort study. Lancet 397:220-232.
11. Grifoni A, Weiskopf D, Ramirez SI, Mateus J, Dan JM, et al., (2020) Targets of T Cell Responses to SARS-CoV-2 Coronavirus in Humans with COVID-19 Disease and Unexposed Individuals. Cell 181:1489-1501.e15.
12. Thevarajan I, Nguyen THO, Koutsakos M, Druce J, Caly L, et al., (2020) Breadth of concomitant immune responses prior to patient recovery: a case report of non-severe COVID-19. Nat Med 26:453-455.
13. Weiskopf D, Schmitz KS, Raadsen MP, Grifoni A, Okba NMA, et al., (2020) Phenotype and kinetics of SARS-CoV-2-specific T cells in COVID-19 patients with acute respiratory distress syndrome. Sci Immunol 5: eabd2071.
14. togoglaise R. Situation au Togo 2023.
15. Adil MT, Rahman R, Whitelaw D, Jain V, Al-Taan O, et al., (2021) SARS-CoV-2 and the pandemic of COVID-19. Postgrad Med J 97:110-116.
16. Diarra M, Barry A, Dia N, Diop M, Sonko I, et al., (2022) First wave COVID-19 pandemic in Senegal: Epidemiological and clinical characteristics. PLoS One 17: e0274783.
17. Pastor-Barriuso R, Pérez-Gómez B, Hernán MA, Pérez-Olmeda M, Yotti R, et al., (2020) Infection fatality risk for SARS-CoV-2 in community dwelling population of Spain: nationwide seroepidemiological study. BMJ 371:m4509.
18. Pantea Stoian A, Pricop-Jeckstadt M, Pana A, Ileanu BV, Schitea R, et al., (2020) Death by SARS-CoV2: a Romanian COVID-19 multi-centre comorbidity study. Sci Rep 10:21613.
19. Long QX, Liu BZ, Deng HJ, Wu GC, Deng K, et al., (2020) Antibody responses to SARS-CoV-2 in patients with COVID-19. Nat Med 26: 845-848.
20. Lee YL, Liao CH, Liu PY, Cheng CY, Chung MY, et al., (2020) Dynamics of anti-SARS-Cov-2 IgM and IgG antibodies among COVID-19 patients. J Infect 81: e55-e58.
21. To KK, Tsang OT, Leung WS, Tam AR, Wu TC, et al., (2020) Temporal profiles of viral load in posterior oropharyngeal saliva samples and serum antibody responses during infection by SARS-CoV-2: an observational cohort study. Lancet Infect Dis 20:565-574.
22. Chen G, Wu D, Guo W, Cao Y, Huang D, et al., (2020) Clinical and immunological features of severe and moderate coronavirus disease 2019. J Clin Invest 130:2620-2629.
23. Wang W, Ye L, Ye L, Li B, Gao B, et al., (2007) Up-regulation of IL-6 and TNF-alpha induced by SARS-coronavirus spike protein in murine macrophages via NF-kappaB pathway. Virus Res 128:1-8.
24. Yao X, Huang J, Zhong H, Shen N, Faggioni R, et al., (2014) Targeting interleukin-6 in inflammatory autoimmune diseases and cancers. Pharmacol Ther 141:125-139.
25. Dhar SK, K V, Damodar S, Gujar S, Das M (2021) IL-6 and IL-10 as predictors of disease severity in COVID-19 patients: results from meta-analysis and regression. Heliyon 7: e06155.
26. Ghazavi A, Ganji A, Keshavarzian N, Rabiemajd S, Mosayebi G (2021) Cytokine profile and disease severity in patients with COVID-19. Cytokine 137:155323.
27. Chen HL, Lo CH, Huang CC, Lu MP, Hu PY, et al., (2021) Galectin-7 downregulation in lesional keratinocytes contributes to enhanced IL-17A signaling and skin pathology in psoriasis. J Clin Invest 131: e130740.
28. Kartasheva-Ebertz D, Gaston J, Lair-Mehiri L, Mottez E, Buivan TP, et al., (2022) IL-17A in Human Liver: Significant Source of Inflammation and Trigger of Liver Fibrosis Initiation. Int J Mol Sci 23:9773.

29. Luetke-Eversloh M, Hammer Q, Durek P, Nordström K, Gasparoni G, et al., (2014) Human cytomegalovirus drives epigenetic imprinting of the IFNG locus in NKG2Chi natural killer cells. PLoS Pathog 10: e1004441.

30. Hasanvand A (2022) COVID-19 and the role of cytokines in this disease. Inflammopharmacology 30:789-798.

31. Kidd P (2003) Th1/Th2 balance: the hypothesis, its limitations, and implications for health and disease. Altern Med Rev 8:223-246.