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Original Research Article

Perturbation of Biological Markers in COVID-19 Positive Type-2 Diabetic Subjects (Delta and Omicron Variant Cases) in Pointe-Noire

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Abstract: ***Introduction***: The SARS-CoV-19 pandemic affected the whole world, with a particularly high and severe incidence in patients with co-morbidities such as type 2 diabetes (T2DM). The SARS-CoV-2 variants, Delta and Omicron, posed an additional challenge in the management of these patients because of their virulence and high transmissibility. ***Objective***: To assess the disturbance of certain biomarkers in COVID-19-positive T2DM patients in Pointe-Noire. ***Methods***: We performed a descriptive cross-sectional study on a cohort of 206 type 2 diabetic patients affected by COVID-19. Blood samples were taken for analysis of biomarkers (CBC, ESR, CRP, GLY, and HbA1c, lipid profile, urea and creatinine). RNA was extracted from nasopharyngeal samples and PCR was performed to determine the presence of Delta and Omicron variants. ***Results***: The mean age of our patients was 56.33 ± 12 years. The Delta variant was the most common 132 (64.08%), followed by the other variants 57 (27.67%) and Omicron 17 (8.25%). We observed significant disturbances ($p<0.05$) in biomarkers as a function of Delta and Omicron variants concerning: TC, TG, HDL, LDL, DDI, VS, CRP, GLY, and HbA1c. ***Conclusion***: We identified the Delta variant as the most frequent in the diabetic population studied. Almost all biological markers studied were disrupted during COVID-19 infection in the type 2 diabetic population.

Keywords: COVID-19, Delta Variant, Omicron Variant, Type 2 Diabetes, Biomarkers.

INTRODUCTION

Type 2 diabetes (T2D) is a chronic disease characterised by insulin resistance and dysfunction of the beta cells of the pancreas, leading to a constant rise in blood sugar levels. Complications associated with T2DM can affect various organs, increasing the risk of cardiovascular disease, kidney failure, retinopathy and other problems in affected individuals [2, 3].

COVID-19, caused by SARS-CoV-2, is an infectious disease that has been spreading worldwide as a pandemic since 2019. Symptoms of infection vary considerably, ranging from no symptoms to severe forms that can be fatal [4]. This situation has highlighted the complexity of the interactions between this new coronavirus and various pre-existing medical conditions.

Type 2 diabetes in particular stands out due to its increasing prevalence worldwide. Studies have indicated that people with T2DM are more likely to develop severe forms of COVID-19 due to chronic inflammation, immunosuppression, hyperglycemia and

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abnormal coagulation [5-7]. This concern is reinforced by the emergence of more contagious variants of the virus, such as Delta and Omicron [8].

Biomarker analysis has played a crucial role in understanding the pathophysiological mechanisms of diabetes and COVID-19, separately [9], and may also contribute to the identification of new diagnostic and treatment strategies.

Few studies to date have looked specifically at biological markers in patients with T2DM and COVID-19 due to the Delta and Omicron variants. This is an area of research that requires further investigation to improve our understanding of the interaction between T2DM and COVID-19, particularly for these SARS-CoV-2 variants. This gap in the literature is all the more notable in resource-limited settings such as Pointe-Noire.

The aim of this article is to add to the literature by closely examining biomarker changes in T2D patients infected with COVID-19 in Pointe-Noire.

MATERIAL AND METHOD

Study Population

We carried out a cross-sectional descriptive study whose data collection was prospective. The study took place from September 2021 to August 2022, a period of 12 months. The study population consisted of T2D patients with COVID-19 hospitalized at the Guenin, Louise Michel clinic and the General Adolphe sicé hospital in Pointe-Noire.

Clinical Investigation: Data such as age, sex, BMI, covid-19 symptoms and comorbidities were collected from medical records.

Biological Investigation:

The laboratory tests were carried out in the HDL Biomedical Analysis Laboratory at the Polyclinic Foundation Marie Madeleine Gombes in Pointe Noire.

1. **Samples:** The blood sample was taken on an EDTA tube, heparinized and citrated and was stored at -20°C until use.

2. **Analysis of Biomarkers:**

The following analyzes were carried out:
Fasting blood glucose (Gly); Glycated hemoglobin (HbA1c); Creatinine (Creat); Urea;
Uric Acid (AU); Lactate dehydrogenase (LDH); D-dimer (DDI); Blood count (CBC); Sedimentation Rate (ESR); Ultra-sensitive C-reactive protein (CRP us); Transaminases (GOT, GPT); Gamma Glutamyl Transferase (GGT); Lipid assessment (CT, TG, HDL, LDL);

3. **Molecular Analysis:**

a) Extractions

We carried out RNA extraction from nasopharyngeal secretions using the Total RNA Purification Insert PI12200-37 kit, Norgen Biotek Corp (CANADA) in accordance with the manufacturer's recommendations.

b) Amplifications

The extracted RNAs were subjected to PCR using the SARS-CoV-2 E Spike Delta/Omicron TaqMan Typing MDx 40-0813-96 kit, TIB MOLBIOL, Germany.

Procedure:

First step: Mix preparation

- 4µl molecular biology water (nuclease free water)
- 1µl primers and probes (PSR)
- 10µl RT polymerase
- 5µl total RNA

Second step: programming the Mic (thermocycler)

RT-PCR Cycle	Step	Temperature	Duration
Cycle 1	**Step** 1 **Step** 2	55°C 95°c	3min 1min
Cycle 1 (40x)	**Step** 1 **Step** 2	95°C 63°C	3sec 10 sec

Choice of fluorochromes and targets:

- FAM (Omicron: Spike ins214EPE)
- HEX (Delta: Spike del157/158)
- ROX (SARS-CoV-2: SARS E-gene)
- Cy5 (UBC Human gene)

Ethical Considerations

This study was conducted in accordance with the guidelines of the Declaration of Helsinki and was approved under number 125/CERS/FMMG-2021/PNR by the Health Research Ethics Committee (CERS) of the Marie Madeleine Gombes Foundation in Pointe Noire.

Statistical Analysis

The categorical data are expressed in numbers (percentage). The $\chi 2$ test was used to compare categorical data. A p value of less than 0.05 was considered to indicate statistical significance. All analyses were conducted using SPSS software (version 26.0; IBM).

RESULTS

Table I shows the distribution of variants according to a number of socio-demographic and clinical parameters. The Delta variant is mainly detected in men (75.75%), as is the Omicron variant (88.24%), while the other variants are more evenly distributed between men (52.63%) and women (47.37%). A significant gender

difference in the distribution of COVID-19 variants ($p < 0.001$***).

A significant difference was also observed in the distribution of variants by age group ($p < 0.001$***). The Delta variant was most prevalent in the 40-69 age group (78.03%) and the 20-39 age group (5.30%).

The Omicron variant was more common in the 40-69 age group (70.58%). The other variants are mainly observed in the 40-69 age group (78.95%) and the 20-39 age group (14.04%).There was a significant difference in the distribution of variants according to body mass ($p < 0.001$***). The Delta variant was found predominantly in overweight individuals (77.27%), while the Omicron variant was relatively distributed between normal-weight (47.06%) and overweight (52.94%) individuals.

The majority of patients (62.13%) had suffered from this condition for between 1 and 10 years. Some had been suffering for more than 20 years (14.8%). There was a significant difference in the distribution of variants in relation to the severity of COVID symptoms ($p < 0.001$***). The majority of individuals with the Delta variant (68.18%, n=90) and the other variants (77.19%, n=44) had severe symptoms while individuals with the Omicron variant had moderate symptoms (76.47%, n=13). There was a significant difference in the distribution of variants according to clinical course ($p < 0.001$***).

Table II shows the profile of the biological markers studied according to the different types of variants identified. Analysis of the biological profile of T2DM patients infected with the Delta and Omicron variants of SARS-CoV-2 reveals significant disturbances in the biomarkers concerned: TG, HDL, LDL, DDI, VS, CRP, GLY, and HbA1c. There was a statistically significant difference ($p<0.05$) in almost all biomarkers between the different variants.

Table I: Variant distribution

	Variant Delta		**Variant Omicron**		**other variants**		**P-value**
Variables	number (n)	Frequency (%)	number (n)	Frequency (%)	number (n)	Frequency (%)	
Sex							<0,001***
male	100	75,75	15	88,24	30	52,63	
Female	32	24,25	02	11,76	27	47,37	
age group							<0,001***
20 – 39 years	07	5,30	03	17,65	08	14,04	
40 – 69 years	103	78,03	12	70,59	45	78,95	
>70 years	22	16,67	02	11,76	04	7,01	
Body mass							<0,001***
Normal	30	27,73	08	47,06	32	56,14	
Overweight	102	77,27	09	52,94	25	43,86	
age of D2T							p=0,044*
1-10	82	62,12	08	47,06	38	66,67	
11-20	34	25,76	05	29,41	10	17,54	
>20	16	12,12	04	23,53	09	15,79	
Severity							<0,001***
Severe	90	68,18	04	23,53	44	77,19	
Moderate	42	31,82	13	76,47	13	22,81	
Symptoms							<0,001
Shortness of breath	89	67,42	15	88,23	22	38,59	
Fatigue	120	90,9	12	70,58	32	56,14	
Cough	121	91,66	17	100	45	78,94	
Fever	130	98,48	15	88,23	28	49,12	
Diarrhea	25	18,93	8	47,05	12	21,05	
Loss of taste	132	100	17	100	55	96,49	
Anorexia	113	85,6	11	100	52	91,22	
Loss of sense of smell	116	87,87	16	94,11	24	42,1	
Comorbidity							<0,001**
No comorbidity	47	35,6	5	29,4	35	61,4	
With comorbidity	85	64,4	12	70,58	22	38,59	
Clinical course							p=0,032*
Survivor	95	71,97	17	100	40	70,17	
Non-survivor	37	28,03	0	0	17	29,82	
TOTAL	**132**	**64,08**	**17**	**8,25**	**57**	**27,67**	

Table II: Biomarker profile according to variants

BIOMARKERS	N	VARIANT DELTA n = 132	VARIANT OMICRON n = 17	OTHER VARIANTS n = 57	P-value
Age (ans)	206	57 ± 13	64 ± 16	48 ±8	<0.001
Hb (g/dl)	206	12.10 ± 2.30	11.67 ± 1.79	13.41 ± 1.75	<0.001
PNN ($x10^3/mm^3$)	206	7,518 ± 6,113	7,054 ± 2,598	12,682 ± 8,655	0.003
PNE (mm^3)	206	45 ± 58	15 ± 43	35 ± 63	0.046
PNB (mm^3)	206	34 ± 6.4	27 ± 7.1	32 ± 3.3	<0.001
LYMPHO ($x10^3/mm^3$)	206	1,099 ± 651	1,328 ± 429	1,581 ± 594	<0.001
MONO (mm^3)	206	543 ± 401	695 ± 584	537 ± 206	0.064
PLQTES ($x10^3/mm^3$)	206	240,137 ± 128,358	157,66 ± 66,287	239,140 ±135,912	0.017
CT (g/L)	206	2.10 ± 0.51	1.79 ± 0.63	2.27 ± 0.49	0.025
HDL (g/L)	206	0.38 ± 0.16	0.26 ± 0.15	0.51 ± 0.27	<0.001
LDL (g/L)	206	1.31 ± 0.48	1.29 ± 0.62	1.53 ± 0.54	0.13
TG (g/L)	206	2.20 ± 1.37	1.49 ± 0.59	1.41 ± 0.52	<0.001
CHOLT/HDL	206	6.6 ± 3.1	7.9 ± 3.5	6.9 ± 5.2	0.12
TG/HDL	206	7.5 ± 7.1	6.7 ± 3.4	3.3 ± 2.8	<0.001
LDH (U/L)	206	306 ± 153	463 ± 152	268 ± 160	<0.001
UREE (g/L)	206	0.33 ± 0.21	0.44 ± 0.18	0.84 ± 0.81	<0.001
AU (mg/L)	206	51 ± 17	56 ± 10	49 ± 11	0.013
Créat (mg/L)	206	12 ± 4	15 ± 7	20 ± 18	<0.001
GGT (U/L)	206	56 ± 34	38 ± 16	42 ± 27	<0.001
GPT(U/L)	206	54 ± 30	76 ± 24	53 ± 50	<0.001
GOT(U/L)	206	64 ± 30	78 ± 24	54 ± 34	0.001
DDI (µg/L)	206	2,116 ± 2,563	1,942 ± 318	3,887 ± 3,133	<0.001
ESR (mm^3)	206	34 ± 28	16 ± 11	69 ± 45	<0.001
CRPus (mg/L)	206	173 ± 76	154 ± 98	247 ± 118	<0.001
HBA1c (%)	206	8.40 ± 1.86	9.34 ± 1.69	7.91 ±2.44	0.025
GLY(g/L)	206	2.34 ± 0.83	2.18 ± 0.61	2.53 ± 1.39	>0.9

DISCUSSION

The aim of our study was to examine changes in biomarkers in T2DM patients infected with COVID-19 in Pointe-Noire, focusing on the Delta and Omicron variants of the virus.

Our results showed that the Delta variant 132 (64.08%) was the most commonly detected in our population, followed by the other variants 57 (27.67%) and the Omicron variant 17 (8.25%). Interestingly, the distribution of variants was significantly affected by gender, age, body mass and other health variables.

In particular, the Delta variant was more frequently detected in men, people aged between 40 and 69 years 103(78.03%) and overweight individuals 102(77.27%), while the Omicron variant was more common in people of normal weight 08(47.06%). The prevalence of symptoms characteristic of omicron infection differed from that of the delta variant of SARS-CoV-2, with a lower incidence of severe symptoms 04 (23.53%), and no deaths. These results are consistent with the existing literature. Several studies have shown that the Delta variant is more transmissible and can cause more severe symptoms than previous variants of the virus (Kirsebom. 2022; Lopez Bernal *et al*., 2021) [17, 18]. Similarly, work suggests that the Omicron variant may be associated with less severe disease, although further research is needed to confirm this observation (Pulliam, 2022; Menni., 2022) [19, 20]. In this study, we did not have access to participant vaccination data, which could influence the distribution of virus variants.

Analysis of the biological profile of patients with COVID-19, specifically the Omicron and Delta variants, reveals significant disturbances. These variations in various biomarkers reflect the complex effects of COVID-19 infection and suggest a potentially important impact on the management of diabetes and other related complications.

In our study, markers of inflammation, specifically CRP and ESR, showed elevated levels in patients, particularly with the Omicron and Delta variants. This observation is consistent with other studies in the literature that report increased systemic inflammation in diabetic patients with COVID-19 (21, 22). This reinforces the hypothesis that inflammation may play a role in the increased severity of COVID-19 in people with type 2 diabetes.

In addition, our study showed a significant increase in GLY and HbA1c levels, suggesting a possible disruption of glucose regulation during infection. This is in line with other studies that have reported a

deterioration in glycemic control in patients with COVID-19 [23].

Furthermore, we observed elevated d-dimer levels in diabetic patients with Omicron and Delta variants, signaling a potential increased risk of thrombotic complications in this population. These results support the conclusions of the studies by Kirsebom *et al*., 2022; Lopez Bernal. 2021 [17, 18], which demonstrated an association between elevated levels of DDI and an increased risk of thrombosis in patients with COVID-19 [24].

Our study also revealed disturbances in lipid (CHOLT, HDL, LDL, TG) and liver (GPT, GOT, GGT) markers. This contrasts with the results found in the study by Yongli Yan. 2020 [22], in which the lipid and liver profiles were balanced. This suggests a possible effect of the virus on liver function and lipid metabolism, a hypothesis that requires further investigation.

Regarding WBC, RBC, PNN and platelet status, some participants had normal levels, although some showed lymphopenia. This result is similar to that found by Alzaid. 2020 and Yu Chen Chen .2020 [25-27]. The latter could indicate a weakened immune system in these patients, which could increase their vulnerability to COVID-19 infection [26].

CONCLUSION

Our study has shown that COVID-19 infection is an aggravating factor leading to significant disruption of the biological markers studied. Disruption of these markers was a poor prognostic factor in the course of COVID-19 in T2DM patients in Pointe-Noire. They also highlight the importance of comprehensive management of these patients, taking into account not only their glycemic control but also their COVID-19 status.

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