Polymorphic Variations of ACE2 and TMPRSS2 Genes in Egyptian COVID-19 Patients: Correlation with Disease Severity and Sex-Related Gene Variations

Faten M. Saad[1], Ahmed Diaf[1], Moustafa A. Sakr[1*], Hisham A. Ismail[1]

[1]Molecular diagnostics and therapeutics department, Genetic Engineering and Biotechnology Research Institute (GEBRI), University of Sadat City, Egypt

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Corresponding author: Prof. Moustafa A. Sakr
E-mail: **mostafa.sakr@gebri.usc.edu.eg**

ORCID of the author(s): 0000-0002-5873-8468

ABSTRACT:

This research pointed to accessing the polymorphic variations of the ACE2 and TMPRSS2 genes in Egyptian COVID-19 patients and their potential correlation with disease severity. The study involved a total of ninety-six participants, with forty-eight experiencing severe symptoms and an equal number exhibiting mild symptoms. Genotyping of single nucleotide polymorphisms (SNPs) in ACE2 and TMPRSS2 was performed using PCR-RFLP analysis.

The median age of individuals with mild cases was 35 years (range: 30-41 years), while for severe cases, it was 65 years (range: 58 to 72 years). The male-to-female ratio in the entire group was 47.9% to 52.1%. All participants tested positive for COVID-19 based on nasal swab qPCR tests.

Regarding genotypic distribution, the TT genotype of TMPRSS2 rs12329760 was found to be statistically more prevalent in the severe group compared to the mild group. However, no significant differences were observed in the distribution of ACE2 rs2285666 genotypes or alleles between the two groups. Concerning gene variations associated with sex in severe cases, males displayed a higher occurrence of the AA genotype, while females exhibited a higher prevalence of the GA genotype.

Keywords: COVID-19, ACE2, TMPRSS2, Single Nucleotide Polymorphisms, PCR-RFLP.

1. INTRODUCTION:

The global impact of SARS-CoV-2, which causes COVID-19 disease, has been significant, leading to the unfortunate loss of millions of lives and imposing a substantial burden on healthcare systems. The illness manifests across a broad clinical spectrum, encompassing asymptomatic instances to severe pneumonia and respiratory distress. Unvaccinated populations, in particular, have borne the brunt of the pandemic, with an approximate fatality rate of 1% and a range of disease manifestations, including asymptomatic infections and benign upper respiratory tract diseases (Zhang *et al*., 2022).

Age, sex, and chronic conditions like heart disease, Hyperglycemia, high blood pressure, renal failure, and malignancy, along with a history of smoking, are among the well-established risk factors that influence the severity of COVID-19. Recent discoveries emphasize that individuals who are not vaccinated are at a greater risk of having a severe disease. Also, genetic factors may determine the severity and outcome of the disease (Martono& Mulyanti, 2023).

ACE2 has emerged as a key player. It serves as the primary receptor for the viral spike glycoprotein, Allowing the passage and infection by SARS-CoV-2. Notably, ACE2

expression within the mucosa of the mouth cavity is seen as a major viral gateway into new hosts, contributing to the increased risk of bronchitis and pneumonia in severe COVID-19 cases. Polymorphic variations within ACE2 may impact its expression and function, and this genetic diversity could influence vulnerability to SARS-CoV-2 infection (Lukassen *et al.,* 2020).

Another critical gene, TMPRSS2 plays a pivotal role in coronavirus infections by separating the virus's spike glycoprotein, enabling cellular entry (Shang *et al.,* 2020).

Polymorphic variations within TMPRSS2 may result in altered protein function, potentially affecting the viral entry process and disease outcomes. The TMPRSS2 variant, rs12329760 (C to T), which leads to a missense mutation in the TMPRSS2 protein, is one such genetic polymorphism that may have implications for COVID-19 susceptibility and severity (Yaghoobi *et al.,* 2023).

Analyzing the genetic variations in the ACE2 and TMPRSS2 genes within the Egyptian population holds a significance importance. Egypt, situated in the Middle East, offers a distinct demographic and genetic landscape, introducing unique genetic variants that may underlie variations in susceptibility and COVID-19 intensity (Schaalan *et al.,* 2022). This study aims to explore the potential associations between specific ACE2 and TMPRSS2 gene variants and the likelihood of severe COVID-19 among Egyptian patients.

Our primary objective is to shed light on the genetic determinants that influence disease outcomes within this specific population. With a dedicated focus on the impact of genetic polymorphism within the ACE2 and TMPRSS2 genes, this research aims to enhance our understanding of the complex genetic factors that shape the clinical course of COVID-19, thereby highlighting the crucial role of genetic variations in influencing disease outcomes.

2. SUBJECTS AND METHODS*:*

2.1. Subjects:

This study involved the inclusion of 96 Egyptian patients who were newly diagnosed with COVID-19 using qPCR analysis of nasopharyngeal samples. These patients were clinically categorized into two subgroups based on the WHO case definition: Mild Cases: Refer to patients exhibiting mild clinical symptoms without any indications of pneumonia manifestations in lung imaging. In contrast, Severe Cases: Encompass patients meeting any of the specified severity criteria: 30 breaths/ min breathing rate, resting oxygen saturation below 93%, oxygen arterial pressure (PaO2)/ inspired oxygen fraction (FiO2) < 300 mm Hg, or those with over 50% of lung lesions detected in imaging within 24 to 48 hours. All patients underwent thorough clinical assessments, including detailed registrations, chest, and general examinations, and comprehensive laboratory examinations to identify potential complications. Patient recruitment took place between March and May 2021 at Menoufia University Hospital. All patients provided written informed consent., The ethical committee of Medical Research, Faculty of Medicine, Menoufia University, approved this study. (no: BIO .191219).

2.2. Sample Collection:

Peripherally blood samples were withdrawn from all subjects for routine COVID-19 assessments, which included CBC, CRP, ALT, AST, prothrombin time assessment, interleukin-6 (IL-6) measurement, lactate dehydrogenase (LDH) levels, ferritin levels, D-dimer analysis, and kidney function tests using commercially available assays.

2.3. Isolation of Genomic DNA:

Genomic DNA was isolated from peripheral whole blood samples utilizing the Genomic DNA Mini Kit from Geneaid (Taiwan), in accordance with the

manufacturer's guidelines. Subsequently, the extracted DNA was stored at -20°C until required for further use.

2.4. Genotyping of ACE2 rs2285666 and TMPRSS2 rs12329760:

Using the PCR-RFLP method, the genotyping of ACE2 rs2285666 and TMPRSS2 rs12329760 was carried out. For this process, PCR fragments encompassing rs2285666 and rs12329760 were first amplified using the DreamTaq Green PCR Master Mix (2x) from Thermo Fisher Scientific Inc. (USA). The primer sequences utilized for the RFLP-PCR analysis of ACE2 rs2285666 and TMPRSS2 rs12329760 are as follows, for the *ACE2* gene the Forward- 5-CATGTGGTCAAAAGGATATC-3′, the Reverse-5′-AAAGTAAGGTTGGCAGACAT-3′ (Srivastava *et al.,* 2020) and for TMPRSS2 Forward-5' CGCCCGTAGTTCTCGTTCC 3', Reverse-3' TTCGCCTCTACGGACCAAAC 5' (29) (Schönfelder *et al.,* 2021). For the *ACE2* rs2285666 polymorphism, the PCR products underwent incubation with the *AluI* restriction enzyme from Thermo Fisher Scientific Inc. (USA) at 37°C overnight. After digestion, the resulting fragment sizes were as follows: 466 bp for the GG genotype, 466 + 281 + 185 bp for the GA genotype, and 281 + 185 bp to identify the AA genotype *(Fig. 1a)*. For the TMPRSS2 rs12329760 polymorphism, the PCR products were subjected to incubation with the *Hpy8I* restriction enzyme from Thermo Fisher Scientific Inc. (USA) at 37°C overnight. Following digestion, The TT genotype exhibited a fragment size of 100 bp, the CT genotype displayed fragments of 100 + 65 + 35 bp, and the CC genotype showed fragments of 65 + 35 bp *(Fig. 1b)*. Subsequently, all digestion products were electrophoresed on a 3% agarose gel and visualized by staining with ethidium bromide from Sigma-Aldrich (Germany). The evaluation of these products was conducted using a gel documentation system.

2.5. Statistical Analysis:

The statistical analysis of the collected data was conducted using **the** IBM SPSS software package, version 20.0 (IBM Corp., Armonk, NY). Percentage and numerical data were used to convey quantitative information. The normal distribution of the data was evaluated through the Shapiro-Wilk test. Descriptive statistics, including the range (minimum and maximum values), mean, standard deviation, and median, were employed to characterize quantitative data. For the comparison of normally distributed quantitative data between the two study groups, the independent t-test was utilized. The correlation coefficient was employed to evaluate the relationship between polymorphism and illness features as well as severity. A significance level of $p < 0.05$ was established for all analyses.

3. RESULTS:

3.1. Demographic and Laboratory Data:

Table 1 presents the demographic, haematological, and biochemical features of the study cohorts. Notably, severe COVID-19 patients had a significantly higher age, with median ages of 65 years, compared to those with milder cases, who had a median age of 35 years. Nevertheless, there were no notable gender differences observed between the mild and severe groups. Severe patients exhibited significantly elevated levels of several serum parameters, including ALT, AST, Ferritin, IL6, LDH, WBC count, Prothrombin time, INR, Urea, and Creatinine, in contrast to mild patients. Conversely, severe patients had significantly lower levels of haemoglobin, RBC count, and platelet count. The lymphocytic count (both Absolute and Relative), CRP, and D-dimers exhibited no significant differences between the studied groups.

3.2. Genotypic and Allelic Frequencies of ACE2 and TMPRSS2 SNPs:

Table 2 investigates the prevalence of ACE2 rs2285666 and TMPRSS2 rs12329760 genotypes in both mild and severe cases of COVID-19. Notably, the prevalence of the TT genotype associated with TMPRSS2 rs12329760 was significantly higher in the severe group compared to the mild group ($p < 0.001$). However, there were no statistically significant differences observed in the genotypic or allelic distribution for ACE2 rs2285666 between the two groups.

3.3. Relation between gene polymorphism and demographic, biochemical and hematological data:

Table 3 analysed the data on ACE2 polymorphism in a severe cases group, no significant associations were found between ACE2 genotypes (GG, GA, and AA) and demographic, biochemical, or haematological parameters, except for a noteworthy gender disparity, with all females possessing the AA genotype. Despite a lack of strong associations, a trend towards significance was observed in D-dimer levels.

As regards the TMPRSS2 gene, data suggests that TMPRSS2 polymorphism might not be strongly associated with demographic, biochemical, or haematological variations in the severe cases group (Table 4).

4. DISCUSSION:

The worldwide COVID-19 pandemic, initiated by SARS-CoV-2, has led to an unparalleled global health emergency characterized by a notable mortality rate. *(*Dong *et al*., 2020). The disease manifests across a broad clinical spectrum, ranging from individuals with no apparent symptoms to those with mild upper respiratory symptoms, and in severe cases, progressing to bilateral pneumonia and ARDS necessitating critical care interventions *(*Huang *et al.*, 2020 and Wu *et al.*, 2020).

Our research concentrated on examining the genetic variations (polymorphism) within the ACE2 and TMPRSS2 genes and their influence on the prognosis of COVID-19 among Egyptian cases receiving treatment at Menoufia University Hospital from March 2021 to May 2021. The patients were categorized into two groups based on the severity of the disease: Mild and Severe.

Regarding the genotypic and allelic frequencies of ACE2 and TMPRSS2 single nucleotide polymorphisms (SNPs), the notable association between the TMPRSS2 rs12329760 TT genotype and severe COVID-19 is a noteworthy discovery. This aligns with recent studies underscoring the potential role of TMPRSS2 in SARS-CoV-2 infection (Asselta *et al*., 2020). However, the absence of significant differences in genotypic or allelic distribution for ACE2 rs2285666 is consistent with other studies (Karakaş *et al.*, 2021). In contrast certain studies propose that ACE2 genetic variations play a role in increasing susceptibility to COVID-19 (Cao *et al.*, 2020).

The noted substantial difference in median age between mild and severe COVID-19 patients corresponds with findings from prior studies that have identified age as a risk factor for severe disease (Richardson *et al*., 2020 and Zhou *et al*., 2020). This observation underscores the role of age as a non-genetic determinant of COVID-19 severity. In contrast, the lack of notable gender differences between the two groups is in accordance with results reported in certain studies. (Jin *et al.*, 2020), suggesting that gender may not be a primary determinant of disease severity.

The elevated levels of various serum parameters, including ALT, AST, Ferritin, IL6, LDH, WBC count, Prothrombin time, INR, Urea, and Creatinine in severe patients, mirror previous research indicating that these markers are related to the intensity and progression of COVID-19 *(*Zhang *et al*., 2020; Wang *et al*., 2020 and Chen *et al.*, 2020). Conversely, the lower levels of haemoglobin,

RBC count, and platelet count in severe patients may reflect the haematological complications in severe COVID-19 cases, as mentioned by *(*Lippi *et al.*, 2020*)*. The lack of significant differences in lymphocytic count, CRP, and D-dimers between the two groups aligns with some prior studies *(*Velavan and Meyer, 2020 and Tan *et al*., 2020).

The observed gender-specific association between ACE2 gene polymorphism and disease severity is intriguing. This finding supports the hypothesis that ACE2 genetic variants may interact differently with the virus in male and female individuals *(*Gemmati *et al.*, 2021*)*. However, the non-significant age differences based on ACE2 genotype align with some previous studies that also found no age-related associations *(*Benetti *et al.*, 2020*)*.

In contrast, TMPRSS2 gene polymorphism did not show significant associations with either gender or age, suggesting that TMPRSS2 genetic variations may not play a prominent role in these demographic factors in COVID-19 severity. This discovery aligns with findings from previous studies that found TMPRSS2 polymorphisms may not be directly linked to disease outcomes *(*Lucas *et al.*, 2020).

The non-significant differences in most biochemical parameters among ACE2 genotypes align with studies by *(*Guan*et al.*, 2020 and Vaduganathan *et al.*, 2020*)* which cast doubt on the direct impact of ACE2 polymorphism on inflammatory markers and liver enzymes. Conversely, the absence of significant differences in CRP, IL6, LDH, Ferritin, D-dimer, ALT, AST, Creatinine, and Urea among TMPRSS2 genotypes prompts questions about the direct influence of TMPRSS2 polymorphism on these biochemical markers. This contradicts studies of *(*Bertram *et al.*, 2011*)* suggesting that TMPRSS2 expression may affect viral entry and subsequent inflammatory responses. The lack of clear associations in our study underscores the complexity of the host-virus interaction, emphasizing the imperative for further investigations into the underlying mechanisms.

The absence of significant differences in haematological parameters suggests that ACE2 polymorphism may not be a primary determinant of variations in blood cell counts and coagulation profiles in severe cases. This observation aligns with studies by Ellinghaus, (2020). Similarly, the lack of significant differences in haematological parameters among TMPRSS2 genotypes challenges the hypothesis of *(*Nienhold *et al.*, 2020*)* that TMPRSS2 polymorphism directly influences blood cell counts and coagulation profiles in severe cases.

5. CONCLUSION:

This study provides valuable insights into the relationship between genetic variations in ACE2 and TMPRSS2 genes and the severity of COVID-19 among Egyptian patients. The findings demonstrate that severe cases of COVID-19 are associated with older age and exhibit significantly elevated levels of several serum parameters, indicating a more severe disease course. Notably, the presence of the TT genotype of TMPRSS2 rs12329760 is significantly more prevalent in severe cases, suggesting a potential association between this genetic variant and disease severity. On the other hand, no significant differences were observed in the genotypic or allelic distribution of ACE2 rs2285666 between mild and severe cases, indicating that this specific genetic variant may not play a major role in disease severity among the Egyptian population.

While no strong associations were found between ACE2 genotypes and demographic, biochemical or hematological parameters, an interesting gender disparity was identified, with all females exhibiting the AA genotype. This observation highlights the potential influence of sex-related gene variations on disease outcomes. Furthermore, the TMPRSS2

polymorphism did not show significant associations with demographic, biochemical, or hematological variations in severe cases, indicating that other factors may be more influential in determining disease severity in this context.

Overall, these findings contribute to our understanding of the genetic factors that contribute to the severity of COVID-19, specifically within the Egyptian population. By identifying specific gene variants, such as the TT genotype of TMPRSS2 rs12329760, that are associated with disease severity, this research opens avenues for further investigations into the underlying mechanisms and potential therapeutic targets. Further studies are needed to validate and expand upon these findings, which may ultimately aid in the development of personalized approaches for managing and treating COVID-19 based on an individual's genetic profile.

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AUTHOR CONTRIBUTIONS:

All authors participated in the conception and design of the study. F.S. collected the samples, while material preparation and data analysis were carried out by F.S., A.D., and M.S. The manuscript was drafted and revised by F.S., A.D., M.S., and H.S. All authors provided feedback on earlier manuscript versions and approval for the final manuscript.

COMPLIANCE WITH ETHICAL STANDARDS:

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Conflict of Interest: The authors affirm that there are no conflicts of interest.

Ethical approval: Approval for this study was granted by the ethics committee of the Faculty of Medicine's Medical Research at Menoufia University (no: BIO .191219).

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Table 1. Profile of Study Group Characteristics: Demographic, Biochemical, and Hematological Data.

Variables	Mean ± SD		P Value
	Mild cases (n = 48)	Severe cases (n = 48)	
Demographic Data			
***Age:* Range (Mean)**	22.0 – 58.0 (36.10)	31.0 – 87.0 (64.19)	<0.001*
Gender: number (%)			
Male	24 (50%)	23 (47.9%)	NS
Female	24 (50%)	25 (52.1%)	
Biochemical parameters			
CRP: number (%)			NS
Positive (> 10 mg/L)	36 (75 %)	40 (83.3 %)	
Negative	12 (25 %)	8 (16.7%)	
IL6 (Pg/ml)	28.28 ± 9.09	36.64 ± 19.78	0.049*
LDH (u/l)	196.59 ± 39.46	351.21 ± 307.41	0.001*
Ferritin (ng/ml)	85.51 ± 75.07	211.41 ± 114.75	<0.001*
D-dimer: number (%)			NS
Positive (>250 ng/ml)	0	6 (12.5%)	
Negative	48 (100%)	42 (87.5%)	
ALT (up to 40 U/L)	25.52 ± 6.99	53.75 ± 40.75	<0.001*
AST (up to 40U/L)	27.29 ± 7.03	57.19 ± 28.84	<0.001*
Creatinine (0.6-1.3 mg/dl)	0.97 ± 0.12	1.18 ± 0.45	0.003*
Urea (20-45 mg %)	31.69 ± 4.78	46.10 ± 15.50	<0.001*
Hematological parameters			
HB	12.88 ± 1.80	12.11 ± 1.57	0.028*
RBCs	4.89 ± 0.45	4.60 ± 0.67	0.015*
WBCs	3.44 ± 0.90	10.17 ± 5.19	<0.001*
Lymphocytes			
Absolute (1000-4000)	628.94 ± 251.98	761.96 ± 375.33	0.056
Relative (20-45 %)	13.53 ± 4.52	11.90 ± 5.90	0.139
Platelets	304.27 ± 95.66	144.33 ± 79.07	<0.001*
Prothrombin time (11-13.5 Sec)	12.04 ± 0.71	14.59 ± 1.26	<0.001*
INR (.8-1.1	0.96 ± 0.09	1.23 ± 0.24	<0.001*

Groups bearing different numbers are significantly different from each other at $P \leq 0.05$.
* Show statistical significant difference.

Table 2. Genotypic and allelic frequencies of ACE2 and TMPRSS2 SNPs in Mild and severe groups.

SNPs	Mild cases (n = 48) %	Severe cases (n = 48) %	P
ACE2 rs2285666			
GG	68.8	77.1	0.690
GA	22.9	16.7	
AA	8.3	6.3	
Allele			
G	80.2	85.4	0.339
A	19.8	14.6	
TMPRSS2 rs12329760			
CC	62.5	70.8	<0.001*
CT	35.4	8.3	
TT	2.1	20.8	
Allele			
C	80.2	75.0	0.387
T	19.8	25.0	

*: Statistically significant at $p \leq 0.05$

Table 3. Relation between ACE2 polymorphism and demographic, biochemical, and hematological data in severe cases group (n = 48).

Variables	Mean ± SD						P Value
	GG (n = 37)		GA (n = 8)		AA (n = 3)		
Demographic Data							
***Age:* Range (Mean)**	64.65 ± 9.69		60.50 ± 14.58		68.33 ± 7.64		0.474
Gender: number (%)							
Male	20	54.1 %	0	0.0 %	3	100.0 %	**0.001***
Female	17	45.9 %	8	100.0 %	0	0.0 %	
Biochemical parameters							
CRP: number (%)							
Positive	29	78.4 %	8	100.0 %	3	100.0 %	0.463
Negative	8	21.6 %	0	0.0 %	0	0.0 %	
IL6	38.57 ± 19.38		30.34 ± 23.23		29.63 ± 15.48		0.372
LDH	348.5 ± 317.9		292.8 ± 229.96		540.7 ± 385.91		0.491
Ferritin	215.5 ± 111.5		143.2 ± 56.54		342.2 ± 173.34		0.094
D-dimer: number (%)							
Positive	4	10.8 %	0	0.0 %	2	66.7 %	0.055
Negative	33	89.2 %	8	100.0 %	1	33.3 &	
ALT	55.65 ± 44.51		37.0 ± 12.75		75.0 ± 30.35		0.078
AST	58.65 ± 30.18		43.75 ± 14.43		75.0 ± 34.39		0.249
Creatinine	1.20 ± 0.48		1.11 ± 0.31		1.07 ± 0.21		0.876
Urea	47.22 ± 15.87		42.50 ± 15.43		42.0 ± 13.45		0.442
Hematological parameters							
HB	12.30 ± 1.59		11.40 ± 1.37		11.60 ± 1.70		0.292
RBCs	4.64 ± 0.69		4.43 ± 0.62		4.51 ± 0.64		0.711
WBCs	10.12 ± 5.63		10.56 ± 3.49		9.70 ± 4.36		0.775
Lymphocytes							
Absolute	727.2 ± 368.65		886.6 ± 463.43		858.0 ± 119.70		0.551
Relative	12.08 ± 5.64		12.65 ± 7.75		7.67 ± 2.08		0.486
Platelets	147.19 ± 85.93		147.50 ± 50.07		100.67 ± 46.70		0.491
Prothrombin time	14.67 ± 1.28		14.04 ± 1.08		15.13 ± 1.40		0.330
INR	1.22 ± 0.21		1.19 ± 0.26		1.45 ± 0.48		0.249

SD: Standard deviation
p: p value for Relation between ACE2 polymorphism and demographic, biochemical and haematological profile

Table 4. Relation between TMPRSS2 polymorphism and demographic, biochemical, and hematological data in severe cases group (n = 48).

Variables	Mean ± SD						P Value
	CC (n = 34)		CT (n = 4)		TT (n = 10)		
Demographic Data							
***Age:* Range (Mean)**	64.71 ± 11.10		60.25 ± 4.79		64.0 ± 10.24		0.730
Gender: number (%)							
Male	14	41.2 %	3	75.0 %	6	60.0 %	0.363
Female	20	58.8 %	1	25.0 %	4	40.0 %	
Biochemical parameters							
CRP: number (%)							
Positive	27	79.4 %	3	75.0 %	10	100.0 %	0.278
Negative	7	20.6 %	1	25.0 %	0	0.0 %	
IL6	38.09 ± 20.94		43.40 ± 18.19		28.99 ± 15.22		0.291
LDH	341.1 ± 226.6		313.5 ± 231.5		400.8 ± 532.5		0.721
Ferritin	193.10 ± 105.10		334.03 ± 175.34		224.6 ± 99.40		0.175
D-dimer: number (%)							
Positive	4	11.8 %	0	0.0 %	2	20.0 %	0.778
Negative	30	88.2 %	4	100.0 %	8	80.0 %	
ALT	53.62 ± 46.13		44.75 ± 15.52		57.80 ± 27.13		0.411
AST	54.21 ± 29.36		53.75 ± 14.06		68.70 ± 30.49		0.210
Creatinine	1.17 ± 0.49		1.33 ± 0.54		1.14 ± 0.18		0.607
Urea	46.38 ± 17.26		46.50 ± 12.40		45.0 ± 10.48		0.837
Hematological parameters							
HB	12.12 ± 1.58		11.0 ± 1.92		12.49 ± 1.31		0.279
RBCs	4.63 ± 0.57		4.18 ± 0.86		4.66 ± 0.91		0.419
WBCs	9.76 ± 4.59		9.28 ± 7.19		11.92 ± 6.45		0.503
Lymphocytes							
Absolute	742.89 ± 385.64		697.73 ± 287.61		852.50 ± 387.93		0.697
Relative	11.70 ± 5.66		10.30 ± 6.20		13.21 ± 6.94		0.732
Platelets	148.56 ± 86.54		144.75 ± 78.73		129.80 ± 53.10		0.997
Prothrombin time	14.62 ± 1.37		14.23 ± 1.18		14.64 ± 0.94		0.834
INR	1.23 ± 0.26		1.25 ± 0.26		1.22 ± 0.18		0.973

SD: Standard deviation

p: p-value for Relation between TMPRSS2 polymorphism and demographic, biochemical, and haematological profile/

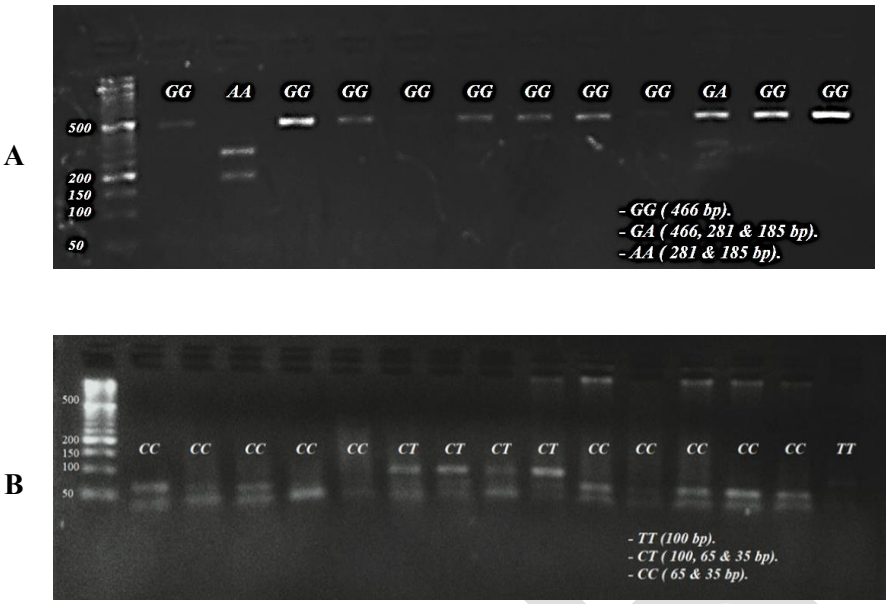

Figure 1: RFLP-PCR genotyping of ACE2 and TMPRSS2 Genes SNPs from Egyptian patients with COVID-19 by agarose gel electrophoresis. A) Genotyping of ACE2 rs2285666; *GG genotype product; 466 bp, while GA genotype; 466, 281 and 185 bp and AA genotype; 281 and 185 bp.* ***B) Genotyping of TMPRSS2 rs12329760;*** *TT genotype product; 100 bp, while CT genotype; 100, 65 and 35 bp and CC genotype; 65 and 35 bp.*