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The suppressor of cytokine signaling-1 (*SOCS1*) gene polymorphism and promoter methylation correlate with the course of COVID-19

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ABSTRACT

The suppressor of the cytokine signaling-1 (SOCS1) gene is a short sequence located on chromosome 16 that functions to induce an appropriate immune response and is an essential physiological regulator of interferon (IFN) signaling. In addition to comparing the global DNA and SOCS1 gene promoter methylation status between our patients with coronavirus disease 2019 (COVID-19) and healthy controls, this study demonstrates the effect of the SOCS1 rs33989964 polymorphism on patients with COVID-19. The study group included 139 patients diagnosed with COVID-19 in our hospital's clinics between June and December 2020, and the control group included 78 healthy individuals. After comparing the initial gene polymorphisms of the patients with the healthy control group, three separate clinical subgroups were formed. The gene polymorphism distribution and the methylation status of SOCS1 were examined in these clinical subgroups. Hypomethylation of the SOCS1 gene was observed in the COVID-19 patient group compared to the healthy control group ($p = 0.001$). Between the patients divided into two separate clinical subgroups, those with severe and mild infections, the Del/Del genotype of the SOCS1 gene was more common in patients with severe infection than in patients with mild infection ($p = 0.018$). Patients with the CA/CA and CA/Del genotypes were 0.201 times more likely to have a severe infection (95% CI: 0.057–0.716, $p = 0.007$). Having a non-Del/Del genotype was a protective factor against severe infection. The effect of the SOCS1 rs33989964 polymorphism and methylation status of the SOCS1 gene throughout the COVID-19 pandemic could be significant contributions to the literature.

KEYWORDS

COVID-19; SOCS1; polymorphism; methylation; prognosis; mortality

Introduction

Coronavirus disease 2019 (COVID-19) is a pandemic that has infected millions of people since the first case has been detected and is among the major infectious events of the century [1]. Severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2) has been identified as the causative agent of COVID-19. Additionally, it was observed that the disease had a mortal course in a significant proportion of patients aged 65 and over and those with co-morbidities [2–4]. Patients with co-

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morbidities such as hypertension, chronic respiratory and heart diseases, diabetes mellitus, renal failure, and malignancy have been among the most severely affected groups [5].

Current COVID-19 research focuses on various factors to understand immune pathophysiology and develop novel treatment modalities. Within this regard, the host-based genetic approach is one of them. The suppressor of the cytokine signaling-1 (*SOCS1*) gene is a short sequence located on chromosome 16 [6]. The coding sequence consists of two exons regulated by a promoter region characterized by a large CpG island spanning the gene from its promoter to the end of exon 2 [6]. *SOCS1* induces an effective immune response and is an essential physiological regulator of interferon (IFN) signaling [6,7]. Activated IFN production is essential for atopic disorders. *SOCS1* directly interacts with Janus kinases (JAKs), the primary intracellular mediators of immune cytokine action, and inhibits their tyrosine kinase activities [7]. Overexpression of *SOCS1* causes viral-mediated end-organ injury in the early stages of infection. Moreover, *SOCS1* inhibits the expression of the antiviral proteins Myxovirus resistance-A and 2–5 oligoadenylate synthetase [8].

DNA methylation, an epigenetic regulatory mechanism frequently studied in many tumors, involves adding a methyl group to the carbon 5 position of cytosine [9]. This reaction is catalyzed by DNA methyltransferases and occurs when cytosine is part of a 5′-CG-3′ sequence known as CpG or CG dinucleotide [10–12]. CpG islands are small DNA regions normally located at the 5′ end of a gene. They range in size from 0.5 to 5 kb and are generally protected from methylation, leading to the downregulation of their expression [10–12].

In this study, COVID-19 patients and healthy controls were compared in terms of global DNA methylation and SOCS1 gene promoter methylation. Furthermore, the present study aims to investigate the effect of the *SOCS1 rs33989964* polymorphism on patients with COVID-19.

Material and methods

In the study, 139 patients diagnosed with COVID-19 in our hospital's clinics between June and December 2020 were included in the study group, while 78 healthy individuals constituted the control group. The control group consisted of volunteers with the same ethnicity, age, and gender, with no active infection and no consanguinity. They were also checked with nasopharyngeal swab PCR to exclude asymptomatic COVID-19 positivity. In addition to demographic information, including age and gender, co-morbidities, clinical findings, physical examination findings, and initial laboratory results were noted down for each patient. After comparing the initial gene polymorphisms of the patients with the healthy control group, three separate clinical subgroups were formed. The distribution of gene polymorphisms and their statistical significance were examined in these patient groups.

Furthermore, the SOCS1 methylation status was checked in samples taken after polymerase chain reaction (PCR) negativity and complete clinical recovery in addition to the samples taken at the time of diagnosis of COVID-19 infection. The methylation status of *SOCS1* in patients with COVID-19 was also examined in these clinical subgroups (Table 1).

In the present study, patients whose nasopharyngeal swab PCR results were positive, or whose computed tomography (CT) results were compatible with typical COVID-19 involvement, were included in the study.

DNA isolation, genotyping, and methylation

Leukocytes isolated from the blood samples taken from the patients and control group individuals were put into 2 ml EDTA tubes and studied. Additionally, DNA isolation was performed from the obtained leukocytes with the Quick-DNA Miniprep Plus Kit (Zymo Research) according to the manufacturer's instructions. DNA samples were stored at −20 °C.

The *SOCS1* −1478 CA/Del polymorphism was analyzed by the PCR-restriction fragment length polymorphism (RFLP) technique. The primer sequences used for the *SOCS1* −1478 CA/Del polymorphism were 5′-TGTCGTCCAGCTGCACCTC-3′ (forward) and 5′-ACCACAGGCTTCAGAGGAAC-3′ (reverse). The size of the PCR product was determined to be 250 bp. The DdeI enzyme was used as a restriction endonuclease. The cut products were kept at 37 °C for one night, run on a 2.5% agarose gel, and visualized under UV light. Fragment lengths were determined as 250 bp, 145 bp, and 105 bp for the CA/Del genotype, 145 bp and 105 bp for the CA/CA genotype, and 250 bp and 105 bp for the Del/Del genotype (Figure 1) [13].

DNA bisulfite conversion was first performed for *SOCS1* gene methylation analysis using the EZ-96 DNA Methylation-Gold™ Kit (Zymo Research) protocol.

Bisulfite-converted DNA samples were analyzed using the real-time quantitative methylation-specific PCR method to measure the methylation level of the *SOCS1* gene. The primer sequences for the *SOCS1* gene were 5-TTCGCGTGTATTTTTAGGTCGGTC-3 (forward) and 5-CGACACAACTCCTACAACGACCG3 (reverse) [12]. The primer sequences for the control gene β-actin were 5′-TGGTGATGGAGGAGGTTTAGTAAGT-3′ (forward) and 5′-AACCAATAAAACCTACTCCTCCCTTAA-3′ (reverse) [14]. The percent methylated reference (PMR) value expresses the percent methylation of the gene of interest. It was calculated using the 2-ΔΔCq method. ΔΔCq was calculated using the following formula: Sample DNA (Cq target gene-Cq ACTBcontrol)-fully methylated DNA (Cq target gene-Cq ACTB control) [15].

Table 1. Clinical subgroups.

Severe/mild infection	• Respiratory rate more than 30/min, • Presence of dyspnea or peripheral oxygen saturation <90%, nasal oxygen requirement more than 5 L/min, PaO2/FiO2 ≤ 300, or lactate >2 mmol/L, • Presence of hypotension (if systolic blood pressure 40 mmHg lower than normal systolic blood pressure) or a heart rate >100 beats/min, • Presence of renal, hepatic, hematological (thrombocytopenia) or cerebral (confusion) dysfunction, • Presence of sepsis or septic shock or skin findings such as cutis marmorata and peripheral coldness, • Presence of mild/severe pneumonia (bilateral infiltration and/or the presence of multiple ground-glass opacities), • Presence of the need for anti-cytokine therapy and/or the presence of broad-spectrum antibacterial therapy
Exitus/alive during the 28-day follow-up	• Patients who were exitus or alive during the 28-day follow-up period
The need for intensive care/being only inpatient	• Patients who needed intensive care follow-up at any period during hospitalization and those who were treated as inpatients

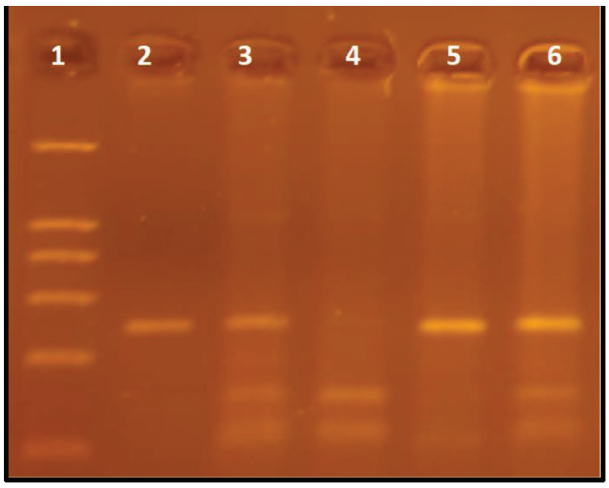

Figure 1. Agarose gel image showing the genotypes against the DNA size ladder. 1: ladder, 2: PCR product, 3,6: CA/Del (250,145,105 bp), 4: CA/CA (145,105 bp), 5: Del/Del (250 bp)

According to the manufacturer's manual, the global methylation status of DNA obtained from blood taken from the patient and control groups was measured using the 5-mC DNA ELISA Kit (Zymo Research). The standard curve was generated with the negative and positive controls included in the kit. The percent 5-mC in each sample was calculated using the y-intercept and slope generated by logarithmically plotting the absorbance values of seven DNA control samples with known 5-mC amounts. In 25% of the samples, measurements were repeated [16].

Ethical committee approval was obtained (xxx University, Faculty of Medicine, approval date and number: 29/05/2020–86529), and the patients and control subjects gave informed consent before the onset of the study. The experimental procedures were based on the Ethical Principles of Declaration of Helsinki and relevant institutional regulations.

Statistical analysis

IBM SPSS Statistics for Windows 21.0 (IBM Corp., Armonk, NY, USA) was used for the statistical analysis. The descriptive statistics were expressed as the mean, standard deviation, median, minimum, and maximum for the continuous variables after assessing their normality, while frequency and percentage were used to express the nominal variables. The Pearson chi-square test or Fisher's exact test was used to comparing the discrete variables, and Bonferroni correction was conducted in the pairwise comparisons to determine which group or groups showed statistically significant results. Multivariate binary logistic regression analyses were conducted to determine the association between different variants of the genes and the study parameters. The results were adjusted for age and sex.

Consequently, the odds ratio (OR) and 95% confidence interval (CI) were used to express the association of the gene variants with the study parameters. The Hardy-Weinberg equilibrium (HWE) was calculated using the De-Finetti program (online HWE and Association Testing – Institut für Humangenetik, Munich, Germany). The results were considered significant at $p < 0.05$ in all analyses. The effect size was determined to be 0.63%. The power of the study with an alpha of 0.05 was calculated to be 88%.

Results

Of the patients diagnosed with COVID-19, 56 (40.3%) were female, and 83 (59.7%) were male. The most common comorbidity was hypertension (32.4%), affecting 45 patients. Of all patients included in the study, 94 (67.6%) had severe infections and 45 (32.4%) had mild infections. A total of 10 patients (7.2%) received tocilizumab due to disease severity. The number of patients who required intensive care was 12.2% (17 patients), and the 28-day mortality was 6.5% (9 patients). Table 2 shows the demographic data and clinical characteristics of the patients.

Distribution of the genotypes and methylation status of *SOCS1*

Statistical analysis of the distribution of SOCS1 gene variants between the patient group and healthy

Table 2. Demographic and clinical features of patients with COVID-19.

			COVID-19	COVID-19
			n[a] (%)	median
Age				55 (22–92)
Gender	Female/Male		56/83 (40.3/59.7)	
Comorbidity	Hypertension		45 (32.4)	
	DM		23 (16.5)	
	COPD		17 (12.2)	
	CAD		10 (7.2)	
	CHF		3 (2.6)	
	Solid malignancy		21 (14.1)	
	Hematologic malignancy		3 (2.6)	
Clinical subgroups	Severe/Mild		94/45 (67.6/32.4)	
	Cough		84 (60.4)	
	Fever		78 (56.1)	
	Myalgia		74 (53.2)	
	Dyspnea		56 (40.3)	
	Nausea-Vomiting		17 (12.2)	
	Diarrhea		12 (8.6)	
	Anosmia		3 (2.6)	
Initial physical examination	Fever			36.7 (35–40)
	spO2			96 (80–100)
	Systolic BP			130 (90–200)
	Diastolic BP			76 (50–110)
	Heart rate			92 (60–160)
	Respiratory rate			16 (13–40)
	pH			7.41 (7–8)
	pO2			63 (35–86)
	pCO2			38 (23–58)
	HCO3			24 (14–30)
	Lactate			1.45 (1–5)
Laboratory	Hemoglobin	gr/dL		13.2 (6.3–18)
	Leukocyte	µL		6770 (2200–28.300)
	Platelet	10^3/µL		240 (66–576)
	Lymphocyte	µL		1240 (290–4500)
	Lymphocyte	<800 µL	32 (23)	
	C-reactive protein	mg/dL		36 (1–363)
	Procalcitonin			0.07 (0.20–50.0)
	Ferritin			239 (9–6656)
	D-Dimer			690 (190–20000)
Anti-cytokine therapy				
	Tocilizumab		10 (7.2)	
28-day mortality			9 (6.5)	
Need for ICU			17 (12.2)	

n[a] = 139.

Abbreviations: DM: Diabetes mellitus, **COPD**: Chronic obstructive pulmonary disease, **CAD**: Coronary artery disease, **CHF**: Congestive heart failure, **sPO_2**: capillary oxygen saturation, **BP**: Blood pressure, **pO_2**: Partial pressure of oxygen, **pCO_2**: Partial pressure of carbon dioxide, **HCO_3**: Bicarbonate, **ICU**: Intensive care unit.

controls was performed. There was no significant difference in genotype distribution ($p > 0.05$). The Del allele was the minor allele, and its frequency was 0.488 in the COVID −19 patient group and 0.442 in the healthy controls. Hypomethylation of the SOCS1 gene was observed in the COVID −19 patient group compared to the healthy controls (PMR value for COVID −19 patients: 16.34, range: 2.11–998.21; PMR value for healthy controls: 43.48, range: 2.76–247.75; $p = 0.001$) (Figure 2a). No significant result was obtained with respect to global DNA methylation (Table 3).

In the post-treatment analysis, although an increase in the PMR value of the SOCS1 gene was observed compared with the pre-treatment measurements, no statistical significance was observed ($p = 0.089$) (Table 4).

The statistical analysis was expanded by dividing the patients into subgroups:

1) Severe/mild infection

According to the symptoms of the disease, patients were divided into two clinical subgroups: Groups with severe infection and with mild infection. The Del/Del genotype of the SOCS1 gene was more common in patients with severe infection than in patients with mild infection ($p = 0.018$). When grouped into Del/Del and other genotypes, the Del/Del genotype was found to be significantly more common in patients with severe infection; patients with CA/CA and CA/Del genotypes were 0.201 times more likely to have a severe infection (95% CI: 0.057–0.716, $p = 0.007$). The presence of a non-Del/Del genotype was a protective factor against severe infection. There was no significant difference in the PMR value of the SOCS1 gene (Figure 2b) and global DNA methylation between the two clinical subgroups ($p > 0.05$ for all) (Table 5).

2) Exitus/alive during the 28-day follow-up

According to their last condition during their 28-day follow-up, patients were divided into two clinical subgroups: exitus and alive groups. There was no significant difference in the distribution of SOCS1 genotypes in both groups ($p > 0.05$ for all). The PMR value of the SOCS1 gene (Figure 2b) and global DNA methylation were not significantly different between the two clinical subgroups ($p > 0.05$ for all) (Table 6).

3) The need for intensive care/being only an inpatient

Regarding the need for intensive care during hospitalization, patients were divided into two clinical subgroups: those who required ICU follow-up and those who did not. There was no significant difference in the distribution of SOCS1 genotypes in both groups ($p > 0.05$ for all). There was no significant difference in the PMR value of the SOCS1 gene (Figure 2b) and global DNA methylation between the two clinical subgroups ($p > 0.05$ for all). (Table 7).

The distribution of *SOCS1* methylation between genotypes of patients and healthy controls is shown in Figure 2c.

Discussion

This study highlights the literature on COVID −19 host factor-based immunogenesis with many aspects.

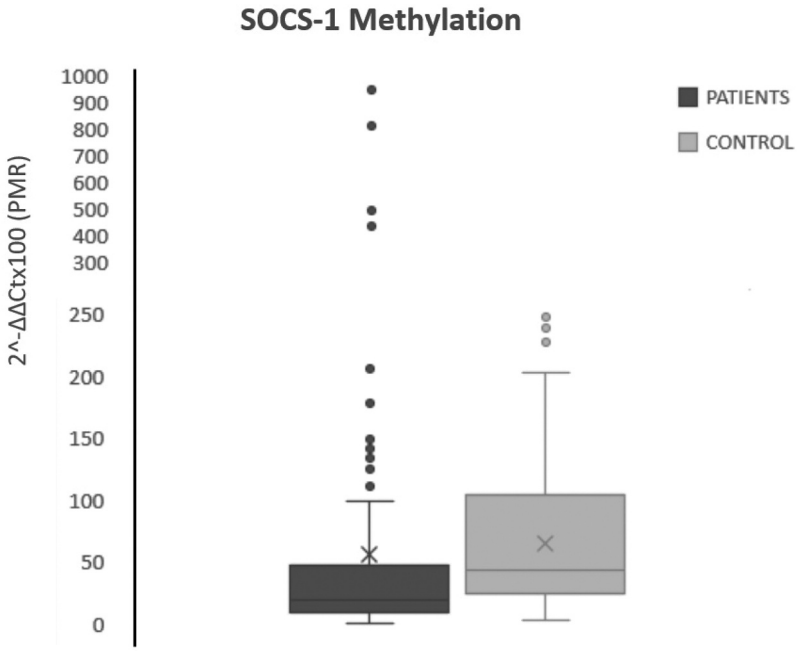

Figure 2a. Distribution of SOCS-1 methylation between patients and healthy controls.

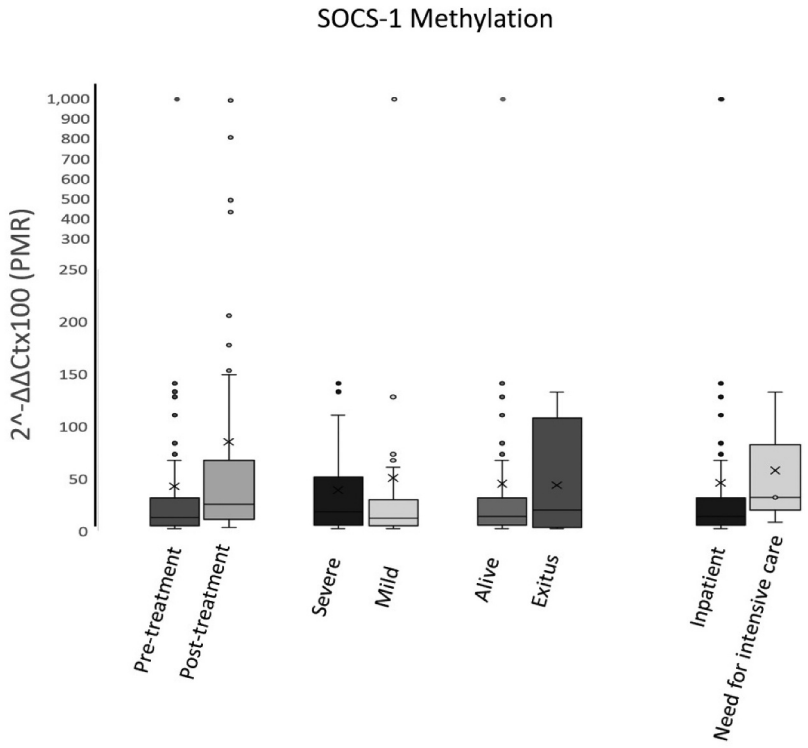

Figure 2b. Distribution of SOCS-1 methylation between subgroups of the study.

Major contributions of the present study are the association of the Del/Del genotype with severe COVID −19 infection and significant hypomethylation of the SOCS1 gene in the COVID −19 patient group compared to healthy controls.

The *SOCS1* (−1478, rs33989964) polymorphism represents a dinucleotide CA insertion/deletion (CA/Del) associated with respiratory immune-inflammatory diseases with high expression levels [7]. Additionally, data from the literature have revealed that hypermethylation reduces *SOCS1* gene expression, while hypomethylation induces gene expression [17, 18, 19]. There are limited data in the *literature about SOCS1* gene polymorphisms and the role of *SOCS1* gene expression in viral infections. In a study investigating the link between viral infection and the SOCS1 gene, it was found that herpes simplex virus-induced expression of SOCS1 and SOCS3 bypasses

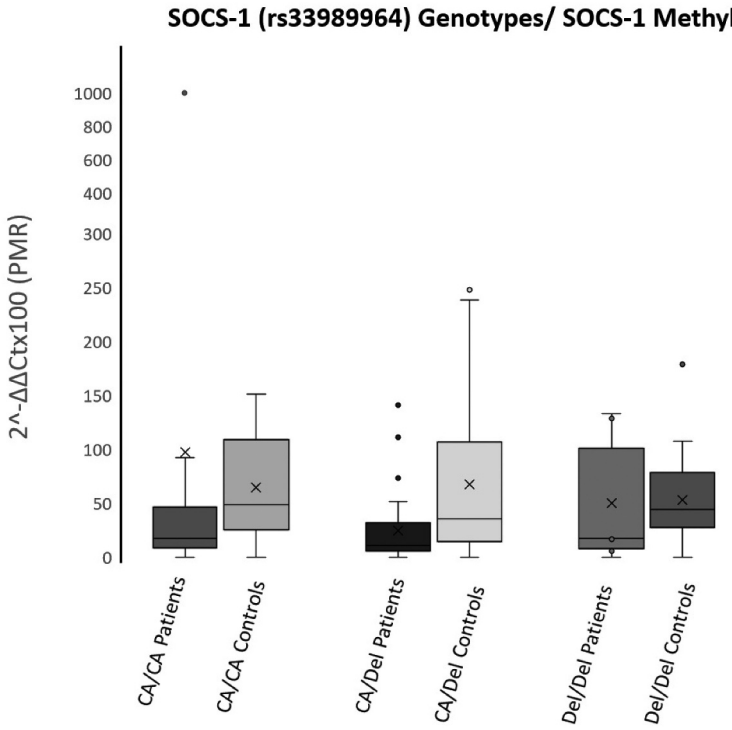

Figure 2c. Distribution of SOCS-1 methylation between genotypes of patients and healthy controls.

Table 3. Distribution of SOCS-1 gene variants and methylation (global DNA and SOCS-1) status between patients with COVID-19 and healthy controls.

SOCS-1	Genotype	COVID-19	Healthy Controls	OR Exp(B)	95% CI	p-value
		n= [a] (%)	n = 78 (%)			
	CA/CA	56 (40.3)	29 (37.2)	0.711*	0.329–1.538*	0.386*
	CA/Del	58 (41.7)	29 (37.2)	0.643*	0.298–1.387*	0.260*
	DEL/Del	25 (18.0)	20 (25.6)	0.595[&]	0.307–1.153&	0.166[&]
Allele						
	CA	170 (61.2)	87 (55.8)			
	Del	108 (48.8)	69 (44.2)	0.801[&]	0.538–1.192&	0.309[&]
HWEp		**0,151**	**0,129**			
2^-ΔΔCt	×100 (PMR)	16.34 (2.11–998.21)	43.48 (2.76–247.75)			**0.001**[#]
Global methylation		10.73 (1.82–45.20)	11.03 (2.77–18.82)			0.851[#]

[a]n = 139, *: OR (95%CI) was adjusted by age and sex, [&]Fisher's Exact Test, [#]median test.
Abbreviations: SOCS-1: The suppressor of cytokine signaling-1, **COVID-19**: Coronavirus disease 2019, **OR**: Odds ratio, **CI**: confidential interval, **PMR**: Percent methylated reference.

Table 4. Change in methylation status (global DNA and SOCS-1): Pretreatment and posttreatment measurements.

SOCS-1	Genotype	Pretreatment	Post-treatment	OR Exp(B)	95% CI	p value
2^-ΔΔCt	×100 (PMR)	16.34 (2.11–998.21)	25.88 (2.72–991.02)			0.089[#]
Global methylation		10.73 (1.82–45.20)	8.99 (3.29–68.23)			0.602[#]

[#]median test.
Abbreviations: SOCS-1: The suppressor of cytokine signaling-1, **OR**: Odds ratio, **CI**: confidential interval, **PMR**: Percent methylated reference.

innate immunity by inhibiting the production of type I and type III interferon [20]. Similar mechanisms have been described for the respiratory syncytial virus, Zika virus, and SARS-CoV-2 [21–24]. Induction of SOCS expression has been demonstrated following SARS-CoV-2 infection [24]. Interferons play an important role in antiviral cytokine mechanisms, and their inhibition is among the important viral escape phenomena [25, 26]. The presence of hypomethylation associated with high *SOCS1* expression was observed in the COVID-19 patient group in our study. However, no significant difference was observed between the

Table 5. Distribution of SOCS-1 gene variants and methylation status (global DNA and SOCS-1) between the clinical subgroups: severe or mild infection.

SOCS-1	Genotype	Severe	Mild	OR Exp (B)	95% CI	p value*
		n= [a] (%)	n = 45 (%)			
	CA/CA	36 (38.3)	20 (44.4)	0.205*	0.051–0.828*	0.026*
	CA/Del	36 (38.3)	22 (48.9)	0.230*	0.058–0.917*	0.037*
	Del/Del	22 (23.4)	3 (6.7)	4.278[&]	1.280–15.155[&]	**0.018[&]**
	CA/CA+CA/Del	72	42	0.201	0.057–0.716	
	Del/Del	22 (23.4)	3 (6.7)		1.280–15.155&	**0.007**
2^-ΔΔCt	×100 (PMR)	18.75 (2.11–141.05)	13.57 (2.76-998-21)			0.364[#]
Global methylation		10.88 (1.82–30.44)	10.58 (3.18–45.2)			1.000[#]

Abbreviations: SOCS-1: suppressor of cytokine signaling-1, **COVID-19**: coronavirus disease 2019, **OR**: odds ratio, **CI**: confidence interval, **PMR**: percent methylated reference.

Table 6. Distribution of SOCS-1 gene variants and methylation status (global DNA and SOCS-1) between clinical subgroups: exitus or alive during the 28-day follow-up.

SOCS-1	Genotype	Exitus	Alive	OR Exp (B)	95% CI	p value*
		n= [a] (%)	n = 130 (%)			
	CA/CA	4 (44.5)	52 (40.0)	1.459*	0.229–9.285*	0.689*
	CA/Del	3 (33.3)	55 (42.3)	1.768*	0.259–12.061*	0.561*
	Del/Del	2 (22.2)	23 (17.7)	0.752[&]	0.147–3.858[&]	0.664[&]
2^-ΔΔCt	×100 (PMR)	18.75 (2.11–141.05)	16.0 (2.76-998-21)			0.458[#]
Global methylation		11.69 (9.58–13.80)	10.73 (1.82–45.2)			0.473[#]

[a]n = 9, *: OR (95%CI) was adjusted by age and sex, [&]Fisher's Exact Test, [#]median test.
Abbreviations: SOCS-1: The suppressor of cytokine signaling-1, **OR**: Odds ratio, **CI**: confidential interval, **PMR**: Percent methylated reference.

Table 7. Distribution of SOCS-1 gene variants and methylation status (global DNA and SOCS-1) between the clinical subgroups: the need for intensive care or being an inpatient.

SOCS-1	Genotype	Need for intensive care	Inpatient	OR Exp (B)	95% CI	p value*
		n= [a] (%)	n = 122 (%)			
	CA/CA	6 (35.3)	50 (41.0)	1.958*	0.466–8.224*	0.359*
	CA/Del	7 (41.2)	51 (41.8)	1.425*	0.357–5.687*	0.616*
	Del/Del	4 (23.5)	21 (17.2)	0.676[&]	0.200–2.278[&]	0.509[&]
2^-ΔΔCt	×100 (PMR)	32.04 (8.51–133.1)	16.18 (2.11–998.21)			0.977[#]
Global methylation		9.58 (6.57–13.8)	10.88 (1.82–45.2)			1.000[#]

[a]n = 17, *: OR (95%CI) was adjusted by age and sex, [&]Fisher's Exact Test, [#]median test.
Abbreviations: SOCS-1: The suppressor of cytokine signaling-1, **OR**: Odds ratio, **CI**: confidential interval, **PMR**: Percent methylated reference.

COVID-19 patient group and healthy controls regarding the distribution of *SOCS1* gene variants associated with different expression levels. This study is the first to demonstrate the escape of antiviral mechanisms and increased susceptibility to viral infections. Additionally, the SOCS1 gene's methylation was significantly increased after COVID-19; however, the statistical significance of this result could not be determined.

The IFN-JAK-STAT pathway induces the cytokine storm that occurs during COVID −19 in contrast to its role in preventing viral infections. Johnson et al [27] investigated the paradoxical clinical situation due to the risk of cytokine storm induction. They indicated that the SOCS-1/3 inhibitor pJAK2 (1001–1013) they developed could be used to prevent COVID −19 infections. JAK-STAT Inhibitors used in COVID −19 related cytokine storms and their clinical utility were a guide for new agents [28–30]. It is important to highlight the fact that the Del/Del genotype associated with low SOCS1 expression and high cytokine secretion was significantly higher in patients with severe COVID-19. The presence of a non-Del/Del genotype was shown to be a protective factor against severe infections, whereas there was no significant factor for other clinical subgroups. This result suggests that in addition to JAK inhibitors, 'SOCS mechanism-related agents' may soon be used for similar purposes. Considering the anti-cytokine treatment options used in the course of COVID −19, it can be hypothesized that SOCS-1 agonists may be involved in the treatment by inhibiting not only a single cytokine but multiple proinflammatory cytokines (such as IFN, interleukin-1, interleukin-6).

There were also some limitations to our study. The most important limitation was that the concomitant expression of SOCS1 could not be detected. In addi-

tion, the increase in SOCS1 methylation after COVID −19 infection could be statistically significant in a larger group of patients.

In conclusion, in this study, hypomethylation of the SOCS1 gene was observed in COVID −19 patients compared with healthy controls. The Del/Del genotype of the SOCS1 gene was more common in patients with severe COVID −19 infection. A non-Del/Del genotype was protective against severe infection. The distribution of the SOCS1 genotype was associated with disease severity, whereas methylation status was related to disease susceptibility. During the course of COVID −19, in which JAK or SOCS agonists could also be used as an anti-cytokine treatment option, hypomethylation of the SOCS1 gene increases susceptibility to COVID-19 but is not associated with disease severity. It is considered that the effects of SOCS1 polymorphism rs33989964 along with methylation status of the SOCS1 gene during the course of COVID −19 may make a significant contribution to the literature regarding the SOCS1 gene.

Acknowledgments

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Authors' contributions

T.T., S.P., A.M., I.S., H.A., N.S., Y.O., and U.I.A. collected the data; M.P., I.S., and S.P. contributed data or analysis tools; M. P. performed the analysis; I.S. and S.P. wrote the paper.

Data availability statement

The authors declare that data supporting the findings of this study are available within the referenced articles.

Ethical approval and consent to participate

Ethical committee approval was received (xxx University, Faculty of Medicine, approval date and number: 29/05/2020–86529), and the patients and control subjects gave informed consent before the beginning of the study. The experimental procedures were based on the Declaration of Helsinki and relevant institutional regulations.

Patient consent for publication

Informed consent was obtained as written forms from all our patients to publish.

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