Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

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In silico identification of potential miRNAs -mRNA inflammatory networks implicated in the pathogenesis of COVID-19

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ABSTRACT

COVID-19 has been found to affect the expression profile of several mRNAs and miRNAs, leading to dysregulation of a number of signaling pathways, particularly those related to inflammatory responses. In the current study, a systematic biology procedure was used for the analysis of high-throughput expression data from blood specimens of COVID-19 and healthy individuals. Differentially expressed miRNAs in blood specimens of COVID-19 vs. healthy specimens were then identified to construct and analyze miRNA–mRNA networks and predict key miRNAs and genes in inflammatory pathways. Our results showed that 171 miRNAs were expressed as outliers in box plot and located in the critical areas according to our statistical analysis. Among them, 8 miRNAs, namely miR-1275, miR-4429, miR-4489, miR-6721-5p, miR-5010-5p, miR-7110-5p, miR-6804-5p and miR-6881–3p were found to affect expression of key genes in NF-KB, JAK/STAT and MAPK signaling pathways implicated in COVID-19 pathogenesis. In addition, our results predicted that 25 genes involved in above-mentioned inflammatory pathways were targeted not only by these 8 miRNAs but also by other obtained miRNAs (163 miRNAs). The results of the current in silico study represent candidate targets for further studies in COVID-19.

1. Introduction

Health care systems across the world have been noticeably influenced by Coronavirus disease 2019 (COVID-19) pandemic (Das et al., 2021; Rakhsha et al., 2020). COVID-19 mortalities and morbidities including post-recovery syndromes mainly result from immune-homeostasis disruption in infected people (Choudhury and Mukherjee, 2020). It has recently been found that spike glycoprotein, which is the major infectious surface protein of SARS-CoV-2, acts as a ligand for human TLR4 (Choudhury et al., 2022), and this protein–protein interaction activates transcription factors such as NF-κB, interferon regulatory factor and activator protein 1 (AP-1) that encode pro-inflammatory cytokines (Patra et al., 2021a) such as CC-chemokine ligand 2 (CCL2), interleukin 6 (IL-6), tumor necrosis factor (TNF), C-reactive protein (CRP), and CXC-chemokine ligand 10 (CXCL10) (Jiang et al., 2022). These inflammatory mediators are capable of activating various adaptive and innate immune cells, which in turn amplifies the inflammatory environment and leads to clear immunopathology, that is 'cytokine storm'. Major organs such as heart, pancreas, and kidney are directly damaged by cytokine storm (Choudhury et al., 2021; Mukherjee, 2022; Patra et al., 2021b). Note that inflammatory molecules not only increase the progress of the disease early during viral infection but also remain in the patients' plasma after acute infection and may impact patients surviving COVID-19 in long term (Patel et al., 2021). It has been found that this hyper-inflammation, known as Multisystem Inflammatory Syndrome (MIS), affects patients 4 to 6 weeks after their infection (Yao

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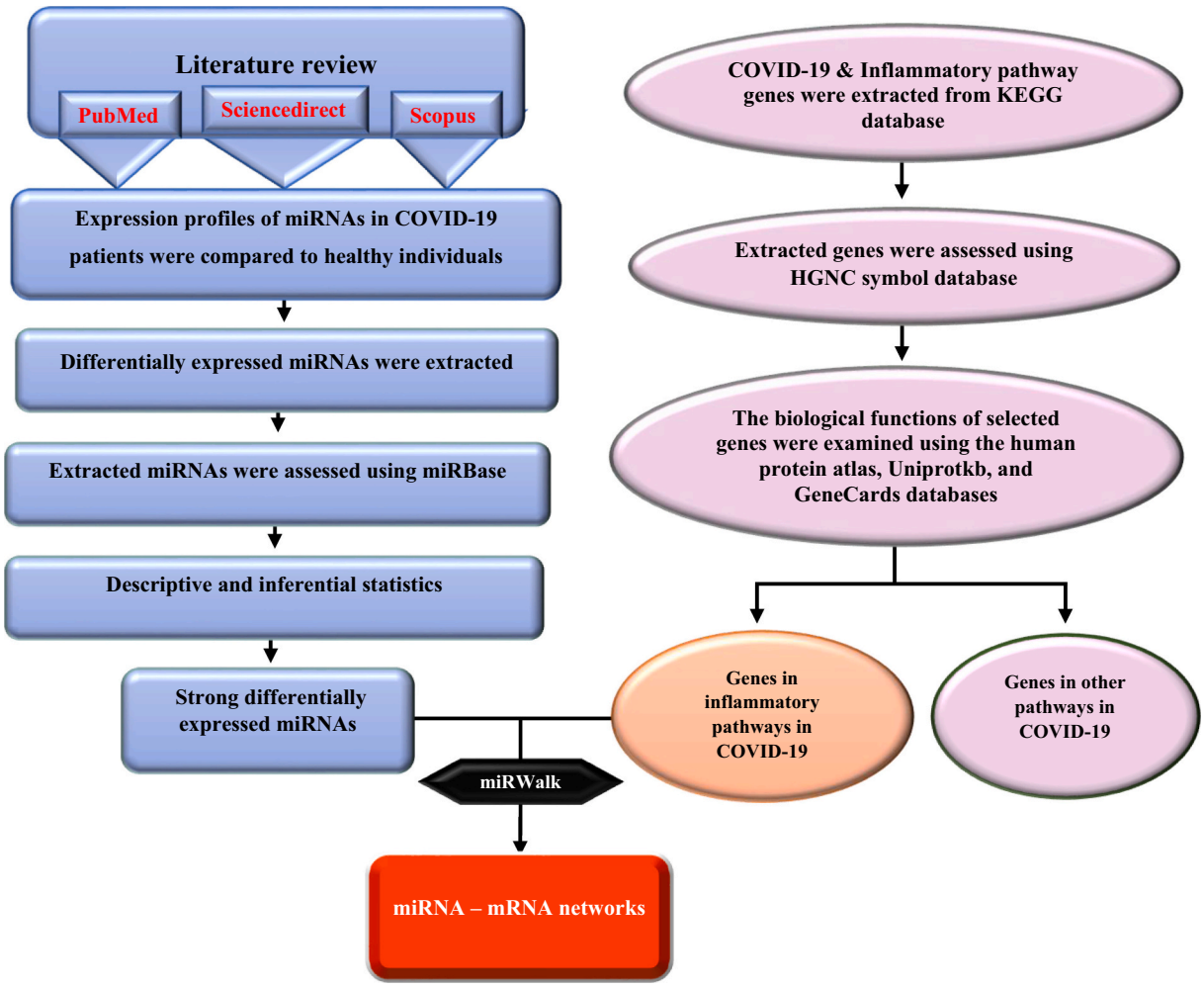

Fig. 1. Study design flowchart.

et al., 2021) and leads to shock, gastrointestinal symptoms, cardiac dysfunction, and increased inflammatory markers (Mezochow et al., 2022).

Despite the existence of multiple vaccine options and treatments across the globe to prevent COVID-19 infection, the majority of these are not sufficiently efficient to target late-stage cytokine storm associated with COVID-19. Hence, in addition to the prevention or blocking of the entrance of the virus, greater attention should be paid to the reduction of hyperinflammation induced by COVID-19 (Zoulikha et al., 2022).

According to the literature, multiple physiological parameters change due to infection and cytokine storm, among which, microRNAs (miRNAs) can represent this poor state given their pivotal contribution to various essential biological processes (Garcia-Giralt et al., 2022). These processes include neuronal development, metabolic control, muscle differentiation, stem cell differentiation, cell cycle, and immune modulation. Thus, miRNA dysfunction can be linked to a diverse range of diseases, such as cancers, immune system disorders, and cardiovascular diseases (Condrat et al., 2020).

As small non-coding RNAs with an approximate length of 20 to 25 nucleotides, miRNAs are responsible for regulating the expression of several target genes via sequence-specific hybridization to the 3′ untranslated region (UTR) of messenger RNAs. These miRNAs can either prevent the translation or directly degrade their target messenger RNAs (Leichter et al., 2017). Regarding the fact that perfect complementarity is not needed for target recognition by miRNAs, a single miRNA is able to regulate several messenger RNAs. Despite the slight impact of miRNAs on each messenger RNA's target, the combined impact is considerable and generates notable phenotypic results. The fact that the majority of miRNAs are conserved across several species of animals indicates the evolutionary significant modulatory role of these molecules in essential biological processes and pathways (Christopher et al., 2016).

These targeting pathways of miRNAs in human diseases represent a novel and potentially strong therapeutic candidate for different pathological afflictions (Christopher et al., 2016). In addition, as reported in many research works, miRNAs can be released into extracellular fluids including plasma and serum, saliva, cerebrospinal fluid, breast milk, tears, urine, peritoneal fluid, colostrum, bronchial lavage, ovarian follicular fluid, and seminal fluid. Extracellular miRNAs, in contrast to cellular RNAs, demonstrate high stability, such that they can resist degradation when exposed to ambient temperature for up to four days and to harmful conditions including low or high pH, several freeze-thaw cycles, and boiling. Hence, extracellular miRNAs can be employed as biomarkers for various diseases(O'Brien et al., 2018).

According to the above discussion, the present in silico research was designed to discover the most relevant regulatory miRNAs of major inflammatory pathways (JAK-STAT, NF-kb, and MAPK) that are dysregulated in COVID-19 infection.

2. Methods

In the current study, a systematic biologic procedure was used for the analysis of high-throughput expression data from blood samples of COVID-19 and healthy individuals. Since it is only an in silico study, we did not include any patients or control samples in this study. We aimed

Table 1
Summary of high-throughput research methodology to assess miRNA expression profiles in COVID-19 patients.

Group	Sample	Method	Reference
COVID-19/control	Whole blood	High-throughput sequencing -Illumina HiseqX Ten platform	Li et al. (2020)
COVID-19/control	Whole blood Plasma cfRNA	*BGISEQ-500 sequencer*	Yang et al. (2021)
COVID-19/control	Whole blood	Multi-transcriptome sequencing -Illumina Hiseq 4000 platform	Tang et al. (2020)
COVID-19/control	Peripheral blood/plasma	MiRCURY LNA miRNome qPCR panels	Fayyad-Kazan et al. (2021)
COVID-19/control	Peripheral blood	Whole-Transcriptome RNA Sequencing-Illumina HiSeq X Ten platform	Li et al. (2021)
COVID-19/control	Plasma sample	Small RNA sequencing- Illumina	Farr et al. (2021)
COVID-19/control	Whole blood/ Plasma	miRNA sequencing- HiSeq 2500 platform Illumina	Nicoletti et al. (2022)
COVID-19/control	Peripheral blood	miRNA sequencing- Illumina	Duecker et al., 2021
COVID-19/control	Blood/plasma	HTG EdgeSeq miRNA Whole Transcriptome Assay	Akula et al. (2022)
COVID-19/control	Plasma	Small RNA sequencing- Illumina	Gutmann et al. (2022)
COVID-19/control	Plasma	Small RNA sequencing- Illumina	Fernández-Pato et al. (2022)

to detect differentially expressed miRNAs in blood samples of COVID-19 vs. healthy specimens to construct and analyze miRNA–mRNA networks and predict key miRNAs and genes in inflammatory pathways. Fig. 1 briefly illustrates the steps applied in the in silico analysis (Fig. 1).

2.1. *miRNA expression profiling*

Firstly, we searched web-based references in which the expression profiles of miRNAs in blood samples of COVID-19 patients are provided in comparison to healthy individuals. The search criteria included COVID-19 patients in all severity-based categories, mild, moderate as well as severe respiratory failure, diagnosed by means of available qPCR Detection Kits. Among all the available sources, miRNA expression profiles of 11 paired datasets which included the profile of COVID-19 patients versus healthy samples were selected for further investigation (Table 1). In the following steps, the expression profiles of the selected miRNAs were processed using statistical methods. To do so, we used Shapiro-Wilks and Kolmogorov-Smirnov tests with 0.01 and 0.001 significant levels for investigating normality of data. After that, some parametric-statistical tests were applied to determine the critical area of data in comparison to mean of data. To investigate how data are distributed, Box-Plot with test, frequency table, as well as some appropriate plots such as histogram and line plot, were depicted.

2.2. *Identification of key genes in inflammatory signaling pathways in COVID-19*

KEGG pathway database (https://www.genome.jp/kegg/) was used to determine key genes involved in inflammatory signaling pathways in COVID-19 pathogenesis. This database provides a set of depicted pathway maps indicating molecular interaction and relation networks for genetic and environmental information processing, cellular processes, organismal systems, metabolism, human diseases, and drug development. Based on KEGG database, genes involved in common inflammatory pathways, i.e. JAK/STAT, NF-KB, and MAPK were obtained. We further assessed our selected genes using HGNC symbol database (https://www.genenames.org/), a web-based resource in which human gene nomenclatures are approved. After that, genes that not only participated in the above-mentioned inflammatory pathways but also made a unique contribution in COVID-19 pathogenesis were chosen for further investigation.

2.3. *miRNA – mRNA networks prediction*

In this report, we applied the miRWalk (http://mirwalk.umm.uni-heidelberg.de/), an open-source high accuracy platform, to predict miRNAs of interest and inflammatory target gene interactions. This online tool provides the most comprehensive collection of predicted and experimentally verified miRNA-target gene interactions based on TarPmiR algorithm which utilizes a random-forest based learning approach to predict miRNA target sites as accurately as possible.

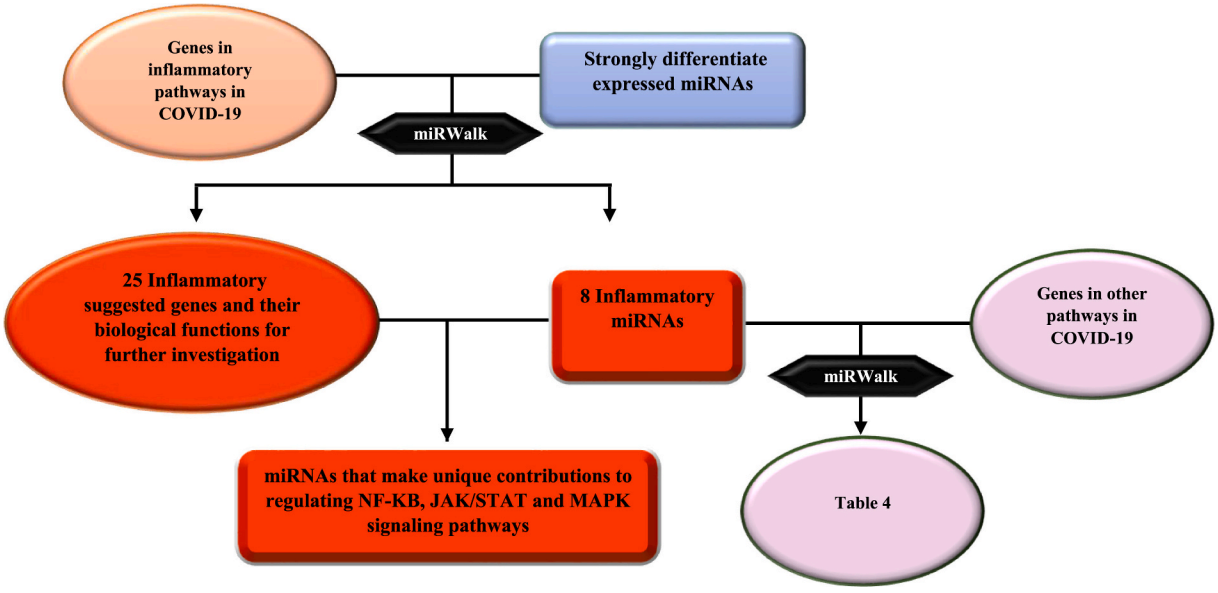

Fig. 2. Short description of obtained results in this study.

Table 2
Demographic features of COVID-19 patients considered in this study.

Group	Population	Sex(male/female)	Age (years)	Symptoms	Reference
COVID-19	10	4/6	44.90 ± 19.94	Mild Moderate	Li et al. (2020)
Control	4	2/2	44.75 ± 11.84	–	
COVID-19	5	–	–	Covid-19 patient	Yang et al. (2021)
Control	3	–	–	–	
COVID-19	6	4/2	50–89	Moderate	Tang et al. (2020)
	6	5/1	60–89	Severe	
Control	4	2/2	50–69	–	
COVID-19	14	7/6	39.29 ± 7.477	Mild	Fayyad-Kazan et al. (2021)
	13	9/5	46.31 ± 8.548	Moderate	
	6	4/2	57.67 ± 2.16	Severe	
Control	10	–	–	–	
COVID-19	10	4/6	44:90 ± 19:94	–	Li et al. (2021)
Control	4	1/3	34:75 ± 11:84	–	
COVID-19	10	4/6	53.5 ± 17.2	–	Farr et al. (2021)
Control	10	4/6	53 ± 17.6	–	
COVID-19	4	2/2	61.8 ± 11.7	Mild/moderate	Nicoletti et al. (2022)
	4	2/2	64.0 ± 8.6	severe/critical	
Control	4	2/2	62.8 ± 14.9	–	
COVID-19	11	9/2	76 (48–91)	Moderate	Duecker et al. (2021)
	10	8/2	69 (52–79)	Severe	
Control	8	5/3	68 (49–89)	–	
COVID-19	12	6/6	47.8 ± 9.8	Moderate Severe	Akula et al. (2022)
Control	8	5/3	46 ± 7.3	–	
COVID-19	18	56٪/44٪	55.0 (36.0, 66.0)	Mild to Moderate	Gutmann et al. (2022)
	18	42٪/58٪	58.0 (39.0, 66.0)	Severe	
Control	11	42٪/58٪	40.0 (30.0, 46.0)	–	
COVID-19	32	23/9	63.4 (52.9–78.3)	Severe	Fernández-Pato et al. (2022)
	52	26/26	59.4 (53.0–71.8)	Moderate	
	12	4/8	66.2 (44.4–72.6)	Asymptomatic/Mild	
Control	13	7/6	66.7 (57–68.9)	–	

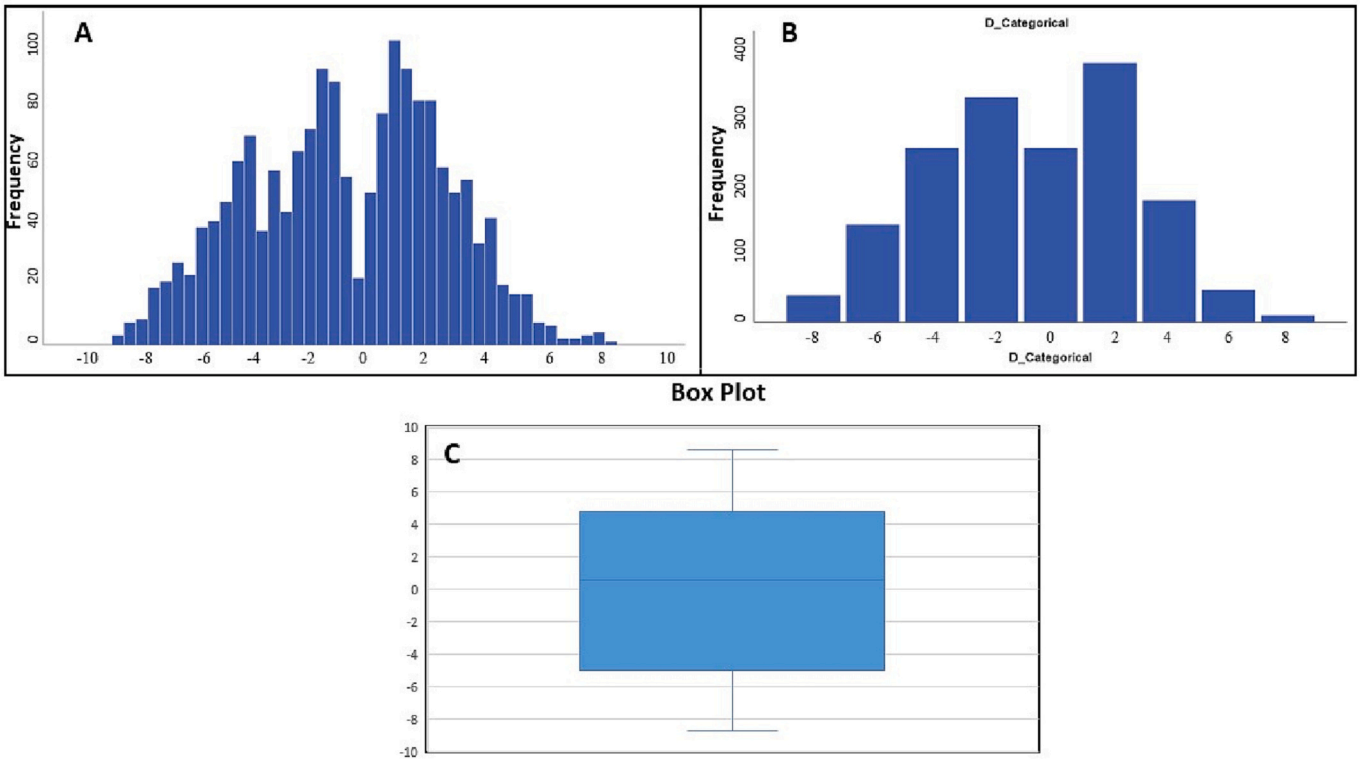

Fig. 3. A) Frequency distribution of miRNAs involved in COVID-19 pathogenesis. X-axis shows the miRNA expression profiling intervals based on fold changes reported for miRNA expression in high-throughput studies. Y-axis shows the frequency of miRNAs in determined intervals B) Frequency distribution of categorical miRNAs involved in COVID-19 pathogenesis. X-axis shows the miRNA expression profiling intervals based on fold changes reported for miRNA expression in high-throughput studies. Y-axis shows the frequency of miRNAs in determined intervals. Y-axis shows the frequency of miRNAs in determined intervals (0–400) C) Box-Plot with test of miRNAs involved in COVID-19 pathogenesis. miRNA expression profiling values (From bottom to top of the chart: Minimum, Quartile 1, Median, Quartile 3, Maximum).

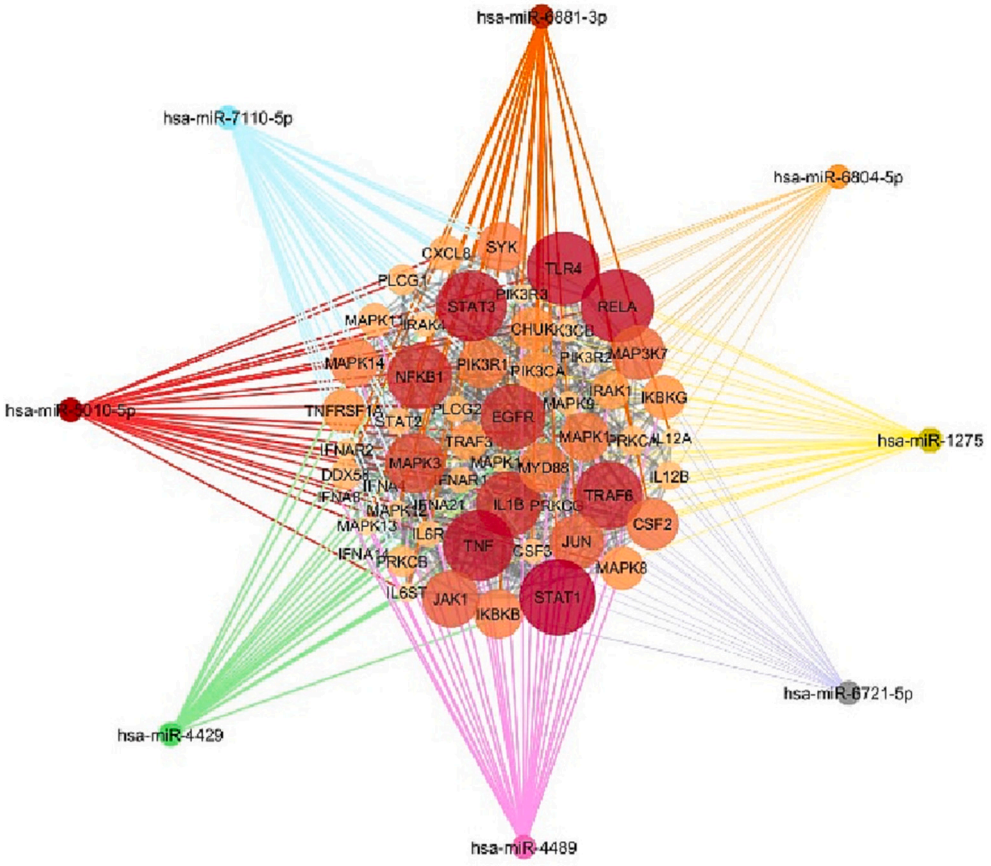

Fig. 4. The potential interactions between potential target genes of miRNAs of interest in NF-KB, JAK/STAT and MAPK signaling pathways participating in COVID-19 pathogenesis and miR-1275, miR-4429, miR-4489, miR-6721-5p, miR-5010-5p, miR-7110-5p, miR-6804-5p and miR-6881–3p were depicted by Cytoscape. Genes that have more interactions are indicated larger in size. Moreover, there is direct relation between the number of protein interactions and color intensity.

Furthermore, miRWalk database is capable of extracting information from popular gene-miRNA prediction databases such as TargetScan (https://www.targetscan.org/vert_80/), miRDB (http://mirdb.org/), and miRTarBase (https://mirtarbase.cuhk.edu.cn/~miRTarBase/miRTarBase_2022/php/index.php). To validate protein-protein interaction networks, we recruited STRING database (https://string-db.org/) in which all known and predicted physical and functional protein associations are integrated. For miRNA–mRNA networks construction and analysis, the Cytoscape software version 1.7.3 was used. To do so, the gained miRNA–protein interactions were chosen and depicted in this open source bioinformatics software.

2.4. Biological functions of genes potentially dysregulated in COVID-19 pathogenesis

Among all genes playing critical roles in COVID-19 pathogenesis, some potentially dysregulated genes were selected and further investigated in terms of molecular and biological functions. For this purpose, the human protein atlas database (https://www.proteinatlas.org/) providing the expression levels of mRNAs and proteins of human genes, as well as their biological roles, according to different genomics technologies such as transcriptomics and systems biology was employed. Moreover, other protein-based databases including UniProtKB (https://www.uniprot.org/help/uniprotkb) as well as GeneCards (https://www.genecards.org/) were utilized for further assessment.

3. Results

3.1. miRNA expression profile processing

Figure 2 briefly illustrates the results obtained in this study (Fig. 2). At first, miRNA expression profiles in blood samples of COVID-19 vs. healthy specimens were obtained according to previously described criteria. The demographic characteristics of selected paired samples were succinctly recorded (Table 2). As shown in Table 2, the enrolled COVID-19 and healthy participants belonged to various sex and age-groups in all severity-based categories, i.e. mild, moderate, and severe respiratory failure. We selected differentially expressed miRNAs in blood samples of COVID-19 vs. healthy samples for further assessment. The statistical processing of the miRNA expression profiles was implemented using descriptive and inferential statistics. The normality of data was strongly confirmed via Shapiro–Wilk test in which the null hypothesis is a sample x1, …, xn coming from a normally distributed population. In different studies, confidence levels (1-α) % of 95 or 99% are frequently used for both tests, with the significance level (α)% equal to 1% or 5%. Considering the main and critical condition for the parametric test, normality, *t*-test with a confidence level of 95% was conducted, which is a statistical hypothesis test used to determine whether an unknown population mean is different from a specific value. The results indicated significant difference between the reported mean of miRNA expression and the mean of existing data. Confidence intervals for the expression levels of miRNA (log2 fold change) were regarded from −5 to 4.8. Based on the outliers in this box plot (Fig. 2.C), miRNAs expressed outside of this interval were in our critical areas of interest (more than max and less than min). Furthermore, Box-Plot with test

results indicated that miRNAs expressed outside of the confidence intervals and entering quartile distance can be selected for further investigation. Our results showed that 171 miRNAs were expressed outside of the defined and entered quartile distance (Fig. 3).

At first, miRNA expression profiles in blood samples of COVID-19 vs. healthy specimens were obtained according to previously described criteria. The demographic characteristics of the selected paired samples were succinctly recorded (Table 2). As shown in Table 2, the enrolled COVID-19 and healthy participants were ranged in various sex and age-groups in all severity-based categories, i.e. mild, moderate as well as severe respiratory failure. We selected differentially expressed miRNAs in blood samples of COVID-19 vs. healthy specimens for further assessment. The statistical processing of the miRNA expression profiles was implemented using descriptive and inferential statistics. The normality of data was strongly confirmed. Considering the main and critical condition for parametric test, normality, t-test was conducted and the results indicated the significant difference between the reported mean of miRNA expression and the mean of existing data. Confidence intervals for the expression levels of miRNAs (log2 fold change) were regarded from −5 to 4.8. Based on the defined confidence intervals, miRNAs expressed out of this interval, were in our critical areas of interest. Furthermore, Box-Plot with test results indicated that miRNAs expressed out of enter quartile distance can be selected for further investigation. Our results have shown that 171 miRNAs are expressed out of defined confidence intervals and entered quartile distance (Fig. 3).

Table 3
Potential targets of miRNAs involved in COVID-19 pathogenesis.

Gene	miRNA
MAPK1	miR-4429, miR-6721-5p, miR-5010-5p, miR-6804-5p, miR-4489, miR-7110-5p
MAPK3	miR-1275, miR-6881–3p, miR-5010-5p, miR-6804-5p, miR-4489, miR-7110-5p
MAPK8	miR-5010-5p, miR-6804-5p, miR-4489
MAPK9	miR-4429, miR-1275, miR-6721-5p, miR-5010-5p, miR-6804-5p, miR-4489, miR-7110-5p
MAPK10	miR-4429, miR-1275, miR-6881–3p, miR-5010-5p, miR-6804-5p,miR-4489, miR-7110-5p, miR-6721-5p
MAPK11	miR-4429, miR-6721-5p,miR-4489, miR-7110-5p
MAPK12	miR-6881–3p, miR-7110-5p
MAPK13	miR-4429, miR-1275, miR-6721-5p, miR-6881–3p, miR-6804-5p, miR-4489, miR-7110-5p, miR-5010-5p
MAPK14	miR-4429, miR-1275, miR-6804-5p, miR-5010-5p, miR-4489
MAP3K7	miR-4429, miR-6721-5p, miR-6804-5p
PRKCG	miR-4429, miR-1275, miR-6721-5p, miR-5010-5p, miR-4489, miR-7110-5p
EGFR	miR-4429, miR-6721-5p, miR-6804-5p, miR-7110-5p
STAT1	miR-4429, miR-6881–3p, miR-6804-5p
DDX58	miR-4429, miR-6721-5p, miR-6881–3p, miR-5010-5p, miR-4489
PLCG2	miR-4429, miR-1275, miR-6721-5p, miR-4489, miR-7110-5p
PIK3CA	miR-4429, miR-1275, miR-6881–3p, miR-5010-5p, miR-4489
IL6R	miR-4429, miR-6881–3p, miR-5010-5p, miR-7110-5p
TNF	miR-4429, miR-1275, miR-6721-5p
IFNAR1	miR-4429, miR-1275, miR-5010-5p, miR-4489, miR-7110-5P
IRAK1	miR-4429, miR-4489
TNFRSF1A	miR-4429
PIK3R3	miR-4429, miR-1275, miR-6721-5p, miR-6881–3p, miR-5010-5p, miR-6804-5p, miR-4489, miR-7110-5p
IRAK4	miR-4429, miR-1275, miR-5010-5p, miR-6804-5p, miR-7110-5p
MYD88	miR-4429, miR-1275, miR-6721-5p, miR-6881–3p, miR-5010-5p, miR-6804-5p
IL6ST	miR-4429, miR-1275, miR-6721-5p, miR-6881–3p, miR-5010-5p, miR-6804-5p, miR-7110-5p
PIK3R1	miR-4429, miR-1275, miR-6881–3p, miR-5010-5p, miR-6804-5p, miR-7110-5p
SYK	miR-1275, miR-7110-5p
IKBKB	miR-1275, miR-6721-5p, miR-6881–3p, miR-6804-5p, miR-4489
TRAF3	miR-1275, miR-5010-5p, miR-6804-5p, miR-4489, miR-7110-5p, miR-6881–3p
PIK3CB	miR-1275, miR-6721-5p, miR-7110-5p
CHUK	miR-1275, miR-5010-5p, miR-6804-5p
IL12B	miR-1275
TLR4	miR-1275, miR-6721-5p, miR-5010-5p, miR-6804-5p, miR-4489, miR-7110-5p
TRAF6	miR-1275, miR-5010-5p, miR-4489, miR-7110-5p
IL1B	miR-1275, miR-6721-5p
CSF3	miR-1275, miR-6881–3p, miR-5010-5p, miR-6804-5p, miR-7110-5p
IFNAR2	miR-1275, miR-6721-5p, miR-6881–3p, miR-7110-5p
PRKCA	miR-6721-5p, miR-6881–3p, miR-5010-5p, miR-4489
STAT2	miR-6721-5p, miR-6881–3p, miR-7110-5p
STAT3	miR-6804-5p, miR-7110-5p, miR-5010-5p, miR-6881–3p
JUN	miR-6881–3p
RELA	miR-5010-5p, miR-7110-5p, miR-6721-5p
IL12A	miR-6881–3p
PLCG1	miR-7110-5p, miR-5010-5p
PRKCB	miR-4489, miR-7110-5p, miR-6804-5p
CXCL8	miR-5010-5p, miR-7110-5p
JAK1	miR-6804-5p
NFKB1	miR-6804-5p
IKBKG	miR-4489, miR-7110-5p
IFNA8	miR-4489
IFNA14	miR-4489
IFNA21	miR-4489
CSF2	miR-6881–3p,miR-1275
PIK3R2	miR-4489
IFNA4	miR-4489

Table 4
Potential target genes of miRNAs of interest in other signaling pathways involved in COVID-19 pathogenesis.

miRNA	Gene	Biological process
hsa-miR-5010-5P	ACE	Renin-Angiotensin Pathway
	ADAR	Antiviral defense, Immunity, Innate immunity, mRNA processing, RNA-mediated gene silencing
	C1QC, C2	Complement pathway, Immunity, Innate immunity
	CASP1	Apoptosis
	CXCL10	Chemotaxis, Inflammatory response
	CGAS	Antiviral defense, DNA damage, DNA repair, Host-virus interaction, Immunity, Innate immunity
	EIF2AK2	Antiviral defense, Host-virus interaction, Immunity, Innate immunity, Transcription, Transcription regulation
	F13A1	Blood coagulation, Hemostasis
	HBEGF	Immunity
	MAS1	ACE Inhibitor Pathway, Peptide ligand-binding receptors
	MASP1	Complement activation lectin pathway, Immunity, Innate immunity
	MAVS	Antiviral defense, Host-virus interaction, Immunity, Innate immunity
	MX2	Antiviral defense, Immunity, Innate immunity, mRNA transport, Protein transport, Translocation, Transport
	NFKBIB	Immunity, Inflammatory response
	NLRP3	Immunity, Inflammatory response, Innate immunity, Transcription, Transcription regulation
	OAS1,2	Antiviral defense, Immunity, Innate immunity
	RPL7,10,13,14,22, 22 L1,27A,32,34, 35A,37,38	Host-virus interaction, rRNA processing, Translation, Gene expression
	RPS4Y1,8,15A, 20,23,29, 27 L	Host-virus interaction, rRNA processing, Translation
	TLR3	Immunity, Inflammatory response, Innate immunity
	UBA52	Host-virus interaction, Translation
hsa-miR-6804-5P	ACE	Renin-Angiotensin Pathway
	ADAR	Antiviral defense, Immunity, Innate immunity, mRNA processing, RNA-mediated gene silencing
	C7	Complement pathway, Immunity, Innate immunity
	CYBB	Electron transport, Ion transport, Transport
	F13A1	Blood coagulation, Hemostasis
	MAS1	ACE Inhibitor Pathway, Peptide ligand-binding receptors
	MASP1	Complement activation lectin pathway, Immunity, Innate immunity
	MAVS	Antiviral defense, Host-virus interaction, Immunity, Innate immunity
	OAS1,2	Antiviral defense, Immunity, Innate immunity
	RPL10,13,14, 22 L1,28,32,37,37A	Host-virus interaction, rRNA processing, Translation, Gene expression
	RPS15A,24,27 L	Host-virus interaction, rRNA processing, Translation
	TLR3	Immunity, Inflammatory response, Innate immunity
	UBA52	Host-virus interaction, Translation
hsa-miR-7110-5P	ADAR	Antiviral defense, Immunity, Innate immunity, mRNA processing, RNA-mediated gene silencing
	ADAM17	Notch signaling pathway
	CASP1	Apoptosis
	C1QA,C1QC	Complement pathway, Host-virus interaction, Immunity, Innate immunity
	C3AR,C5AR1	Chemotaxis
	CXCL10	Chemotaxis, Inflammatory response
	CGAS	Antiviral defense, DNA damage, DNA repair, Host-virus interaction, Immunity, Innate immunity
	C2,6,7,C8B	Complement pathway, Immunity, Innate immunity
	CYBB	Electron transport, Ion transport, Transport
	EIF2AK2	Antiviral defense, Host-virus interaction, Immunity, Innate immunity, Transcription, Transcription regulation
	FGB, FGA	Adaptive immunity, Blood coagulation, Hemostasis, Immunity, Innate immunity
	HBEGF	Immunity
	MAS1	ACE Inhibitor Pathway, Peptide ligand-binding receptors
	MASP1,2	Complement activation lectin pathway, Immunity, Innate immunity
	MAVS	Antiviral defense, Host-virus interaction, Immunity, Innate immunity
	MX2	Antiviral defense, Immunity, Innate immunity, mRNA transport, Protein transport, Translocation, Transport
	NFKBIB	Immunity, Inflammatory response
	OAS1,2,3	Antiviral defense, Immunity, Innate immunity
	RPL10L,11,13, 14,22 L1,23,23A, 27A,28,32, 36, 37,37A,38, 41	Host-virus interaction, rRNA processing, Translation, Gene expression
	RPS3A,9,14, 15A,16,19,29,27 L	Host-virus interaction, rRNA processing, Translation
	RPS10-NUDT3	Transcription
	STING1	Autophagy, Host-virus interaction, Immunity, Innate immunity
	SELP	Cell adhesion
	TMPRSS2	Regulation of Androgen receptor activity, Host-virus interaction
	TLR2,3	Immunity, Inflammatory response, Innate immunity
	UBA52	Host-virus interaction, Translation
hsa-miR-6881–3P	ACE	Renin-Angiotensin Pathway
	CASP1	Apoptosis
	C3AR1	Chemotaxis
	CFD	Complement alternate pathway, Immunity, Innate immunity
	CYBB	Electron transport, Ion transport, Transport
	EIF2AK2	Antiviral defense, Host-virus interaction, Immunity, Innate immunity, Transcription, Transcription regulation
	FCGR2A	Immunity

(*continued on next page*)

Table 4 (*continued*)

miRNA	Gene	Biological process
	MAS1	ACE Inhibitor Pathway, Peptide ligand-binding receptors
	MAVS	Antiviral defense, Host-virus interaction, Immunity, Innate immunity
	NRP1	Angiogenesis, Differentiation, Host-virus interaction, Neurogenesis
	NFKBIB	Immunity, Inflammatory response
	OAS1,3	Antiviral defense, Immunity, Innate immunity
	RPS19,14	Host-virus interaction, rRNA processing, Translation
	RPL13, 14,22, 36A,37,38	Host-virus interaction, rRNA processing, Translation, Gene expression
	UBA52	Host-virus interaction, Translation
	ACE	Renin-Angiotensin Pathway
	CFD	Complement alternate pathway, Immunity, Innate immunity
	EIF2AK2	Antiviral defense, Host-virus interaction, Immunity, Innate immunity, Transcription, Transcription regulation
	FCGR2A	Immunity
	MASP1	Complement activation lectin pathway, Immunity, Innate immunity
	MMP1	Collagen degradation, Host-virus interaction
	MX1	Antiviral defense, Immunity, Innate immunity
	MX2	Antiviral defense, Immunity, Innate immunity, mRNA transport, Protein transport, Translocation, Transport
hsa-miR-6721-5P	OAS1,2	Antiviral defense, Immunity, Innate immunity
	RPL4,7,10,13,22, 23,27A,28,31,32, 37,37A	Host-virus interaction, rRNA processing, Translation, Gene expression
	RPS3,4×,15A, 16,19,24,29	Host-virus interaction, rRNA processing, Translation
	RPS10-NUDT3	Transcription
	RSL24D1	RSL24D1
	TLR2	Immunity, Inflammatory response, Innate immunity
	CYBB	Electron transport, Ion transport, Transport
	C6,7	Complement pathway, Immunity, Innate immunity
	FAU	Host-virus interaction, RNA processing, Transcription and Translation
	MAS1	ACE Inhibitor Pathway, Peptide ligand-binding receptors
	MAVS	Antiviral defense, Host-virus interaction, Immunity, Innate immunity
	OAS2	Antiviral defense, Immunity, Innate immunity
hsa-miR-4429	RPL3,14,15,22 L1, 28,32,34,37,38	Host-virus interaction, rRNA processing, Translation, Gene expression
	RPS3,4×,19,26,29	Host-virus interaction, rRNA processing, Translation
	RSL24D1	Ribosome biogenesis
	STING1	Autophagy, Host-virus interaction, Immunity, Innate immunity
	TLR7	Immunity, Inflammatory response, Innate immunity
	UBA52	Host-virus interaction, Translation
	ACE	Renin-Angiotensin Pathway
	ADAR	Antiviral defense, Immunity, Innate immunity, mRNA processing, RNA-mediated gene silencing
	C7	Complement pathway, Cytolysis, Immunity, Innate immunity
	C1QC	Complement pathway, Immunity, Innate immunity
	CASP1	Apoptosis
	HBEGF	Immunity
	IKBKE	DNA damage, Host-virus interaction
	MAS1	ACE Inhibitor Pathway, Peptide ligand-binding receptors
	MASP2	Complement activation lectin pathway, Immunity, Innate immunity
	MAVS	Antiviral defense, Host-virus interaction, Immunity, Innate immunity
hsa-miR-1275	MBL2	Complement activation lectin pathway, Complement pathway, Immunity, Innate immunity
	MX1	Antiviral defense, Immunity, Innate immunity
	MX2	Antiviral defense, Immunity, Innate immunity, mRNA transport, Protein transport, Translocation, Transport
	OAS2,3	Antiviral defense, Immunity, Innate immunity
	RPL4,13,14, 23A,27A,28,31	Host-virus interaction, rRNA processing, Translation, Gene expression
	RPS3A,4×,19, 14,29	Host-virus interaction, rRNA processing, Translation
	RPS10-NUDT3	Transcription
	STING1	Autophagy, Host-virus interaction, Immunity, Innate immunity
	TMPRSS2	Regulation of Androgen receptor activity, Host-virus interaction
	UBA52	Host-virus interaction, Translation
	ACE	Renin-Angiotensin Pathway
	ACE2	Host-virus interaction
	AGTR1	Renin-Angiotensin Pathway
	C2,7	Complement pathway, Immunity, Innate immunity
	EIF2AK2	Antiviral defense, Host-virus interaction, Immunity, Innate immunity, Transcription, Transcription regulation
	FGB	Adaptive immunity, Blood coagulation, Hemostasis, Immunity, Innate immunity
	MAS1	ACE Inhibitor Pathway, Peptide ligand-binding receptors
hsa-miR-4489	MASP1	Complement activation lectin pathway, Immunity, Innate immunity
	MX1,MX2	Antiviral defense, Immunity, Innate immunity, mRNA transport, Protein transport, Translocation, Transport
	OAS1,3	Antiviral defense, Immunity, Innate immunity
	RPL14,15,10,13A, 19,28,31,35A,37	Host-virus interaction, rRNA processing, Translation, Gene expression
	RPL36A-HNRNPH2	Transcription
	RPS4X,19,14,29	Host-virus interaction, rRNA processing, Translation
	TMPRSS2	Regulation of Androgen receptor activity, Host-virus interaction
	UBA52	Host-virus interaction, Translation

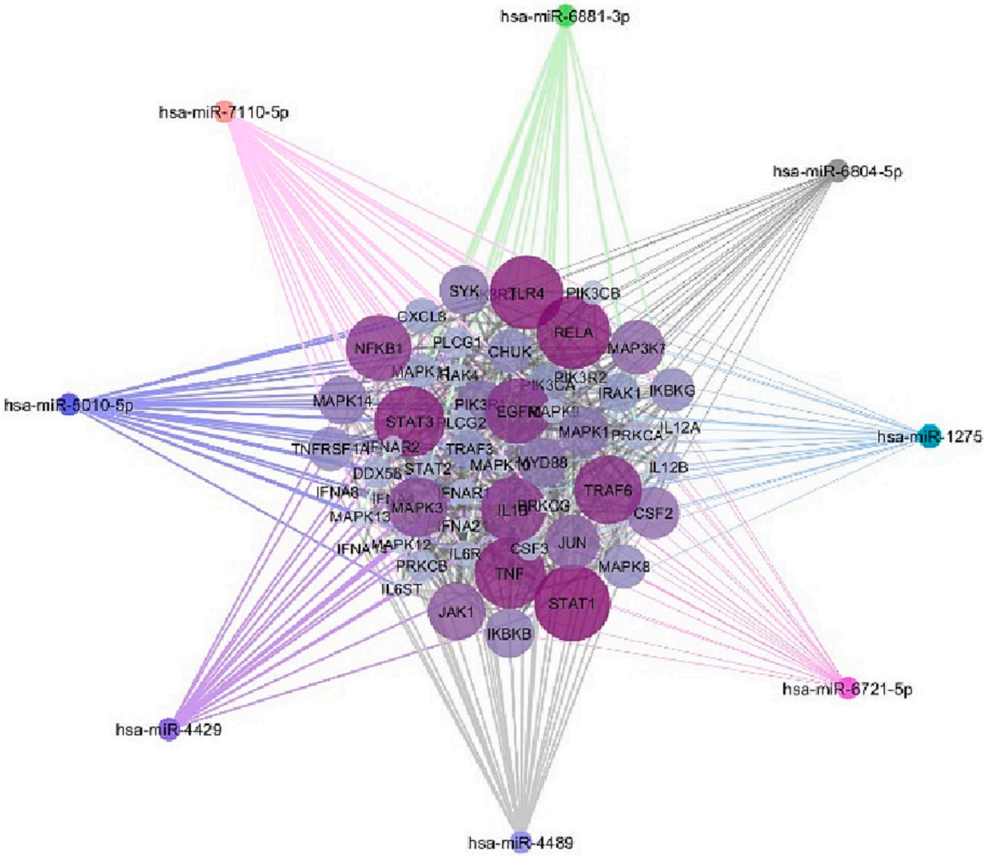

Fig. 5. Predicted interactions between target genes of miRNAs of interest in other signaling pathways involved in COVID-19 pathogenesis and miR-1275, miR-4429, miR-4489, miR-6721-5p, miR-5010-5p, miR-7110-5p, miR-6804-5p and miR-6881–3p were depicted via Cytoscape. There is direct relation between the number of protein interactions and color intensity.

3.2. Potential capability of dysregulated miRNAs in regulating inflammatory signaling pathways in COVID-19

Looking into KEGG database, genes encoding proteins that are related to COVID-19 pathogenesis as well as genes involved in common inflammatory signaling pathways, JAK/STAT, NF-KB, and MAPK were gained. HGNC was recruited and extracted gene names were checked and corrected, accordingly. As a result, we obtained 74 genes playing roles in so-called inflammatory pathways in COVID-19 pathogenesis. To evaluate whether selected inflammatory genes are the genuine targets of determined miRNAs (miRNAs that were expressed outside of the defined confidence intervals and entered quartile distance), miRWalk database was used. Our results revealed that many components of inflammatory pathways associated with COVID-19 pathogenesis were the targetome of determined miRNAs. Considering predicted miRNA-gene interactions, 8 strong differentially expressed miRNAs, including miR-1275, miR-4429, miR-4489, miR-6721-5p, miR-5010-5p, miR-7110-5p, miR-6804-5p and miR-6881–3p, affecting about one third of inflammatory genes, were selected as candidates for further investigation.

3.3. miRNA – mRNA networks analysis and construction

Altogether, miRWalk web tool predicted 202 interactions between inflammatory genes and miRNAs of interest. Protein-protein interaction network was gained using STRING database with confidence edges and high confidence score (0.700). In the following step, the complete list of the interactions of the determined miRNAs of interest and inflammatory genes along with protein-protein interactions was imported into Cytoscape software version 1.7.3, analyzed, and then visualized (Fig. 4, Table 3). As shown in Fig. 4, genes that have more interactions are shown larger in size. Moreover, there is direct relation between the number of protein interactions and color intensity. Regarding the striking roles of miRNAs of interest in regulating inflammatory genes in COVID-19 pathogenesis, we focused on the effects of miRNAs of interest in regulating other genes associated with COVID-19 pathogenesis. Potential target genes of miRNAs of interest in other signaling pathways involved in COVID-19 pathogenesis are summarized in Table 4. Accordingly, 252 miRNA-gene interactions were predicted and pictured in Cytoscape software (Fig. 5). Apart from this, the role of miRNAs of interest (miR-1275, miR-4429, miR-4489, miR-6721-5p, miR-5010-5p, miR-7110-5p, miR-6804-5p and miR-6881–3p) in regulating key genes in NF-KB, JAK/STAT and MAPK signaling pathways involved in COVID-19 pathogenesis was pictured based on KEGG data base (Figs. 6 and 7). (See Fig. 8.)

3.4. Predicting marked dysregulated genes in inflammatory signaling pathways participating in COVID-19 pathogenesis

Our results predicted that among many genes (509) of JAK/STAT, NF-KB, and MAPK signaling pathways, 25 genes were not only targeted by 171 miRNAs that were expressed outside of the defined confidence intervals and entered quartile distance but also strongly influenced by miRNAs of interest (miR-1275, miR-4429, miR-4489, miR-6721-5p, miR-5010-5p, miR-7110-5p, miR-6804-5p and miR-6881–3p). Potential target genes regulated by more than twenty miRNAs and their biological functions are represented in Table 5. The role of 25 key genes in other inflammatory pathways and their interactions are shown in Table 6 and Fig. 7. It can be seen that a total of 11 out of 25 genes are targeted by 30 miRNAs: IL6R, IL6ST, MAPK9, MAPK10, MAPK13, TRAF3, PIK3R1, PIK3R3, TLR4, SYK, and IRAK4. Apart from this, our

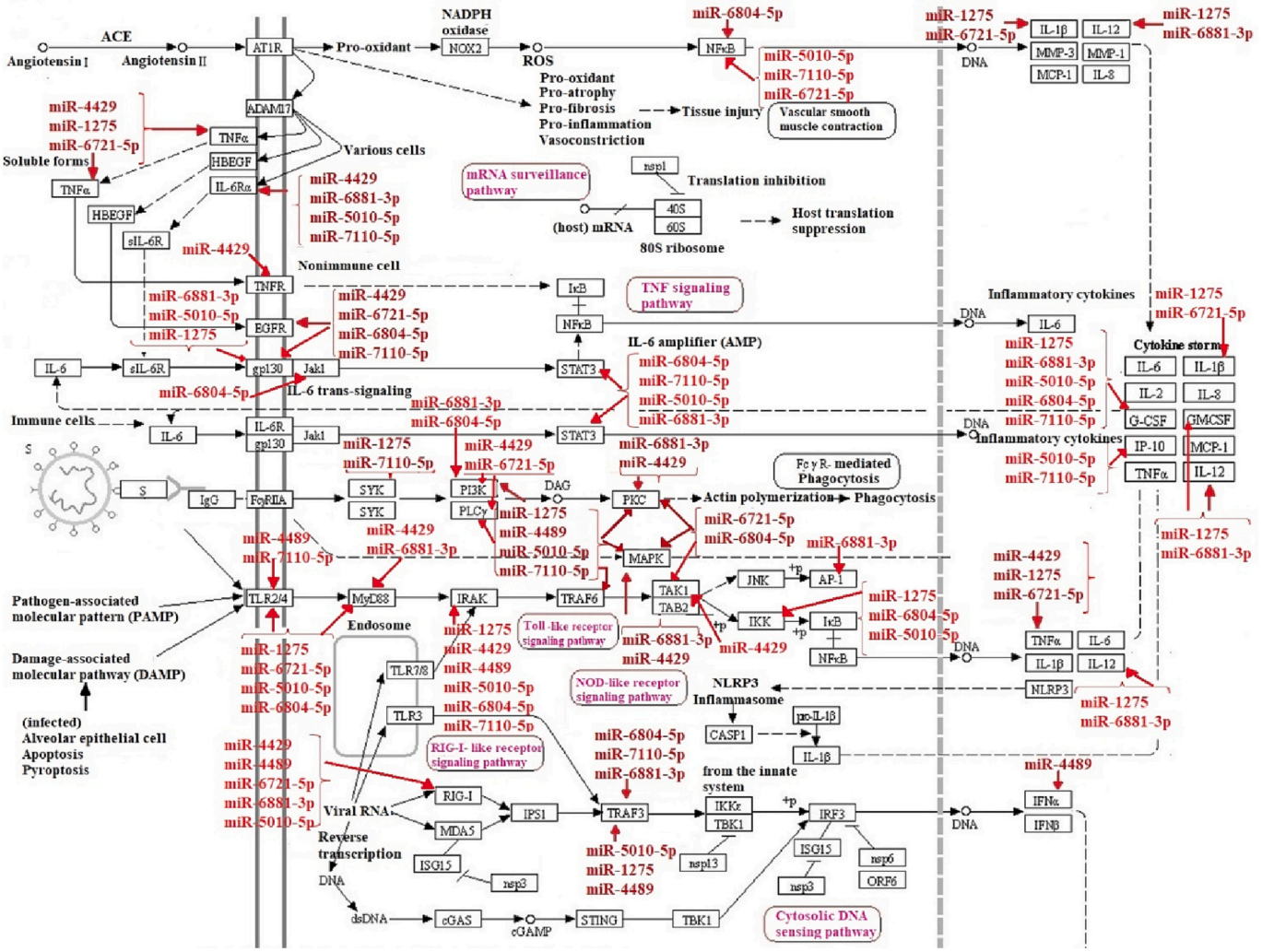

Fig. 6. The role of miRNAs of interest (miR-1275, miR-4429, miR-4489, miR-6721-5p, miR-5010-5p, miR-7110-5p, miR-6804-5p and miR-6881–3p) in regulating key genes in NF-KB and MAPK signaling pathways contributing to COVID-19 pathogenesis.

results highlighted that miR-5010-5p and miR-7110-5p make a unique contribution to regulating NF-KB signaling pathway. Strikingly, miR-6805-5p and miR-4489 strongly affected MAPK signaling pathway. Moreover, the components of JAK/STAT signaling pathway are significant targets of miR-7110-5p and miR-6881–3p. Based on this result, we found very significant candidates for prospective studies in COVID-19.

4. Discussion

Given the fact that mechanisms underlying the role of mis-expressed miRNAs in the regulation of inflammatory pathways in COVID-19 have not been fully understood, a comprehensive literature review and a bioinformatics approach were employed to predict miRNA–mRNA networks and identify potential key inflammatory miRNAs in the COVID-19 pathogenesis. This approach highlighted the importance of eight miRNAs, namely miR-1275, miR-4429, miR-4489, miR-6721-5p, miR-5010-5p, miR-7110-5p, miR-6804-5p, and miR-6881–3p, and 25 target genes in COVID-19 pathogenesis. Note that the expression of the above-mentioned miRNAs and their underlying mechanisms had not been completely investigated in COVID-19 patients. However, in line with the obtained results, the significant down-regulation of miR-1275 and miR-4429 in two separate COVID-19 patient cohorts had been previously confirmed (Duecker et al., 2021). In this study, we aimed to investigate the participation of these miRNAs in regulating inflammatory signaling pathways. The results of this study predicted the significant participation of the mentioned miRNAs in regulating inflammatory pathways.

The role of the JAK/STAT, NF-KB, and MAPK signaling pathways in generating inflammatory responses in COVID-19 was established. SARS-CoV-2 infection employs the JAK/STAT pathway to trigger an inflammatory response, which leads to serving dendritic cells, endothelial cells, monocytes, natural killer cells, lymphocytes, macrophages, and pneumocytes and proceeding towards cytokine storm. As a result of this process, different inflammatory markers are produced in the host that specify the severity of the disease. In addition, JAK/STAT pathway utilizes B cell and T cell differentiation to mediate immune responses (Satarker et al., 2021). For instance, an association was found between components of JAK/STAT signaling pathway such as JAK1–3 and STAT1–6 and different immune response roles, which include IL-12, IL-6, IL-4, IL-2 signaling, Th (T-helper)1, Th2, Treg cell and Th9, Th 17 cell differentiation, viral selective CD8+ T cell proliferation and B-cell lymphoma 2 (Bcl-2) (Banerjee et al., 2017; O'Shea and Plenge, 2013). Furthermore, NF-κB governs the expression of pro-inflammatory cytokines, which leads to an increased activation of NF-κB through a positive feedback loop (Chen and Greene, 2004). In COVID-19, pro-inflammatory cytokines with high expression levels further stimulate NF-KB (Neufeldt et al., 2022). The upregulation of NF-κB may contribute to inflammatory responses, as observed in patients with COVID-19. Intriguingly, some proteins belonging to SARS-CoV-2 are recruited as the NF-κB activators. For instance, as the most potent NF-kB inducer,

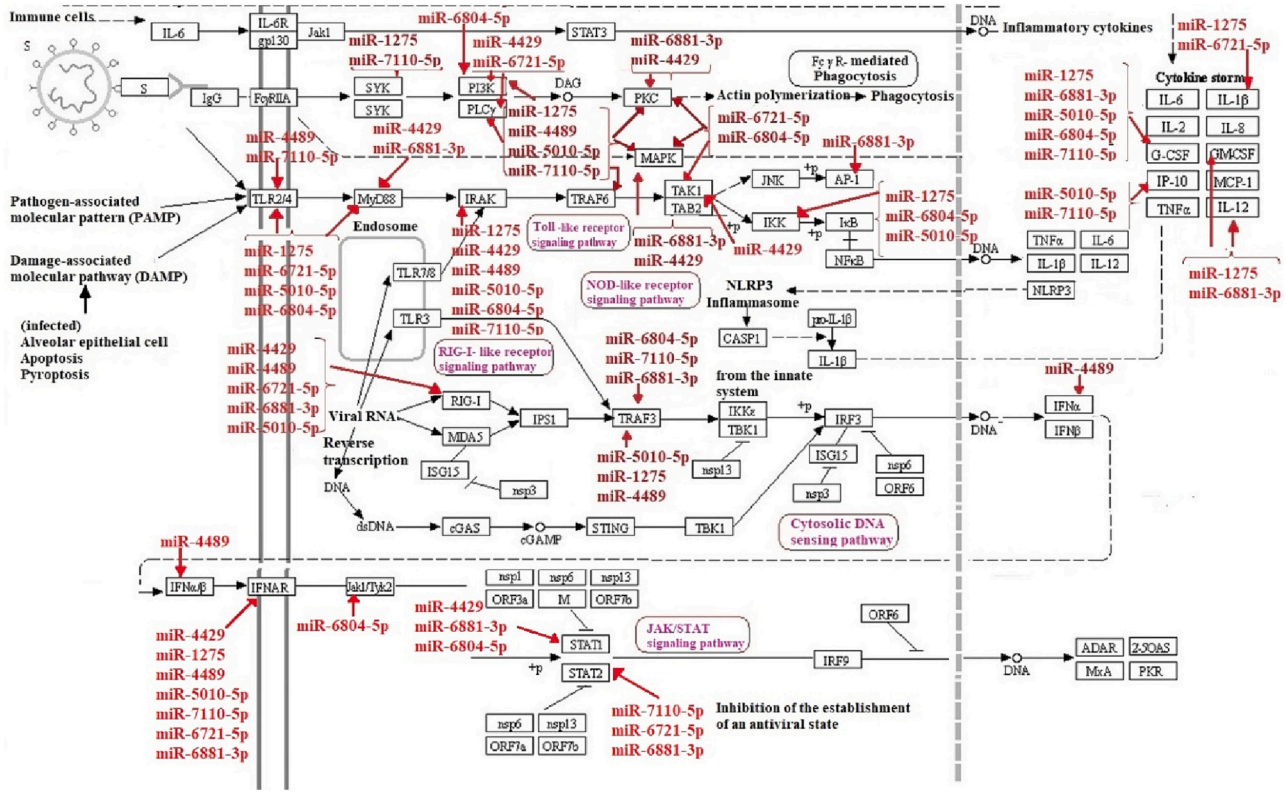

Fig. 7. The role of miRNAs of interest (miR-1275, miR-4429, miR-4489, miR-6721-5p, miR-5010-5p, miR-7110-5p, miR-6804-5p and miR-6881–3p) in regulating key genes in JAK/STAT signaling pathway involved in COVID-19 pathogenesis.

ORF7a is thus a producer of pro-inflammatory cytokines (Su et al., 2021). However, by up-regulating the p38 MAPK signaling pathway, several pro-inflammatory cytokines, like IL-1β, TNF-α, and IL-6, are activated (Zarubin and Han, 2005). The lost activity of ACE2 upon the entrance of the virus, which converts Angiotensin II into Angiotensin 1–7, potentially enables the predomination of Ang II-mediated activation of p38 in the heart and lungs when Ang 1–7 is down-regulated. This enables unrestrained inflammation and generates a positive feedback loop since the activation of p38 also upregulates ADAM17, a protease found to cleave the ACE2 ectodomain to further decrease local ACE2 protective activity (Scott et al., 2011). In addition, SARS-CoV utilizes a viral protein to upregulate p38 activity directly, like other RNA respiratory viruses that may be capable of hijacking p38 activity to promote replication (Grimes and Grimes, 2020).

miRNA target databases indicate that miRNAs regulate most human genes. miRNAs also affect the expression levels of genes involved in the NF-KB, JAK/STAT, and MAPK signaling pathways participating in inflammatory responses in COVID-19. In line with the results of our study, studies have demonstrated that some miRNAs obtained in this study can affect signaling pathways genes involved in inflammatory responses. For example, the combined influence of miR-146a and miR-1275 as well as miR-132 resulted in mRNA degradation and protein inhibition of IRAK1 and MAPK3, respectively (Betáková and Švančarová, 2013). In addition, miR-1275 was implicated in the regulation of NF-κB and IL-6 (Plowman and Lagos, 2021). The involvement of miR-1275 down-regulation has been established in cancers, infections, and obesity (Zhou et al., 2017). MiR-1275 silences ELK1, an *E*-twenty-six-domain transcription factor related to adipocyte differentiation, and thus suppresses human visceral pre-adipocyte differentiation (Pang et al., 2016). Also, IL6 and TNFα can down-regulate miR-1275 expression in mature human adipocytes. Furthermore, it has been established that NF-κB participates in regulating miR-1275 transcription via attaching to its promoter region. Responding to TNF-α, NF-κB attached to the promoter region of miR-1275 and suppressed its transcription. This somewhat explains the miR-1275 reduction in mature human adipocytes in obesity (Zhou et al., 2017). Fawzy et al. (2015) demonstrated that IGF1R is the direct target of miR-1275 and that this miRNA is able to somewhat control the development of hepatocellular carcinoma by directly targeting IGF1R and IGF2BPs genes (Fawzy et al., 2015). By targeting IGF1R, AKT3, MAPK1, and PTEN genes involved in mTOR and PI3K/Akt signaling pathways, miR-4429 plays a role in the appearance of Biliary atresia (BA) disease. In this disease, the progressive inflammation, extrahepatic bile ducts and fibrosis of intrahepatic, resulting in cirrhosis were observed (Dong et al., 2016). Expression profiling study is the most cost-effective tool with greatest promise for identifying new biomarkers for diseases and drug responses (Gurwitz, 2013). Comparing 10 patients with COVID-19 and 10 healthy individuals as the control in a profiling study found several interesting differential miRNA expression signatures. The results indicated a notable down-regulation of miR-1275 in the COVID-19 patient cohort. Hence, miR-1275 can serve as a beneficial biomarker to identify an inflammatory response in COVID-19 (1, D5). Moreover, the expression profiling of miRNAs in sepsis secondary to pneumonia patients showed the up-regulated expression levels of miR-7110-5p in patients compared with healthy individuals. In addition, research showed the promise of these miRNAs for use as sepsis biomarkers (Zhang et al., 2019).

As far as we know, this is the first report to introduce a set of miRNAs regulating inflammatory signaling pathways by conducting a comprehensive profiling investigation on COVID-19 patients. Among these miRNAs, the role of miR-1275 in regulating inflammatory pathways has received the attention of many studies; this confirms the results of our study. Although the relationship of some of miRNAs obtained in this study with genes in inflammatory signaling pathways has not been investigated, bioinformatics predictions of this study indicate that miR-4429, miR-4489, miR-6721-5p, miR-5010-5p, miR-7110-5p, miR-6804-5p and miR-6881–3p can affect the expression of a significant number of

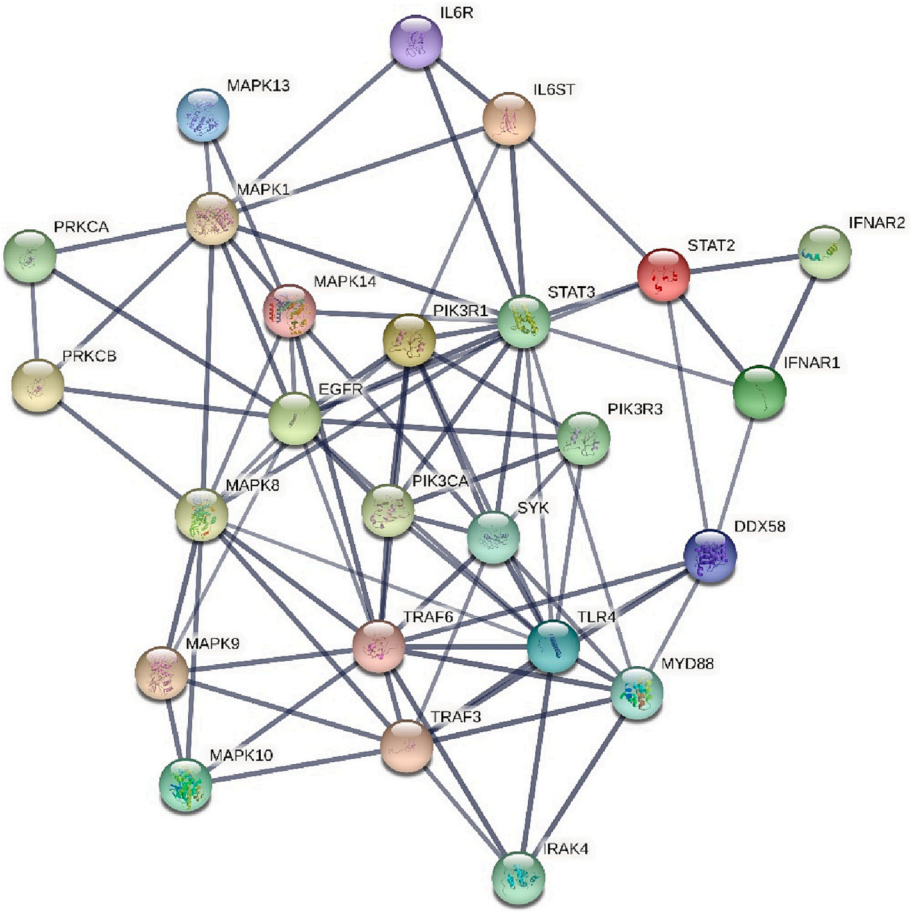

Fig. 8. Potential interactions between target genes of miRNAs of interest in other signaling pathways involved in COVID-19 pathogenesis and miR-1275, miR-4429, miR-4489, miR-6721-5p, miR-5010-5p, miR-7110-5p, miR-6804-5p and miR-6881–3p were depicted via Cytoscape. There is direct relation between the number of protein interactions and color intensity.

Table 5
Strong possible target genes of investigated miRNAs.

Gene symbol	Molecular function	Protein function
DDX58	Helicase Hydrolase RNA-binding	Production of proinflammatory cytokines and IFN-I, RIG-I-mediated antiviral signaling, Transcription of antiviral immunological genes, Detection of both positive and negative strand RNA viruses
EGFR	Host cell receptor for virus entry	Activates downstream signaling cascades such as the RAS-RAF-MEK-ERK, PI3K /AKT, PLCγ1, STATs modules and NF-kappa-B
IFNAR1	Receptor	A component of IFN-I receptor Activation of JAK-STAT pathway
IFNAR2	Receptor	A component of IFN-I receptor Activation of JAK-STAT pathway
IL6ST	Receptor	A signal transduction molecule associated with IL6R and activates JAK-STAT3 and JAK-MAPK signaling pathways.
IL6R	Receptor	Part of the receptor for IL6 that associated with IL6ST and leads to the immune response regulation.
IRAK4	Serine/threonine-protein kinase	Activation of NF-kappa-B in both TLR and TCR signaling pathways.
MAPK1	Serine/threonine-protein kinase	MAPK1/ERK2 participates in the MAPK/ERK cascade. Depending on the cellular context, this cascade is involved in various biological processes such as cell growth, adhesion, survival and differentiation. Moreover, It plays key roles in apoptosis, cell cycle, and host-virus interaction.
MAPK8	Serine/threonine-protein kinase	Stimulated MAPK8/JNK1 via extracellular stimuli such as proinflammatory cytokines or physical stress phosphorylates primarily components of AP-1 (JUN, JDP2 and ATF2), resulting in regulation of AP-1 transcriptional activity.
MAPK9	Serine/threonine-protein kinase	Stimulated MAPK9/JNK2 via proinflammatory cytokines or physical stress phosphorylates primarily components of AP-1 (JUN, JDP2 and ATF2), resulting in regulates AP-1 transcriptional activity.
MAPK10	Serine/threonine-protein kinase	Stimulated MAPK10/JNK3 via proinflammatory cytokines or physical stress phosphorylates primarily components of AP-1 (JUN, JDP2 and ATF2), resulting in regulates AP-1 transcriptional activity.
MAPK13	Serine/threonine-protein kinase	Stimulated MAPK13 via proinflammatory cytokines or physical stress leads to direct activation of transcription factors such as ELK1 and ATF2.
MAPK14	Serine/threonine-protein kinase	Stimulated MAPK14 via proinflammatory cytokines or physical stress leads to direct or indirect activation of transcription factors such as RPS6KA5/MSK1, RPS6KA4/MSK2, CREB1, ATF1, the NF-kappa-B isoform RELA/NFKB3, STAT1 and STAT3.
MYD88	Cytosolic adapter	Adapter protein involved in the innate immune response, NF-kappa-B activation, cytokine production and inflammatory responses.

(*continued on next page*)

Table 5 (*continued*)

Gene symbol	Molecular function	Protein function
PIK3CA	Serine/threonine-protein kinase	Inflammation
PIK3R1	Adaptor, Regulatory subunit	Host-virus interaction, IL-4 Signaling Pathways, IL-2 Pathways Hypoxia-inducible factor regulation
PIK3R3	Regulatory subunit	IL-4 Signaling Pathways, IL-2 Pathways
PRKCA	Serine/threonine-protein kinase	Positive and negative regulation of platelet function and inflammation
PRKCB	Serine/threonine-protein kinase	B-cell activation, Activation of the NF-kappa-B pathway
STAT2	Signal transducer and transcription activator	Signal transducer and activator of transcription mediating signaling by type I interferons. Moreover, recruiting USP18 to the type I IFN receptor subunit IFNAR2, STAT2 has a negative regulatory role in the type I IFN signaling.
STAT3	Signal transducer and transcription activator	It mediates cellular response to interleukins, KITLG/SCF, LEP and other growth factors. It is Involved in inflammatory response by regulating differentiation of naive CD4(+) T-cells into T-helper Th17 or regulatory T-cells. Moreover, STAT3 is capable of binding to the IL6-responsive elements in the promoter region of different acute-phase protein genes.
SYK	Non-receptor tyrosine kinase	It mediates signal transduction downstream of a variety of transmembrane receptors including BCR. It plays a key role in stimulation of neutrophil phagocytosis by IL15. It is involved in dendritic and mast cells activation as well as IL-3/IL3-mediated signaling in basophils. It strengthens integrin-mediated activation of neutrophils and macrophages and P-selectin receptor/ SELPG-mediated leukocyte recruitment to inflammatory sites.
TRAF3	Transferase	It regulates pathways resulting in the activation of NF-kappa-B and MAP kinases. It plays critical roles in B-cell survival, normal antibody isotype switching from IgM to IgG, T-cell dependent immune responses, antiviral responses as well as the production of cytokines and interferon.
TRAF6	Hydrolase, Receptor	It mediates dendritic cells (DCs) maturation and/or activation as well as NF-kappa-B and JUN activation. It plays a role in signal transduction initiated via TNF receptor, IL-1 receptor and IL-17 receptor. TRAF6 and MAP3K8 regulate immunoglobulin production via CD40 signals that activate ERK in B-cells and macrophages.
TLR4	Receptor	Receptor protein involved in the innate immune responses, activation of NF-kappa-B, production of cytokines and inflammatory responses

Table 6
The inflammatory pathways of strong potential target genes.

Gene symbol	Inflammatory pathway
DDX58	NF-KB
EGFR	JAK-STAT,MAPK
IFNAR1	JAK-STAT
IFNAR2	JAK-STAT
IL6ST	JAK-STAT
IL6R	JAK-STAT
IRAK4	NF-KB
MAPK1	MAPK
MAPK8	MAPK
MAPK9	MAPK
MAPK10	MAPK
MAPK13	MAPK
MAPK14	MAPK
MYD88	NF-KB
PIK3CA	JAK-STAT
PIK3R1	JAK-STAT
PIK3R3	JAK-STAT
PRKCA	MAPK
PRKCB	NF-KB, MAPK
STAT2	JAK-STAT
STAT3	JAK-STAT
SYK	NF-KB
TRAF3	NF-KB
TRAF6	NF-KB
TLR4	NF-KB

genes involved in inflammatory pathways in COVID-19. It is possible to use these miRNAs as therapeutic targets and potential biomarkers for this and other inflammatory disorders.

Note that the results of this study were obtained via investigating high-throughput studies using in silico tools to predict potential miRNAs-mRNA interactions in inflammatory networks. Therefore, using the results, researchers can develop hypotheses for future experimental studies in the area of inflammation in relation to not only COVID-19 but also other inflammatory disease.

Ethics approval and consent to participant

Not applicable.

Consent of publication

Not applicable.

Authors' contributions

SGF and SHS wrote the draft and revised it. SHS, BMH and MDO designed and supervised the study. ZAF, PM, ZaAF, ShKh and NS collected the data and performed the bioinformatic analysis. All the authors read and approved the submitted version.

Author statement

The authors declare that all persons who meet authorship criteria are listed and actively participated in the design and writing of this work. We also declare that we have no any financial conflict of interest that could have appeared to influence the work.

Declaration of Competing Interest

The authors declare they have no conflict of interest.

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