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Biomarkers: A Potencially Networthy Tool in Covid-19

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I. INTRODUCTION:

The novel coronavirus disease COVID-19 has become the fifth documented pandemic in 2019 since the 1918 flu (influenza) pandemic. Coronavirus is the virus from coronaviridae family and it is of zoonotic origin.

In 2019, corona virus epidemic has been found which was first reported in wuhan, china Hubei and subsequently spread worldwide. In July 2020, WHO and china began the studies to better understand the origin of virus. The working group examined that COVID-19 is affecting mainly the respiratory system. Furthermore documented data showed that the virus has rapid rate of mortality and pneumonia specific deaths were occurred in wuhan 2020 and thus the studies has shown up that the virus transmission widespread among the humans via human to human transmission rather it would be considered to be harmful only in animals.

The molecular epidemiology and bioinformatics working groups examined the genomic data of virus and shown that the virus is highly related to SARS-CoV-2 (Severe acute respiratory syndrome-corona virus-2) and also MERS (Middle East respiratory syndrome). The animal and environmental working group shown that the virus is phylogenetically related to SARS-CoV-2 identified in different animals include horseshoe bats (rhinolopus) and pangolins and also declared the virus to be of zoonotic origin by Joint International team. The findings showed that it was a positive stranded RNA virus belonging to the family coronaviridae a subgroup B beta coronavirus and was new to humans.The World Health Organization declare the outbreak to be a Public health emergency on International concern on 1 January 2020 and recognized it as a Pandemic on 3 January 2020.

CORONA VIRUS:

Corona viruses are a family of enveloped, non-segmented, single stranded, positive sense RNA viruses classified within the nidovirales order (2). Corona virus have been described for more than 50 years, which is the strain murine virus the animal strain reported in 1949. In spring 2003, it became clear that corona virus also affects humans as a first symptom of common cold. In SARS-CoV human coronavirus there were two prototype human coronaviruses, OC43 and 229E are found. Coronaviruses contain a non-segmented, positive-sense RNA genome of ~30 kb. The genome contains a 5′ cap structure along with a 3′ poly (A) tail, allowing it to act as an mRNA for translation of the replicase polyproteins. The replicase gene encoding the non-structural proteins (nsps) occupies two-thirds of the genome, about 20 kb, as opposed to the structural and accessory proteins, which make up only about 10 kb of the viral genome. The 5′ end of the genome contains a leader sequence and untranslated region (UTR) that contains multiple stem loop structures required for RNA replication and transcription. Additionally, at the beginning of each structural or accessory gene are transcriptional regulatory sequences (TRSs) that are required for expression of each of these genes. The 3′ UTR also contains RNA structures required for replication and synthesis of viral RNA. The organization of the coronavirus genome is 5′-leader-UTR- replicase-S (Spike)-E (Envelope)-M (Membrane)-N (Nucleocapsid)-3′ UTR-poly (A) tail with accessory genes interspersed within the structural genes at the 3′ end of the genome. The accessory proteins are almost exclusively nonessential for replication in tissue culture; however, some have been shown to have important roles in viral pathogenesis 3).

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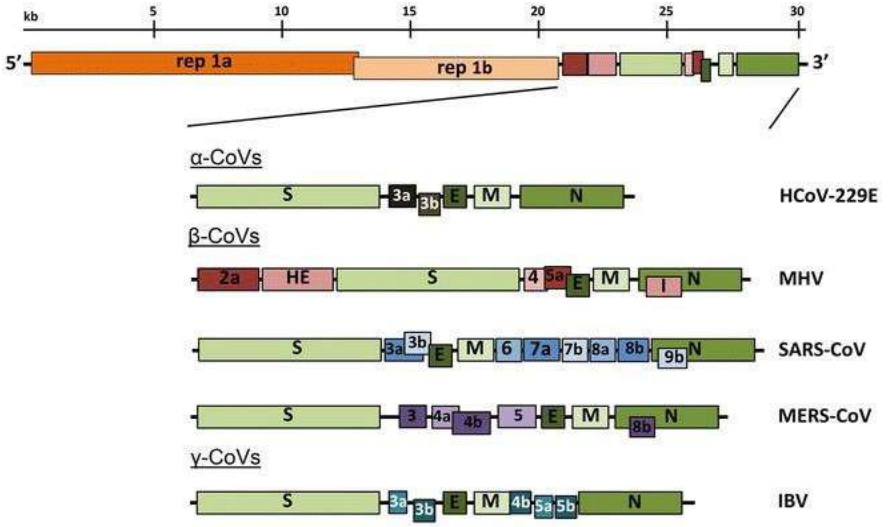

Fig 1. Genomic organization of representative α, β, and γ CoV

There are various type of corona viruses which are:

1. Murine coronavirus (mouse virus)
2. Porcine coronavirus (neonatal)
3. Avian coronavirus (chickens)
4. Feline coronavirus (cats)
5. Bovine coronavirus(calf)
6. SARS-CoV-2 (Humans)

In the structure of corona virus following proteins are seen:

Spike protein: The coronavirus spike protein is a type I glycoprotein that forms the peplomers on coronavirus particles. Some coronavirus spike proteins are cleaved into two subunits by a furin-like enzymatic activity during processing in the Golgi apparatus (2).

M protein: M Protein is associated with the rigidity, clusters of spikes and a narrow range of membrane curvature. It is also associated with flexibility and low spike density.

E Protein: E Protein has ion channel activity and it interact with host proteins and may have multiple membrane topologies.

Haemagglutinin-esterase: It mediate reversible attachment to O-acylated sialic acid by acting both as lectins and as receptor destroying enzymes (3).

N Protein: It stands for nucleocapsid protein. These are basic proteins that encapsulate viral genome RNA to form the part of viral structure. It is highly antigenic and associated with several host-cell interactions.

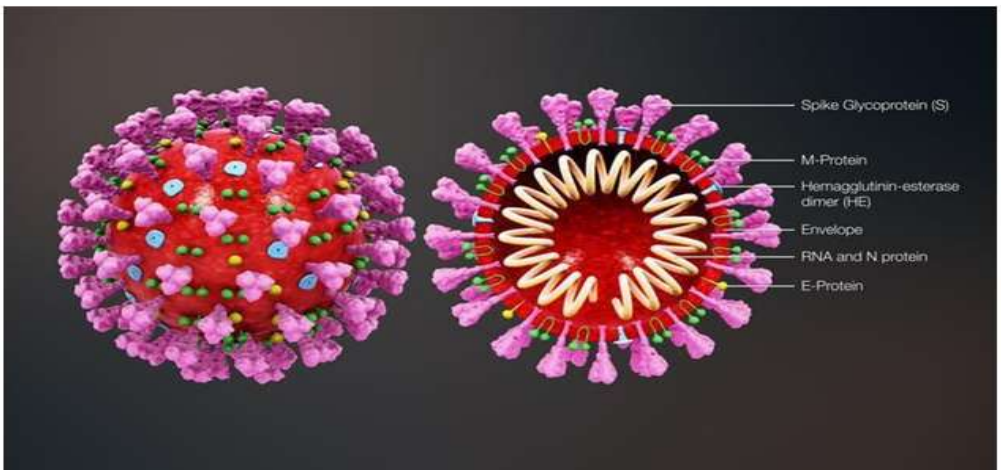

Fig.1 Corona virus structure

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REPLICATION CYCLE AND PATHOGENESIS:

The replication cycle of SARS-CoV-2 provide viral transmission mechanism and reveal therapeutic targets (4). Viral genomic replication is initiated by the synthesis of full-length negative-sense genomic copies, which
function as templates for the generation of new positive-sense genomic RNA. These newly synthesized genomes are used for translation to generate more nsps and RTCs or are packaged into new virions . Stages of the cycle are:

1. Virus entry
2. Translation of viral replication machinery
3. Replication
4. Translation of viral structure protein
5. Virion assembly
6. Release of virus

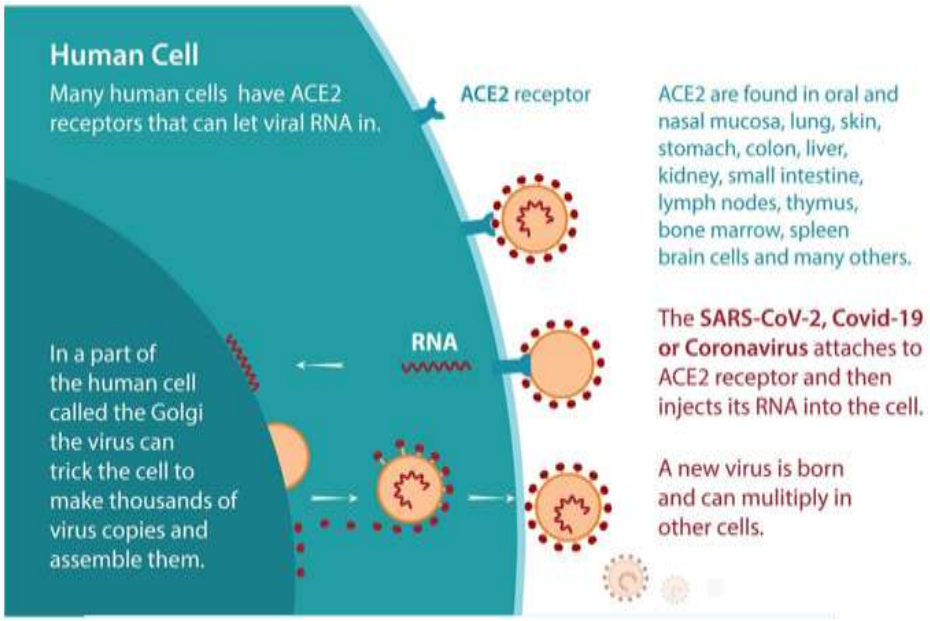

Fig 2. Corona virus replication cycle (4)

The next step in the coronavirus lifecycle is the translation of the replicase gene from the virion genomic RNA. The replicase gene encodes two large ORFs, rep1a and rep1b, which express two co-terminal polyproteins, pp1a and pp1ab. In order to express both polyproteins, the virus utilizes a slippery sequence (5′-UUUAAAC-3′) and an RNA pseudoknot that cause ribosomal frameshifting from the rep1a reading frame into the rep1b ORF. Polyproteins pp1a and pp1ab contain the nsps 1–11 and 1–16, respectively. In pp1ab, nsp11 from pp1a becomes nsp12 following extension of pp1a into pp1b. However, γ-coronaviruses do not contain a comparable nsp1. These polyproteins are subsequently cleaved into the individual nsps. Coronaviruses encode either two or three proteases that cleave the replicase polyproteins. They are the papain-like proteases (PLpro), encoded within nsp3, and a serine type protease, the main protease, or Mpro, encoded by nsp5. Most coronaviruses encode two PLpros within nsp3, except the γ-coronaviruses, SARS-CoV and MERS-CoV, which only express one PLpro. Next, many of the nsps assemble into the replicase–transcriptase complex (RTC) to create an environment suitable for RNA synthesis, and ultimately are responsible for RNA replication and transcription of the sub-genomic RNAs (4).

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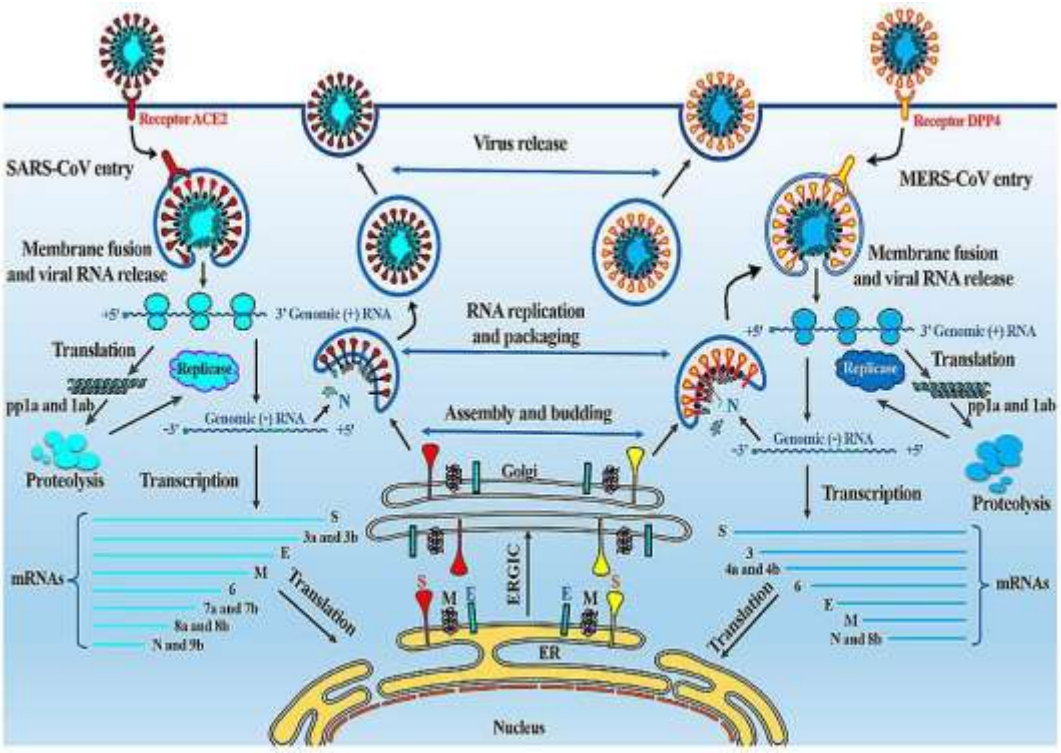

Fig.3 Corona virus replication cycle (4)

Coronavirus receptors are:

Virus	Receptors
Alpha corona viruses	
HCov-229E	Amino peptidase
HCoV-NL63	Angiotensinogen converting enzyme
TGEV(transmissible gastroenteritis virus)	Amino peptidase
PEDV(Porcine epidemic diarrhea virus)	Amino peptidase
FIPV(Feline infectious peritonitis virus)	Amino peptidase
CCoV(Canine corona virus)	Amino peptidase
Betacoronaviruses	
MHV(Murine hepatitis virus)	Murine carcinoembryogenic antigen virus
SARS-CoV	Angiotensin converting enzyme-2

Table 1. Corona virus receptors

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Biomarkers-

A biomarker is a characteristic that is objectively measured and evaluated as an indicator of normal biological processes, pathogenic processes or pharmacological responses to a therapeutic intervention(5). Biomarkers can be classified based on different parameters, including their characteristics, such as imaging biomarkers (computed tomography, positron emission tomography, magnetic resonance imaging) or molecular biomarkers. Molecular biomarkers can be used to refer to nonimaging biomarkers that have biophysical properties, which allow them to be measured in biological samples, and include nucleic acid–based biomarkers such as gene mutations or polymorphisms and quantitative gene expression analysis, peptides, proteins, lipids metabolites, and other small molecules. Biomarkers can also be classified based on their application, such as diagnostic biomarkers, staging of disease biomarkers, disease prognosis biomarkers (cancer biomarkers), and biomarkers for monitoring the clinical response to an intervention. Another category of biomarkers includes those used in decision making during early drug development. For instance, pharmacodynamic biomarkers are markers of a certain pharmacological response and are of special interest in dose optimization studies. They can be categorized into five categories based on their application in different disease stage:

1. Antecedent biomarkers to identify the risk of developing an illness
2. Screening biomarkers to screen for subclinical diseases
3. Diagnostic biomarkers to recognize overt disease
4. Staging biomarkers to categorise disease severity
5. Prognostic biomarkers to predict future disease course

Biomarkers are further categorised as:

Biomarker	Acceptance date	Type	Proposed utility and Qualification stage (QS)
Total hip bone mineral density (BMD)	2016	Response	Efficacy response QS: consultation and advice
Serum glutamate dehydrogenase (GLDH)	2016	Safety	Safety assessment QS: Consultation and advice
Plasma fibrinogen in COPD	2012	Prognostic	Patient selection, QS: Qualified

Table 3: List of current biomarkers at FDA for approval (6)

Development of Biomarkers:

Depending on the specific biomarker, it will have specific ways of detecting as well. Some of these biomarkers can be detected through:

1. Positron emission tomography
2. Diffusion magnetic resonance
3. Gas chromatography with mass spectrometry
4. And more.

Application of Biomarkers with examples:

1. Prostate Specific Antigen (PSA)
   - Was used as a biomarker for prostate cancer

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- Recent concerns regarding its diagnostic value
- Great for prognosis

2. CA-125
- Not useful as a screening tool
- Useful for guidance of treatment options
- Great for prognosis

3. Inflammation
- Prognosis for ischemic stroke (7).

Current research and studies are focused on the identification, validation, and implementation of specific biopolymers which can then be used as biomarkers for population screening and prognostic purposes.

Biomarkers have ALWAYS played an important role in clinical decision making in various infectious diseases. For the diagnosis of covid-19 infection various biomarkers are used with a different aspects which are studied on various participants of the infection. They are as follows:
- Lymphocytes
- Platelets
- D dimer
- Lactate dehydrogenase (LDH)
- C reactive protein (CRP)
- Aspartate aminotransferase (AST)
- Creatinine
- Procalcitonin (PCT)
- Creatinine kinase
- Liver function test (LFT)
- Kidney function test (KFT)

1. Analysis of Lymphocyte count (ALC):

The immune system is a complex network of cells known as immune cells that include lymphocytes. These cells work together to defend the body against foreign substances such as bacteria, viruses and cancer cells that can threaten its functioning. Lymphocytes are responsible for both the specificity and memory that are the defining characteristics of the adaptive immune response (9). In the diagnosis of covid-19 infection lymphocyte count is measured as in the outbreak of covid-19 several studies have determined correlation with disease severity and lymphopenia (decrease in lymphocyte count). COVID-19 patients suffering from lymphopenia almost always exhibit significant decreases in T cell counts and showed drastic decrease in CD8 T-cells. The normal range of ALC is 1.10– 4.00 K/uL

Following are the causes for the observed lymphopenia in severe COVID-19 patients, especially the decrease in T cell counts:

1. The SARS-CoV-2 virus may infect T cells. A study reported that two human T cell lines (MT-2 and A3.01) with a very low level of human ACE2 mRNA, the receptor that the virus uses to enter the host, can be infected with the virus in vitro. However, the virus could not replicate within the infected cells, as measured by qPCR expression of the viral N gene.

2. The SARS-CoV-2 infection can interfere with T cell expansion. A report suggests that some genes involved in T cell activation and function, such as MAP2K7 and SOS1 are downregulated in the T cells of severe COVID-19 patients. The expression of most of these genes returned to normal levels upon recovery.

3. COVID-19 infection can result in exhaustion of T cells. A study found both $CD4^+$ and $CD8^+$ T cells from COVID-19 patients had increased cell surface expression of programmed cell death protein 1 (PD-1) and T cell immunoglobulin and mucin domain 3 (Tim-3), two markers of T cell exhaustion.

4. The serum level of pro-inflammatory cytokines, such as TNF-α and IL-6, have been closely correlated with lymphopenia, while recovered patients show close to normal levels of such cytokines(8).

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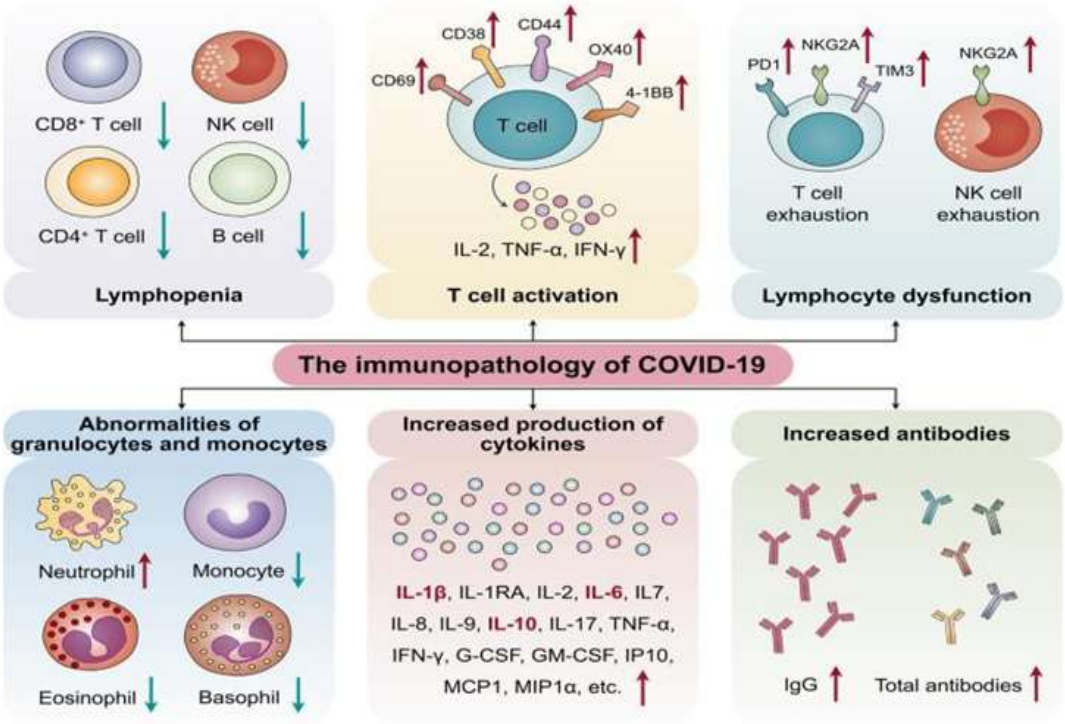

Fig.4 The immunopathology of COVID-19 and lymphocyte count

2. Platelet count:

Platelets are also called thrombocytes which are a component of blood whose function is to react to bleeding from blood vessel injury by clumping, thereby initiating a blood clot. Platelets also perform profound immune functions during infection with various pathogens. It can also mediate a response to various RNA viruses such as influenza and that many viral infections, including severe acute respiratory syndrome coronavirus 2 (SARS- CoV- 2), can affect platelet count. Thrombocytopenia and increased coagulation have been independently associated with increased mortality. In COVID- 19 patients, disseminated intravascular coagulation and reduced platelet count are associated with poor prognosis and increased risk of mortality.

Due to their abundance, platelets may be the first blood component to internalize viral particles and induce a response once the pathogen reaches circulation. In human platelets, the initial response to single- stranded viral RNA is mediated predominantly by Toll- like receptor

7 (TLR7). TLR7 is located in the endolysosomes of platelets and requires both internalization of the viral particle and the acidic pH of the endolysosome for proper activation and signalling. Activation of TLR7 leads to α- granule release in an AKT- and p38- dependent manner and consequently leads to interaction of platelets with neutrophils via P- selectin and CD40L. In addition, platelet TLR7 leads to complement C3 release, which pushes neutrophils to release their DNA in the process of netosis. C3- mediated netosis does not require the attachment of neutrophils to the vascular bed, and netting neutrophils can circulate in blood. Neutrophil extracellular traps (NETs) capture and protect from viral challenge but are also highly prothrombotic and, when dysregulated, may induce intravascular coagulation. Thrombin generated from coagulation in turn can activate C3 and, consequently, the entire proinflammatory complement cascade. In support of intravascular DNA release from neutrophils and potential applicability to this mechanism, sera from patients with COVID- 19 contain highly specific markers for netosis (10).

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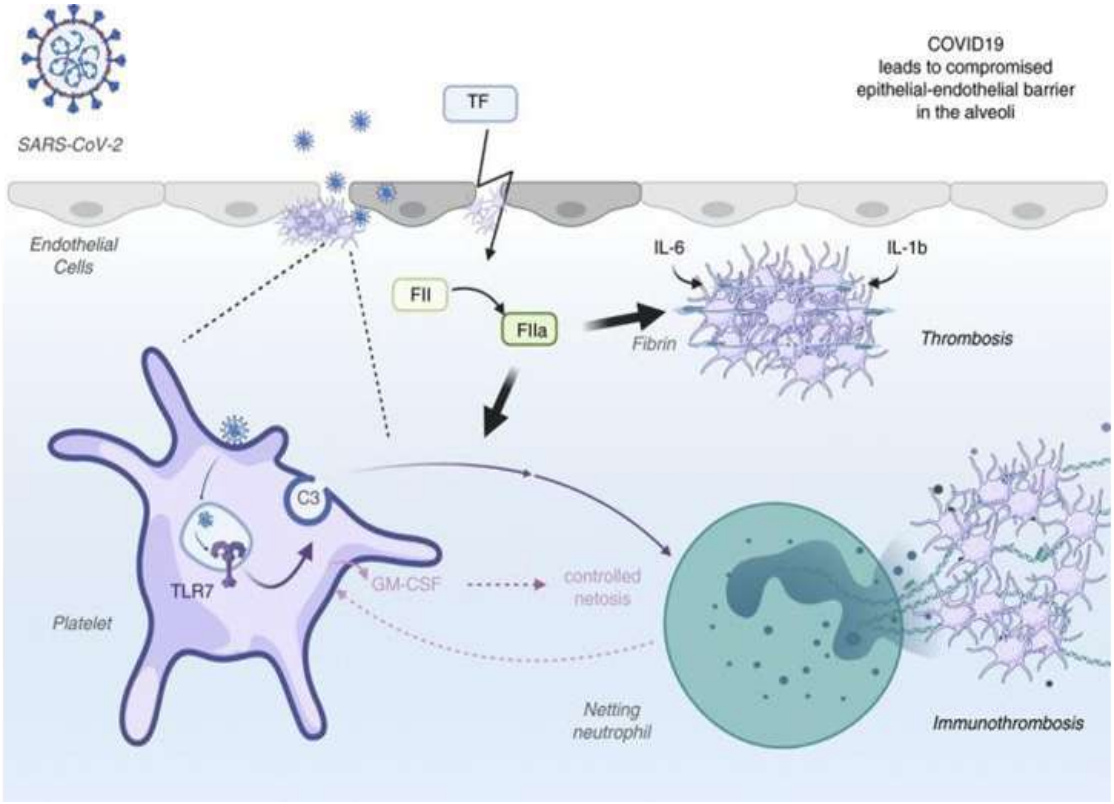

Fig.5 Role of platelet in diagnosis of COVID-19

3. D-Dimer:

D-Dimer is a fibrin degradation product, a small protein fragment present in the blood after a clot is degraded by fibrinolysis. D-dimer concentration may be determined by a blood test to help diagnose thrombosis. Since its introduction in the 1990s, it has become an important test performed in patients with suspected thrombotic disorders. While a negative result practically rules out thrombosis, a positive result can indicate thrombosis but does not rule out other potential causes. Its main use, therefore, is to exclude thromboembolic disease where the probability is low. In addition, it is used in the diagnosis of the blood disorder disseminated intravascular coagulation. A four-fold increase in the protein is a strong predictor of mortality in those suffering from COVID-19.

4.Lactate Dehydrogenase:-

In glucose metabolism, the enzyme LDH converts pyruvate to lactate. LDH secretion is triggered by necrosis of the cell membrane, hinting to viral infection or lung damage, such as the pneumonia induced by SARS-CoV-2. There is convincing evidence linking LDH levels to the development of COVID-19 disease.

A study found significantly higher levels of LDH in ICU patients than non-ICU patients (248 U/L vs 151 U/L, p=0.002). Since high levels of LDH continued in the ICU patients number of days post-admission (160 U/L vs 218 U/L, p=0.002), LDH may be a predictive biomarker of severe disease. However, the one center study may be prone to selection bias which could potentially reduce its validity.

A multi-center study involving 1099 patients reported supporting evidence correlating extent of tissue damage and inflammation with increasing levels of LDH. Furthermore, when LDH levels were correlated with CT scans, significantly higher levels reflected the severity of pneumonia.

There is increasing confidence in using LDH as a biomarker to measure severity of COVID- 19 infection. Another study found that there was a significant rise in LDH levels among refractory COVID-19 patients.

5.C-reactive protein:-

CRP is a plasma protein produced by the liver and induced by various inflammatory

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mediators such as IL-6. Despite being non-specific, this acute phase reactant is used clinically as a biomarker for various inflammatory conditions; a rise in CRP levels are associated with an increase in disease severity.

The application of CRP in COVID-19 has been highlighted by a retrospective single-centre study in Wuhan, China, where the majority of patients in the severe cohort showed significantly higher levels compared to the non-severe cohort (57.9 mg/L vs 33.2 mg/L, P < 0.001) [8]. A second retrospective cohort study found the likelihood of progressing to severe COVID-19 disease increased in patients with CRP levels >41.8 mg/L. Both studies suggest CRP levels are a strong indicator to reflect the presence and severity of COVID-19 infection.

Normal Range:-

- **Lower risk.** You have an hs-CRP level of less than 2.0 milligram per liter (mg/L).
- **Higher risk.** You have an hs-CRP level greater than 2.0 mg/L.

Furthermore, a study from unpublished observations suggests CRP is one of the first biomarkers within blood plasma that changes to reflect physiological complications; if accepted CRP will be the most effective biomarker to predict the progression of COVID-19 infection. Contrastingly, the same study illustrated some cases of infection which showed changes in serum amyloid A (SAA) instead of evoking significant CRP changes thus requiring further evaluation.

Whilst the use of SAA as a biomarker for COVID-19 requires further research, CRP and SAA are commonly used in conjunction to monitor inflammatory diseases. Though SAA is another acute phase reactant, it is responsive to both viral and bacterial infections.

6.Aspartate Aminotransferase:-

Aspartate Aminotransferase (AST) is an enzyme that's present in various tissues of your body. An enzyme is a protein that helps trigger chemical reactions that your body needs to function.

AST is found in the highest concentrations in your liver, muscles, heart, kidney, brain and red blood cells. A small amount of AST is typically in your bloodstream. Higher-than-normal amounts of this enzyme in your blood may be a sign of a health problem. Abnormal levels can be associated with liver injury.

AST levels increase when there's damage to the tissues and cells where the enzyme is found. AST levels can rise as soon as six hours Trusted Source after damage to tissue occurs. The normal range for AST is higher from birth to age 3 compared to the normal ranges for older children and adults.

The AST test measures the amount of AST in your blood that has been released from injured tissue. An older name for the test is serum glutamic-oxaloacetic transaminase (SGOT).

You should have the results in about a day. They are given in units per liter (units/L). Normal ranges are:

Males: 10 to 40units/L Females: 9 to 32units/L

An AST blood test is often included in a routine blood screening. The test may also be used to help diagnose or monitor liver problems.

7.Creatinine:-

A creatinine test is a measure of how well your kidneys are performing their job of filtering waste from your blood.

Creatinine is a chemical compound left over from energy-producing processes in your muscles. Healthy kidneys filter creatinine out of the blood. Creatinine exits your body as a waste product in urine.

A measurement of creatinine in your blood or urine provides clues to help your doctor determine how well the kidneys are working.

Serum creatinine level

Creatinine usually enters your bloodstream and is filtered from the bloodstream at a generally constant rate. The amount of creatinine in your blood should be relatively stable. An increased level of creatinine may be a sign of poor kidney function.

Serum creatinine is reported as milligrams of creatinine to a deciliter of blood (mg/dL) or micromoles of creatinine to a liter of blood (micromoles/L). The typical range for serum creatinine is:

Normal Range:-

For adult men, 0.74 to 1.35 mg/dL (65.4 to 119.3 micromoles/L) For adult women, 0.59 to 1.04 mg/dL (52.2 to 91.9 micromoles/L)

8.Procalcitonin:-

Procalcitonin(PCT) is the 116-amino acid precursor of the hormone calcitonin.

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A hormone that is synthesized by the parafollicular cells of the thyroid and involved in calcium homeostasis. Procalcitonin arises from endopeptidase-cleaved preprocalcitonin.

The biomarker Procalcitonin (PCT) is widely used to assess the risk of bacterial infection and progression to severe bacterial sepsis and septic shock in conjunction with other laboratory findings and clinical assessment. Further, the change of PCT over time is used to determine the mortality risk in patients with bacterial sepsis.

In patients with suspected or confirmed lower respiratory tract infections (LRTI), including communityacquired pneumonia (CAP), acute bronchitis and acute exacerbations of COPD (AECOPD), PCT is an aid in decision making on antibiotic therapy for inpatients or patients presenting in the emergency department (ED).

Normal Range:-o.15ng/ml or less

High risk or severe sepsis -2.00ng/ml or more (High risk or severe sepsis)

Procalcitonin has now been shown, in evolving descriptive studies, to be an additional valuable tool in the current COVID-19 pandemic to early identify patients at low risk for bacterial coinfection and adverse outcome.

9. Creatine Kinase:-

Creatine kinase (CK), formerly known as creatine phosphokinase, is an intracellular enzyme present in greatest amounts in skeletal muscle, myocardium, and brain; smaller amounts occur in other visceral tissues.

Disruption of cell membranes due to hypoxia or other injury releases CK from the cellular cytosol into the systemic circulation. On this basis, elevated serum levels of CK have been used as a sensitive but nonspecific test for myocardial infarction. The poor specificity reflects the ubiquity of CK in many tissues other than the myocardium.

Because of the many current assay methods in use, there is no standard reference value for serum CK. Normal values are best determined locally based on the method employed and the ranges for healthy controls. Values are expressed in international units per liter.

CK is a dimeric molecule composed of two subunits designated M and B. Combinations of these subunits form the isoenzymes CK–MM, CK–MB, and CK–BB. A significant concentration of CK–MB isoenzyme is found almost exclusively in the myocardium, and the appearance of elevated CK–MB levels in serum is highly specific and sensitive for myocardial cell wall injury. Normal reference values for serum CK–MB range from 3 to 5% (percentage of total CK) or 5 to 25 IU/L.

Normal Range is 22-192 U/L (units/liter)

CPK blood tests the different forms of CPK in the bloodstream and the CPK normal range varies from a male to female.

The CPK normal range for a male is between 39 – 308 U/L, while in females the CPK normal range is between 26 – 192 U/L.

10.Liver function tests:-

Liver function tests are blood tests used to help diagnose and monitor liver disease or damage. The tests measure the levels of certain enzymes and proteins in your blood.

Some of these tests measure how well the liver is performing its normal functions of producing protein and clearing bilirubin, a blood waste product. Other liver function tests measure enzymes that liver cells release in response to damage or disease.

Abnormal liver function test results don't always indicate liver disease. Your doctor will explain your results and what they mean.

Liver function tests can be used to:

- Screen for liver infections, such as hepatitis
- Monitor the progression of a disease, such as viral or alcoholic hepatitis, and determine how well a treatment is working
- Measure the severity of a disease, particularly scarring of the liver (cirrhosis)
- Monitor possible side effects of medications

Liver function tests check the levels of certain enzymes and proteins in your blood. Levels that are higher or lower than normal can indicate liver problems. Some common liver function tests include:

- **Alanine transaminase (ALT).** ALT is an enzyme found in the liver that helps convert proteins into energy for the liver cells. When the liver is damaged, ALT is released into the bloodstream and levels increase.
- **Aspartate transaminase (AST).** AST is an enzyme that helps metabolize amino acids.

Like ALT, AST is normally present in blood at low levels. An increase in AST levels may indicate liver damage, disease or muscle damage.

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- **Alkaline phosphatase (ALP).** ALP is an enzyme found in the liver and bone and is important for breaking down proteins. Higher-than-normal levels of ALP may indicate liver damage or disease, such as a blocked bile duct, or certain bone diseases.
- **Albumin and total protein.** Albumin is one of several proteins made in the liver. Your body needs these proteins to fight infections and to perform other functions. Lower- than-normal levels of albumin and total protein may indicate liver damage or disease.

- **Bilirubin.** Bilirubin is a substance produced during the normal breakdown of red blood cells. Bilirubin passes through the liver and is excreted in stool. Elevated levels of bilirubin (jaundice) might indicate liver damage or disease or certain types of anemia.
- **Gamma-glutamyltransferase (GGT).** GGT is an enzyme in the blood. Higher-than- normal levels may indicate liver or bile duct damage.
- **L-lactate dehydrogenase (LD).** LD is an enzyme found in the liver. Elevated levels may indicate liver damage but can be elevated in many other disorders.
- **Prothrombin time (PT).** PT is the time it takes your blood to clot. Increased PT may indicate liver damage but can also be elevated if you're taking certain blood-thinning drugs, such as warfarin.

Normal blood test results for typical liver function tests include:

- **ALT.** 7 to 55 units per liter (U/L)
- **AST.** 8 to 48 U/L
- **ALP.** 40 to 129 U/L
- **Albumin.** 3.5 to 5.0 grams per deciliter (g/dL)
- **Total protein.** 6.3 to 7.9 g/dL
- **Bilirubin.** 0.1 to 1.2 milligrams per deciliter (mg/dL)
- **GGT.** 8 to 61 U/L
- **LD.** 122 to 222 U/L
- **PT.** 9.4 to 12.5 seconds

11.Kidney Function Test:-

Kidneys play an important role in the removal of waste products and maintenance of water and electrolyte balance in the body. Kidney Function Test (KFT) includes a group of blood tests to determine how well the kidneys are working.

These are simple **blood** and urine **tests** that can identify problems with your **kidneys**. You may also need **kidney function testing done** if you have other conditions that can harm the **kidneys**, such as diabetes or high **blood** pressure. They can help doctors monitor these conditions.

Some people suffering with severe cases of COVID-19 are showing signs of kidney damage, even those who had no underlying kidney problems before they were infected with the coronavirus. Early reports say that up to 30% of patients hospitalized with COVID-19 in China and New York developed moderate or severe kidney injury.

A new comprehensive report shows that people hospitalized with COVID-19 are at significant risk of AKI, which can lead to serious illness, dialysis, and even death

"Many patients with severe COVID-19 are those with co-existing, chronic conditions, including high blood pressure and diabetes. Both of these increase the risk of kidney disease,".

Urinalysis

A urinalysis screens for the presence of protein and blood in the urine. There are many possible reasons for protein in your urine, not all of which are related to disease. Infection increases urine protein, but so does a heavy physical workout. Your doctor may want to repeat this test after a few weeks to see if the results are similar.

Your doctor may also ask you to provide a 24-hour urine collection sample. This can help doctors see how fast a waste product called creatinine is clearing from your body. Creatinine is a breakdown product of muscle tissue.

Serum creatinine test

This blood test examines whether creatinine is building up in your blood. The kidneys usually completely filter creatinine from the blood. A high level of creatinine suggests a kidney problem.

According to the National Kidney Foundation (NKF), a creatinine level higher than 1.2 milligrams/deciliter (mg/dL) for women and 1.4 mg/dL for men is a sign of a kidney problem.

Blood urea nitrogen (BUN)

The blood urea nitrogen (BUN) test also checks for waste products in your blood. BUN tests measure the amount of nitrogen in the blood. Urea nitrogen is a breakdown product of protein.

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A normal BUN level is between 7 and 20 mg/dL. A higher value could suggest several different health problems.

Estimated GFR

This test estimates how well your kidneys are filtering waste. The test determines the rate by looking at factors, such as:

- Test results, specifically creatinine levels
- Age
- Gender
- Race
- Height
- Weight

Any result lower than 60 milliliters/minute/1.73m^2 may be a warning sign of kidney disease.

Normal levels:

Urea:-2.5-7.8 mol/L BUN:-7 to 20 mg/dL
Uric Acid:

Adult Female: 2.4 to 6.0 mg/dL Adult Male: 3.4 to 7.0 mg/dL Children: 3 to 4 mg/dL
Creatinine:

Adult males: 0.6 to 1.2 mg/dL Adult females: 0.5 to 1.1 mg/dL
BUN/Creatinine Ratio:-12:1 to 20:1

12.Cycle Threeshold (CT) value:-

A CT value refers to the number of cycles after which the virus can be detected.
CT refers to the number of cycles needed to amplify the viral RNA to a detectable level. The lower the CT value means the higher is the virus level is in your body. And the higher
the CT value means the lower is the virus load in your body. During the RT-PCR test, the CT
value is considered very important.

LOWER CT VALUE = HIGHER viral load
HIGHER CT VALUE = LOWER VIRAL LOAD

An individual is considered COVID-19 negative, only if the CT value is 35 in the RT-PCR test. So, if the CT value is below 35 in the RT-PCR test, then the patient is coronavirus positive.

In RT-PCR testing, RNA is extracted from the swab collected from the patients. RNA is later converted into DNA, which is then amplified. And amplifications refer to the process of creating multiple copies of the genetic material.

In simple words, CT value also gives clarity on the transmission of the virus from one another. CT value below 24 are highly infectious.

13.Interleukin-6:-

Cytokine release syndrome (CRS) is an over-exaggerated immune response involving an overwhelming release of pro-inflammatory mediators. This mechanism underlies several pathological processes including acute respiratory distress syndrome (ARDS). Studies investigating the role of cytokines in SARS and MERS have had also found a link between CRS and disease severity. Understanding their role in COVID-19 disease may help facilitate the design of novel immunotherapies.

Studies have revealed that levels of IL-6, the most common type of cytokine released by activated macrophages, rise sharply in severe manifestations of COVID-19 [15]. However, since most studies to date have been observational, it is difficult to extrapolate if the rise is significant enough to cause the manifestations seen in severe forms.

Normal range-[0-16.4 pg/ml]

One meta-analysis reviewing six studies show mean IL-6 concentrations were 2.9-fold higher in patients with complicated COVID-19 compared to those with non-complicated disease (n= 1302; 95% CI 1.17–7.19). In its analysis, the outcomes of the studies include ICU admission, onset of ARDS and mortality. Since the proportionate rise of IL-6 is correlated with disease severity, this study can prove ground-breaking. Although clinicians can use this to identify severity earlier and commence oxygen therapy sooner, the varying outcomes makes it somewhat difficult to ascertain what level of IL-6 corresponds to what negative outcome. Furthermore, many studies recruited participants from the same centre, giving rise to the potential of selection bias.

DIAGNOSTIC TEST:-

Reverse Transcriotion-polymerase chain reaction (RT-PCR):-

Reverse transcription polymerase chain reaction (**RT-PCR**) is a laboratory technique combining reverse transcription of RNA into DNA (in this context called complementary DNA or cDNA) and amplification of specific DNA targets using polymerase chain reaction (PCR).

A Reverse Transcription-polymerase chain reaction (RT-PCR) test is performed to detect

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genetic material from a specific organism, such as a virus. The test detects the presence of a virus if you are infected at the time of the test. The test could also detect fragments of virus even after you are no longer infected.

A RT-PCR test for COVID-19 is a test used to diagnosis people who are currently infected with SARS-CoV-2, which is the coronavirus that causes COVID-19. The PCR test is the "gold standard" test for diagnosing COVID-19 because it's the most accurate and reliable test.

The COVID-19 RT-PCR Test is a real-time reverse transcription polymerase chain reaction (RT-PCR) test for the qualitative detection of nucleic acid from SARS-CoV-2 in upper and lower respiratory specimens (nasopharyngeal or oropharyngeal swabs, sputum, lower respiratory tract aspirates, broncho alveolar lavage, and nasopharyngeal wash/aspirate) collected from individuals suspected of COVID-19 by their healthcare provider (HCP), as well as upper respiratory specimens (nasopharyngeal or oropharyngeal swabs, anterior nasal swabs, or mid-turbinate swabs) collected from any individual, including individuals without symptoms or other reasons to suspect COVID19.

An RT-PCR test is a laboratory test that combines reverse transcription of RNA into DNA for the detection of the virus. RT-PCR test is the most preferred test for COVID-19; however, this test is time-consuming and costly as it has an elaborate kit. An RT-PCR test uses nose or throat swabs for the detection of viruses. Also, only trained professionals who are instructed for the use of the RT-PCR kit can carry out the RT-PCR test. RT-PCR requires a full set-up that includes the trained practitioners, laboratory, and RT-PCR machine for detection and analysis.

RT-PCR is one of the most accurate laboratory testing method for the current coronavirus pandemic

Real-time RT-PCR stands for Real-Time Reverse Transcription – Polymerase Chain. Reaction enables detection and presence of specific genetic material in the pathogens that include viruses or bacteria. The method utilizes radioactive isotope markers for the detection of targeted genetic materials; however, with advancements, the isotopic labeling is replaced with special markers such as fluorescent dyes. This technique enables professionals to review the results immediately.

Real-time RT-PCR is one of the widely used methods for the detection of COVID-19. This method also helps in increasing the testing volume to meet the increasing number of sample pools.

CT (Computed Tomography) Scan:-

A computerized tomography scan (CT or CAT scan) uses computers and rotating X-ray machines to create cross-sectional images of the body. These images provide more detailed information than normal X-ray images. They can show the soft tissues, blood vessels, and bones in various parts of the body. A CT scan may be used to visualize the:

- Head
- Shoulders
- Spine
- Heart
- Abdomen
- Knee
- Chest

During a CT scan, you lie in a tunnel-like machine while the inside of the machine rotates and takes a series of X-rays from different angles. These pictures are then sent to a computer, where they're combined to create images of slices, or cross-sections, of the body. They may also be combined to produce a 3-D image of a particular area of the body.

II. CONCLUSION:-

In conclusion, the work to date suggests that there is clear evidence of how the levels of biomarkers may change according to severity of COVID-19 infection. This can be used as an adjunct in clinical practice to guide treatment and admission to ICU. By doing so, it may improve prognosis and minimise the mortality rates. However, being in the infant stages of understanding the pathology of this infectious disease, we urge for further research worldwide to better understand the changes noted in this review.

Covid-19 is a heterogeneous disease 4spectrum with manifestation varying with age and presence of co-morbidities. Biomarkers will play a crucial role in early recognition of complications, management and disposition of patients. Each of these components in turn can have crucial implications on the healthcare system and the administrative machinery, directly impacting patient care. Biomarker panels rather than single biomarkers may provide more reliable information.

This study demonstrates that PCT may be an indicator of disease severity and may contribute to determining the severity of patients with

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COVID-19. In addition, serial PCT measurements may be useful in predicting the prognosis. Additional investigation is needed to further illustrate the mechanisms by which increased PCT is synthesised and released in patients infected with SARS-CoV-2.

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