Chapter 12 Characteristics of Healthy Blood



Geetika Garg, Sandeep Singh, Abhishek Kumar Singh, and Syed Ibrahim Rizvi

Abstract Blood is a specialized fluid consisting of plasma and cells that circulate through the entire body. Blood also contains essential nutrients, oxygen, and hormones in adequate quantity that makes the blood healthy. Some infections in the blood affect its overall health. These are bacteria and blood borne viruses that make the blood infected. Healthy blood is free from all kind of such infections. Blood plays an important role in regulating the body's systems and in maintaining the dynamic homeostasis. It carries oxygen, nutrients and hormones to living cells and takes away their waste products. Blood delivers immune cells to fight infections and contains platelets that can form a plug in a damaged blood vessel to prevent blood loss. Clinical markers associated with blood are used for the diagnosis of various health issues or pathological conditions. Biomarkers of oxidative stress in erythrocytes and plasma are extensively used to study physiological and metabolic processes. During the course of their natural aging erythrocytes can undergo an apoptosis-like cell death, termed eryptosis. This chapter provides an account of the composition and markers of healthy blood and its role in the maintenance of overall health.

Keywords Blood · Plasma · Erythrocytes · WBC · Health

12.1 Introduction

Blood is a specialized fluid connective tissue and a lifesaving liquid organ. Blood plays an important role in regulating the body's systems and maintaining homeostasis. It carries oxygen, nutrients and hormones to living cells and takes away their waste

G. Garg · S. Singh · S. I. Rizvi (🖂)

G. Garg

A. K. Singh

© Springer Nature Switzerland AG 2020

Department of Biochemistry, University of Allahabad, Allahabad 211002, India e-mail: sirizvi@gmail.com

Department of Zoology, Savitribai Phule Pune University, Pune 411007, India

Amity Institute of Neuropsychology and Neurosciences, Amity University Uttar Pradesh, Noida, India

J. Sholl and S. Rattan (eds.), *Explaining Health Across the Sciences*, Healthy Ageing and Longevity 12, https://doi.org/10.1007/978-3-030-52663-4_12

products. Blood delivers immune cells to fight infections and contains platelets that can form a plug in a damaged blood vessel to prevent blood loss. The blood that runs through the veins, arteries, and capillaries is known as whole blood. Whole blood is a mixture of cellular elements, colloids and crystalloids. Blood is circulated around the body through blood vessels by the pumping action of the heart. The arteries deliver oxygenated blood, glucose and other nutrients to the brain and the veins carry deoxygenated blood back to the heart, removing carbon dioxide, lactic acid, and other metabolic products.

The average human adult has more than 5 L of blood in their body, which is composed of plasma and several kinds of cells. Some of the most common blood tests determine which substances are present within blood and in what quantities. Other blood tests check for the composition of the blood itself, including the quantities and types of formed elements.

12.2 Characteristics of Healthy Blood

The first characteristic of blood is its colour. Blood that has taken up oxygen in the lungs is bright red, and blood that has released oxygen in the tissues is a dusky red or dark red. This is because of binding capacity of hemoglobin to oxygen. Another characteristic of blood is its viscosity which is five times greater than water. It is a measure of fluid's thickness and is influenced by the presence of plasma proteins and formed elements. Viscosity affects the blood pressure and blood flow. The pH of blood also determines its quality, and it ranges from 7.35 to 7.45 in a healthy person. Buffers present in the blood help to regulate pH.

12.2.1 Components of the Blood

Blood has four major components: plasma, red blood cells (RBCs), white blood cells (WBCs), and platelets. A brief description of the constituents of normal healthy blood is given below, and are listed in Table 12.1.

Plasma: Blood plasma is the yellowish liquid part of the blood that carries cells and proteins throughout the body. It makes up about 55% of the body's total blood volume. Plasma serves as a transport medium for delivering nutrients to the cells of the various organs of the body and for transporting waste products derived from cellular metabolism to the kidneys, liver, and lungs for excretion. Plasma contains proteins that help blood to clot, transport blood cells throughout the body along with nutrients, antibodies, hormones and proteins that help to maintain homoeostasis. Blood plasma also contains glucose and other dissolved nutrients that makes the blood healthy. Clinical diagnostic markers in plasma and serum are listed in Table 12.2.

Red blood cells (RBC): The percentage of whole blood volume that is made up of red blood cells is called the hematocrit and is a common measure of red blood cell

S. No.	Blood component	Reference range	
		Male	Female
1.	Red blood cells (RBC)	4.3–5.9 million/mm ³	3.5–5.5 million/mm ³
2.	Hemoglobin (HGB)	13.5–17.5 g/dL	12.0–16.0 g/dL
3.	Hematocrit (HT)	41-53%	36–46%
4.	White blood cells (WBC)	4500–11,000/mm3	4500–11,000/mm3
5.	Mean corpuscular volume (MCV)	80–100 μm ³	
6.	Mean corpuscular haemoglobin (MCH)	25.4–34.6 pg/cell	
7.	Mean corpuscular hemoglobin concentration (MCHC)	31–36% Hb/cell	
8.	Platelets	150,000–400,000/mm ³	

 Table 12.1
 Constituents of normal blood

levels. Production of red blood cells takes place in the bone marrow under the control of the hormone erythropoietin and after approximately seven days of maturation they are released into the bloodstream. The morphology of RBC is essentially based on the size which varies in different animals. Generally, erythrocytes have a diameter of 4-10 µm. All non-mammalian (birds, reptiles, amphibians and fish) erythrocytes with a few isolated exceptions are nucleated and contain organelles in their cytoplasm. In humans, the mature form of healthy erythrocyte is normally a non-nucleated, yellowish and biconcave disk shaped (discocyte) when not subjected to external stress (Hillman and Finch 1996). The biconcave shape provides a large surface-to-volume ratio for oxygen delivery and better flexibility in narrow capillaries, and thereby RBCs can easily change their shape, which help them to fit through the various blood vessels in the body. However, while the lack of a nucleus makes a red blood cell more flexible, it also limits the life of the cell as it travels through the smallest blood vessels, damaging its cell membrane and depleting its energy supplies (Diez-Silva et al. 2010; Kuhn et al. 2017). Erythrocyte longevity varies across the major vertebrate groups, in humans the cellular half-life of erythrocytes is about 120 days, and is about 40, 600-800, 300-1400 and 80-500 days in birds, reptiles, amphibians and fish, respectively. These characteristics of RBC are essential for biological functions and can be affected by genetic or acquired pathological conditions. Healthy blood meets all these conditions to maintain fluidity and elasticity of membrane.

White Blood Cells (WBCs or Leukocytes): The WBCs (also called leukocytes) are of two types (Greek "leukos" meaning "white" and "kytos," meaning "cell"). The granular leukocytes (eosinophils, neutrophils, and basophils) are named for the granules in their cytoplasm; the agranular leukocytes include monocytes and lymphocytes which lack cytoplasmic granules (Feher 2012). Pluripotent stem cells in the bone marrow produce myeloid and lymphoid progenitors. The myeloid progenitor differentiates further into a granulocyte/macrophage progenitor that further differentiates into the granulocytes and the monocytes while lymphoid progenitor produces while lymphoid progenitor produces while lymphoid progenitor produces.

G . NI			D.C	D.C
S. No.	Markers	Clinical significance	Reference range	Reference
1.	Blood glucose	Supply energy to all cells in the body. Blood glucose higher than that of normal level indicate hyperglycemia and lower level indicates the hypoglycaemia	70–90 mg/dL fasting, 140 mg/dL 2 h after eating	(Duckworth 2001; Kalra et al. 2013)
2.	Total cholesterol	Used to build the structure of cell membrane and hormones. Help the metabolism to work efficiently	<200 mg/dL	(Lin et al. 2015)
3.	Triglyceride	Stored in fat cells. Contribute to measure the heart health. Harden the artery wall, which increases risk for stroke, heart attack, and cardiovascular disease. High triglycerides are a sign of other conditions such as obesity, diabetes, hypothyroidism, and liver or kidney disease	<150 mg/dL or < 1.7 mmol/L	(Teixeira et al. 2019)
4.	HDL	Helps to remove LDL from blood	60 mg/dL	(Després et al. 2000)
5.	LDL	Indicate the risk of heart attack, stroke, and atherosclerosis	<100 mg/dL	(Ivanova et al. 2017)
6.	Troponin	Used to detect chest pain or heart attack	0-0.4 ng/mL	(Al-Otaiby et al. 2011)
7.	Total protein	Necessary for body's growth, evelopment, and health. Level indicated about disease status in different organs	6–8 g/dL	(Krisko and Radman 2019)
8.	Albumin	Help to diagnose liver and kidney dysfunction	3.5-5.0 g/dL	(Chien et al. 2017)

 Table 12.2
 Clinical diagnostic markers in plasma and serum

S. No.	Markers	Clinical significance	Reference range	Reference
9.	Bilirubin	Used to assess liver function. It helps to determine the cause of jaundice and diagnose conditions such as liver disease, hemolyticanemia, and blockage of the bile ducts	0.2–1.2 mg/dL	(Vítek 2017)
10.	SGPT (ALT)	Specific indicator of liver inflammation. Elevated level indicates the medical problems such as viral hepatitis, diabetes, congestive heart failure, liver damage, bile duct problems, infectious mononucleosis, or myopathy	7–56 U/Liter of serum	(Ramaty et al. 2014)
11.	SGOT (AST)	Commonly measured as a part of liver function test. Its level elevated also in diseases such as myocardial infarction, acute pancreatitis, acute hemolyticanemia, severe burns, renal disease, muscular dystrophyand trauma	5–40 U/Liter of serum	(Mavis and Alonso 2015)
12.	Urea	Serum urea concentration reflects the balance between urea production in the liver and urea elimination by the kidneys	5–20 mg/dL or 1.8–7.1 mmol urea/liter	(Bowker et al. 1992; Vanholder et al. 2018),
13.	Creatinine	Level elevated when there is a significant reduction in the glomerular filtration rate or when urine elimination is obstructed	0.6–1.2 mg/dL in adult male and 0.5–1.1 mg/dL in adult female	(Winnett et al. 2011),

Table 12.2 (continued)

S. No.	Markers	Clinical significance	Reference range	Reference
14.	Uric acid	High blood concentrations of uric acid can lead to gout and are associated with other medical conditions, including diabetes and the formation of ammonium acid urate kidney stones	2.4–6.0 mg/dL (female) and 3.4–7.0 mg/dL (male)	(Jin et al. 2012; MacFarlane and Kim 2014)
15.	Creatine kinase	Used to detect muscle dystrophy and myocardial infarction	22–198 U/L	(Blanke et al. 1984)
16.	Sodium	Sodium concentration on mortality in patients hospitalized with heart failure and hyponatremia	135–145 mEq/L	(Madan et al. 2011)
17.	Chloride	Hyperchloremia, hypochloremia is more closely associated with increased mortality and should certainly be considered by intensive care physicians	96–106 mEq/L	(Pfortmueller et al. 2018)
18.	Phosphorus	Risk factor for cardiovascular disease	2.5–4.5 mg/dL	(Gutiérrez 2013)
19.	Lactate dehydrogenase	Lactate dehydrogenase and lactate dehydrogenase isoenzyme measurements in serum in the following main clinical fields: cardiology, hepatology, haematology and oncology	140–280 U/L	(Huijgen et al. 1997)

Table 12.2 (continued)

S. No.	Markers	Clinical significance	Reference range	Reference
20.	C-Reactive protein	Acute-phase marker in tissue injury, infection and inflammation and atherosclerosis. It now has a distinct status of a disease marker in cardiovascular diseases and is well known of its clinical and pathological significance	<3.0 mg/L	(Ansar and Ghosh 2013)
21.	Thyroid hormone	Potent regulators of multiple physiological activities, including cellular metabolic rate, heart and digestive functions, muscle function, brain development, and bone maintenance	Adult: 2–10 μ U/mL Newborn: 3–18 μ U/mL	(Premachandra and Walfish 1982)
22.	Steroid hormone	Help control metabolism, inflammation, immune functions, salt and water balance, development of sexual characteristics, and the ability to withstand illness and injury	NA	(Holst et al. 2004)

Table 12.2 (continued)

lymphocytes. WBCs are an important part of the body's defense against infectious organisms and foreign substances. To defend the body adequately, a sufficient number of WBC receive a message that an infectious organism or foreign substance has invaded the body, and that they should get to where they are needed, and then kill and digest the harmful organism or substance. Adequate defense include different blood cells having different functions: some fight intruders such as bacteria, viruses, parasites or fungi themselves and render them harmless. Others produce antibodies, which specifically target foreign objects or viruses. Certain lymphocytes can also kill cancerous cells that have been produced elsewhere in the body. There are five types of WBC:

- 1. Neutrophils: each mm³ of blood contains 4000 to 11000 WBCs of which neutrophils comprise of 50–70% of the white cell count. The neutrophils are the most numerous type of the leukocytes. During tissue injury, they leave the circulatory system early in the inflammatory response to bacterial invasions.
- 2. Basophils: These are the least in number comprising <1% of total WBCs. Structurally and functionally they resemble mast cells. However, basophils originate in the marrow whereas mast cells originate from precursor cells in the connective tissue. Both basophils and mast cells contain secretory granules that store histamine and heparin, among other chemicals.
- 3. Eosinophils: These cells make 4% of total WBCs but are readily identifiable in blood smears because their cytoplasmic granules take on an orange-red to bright yellow color when stained with eosin. They have roles in hypersensitivity and allergy and their number increases during allergic conditions such as hay fever and asthma.
- 4. Monocytes: These have several functions, including bacterial removal, are active in inflammation and in repair of damaged tissues, and are the largest of the leukocytes. They are released into the blood from the marrow in an immature form with little phagocytic ability. They circulate in the blood until they find a suitable home in the tissues, where they greatly enlarge to become tissue macrophages. Their life span varies from months to years, depending on their activity.
- 5. Lymphocytes: These are the second most common WBC (approximately 20–25% of the white cell count), and are divided into two major types: B lymphocytes, which make antibodies, and T lymphocytes, which destroy cells infected with viruses. All three cell types derive from a single lymphoid stem cell. The thymus modifies the cells that become T-lymphocytes and these cells promote cell-mediated immunity. The bone marrow influences the cells that become B cells, and these cells differentiate to form plasma cells which produce circulating antibodies that comprise humoral immunity. These cells work together to defend the body against foreign substances, such as bacteria, viruses, and cancer cells.

Platelets: These are small enucleate cell fragments that circulate in blood and play a crucial role in managing vascular integrity and regulating homeostasis. Primarily they are associated with hemostasis, which is to initiate blood coagulation. Although very dynamic, they usually remain in an inactive state and get activated only when a blood vessel is damaged. Hemostasis or blood coagulation is not the sole function of platelets; rather it is employed in several multifunctional attributes monitoring the homeostasis of the body.

12.3 Clinical Markers for Health Associated with Blood

Blood is continuously exposed to plenty of metabolites and free radicals. The overall health of organisms and a wide range of disorders associated with blood can be detected by complete blood count (CBC). A CBC test measures several components

and features of the blood, including RBCs, WBCs, hemoglobin, hematocrit and platelets. Abnormal increases or decreases in cell counts may indicate that a person has some kind of ailment that calls for further evaluation. The disorders of RBC can be divided into those of decreased RBC mass (anemias) and those of increased RBC mass (erythrocytoses). The excess RBC usually create no problems but may cause blood clots in some people.

A higher than normal count of WBC leads to a condition known as leukocytosis, which is usually caused by bacterial infection, tissue damage, and inflammatory diseases (Wahed and Dasgupta 2015). A lower count, a condition known as leukopenia, is often associated with bone marrow deficiency, certain viral infection, and severe bacterial infection. Platelets are also involved in the fundamental biological process of chronic inflammation associated with disease pathology. Primarily, platelet activity is associated with the initiation of coagulation cascades. The decrease in the number of platelets in the blood is known as thrombocytopenia. An increase in platelet count in which the platelets do not work properly is the condition known as thrombocythemia.

12.4 Blood Components as Markers of Health

Blood provides the necessary biological information for the diagnosis of various pathological conditions. In these conditions, biomarkers are used as indicators of a biological factor that represents the health status.

12.4.1 Role of RBC in the Maintenance of Health

The most important and well-known function of erythrocytes is the transport of oxygen from lungs to tissues. Erythrocytes are also essential in maintaining blood pH and carbon dioxide transport. In addition, RBCs are well equipped with antioxidant systems, which essentially contribute to their function and integrity. Damage of red cell integrity, defined as hemolysis, has been shown to significantly contribute to severe pathologies, including endothelial dysfunction (Crawford et al. 2004). Erythrocytes are also involved in tissue protection and the regulation of cardiovascular homeostasis through NO metabolism and release of bioactive molecules (Cortese-Krott et al. 2012). RBCs contain numerous sources of oxygen along with high levels of iron, which in its free form acts as a catalyst of ROS production. RBCs also have limited capacity to restore damaged elements due to loss of protein expression during erythropoietic maturation (Zivot et al. 2018). The combined action of all endogenous antioxidant systems makes RBCs very resistant against oxidation as well as an efficient systemic redox buffering system. These properties help to keep RBC healthy and well functioned. The malfunction of antioxidant defense or conditions of increased oxidant production have severe consequences for RBCs at

subcellular level. This includes the degradation of Hb and other proteins, disturbance of ionic homeostasis, hindered RBC deformation, interference with erythropoiesis and enhanced exposure of phosphatidylserine (Mohanty et al. 2014). Furthermore, RBC membranes consist of high concentration of PUFA that make them susceptible to lipid peroxidation leading to loss of membrane integrity and decreased activity of enzymes associated with erythrocyte membrane (Kaestner and Minetti 2017).

12.4.2 Role of WBC in Maintenance of Health

Normal WBC count is important for determining health status as it helps to understand what is going on inside the body during a variety of health situations. Besides acting as an indicator of current health status, white blood cell count has also been suggested as a predictive and prognostic marker for a number of chronic diseases (Madjid and Fatemi 2013; Wang et al. 2018). As the WBC count goes up, it could mean inflammation somewhere in the body. The role of white blood cell count in pathogenesis of various diseases such as diabetes, cardiovascular disease, and obesity-related disorders has been reported (Twig et al. 2012; Veronelli et al. 2004). Recent studies reveal that higher WBC contributes to atherosclerotic progression and impaired fasting glucose. Most white blood cell disorders are either a type of cancer or proliferative disorder.

12.4.3 Role of Platelets in the Maintenance of Health

Platelet activity is associated with coagulation cascades. Blood vessel damage causes the sub-endothelial surface to be the primary target site for platelet action. Proaggregatory stimuli (platelet agonists) promote the action of platelet adhesion to the sub-endothelial surfaces. During this process, platelets change their shape, release their granule contents, and gradually form aggregates by adhering with each other (Vinik et al. 2001). Thus, platelets primarily function to minimize blood loss. Platelets are also involved in the fundamental biological process of chronic inflammation associated with disease pathology (Ghoshal and Bhattacharyya 2014). Platelets are actively involved in secretion of molecules like GPIIb, IIIa, fibrinogen, catecholamine, serotonin, calcium, ATP, ADP, which are involved in aggregation. Differential expressions of surface receptors like CD36, CD41, CD61 have also been measured in several diseases. Platelet activation and dysfunction have been implicated in diabetes, renal diseases, tumorigenesis, Alzheimer's, and CVD.

12.5 RBC and Aging

The aging process of RBC is considered an issue of special scientific and clinical interest. It represents a total of unidirectional, time-dependent but not-necessarily linear series of molecular events that finally lead to cell clearance (Aminoff et al. 1992). Under normal circumstances, human RBCs live approximately 120 ± 4 days in blood circulation, implying the existence of tightly regulated molecular mechanism(s), responsible for the programming of the lifespan and the nonrandom removal of senescent RBCs (Badior and Casey 2018; Franco 2009; Walsh et al. 2002). RBC is a favorite subject of investigation of cellular senescence (Clark 1988; Singh et al. 2016b). Although RBCs lose their subcellular organelles, they maintain a plethora of cellular functions like anaerobic glycolysis, the pentose phosphate shunt, cellular signaling and possibly even a variant of programmed cell death called eryptosis (Lang et al. 2005; Minetti and Low 1997). RBCs are maintained in a functional state until the very end of their life and go back to the bone marrow to die (Bernhardt and Ellory 2003). RBCs experience a range of continuous metabolic and physical damages as they age, such as membrane vesiculation, haemoglobin (Hb) modifications and progressive failure of both, cellular homeostasis and antioxidant defenses (Piomelli and Seaman 1993; Willekens et al. 2003). The increase in RBCs density, the nonenzymatic glycation of Hb and the deamidation of protein 4.1b to 4.1a have been widely used as sensitive RBC age markers (Bosch et al. 1992; Lutz et al. 1992; Mueller et al. 1987).

12.6 Markers of Oxidative Stress in Erythrocytes and Plasma

RBC and their membrane have always been important media for study due to the important role they play in various physiological and metabolic processes. Erythrocytes have been increasingly studied as they are the easiest available human cell type. Throughout its entire life, the organism is confronted with oxidative stress due to the production of ROS and reactive nitrogen species (RNS). ROS are normally generated as by-products of oxygen metabolism and generate free radical chain reaction. High levels of oxidative stress have been linked with the increased incidence of a variety of health issues. At moderate concentration ROS play several beneficial physiological roles in cell signaling and induce mitogenic response (Genestra 2007). They are needed to synthesize some cellular structures and to be used by the host defense system to fight pathogens.

Various markers of oxidative stress in erythrocytes and plasma are listed in Table 12.3. One of the most putative markers of oxidative stress is the measure of total antioxidant status. Total antioxidant status is measured in terms of 1,1-diphenyl-2-picrylhydrazyl free radical (DPPH) assay, 2,2-azobis-3-ethylbenzthiazoline-6-sulfonic acid (ABTS) assay and ferric reducing ability of plasma (FRAP) assay.

S.No.	Biomarkers	Clinical significance	Reference
1.	Protein Carbonyls	Marker of plasma and membrane protein oxidation. Formed due to the protein-pro cross linking and oxidation of protein backbone	(Sangeetha et al. 2005; Singh et al. 2016a)
2.	Advanced oxidation of protein products	Dityrosine-containing cross-linked protein products Plasma level of AOPP elevated during various diseases	(Garg et al. 2017)
3.	Total thiols	Product of S-thiolation reaction, act as an antioxidant. Alteration in thiol-disulphide redox status has been observed in specific groups of diseases such as cardiovascular, cancer, and neurodegenerative	(Oliveira and Laurindo 2018; Singh et al. 2019)
4.	Reduced Glutathione	Most abundant non protein thiol helps in maintaining intracellular redox environment. Erythrocyte GSH level get reduced during oxidative stress	(Singh et al. 2017, 2018)
5.	Plasma Membrane Redox system	Oxidoreductase system, tranfers electron from intracellular donor to extracellular acceptor and provide antioxidant protection against induced oxidative stress	(Adlard and Bush 2011; Hyun et al. 2006; Rodríguez-Aguilera et al. n.d.; Singh et al. 2017)
6.	Malondialdehyde	Byproduct of lipid peroxidation, play an important role in the pathogenesis and progression of several diseases. Affects the variety of membrane related functions, alteration in membrane fluidity, permeability and loss of function	(Garg et al. 2017; Singh et al. 2016a)

 Table 12.3
 Oxidative stress biomarkers in erythrocytes and plasma

S.No.	Biomarkers	Clinical significance	Reference
7.	Lipid hydroperoxides	Formed from lipid autooxidation and photooxidation. Plama and membrane level of LHP get elevated during diseases associated with oxidative stress	(Gönenç et al. 2006; Peña-Bautista et al. 2019; Singh et al. 2018)
8.	Acetylcholine esterase	Maintains the erythrocyte membrane potential. Marker of erythrocyte aging and RBC membrane integrity	(Suhail and Rizvi 1989)
9.	Sodium potassium ATPase (Na ⁺ /K ⁺ -ATPase) and Plasma membrane calcium ATPase (PMCA)	Maintains the intracellular ionic homeostasis and electrochemical gradients across the membrane. The activity of NKApump is considerably impaired during the alteration in homeostasis mediated by oxidative stress	(Marchesi 2008)
10.	Sodium Hydrogen exchanger (NHE)	NHE activity contributes to overall cell damage. Play vital housekeeping roles in the maintenanceof intracellular ionic homeostasis	(Dubyak 2004; Singh et al. 2016b; Várady et al. 2015)

Table 12.3 (continued)

There is a strong correlation between antioxidant capacity and oxidative damage during aging (Pandey and Rizvi 2010).

Under physiological aerobic conditions, erythrocytes are continuously exposed to oxidants derived from endogenous as well as exogenous sources. Exposure of erythrocytes to physiological oxidative stress leads to lipid peroxidation that could change the membrane composition, inducing conformational changes and protein cross-linking in membrane proteins and, these changes may lead to abnormal cell morphology and hemolysis (Garg et al. 2019; Berzosa et al. 2011; Ciccoli et al. 2013; Freikman et al. 2011; Pytel et al. 2013). Lipid peroxidation is usually measured in terms of malondialdehyde (MDA) and lipid hydroperoxides (LHP). RBCs have also been reported to be associated with a number of biomarkers for age and senescence, these include reduced glutathione (GSH), the plasma membrane redox system (PMRS), rate of cysteine influx and antioxidant enzymatic activity. Due to these robust and reproducible age biomarkers, erythrocytes have become a suitable model for aging research (Kumar and Rizvi 2014; Rizvi et al. 2006, 2009; Rizvi and Maurya 2008). PMRS is an oxidoreductase system that transfers electrons from intracelular donors to extracellular acceptors such as ascorbate free radical and convert it

into ascorbate. Erythrocyte PMRS provides antioxidant protection against oxidative stress. RBCs possess effective enzymatic antioxidant systems that neutralize the ROS into non/less reactive species. Superoxide dismutase (SOD), glutathione peroxidase (GPx), glutathione reductase (GR) and catalase (CAT) are some of the endogenous enzymatic defense systems in all aerobic cells which get affected by advancing age (Finkel and Holbrook 2000; Wojciech et al. 2010). They give protection by directly scavenging superoxide radicals and hydrogen peroxide (Pandey and Rizvi 2010; Scandalios 2005). These antioxidants counteract oxidative stress and mitigate its effects on individuals' health as they are free from important side effects. On the other hand, some prooxidants can be as well useful to human health particularly in cancer (Pizzino et al. 2017). Thus oxidative stress, although being one of the major harms to individual's wellness and health, can also be exploited as a treatment monitoring tool when finely tuned inside human organism.

12.7 Conclusion

In this chapter, we have reviewed the basic components of blood, including the different factors that can be used to determine the health status of the blood. Various clinical and oxidative stress biomarkers in the blood help to understand the dynamic homeostasis at the physiological level. Furthermore, blood analysis provides an array of the minimal-invasive procedures that can be used to assess important clinical biomarkers of health and deviations from it.

Acknowledgements SIR is a recipient of DST-SERB grant, Government of India. Geetika Garg is a recipient of SERB-NPDF fellowship from DST, India. Sandeep Singh acknowledges a Senior Research Fellowship from Indian Council of Medical Research, India.

Conflict of interest Authors have no conflict of interest.

References

- Adlard PA, Bush AI (2011) The plasma membrane redox system in Alzheimer's disease. Exp Neurol 228:9–14. https://doi.org/10.1016/j.expneurol.2010.12.009
- Al-Otaiby MA, Al-Amri HS, Al-Moghairi AM (2011) The clinical significance of cardiac troponins in medical practice. J Saudi Heart Assoc 23:3–11 https://doi.org/10.1016/j.jsha.2010.10.001
- Aminoff D, Rolfes-Curl A, Supina E (1992) Molecular biomarkers of aging: the red cell as a model. Arch Gerontol Geriatr 15 Suppl 1:7–15. https://doi.org/10.1016/s0167-4943(05)80002-0
- Ansar W, Ghosh S (2013) C-reactive protein and the biology of disease. Immunol Res 56:131–142. https://doi.org/10.1007/s12026-013-8384-0
- Badior KE, Casey JR (2018) Molecular mechanism for the red blood cell senescence clock. IUBMB Life 70:32–40. https://doi.org/10.1002/iub.1703
- Bernhardt I, Ellory JC (Eds) (2003) Red cell membrane transport in health and disease. Springer, Berlin Heidelberg. https://doi.org/10.1007/978-3-662-05181-8

- Berzosa C, Gómez-Trullén EM, Piedrafita E, Cebrián I, Martínez-Ballarín E, Miana-Mena FJ, Fuentes-Broto L, García JJ (2011) Erythrocyte membrane fluidity and indices of plasmatic oxidative damage after acute physical exercise in humans. Eur J Appl Physiol 111:1127–1133. https:// doi.org/10.1007/s00421-010-1738-6
- Blanke H, von Hardenberg D, Cohen M, Kaiser H, Karsch KR, Holt J, Smith H, Rentrop P (1984) Patterns of creatine kinase release during acute myocardial infarction after nonsurgical reperfusion: comparison with conventional treatment and correlation with infarct size. J Am Coll Cardiol 3:675–680. https://doi.org/10.1016/s0735-1097(84)80242-9
- Bosch FH, Werre JM, Roerdinkholder-Stoelwinder B, Huls TH, Willekens FL, Halie MR (1992) Characteristics of red blood cell populations fractionated with a combination of counterflow centrifugation and Percoll separation. Blood 79:254–260
- Bowker LK, Briggs RS, Gallagher PJ, Robertson DR (1992) Raised blood urea in the elderly: a clinical and pathological study. Postgrad Med J 68:174–179. https://doi.org/10.1136/pgmj.68. 797.174
- Chien S-C, Chen C-Y, Lin C-F, Yeh H-I (2017) Critical appraisal of the role of serum albumin in cardiovascular disease. Biomark Res 5:31. https://doi.org/10.1186/s40364-017-0111-x
- Ciccoli L, De Felice C, Paccagnini E, Leoncini S, Pecorelli A, Signorini C, Belmonte G, Guerranti R, Cortelazzo A, Gentile M, Zollo G, Durand T, Valacchi G, Rossi M, Hayek J (2013) Erythrocyte shape abnormalities, membrane oxidative damage, and β-actin alterations: an unrecognized triad in classical autism. Mediators Inflamm 2013:432616. https://doi.org/10.1155/2013/432616
- Clark MR (1988) Senescence of red blood cells: progress and problems. Physiol Rev 68:503-554
- Cortese-Krott MM, Rodriguez-Mateos A, Sansone R, Kuhnle GGC, Thasian-Sivarajah S, Krenz T, Horn P, Krisp C, Wolters D, Heiß C, Kröncke K-D, Hogg N, Feelisch M, Kelm M (2012) Human red blood cells at work: identification and visualization of erythrocytic eNOS activity in health and disease. Blood 120:4229–4237. https://doi.org/10.1182/blood-2012-07-442277
- Crawford JH, Chacko BK, Kevil CG, Patel RP (2004) The red blood cell and vascular function in health and disease. Antioxid. Redox Signal. 6:992–999. https://doi.org/10.1089/ars.2004.6.992
- Després JP, Lemieux I, Dagenais GR, Cantin B, Lamarche B (2000) HDL-cholesterol as a marker of coronary heart disease risk: the Québec cardiovascular study. Atherosclerosis 153:263–272. https://doi.org/10.1016/s0021-9150(00)00603-1
- Diez-Silva M, Dao M, Han J, Lim C-T, Suresh S (2010) Shape and biomechanical characteristics of human red blood cells in health and disease. MRS Bull 35:382–388. https://doi.org/10.1557/ mrs2010.571
- Dubyak GR (2004) Ion homeostasis, channels, and transporters: an update on cellular mechanisms. Adv Physiol Educ 28:143–154. https://doi.org/10.1152/advan.00046.2004
- Duckworth WC (2001) Hyperglycemia and cardiovascular disease. Curr Atheroscler Rep 3:383–391. https://doi.org/10.1007/s11883-001-0076-x
- Feher J (2012) White Blood Cells and inflammation. In: Quantitative human physiology. Elsevier, pp 507–515. https://doi.org/10.1016/B978-0-12-800883-6.00046-X
- Finkel T, Holbrook NJ (2000) Oxidants, oxidative stress and the biology of ageing. Nature 408:239–247. https://doi.org/10.1038/35041687
- Franco RS (2009) The measurement and importance of red cell survival. Am J Hematol 84:109–114. https://doi.org/10.1002/ajh.21298
- Freikman I, Ringel I, Fibach E (2011) Oxidative stress-induced membrane shedding from RBCs is Ca flux-mediated and affects membrane lipid composition. J Membr Biol 240:73–82. https://doi. org/10.1007/s00232-011-9345-y
- Garg G, Singh S, Singh AK, Rizvi SI (2019) Erythrocyte as a cellular model of aging research. In: Reference module in biomedical sciences. Elsevier, p. B9780128012383113000. https://doi.org/ 10.1016/B978-0-12-801238-3.11401-1
- Garg G, Singh S, Singh AK, Rizvi SI (2017) Metformin alleviates altered Erythrocyte Redox status during aging in Rats. Rejuvenation Res 20:15–24. https://doi.org/10.1089/rej.2016.1826
- Genestra M (2007) Oxyl radicals, redox-sensitive signalling cascades and antioxidants. Cell Signal 19:1807–1819. https://doi.org/10.1016/j.cellsig.2007.04.009

- Ghoshal K, Bhattacharyya M (2014) Overview of platelet physiology: its hemostatic and nonhemostatic role in disease pathogenesis. Sci World J 2014:1–16. https://doi.org/10.1155/2014/ 781857
- Gönenç A, Erten D, Aslan S, Akinci M, Simşek B, Torun M (2006) Lipid peroxidation and antioxidant status in blood and tissue of malignant breast tumor and benign breast disease. Cell Biol Int 30:376–380. https://doi.org/10.1016/j.cellbi.2006.02.005
- Gutiérrez OM (2013) The connection between dietary phosphorus, cardiovascular disease, and mortality: where we stand and what we need to know. Adv Nutr 4:723–729. https://doi.org/10. 3945/an.113.004812
- Hillman RS, Finch CA (1996) Red cell manual. In: Davis FA, 7 edn. Philadelphia
- Holst JP, Soldin OP, Guo T, Soldin SJ (2004) Steroid hormones: relevance and measurement in the clinical laboratory. Clin Lab Med 24:105–118. https://doi.org/10.1016/j.cll.2004.01.004
- Huijgen HJ, Sanders GT, Koster RW, Vreeken J, Bossuyt PM (1997) The clinical value of lactate dehydrogenase in serum: a quantitative review. Eur J Clin Chem Clin Biochem J Forum Eur Clin Chem Soc 35:569–579
- Hyun D-H, Hernandez JO, Mattson MP, de Cabo R (2006) The plasma membrane redox system in aging. Ageing Res Rev 5:209–220. https://doi.org/10.1016/j.arr.2006.03.005
- Ivanova EA, Myasoedova VA, Melnichenko AA, Grechko AV, Orekhov AN (2017) Small dense lowdensity Lipoprotein as biomarker for Atherosclerotic diseases. Oxid Med Cell Longev 2017:1–10. https://doi.org/10.1155/2017/1273042
- Jin M, Yang F, Yang I, Yin Y, Luo JJ, Wang H, Yang X-F (2012) Uric acid, hyperuricemia and vascular diseases. Front Biosci Landmark 17:656–669. https://doi.org/10.2741/3950
- Kaestner L, Minetti G (2017) The potential of erythrocytes as cellular aging models. Cell Death Differ 24:1475–1477. https://doi.org/10.1038/cdd.2017.100
- Kalra S, Mukherjee JJ, Venkataraman S, Bantwal G, Shaikh S, Saboo B, Das AK, Ramachandran A (2013) Hypoglycemia: the neglected complication. Indian J Endocrinol Metab 17:819–834. https://doi.org/10.4103/2230-8210.117219
- Krisko A, Radman M (2019) Protein damage, ageing and age-related diseases. Open Biol 9:180249. https://doi.org/10.1098/rsob.180249
- Kuhn V, Diederich L, Keller TCS, Kramer CM, Lückstädt W, Panknin C, Suvorava T, Isakson BE, Kelm M, Cortese-Krott MM (2017) Red blood cell function and dysfunction: Redox regulation, Nitric Oxide metabolism. Anemia Antioxid Redox Signal 26:718–742. https://doi.org/10.1089/ ars.2016.6954
- Kumar D, Rizvi SI (2014) A critical period in lifespan of male rats coincides with increased oxidative stress. Arch Gerontol Geriatr 58:427–433. https://doi.org/10.1016/j.archger.2013.11.006
- Lang KS, Lang PA, Bauer C, Duranton C, Wieder T, Huber SM, Lang F (2005) Mechanisms of suicidal erythrocyte death. Cell Physiol Biochem Int J Exp Cell Physiol Biochem Pharmacol 15:195–202. https://doi.org/10.1159/000086406
- Lin C-J, Lai C-K, Kao M-C, Wu L-T, Lo U-G, Lin L-C, Chen Y-A, Lin H, Hsieh J-T, Lai C-H, Lin C-D (2015) Impact of cholesterol on disease progression. BioMedicine 5:7. https://doi.org/ 10.7603/s40681-015-0007-8
- Lutz HU, Stammler P, Fasler S, Ingold M, Fehr J (1992) Density separation of human red blood cells on self forming Percoll gradients: correlation with cell age. Biochim Biophys Acta 1116:1–10. https://doi.org/10.1016/0304-4165(92)90120-j
- MacFarlane LA, Kim SC (2014) Gout: a review of nonmodifiable and modifiable risk factors. Rheum Dis Clin North Am 40:581–604. https://doi.org/10.1016/j.rdc.2014.07.002
- Madan VD, Novak E, Rich MW (2011) Impact of change in serum sodium concentration on mortality in patients hospitalized with heart failure and hyponatremia. Circ Heart Fail 4:637–643. https://doi.org/10.1161/CIRCHEARTFAILURE.111.961011
- Madjid M, Fatemi O (2013) Components of the complete blood count as risk predictors for coronary heart disease: in-depth review and update. Tex Heart Inst J. 40:17–29

- Marchesi VT (2008) The relevance of research on red cell membranes to the understanding of complex human disease: a personal perspective. Annu Rev Pathol 3:1–9. https://doi.org/10.1146/ annurev.pathmechdis.3.121806.154321
- Mavis AM, Alonso EM (2015) Liver disease in the adolescent. Clin Liver Dis 19:171–185. https:// doi.org/10.1016/j.cld.2014.09.010
- Minetti G, Low PS (1997) Erythrocyte signal transduction pathways and their possible functions. Curr Opin Hematol 4:116–121. https://doi.org/10.1097/00062752-199704020-00007
- Mohanty JG, Nagababu E, Rifkind JM (2014) Red blood cell oxidative stress impairs oxygen delivery and induces red blood cell aging. Front Physiol 5:84. https://doi.org/10.3389/fphys. 2014.00084
- Mueller TJ, Jackson CW, Dockter ME, Morrison M (1987) Membrane skeletal alterations during in vivo mouse red cell aging. Increase in the band 4.1a:4.1b ratio. J Clin Invest 79:492–499. https://doi.org/10.1172/JCI112839
- Oliveira PVS, Laurindo FRM (2018) Implications of plasma thiol redox in disease. Clin Sci Lond Engl 1979(132):1257–1280. https://doi.org/10.1042/CS20180157
- Pandey KB, Rizvi SI (2010) Markers of Oxidative stress in Erythrocytes and plasma during aging in humans. Oxid Med Cell Longev 3:2–12. https://doi.org/10.4161/oxim.3.1.10476
- Peña-Bautista C, Baquero M, Vento M, Cháfer-Pericás C (2019) Free radicals in Alzheimer's disease: Lipid peroxidation biomarkers. Clin Chim Acta Int J Clin Chem 491:85–90. https://doi. org/10.1016/j.cca.2019.01.021
- Pfortmueller CA, Uehlinger D, von Haehling S, Schefold JC (2018) Serum chloride levels in critical illness-the hidden story. Intensive Care Med Exp 6:10. https://doi.org/10.1186/s40635-018-0174-5
- Piomelli S, Seaman C (1993) Mechanism of red blood cell aging: relationship of cell density and cell age. Am J Hematol 42:46–52. https://doi.org/10.1002/ajh.2830420110
- Pizzino G, Irrera N, Cucinotta M, Pallio G, Mannino F, Arcoraci V, Squadrito F, Altavilla D, Bitto A (2017) Oxidative Stress: Harms and Benefits for Human Health. Oxid Med Cell Longev 2017:8416763. https://doi.org/10.1155/2017/8416763
- Premachandra BN, Walfish PG (1982) Effects and clinical significance of exogenous thyroxine therapy in patients with circulating thyroid hormone autoantibodies. Am J Clin Pathol 78:63–68. https://doi.org/10.1093/ajcp/78.1.63
- Pytel E, Olszewska-Banaszczyk M, Koter-Michalak M, Broncel M (2013) Increased oxidative stress and decreased membrane fluidity in erythrocytes of CAD patients. Biochem Cell Biol Biochim Biol Cell 91:315–318. https://doi.org/10.1139/bcb-2013-0027
- Rodríguez-Aguilera JC, López-Lluch G, Santos-Ocaña C, Villalba JM, Gómez-Díaz C, Navas P (n.d.) Plasma membrane redox system protects cells against oxidative stress 4
- Ramaty E, Maor E, Peltz-Sinvani N, Brom A, Grinfeld A, Kivity S, Segev S, Sidi Y, Kessler T, Sela BA, Segal G (2014) Low ALT blood levels predict long-term all-cause mortality among adults. A historical prospective cohort study. Eur J Intern Med 25:919–921. https://doi.org/10.1016/j.ejim. 2014.10.019
- Rizvi SI, Jha R, Maurya PK (2006) Erythrocyte plasma membrane redox system in human aging. Rejuvenation Res 9:470–474. https://doi.org/10.1089/rej.2006.9.470
- Rizvi SI, Maurya PK (2008) L-Cysteine Influx in Erythrocytes as a function of human age. Rejuvenation Res 11:661–665. https://doi.org/10.1089/rej.2007.0652
- Rizvi SI, Pandey KB, Jha R, Maurya PK (2009) Ascorbate recycling by Erythrocytes during aging in humans. Rejuvenation Res 12:3–6. https://doi.org/10.1089/rej.2008.0787
- Suhail M, Rizvi SI (1989) Erythrocyte membrane acetylcholinesterase in type 1 (insulin-dependent) diabetes mellitus. Biochem. J 259:897–899
- Sangeetha P, Balu M, Haripriya D, Panneerselvam C (2005) Age associated changes in erythrocyte membrane surface charge: Modulatory role of grape seed proanthocyanidins. Exp Gerontol 40:820–828. https://doi.org/10.1016/j.exger.2005.07.008

- Scandalios JG (2005) Oxidative stress: molecular perception and transduction of signals triggering antioxidant gene defenses. Braz J Med Biol Res Rev Bras Pesqui Medicas E Biol 38:995–1014. https://doi.org//S0100-879X2005000700003
- Singh AK, Singh S, Garg G, Rizvi SI (2016a) Rapamycin alleviates oxidative stress-induced damage in rat erythrocytes. Biochem Cell Biol Biochim Biol Cell 94:471–479. https://doi.org/10.1139/ bcb-2016-0048
- Singh S, Pandey KB, Rizvi SI (2016b) Erythrocyte senescence and membrane transporters in young and old rats. Arch Physiol Biochem 1–7. https://doi.org/10.1080/13813455.2016.1190761
- Singh AK, Garg G, Singh S, Rizvi SI (2017) Synergistic effect of Rapamycin and Metformin against age-dependent Oxidative Stress in Rat Erythrocytes. Rejuvenation Res 20:420–429. https://doi. org/10.1089/rej.2017.1916
- Singh S, Garg G, Singh AK, Tripathi SS, Rizvi SI (2019) Fisetin, a potential caloric restriction mimetic, modulates ionic homeostasis in senescence induced and naturally aged rats. Arch Physiol Biochem 1–8. https://doi.org/10.1080/13813455.2019.1662452
- Singh S, Singh AK, Garg G, Rizvi SI (2018) Fisetin as a caloric restriction mimetic protects rat brain against aging induced oxidative stress, apoptosis and neurodegeneration. Life Sci 193:171–179. https://doi.org/10.1016/j.lfs.2017.11.004
- Teixeira RS, Arriaga MB, Terse-Ramos R, Ferreira TA, Machado VR, Rissatto-Lago MR, Silveira-Mattos PS, Boa-Sorte N, Ladeia AMT, Andrade BB (2019) Higher values of triglycerides: HDLcholesterol ratio hallmark disease severity in children and adolescents with sickle cell anemia. Braz J Med Biol Res 52. https://doi.org/10.1590/1414-431x20198833
- Twig G, Afek A, Shamiss A, Derazne E, Tzur D, Gordon B, Tirosh A (2012) White blood cell count and the risk for coronary artery disease in young adults. PLoS ONE 7:e47183. https://doi.org/10. 1371/journal.pone.0047183
- Vanholder R, Gryp T, Glorieux G (2018) Urea and chronic kidney disease: the comeback of the century? (in uraemia research). Nephrol Dial Transplant 33:4–12. https://doi.org/10.1093/ndt/gfx039
- Várady G, Szabó E, Fehér Á, Németh A, Zámbó B, Pákáski M, Janka Z, Sarkadi B (2015) Alterations of membrane protein expression in red blood cells of Alzheimer's disease patients. Alzheimers Dement Diagn Assess Dis Monit 1:334–338. https://doi.org/10.1016/j.dadm.2015.06.007
- Veronelli A, Laneri M, Ranieri R, Koprivec D, Vardaro D, Paganelli M, Folli F, Pontiroli AE (2004) White blood cells in obesity and diabetes: effects of weight loss and normalization of glucose metabolism. Diabetes Care 27:2501–2502. https://doi.org/10.2337/diacare.27.10.2501
- Vinik AI, Erbas T, Park TS, Nolan R, Pittenger GL (2001) Platelet dysfunction in type 2 diabetes. Diabetes Care 24:1476–1485. https://doi.org/10.2337/diacare.24.8.1476
- Vítek L (2017) Bilirubin and atherosclerotic diseases. Physiol Res 66:S11-S20
- Wahed A, Dasgupta A (2015) Benign white blood cell and platelet disorders. In: Hematology and coagulation. Elsevier, pp 81–92. https://doi.org/10.1016/B978-0-12-800241-4.00005-X
- Walsh M, Lutz RJ, Cotter TG, O'Connor R (2002) Erythrocyte survival is promoted by plasma and suppressed by a Bak-derived BH3 peptide that interacts with membrane-associated Bcl-X(L). Blood 99:3439–3448. https://doi.org/10.1182/blood.v99.9.3439
- Wang Y-L, Ge X-X, Wang Y, Xu M-D, Gong F-R, Tao M, Wang W-J, Shou L-M, Chen K, Wu M-Y, Li W (2018) The values of applying classification and counts of white blood cells to the prognostic evaluation of resectable gastric cancers. BMC Gastroenterol 18:99. https://doi.org/10. 1186/s12876-018-0812-0
- Willekens FLA, Roerdinkholder-Stoelwinder B, Groenen-Döpp YAM, Bos HJ, Bosman GJCGM, van den Bos AG, Verkleij AJ, Werre JM (2003) Hemoglobin loss from erythrocytes in vivo results from spleen-facilitated vesiculation. Blood 101:747–751. https://doi.org/10.1182/blood-2002-02-0500
- Winnett G, Cranfield L, Almond M (2011) Apparent renal disease due to elevated creatinine levels associated with the use of boldenone. Nephrol Dial Transplant 26:744–747. https://doi.org/10. 1093/ndt/gfq663

Wojciech Ł, Ewa Z, Elżbieta S (2010) Influence of green tea on erythrocytes antioxidant status of different age rats intoxicated with ethanol. Phytother Res 24:424–428. https://doi.org/10.1002/ ptr.2986

Zivot A, Lipton JM, Narla A, Blanc L (2018) Erythropoiesis: insights into pathophysiology and treatments in 2017. Mol Med 24:11. https://doi.org/10.1186/s10020-018-0011-z