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DEPARTMENT OF BIOMEDICAL ENGINEERING

QUANTITATIVE PHYSIOLOGY

Laboratory workshop

Workshop *on discipline for students*
of specialties 163 "Biomedical Engineering"

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МІНІСТЕРСТВО ОСВІТИ І НАУКИ УКРАЇНИ
НАЦІОНАЛЬНИЙ ТЕХНІЧНИЙ УНІВЕРСИТЕТ УКРАЇНИ
«КИЇВСЬКИЙ ПОЛІТЕХНІЧНИЙ ІНСТИТУТ
імені Ігоря СІКОРСЬКОГО»

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QUANTITATIVE PHYSIOLOGY

LABORATORY WORKSHOP

Complier: *Svitlana Vovianko, PhD,* Associate Professor of the Department of Biomedical Engineering of Igor Sikorsky Kyiv Polytechnic Institute,
Ganna Ovcharenko, Senior Lecturer of the Department of Biomedical Engineering of Igor Sikorsky Kyiv Polytechnic Institute

Reviewer: *Vasily Kozyar, PhD,* Associate Professor of the Department of Biomedical Engineering of Igor Sikorsky Kyiv Polytechnic Institute
Maryna Sychyk, PhD, Senior Engineer of the State Institution “M.M. Amosov National Institute of Cardiovascular Surgery Ukraine NAMS of Ukraine”

Editor-in-Chief: *Olena Bespalova, PhD,* Associate Professor of the Department of Translation Medical Bioengineering of Igor Sikorsky Kyiv Polytechnic Institute

Laboratory workshop on the subject "Quantitative Physiology" contains laboratory work performed using modern diagnostic methods, which will use methods and tools for quantitative assessment of the functioning of physiological systems in practical engineering. The educational publication is recommended for higher educational institutions of Ukraine with the teaching of specialty 163 "Biomedical Engineering".

CONTENTS

PREFACE

The purpose of practical classes and laboratory work in the course of "Quantitative Physiology", performed with the use of modern diagnostic methods with appropriate equipment and software and Microsoft Windows to process experimental data, is to form students' ability to use methods and tools of quantitative assessment of physiological systems functioning in practical engineering activities. When performing laboratory work, students receive direct confirmation of theoretical material, which is taught in lectures, and acquire skills of research and work with equipment.

The peculiarity of this works is that most of the laboratory work offered in it is performed on humans. In this case, students themselves become researchers and subjects. When conducting research, students should take into account that the necessary prerequisite for obtaining clear and identical results are the same conditions.

As a result of laboratory work, the student must master the practical skills of measuring and calculating physiological parameters, namely: record the electrocardiogram in three standard leads and analyze it; understand the causes of interference in electrophysiological studies and how to avoid them; determine the parameters of systemic hemodynamics and pumping function of the heart by indicators of blood pressure and heart rate; use functional tests to study the function of the cardiovascular and respiratory systems; determine the indicators of oxygen transport function of the blood; measure the quantitative characteristics of blood cells. Specialized AxioVision software is used for image analysis of biological objects.

The results of research and measurements should be documented and presented to the teacher at the end of the lesson.

The laboratory work report should contain:

- title page with the name of the course, lab name, student surname and first name;
- purpose of the work,
- lab performance with all the experimental conditions;
- experimental data;
- calculations and data analysis;
- answers to the control questions.

1 Physiological Measurements Instrumentation

A preferred way to understand the status of a particular physiological system is to study the intrinsic signals, generated by the body itself, without interaction with sampling methods. Most of such methods are well known and as old as the medical science itself. The electrophysiological methods to investigate the heart (electrocardiogram [ECG]), brain (electroencephalogram [EEG]), and muscles (electromyogram [EMG]) are all familiar. Pressure recordings from the vascular system have been used since the beginning of the eighteenth century. Sounds from the airways, lungs, fetus, and the heart have been used as diagnostic tools long before the invention of the stethoscope in the early nineteenth century. Body heat production and temperature changes can be used to understand infectious diseases or inflammatory disorders. The beauty of intrinsic energy analysis is that these methods do not interfere with the physiological systems under study, nor do they cause the patient discomfort.

1.1 Generalized Medical Instrumentation System

Every instrumentation system has at least some of the functional components shown in Figure 1.1.

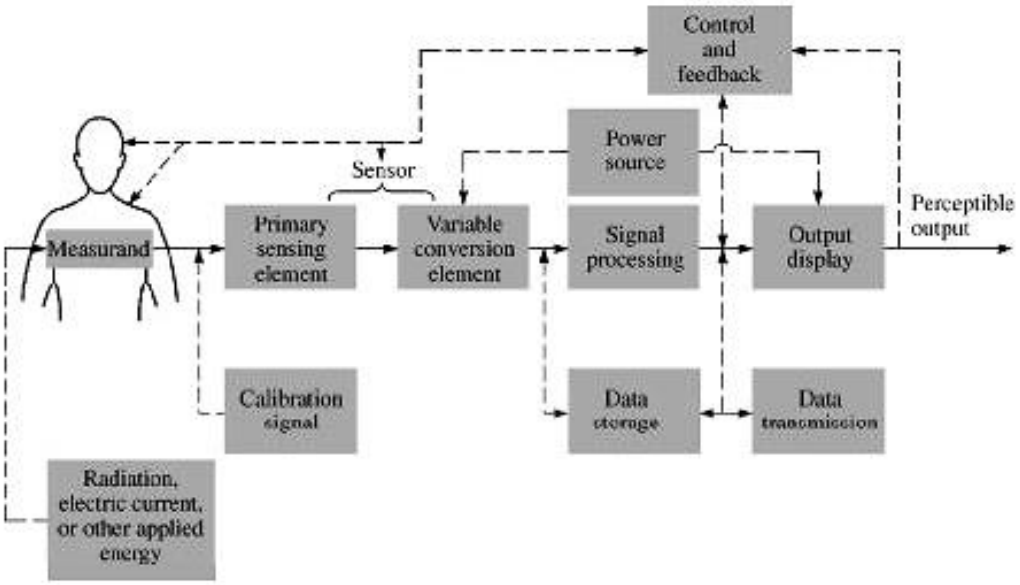


Figure 1.1 – Generalized instrumentation system

(W.Olson, 2004)

The primary flow of information is from left to right. Elements and relationships depicted by dashed lines are not essential. The major difference between this system of medical instrumentation and conventional instrumentation systems is that the source of the signals is living tissue or energy applied to living tissue.

The sensor converts energy or information from the measurand to another form (usually electric). This signal is then processed and displayed so that humans can perceive the information. Elements and connections shown by dashed tines are optional for some applications.

1.2 Functional Components of the Physiological Measurements Instrumentation Systems

Measurand. The physical quantity, property, or condition that the system measures is called the measurand. The accessibility of the measurand is important because it may be internal (blood pressure), it may be on the body surface (electrocardiogram potential), it may emanate from the body (infrared radiation), or it may be derived from a tissue sample (such as blood or a biopsy) that is removed from the body. Most medically important measurands can be grouped in the following categories: biopotential, pressure, flow, dimensions (imaging), displacement (velocity, acceleration, and force), impedance, temperature, and chemical concentrations. The measurand may be localized to a specific organ or anatomical structure.

Electrodes.

Electrodes are used for measuring the ECG, EEG, EMG and intracellular potentials. In order to measure and record potentials and. hence, currents in the body, it is necessary to provide some interface between the body and the electronic measuring apparatus. Biopotential electrodes carry out this interface function. In any practical measurement of potentials, current flows in the measuring circuit for at least a fraction of the period of time over which the measurement is made. Ideally this current should be very small. However, in practical situations, it is never zero. Biopotential electrodes must therefore have the capability of conducting a current across the interface between the body and the electronic

measuring circuit. Our first impression is that this is a rather simple function to achieve and that biopotential electrodes should be relatively straightforward. But when we consider the problem in more detail, we see that the electrode actually carries out a transducing function, because in the body current is carried by ions, whereas in the electrode and its lead wire it is carried by electrons. Thus the electrode must serve as a transducer to change an ionic current into an electronic current.

There are two classifications of ideal electrodes: polarizable and nonpolarizable. With a polarizable electrode, the behavior of the electrode is similar to that of a capacitor. Current flowing through the electrode will charge the electrode causing "polarization." The current responsible for charging (or discharging) the electrode is known as "capacitive current." In a polarizable electrode, there is no "faradaic" current, which is the current due to chemical reaction within the electrode, and no charge actually passes through the electrode– electrolyte interface. The potential of nonpolarizable electrodes, on the other hand, will not change from its equilibrium potential even with large current through the electrode. This behavior is attributed to the extremely fast electrode reaction (has an almost infinite exchange current density). Polarizable electrodes are thus more suitable for recording, while nonpolarizable electrodes are more suited for pacing and stimulation.

With the unipolar method of potential record, stimulation and polarization of tissues, there are active electrode (differential) and passive (indifferent). The active electrode is placed in the area of potential record or in the area of tissue that needs to be to influence. The passive electrode is placed at some distance from the active, usually on a section of tissue having a low and relatively constant potential, or on a dead section of tissue, or in a liquid conductive medium that surrounds the object. In some cases, it is necessary that the surface area in contact with the object, the passive electrode was several times larger than the active.

Sensor. The conversion of non-electrical processes into electrical ones can be done with the help of various devices called sensors.

Generally, the term transducer is defined as a device that converts one form of energy to another. A sensor converts a physical measurand to an electric output. The sensor should respond only to the form of energy present in the measurand, to the exclusion of all others. The sensor should interface with the living system in a way that

minimizes the energy extracted, while being minimally invasive. Many sensors have a primary sensing element such as a diaphragm, which converts pressure to displacement. A variable-conversion element, such as a strain gage, then converts displacement to an electric voltage. Sometimes the sensitivity of the sensor can be adjusted over a wide range by altering the primary sensing element. Many variable-conversion elements need external electric power to obtain a sensor output.

Amplifiers. The electrical activity of biological objects and the electrical parameters of many sensors that convert non-electrical processes into electrical ones are characterized by relatively small values: micro- and milliamperes for the current, and micro- and millivolts for the voltage. Therefore, it is extremely difficult or even impossible to register them without preliminary amplification. Amplifiers are used to pre-amplify small electrical signals. They are required for many measurement circuits. For physiological research, two types of amplifiers are used: AC amplifiers and DC amplifiers.

Signal conditioners. Usually the sensor output cannot be directly coupled to the display device. Simple signal conditioners may only amplify and filter the signal or merely match the impedance of the sensor to the display. Often sensor outputs are converted to digital form and then processed by specialized digital circuits or a microcomputer. For example, signal filtering may reduce undesirable sensor signals. It may also average repetitive signals to reduce noise, or it may convert information from the time domain to the frequency domain (for example, by, Fourier transform).

Output display. The results of the measurement process must be displayed in a form that the human operator can perceive. The best form for the display may be numerical or graphical, discrete or continuous, permanent or temporary — depending on the particular measurand and how the operator will use the information. Most displays rely on our visual sense.

2 ELECTROCARDIOGRAPHY

2.1 Purpose and basic theoretical information

Purpose: Get acquainted with the principles and methods of a electrocardiogram registration and analysis.

Instruments and materials: ECG machine, electrodes, electrolyte gel.

Work task

1. Record the electrocardiogram.
2. Analyze the ECG, compare it with the normal values.

Theoretical information.

The ECG is currently the fundamental technology used to evaluate the electrical activity of the heart.

The pumping action of the heart is coordinated by electrical activity that propagates through the heart's four chambers. When the cardiac impulse passes through the heart, electrical current also spreads from the heart into the adjacent tissues surrounding the heart. A small portion of the current spreads all the way to the surface of the body. If electrodes are placed on the skin on opposite sides of the heart, electrical potentials generated by the current can be recorded; the recording is known as an electrocardiogram.

The ECG is the most important test for interpretation of the cardiac rhythm, conduction system abnormalities, and the detection of myocardial ischemia. The ECG is also of great value in the evaluation of other types of cardiac abnormalities including valvular heart disease, cardiomyopathy, pericarditis, and hypertensive disease. Finally, the ECG can be used to monitor drug treatment (specifically antiarrhythmic therapy) and to detect metabolic disturbances.

Electrodes.

Electrodes are transducers that convert the ionic currents to an electron current from which the ECG potentials can be amplified, recorded, and displayed. Electrodes are

attached to the skin with an electrolyte gel serving as an interface between the electrode and the skin.

Lead systems.

Activation and repolarization of the heart is a 4D process. Therefore, multiple leads are needed to capture directional information of activation and repolarization.

The 12-lead ECG tracings in this configuration are derived from electrodes placed in 10 standardized locations. The 12 leads are also commonly classified into 3 categories: the limb leads, the augmented limb leads, and the precordial leads. Six of the leads are considered “limb leads” because they are placed on the arms and/or legs of the individual (Figure 2.1a). The other six leads are considered “precordial leads” because they are placed on the torso (precordium) as shown in Figure 2.1b.

The six limb leads are called lead I, II, III, aVL, aVR and aVF. The letter “a” stands for “augmented,” as these leads are calculated as a combination of leads I, II and III. The six precordial leads are called leads V1, V2, V3, V4, V5 and V6.

The limb leads (leads I, II, and III) are recorded from electrodes placed on the arms and legs, as shown in Figure 2.1a. The bipolar recording measuring the difference across the left arm (+) and the right arm (−) is designated as lead I. The bipolar recording across the left leg (+) and the right arm (−) is designated as lead II. Lead III is recorded between the left leg (+) and the left arm (−). The right leg is used as a reference for the amplifier circuitry.

Lead I. The negative terminal of the electrocardiograph is connected to the right arm and the positive terminal to the left arm. Therefore, when the point where the right arm connects to the chest is electronegative with respect to the point where the left arm connects, the electrocardiograph records positively, that is, above the zero voltage line in the electrocardiogram. When the opposite is true, the electrocardiograph records below the line.

Lead II. The negative terminal of the electrocardiograph is connected to the right arm and the positive terminal to the left leg. Therefore, when the right arm is negative with respect to the left leg, the electrocardiograph records positively.

Lead III. The negative terminal of the electrocardiograph is connected to the left arm and the positive terminal to the left leg. This means that the electrocardiograph records positively when the left arm is negative with respect to the left leg.

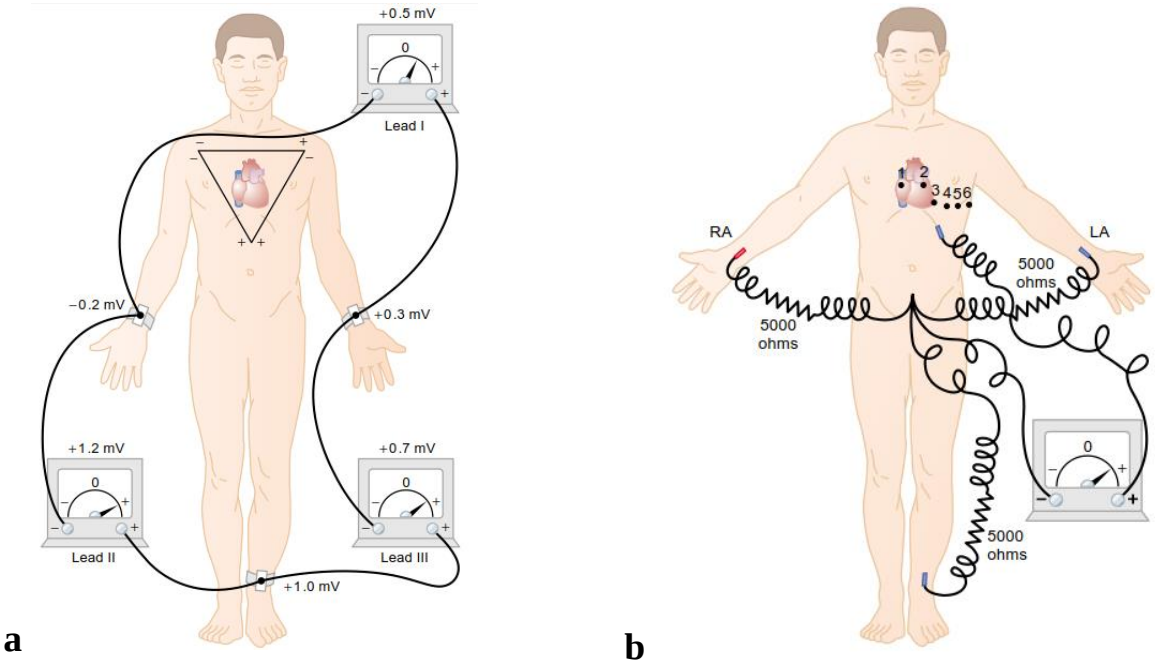


Figure 2.1 – Conventional arrangement of electrodes for recording the standard electrocardiographic leads: a – three bipolar limb leads, b - six precordial leads.

The vector representation of the limb leads known as Einthoven's triangle is shown in Figure 2.3.

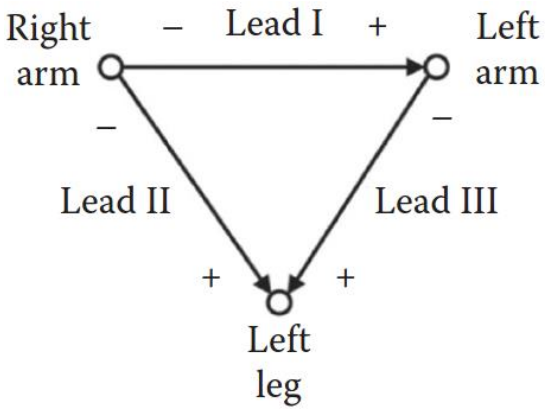


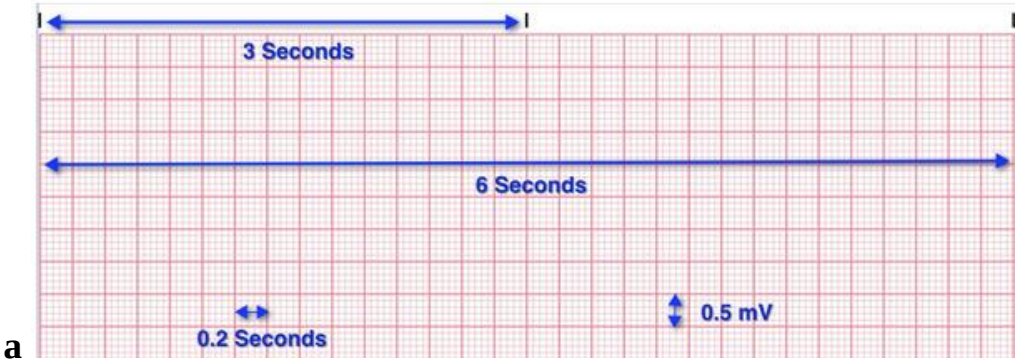
Figure 2.1 – Einthoven's triangle.

According to the 2nd Kirchhoff's law lead III can be obtained by subtracting lead I from lead II.

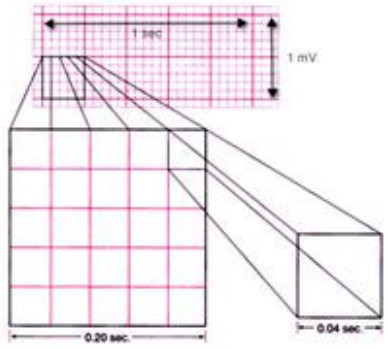
ECG grid

The electrocardiogram (ECG) is a plot of voltage on the vertical axis against time on the horizontal axis. The electrodes are connected to a galvanometer that records a potential difference. The needle (or pen) of the ECG is deflected a given distance depending upon the voltage measured.

The ECG waves are recorded on special graph paper that is divided into 1 mm^2 grid-like boxes (Figure 2.4). The ECG paper speed is ordinarily 25 mm/sec. As a result, each 1 mm (small) horizontal box corresponds to 0.04 second (40 ms), with heavier lines forming larger boxes that include five small boxes and hence represent 0.20 sec (200 ms) intervals. On occasion, the paper speed is increased to 50 mm/sec to better define waveforms. In this situation, there are only six leads per sheet of paper. Each large box is therefore only 0.10 sec and each small box is only 0.02 sec. In addition, the heart rate appears to be one-half of what is recorded at 25 mm/sec paper speed, and all of the ECG intervals are twice as long as normal. Other paper speeds are occasionally used.



a



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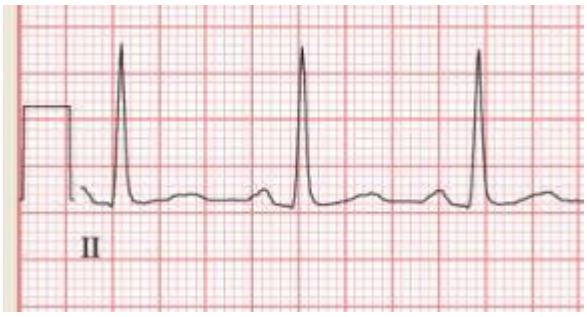


c

Figure 2.4 – ECG grid pattern (**a**) consisting of larger 5 by 5 mm boxes, which are in turn made up of smaller 1 by 1 mm boxes (**b**); **c**- calibration pulse, which indicates the time scale (width of 200 ms) and amplitude scale (height of 1 mV)

Vertically, the ECG graph measures the height (amplitude) of a given wave or deflection, as 10 mm (10 small boxes) equals 1 mV with standard calibration. On occasion, particularly when the waveforms are small, double standard is used (20 mm equals 1 mv). When the wave forms are very large, half standard may be used (5 mm equals 1 mv). The rectangular waveforms preceding each row are calibration pulses (Figure 2.4c), which indicate the time scale (width of 200 ms) and amplitude scale (height of 1 mV). Paper speed and voltage are usually printed on the bottom of the ECG.

ECG tracings are recorded on grid paper. The horizontal axis of the ECG paper records time, with black marks at the top indicating 3 second intervals. Within the large blocks are 5 small blocks, each representing 0.04 seconds/

The Normal Electrocardiogram.

The pumping action of the heart is coordinated by electrical activity that propagates through the heart's four chambers as shown in Figure 2.5.

A normal ECG contains waves, intervals, segments and one complex, as defined below.

Wave: A positive or negative deflection from baseline that indicates a specific electrical event. The waves on an ECG include the P wave, Q wave, R wave, S wave, T wave and U wave.

Interval: The time between two specific ECG events. The intervals commonly measured on an ECG include the PR interval, QRS interval (also called QRS duration), QT interval and RR interval.

Segment: The length between two specific points on an ECG that are supposed to be at the baseline amplitude (not negative or positive). The segments on an ECG include the PR segment, ST segment and TP segment.

Complex: The combination of multiple waves grouped together. The only main complex on an ECG is the QRS complex.

Point: There is only one point on an ECG termed the J point, which is where the QRS complex ends and the ST segment begins.

The main part of an ECG contains a P wave, QRS complex and T wave.

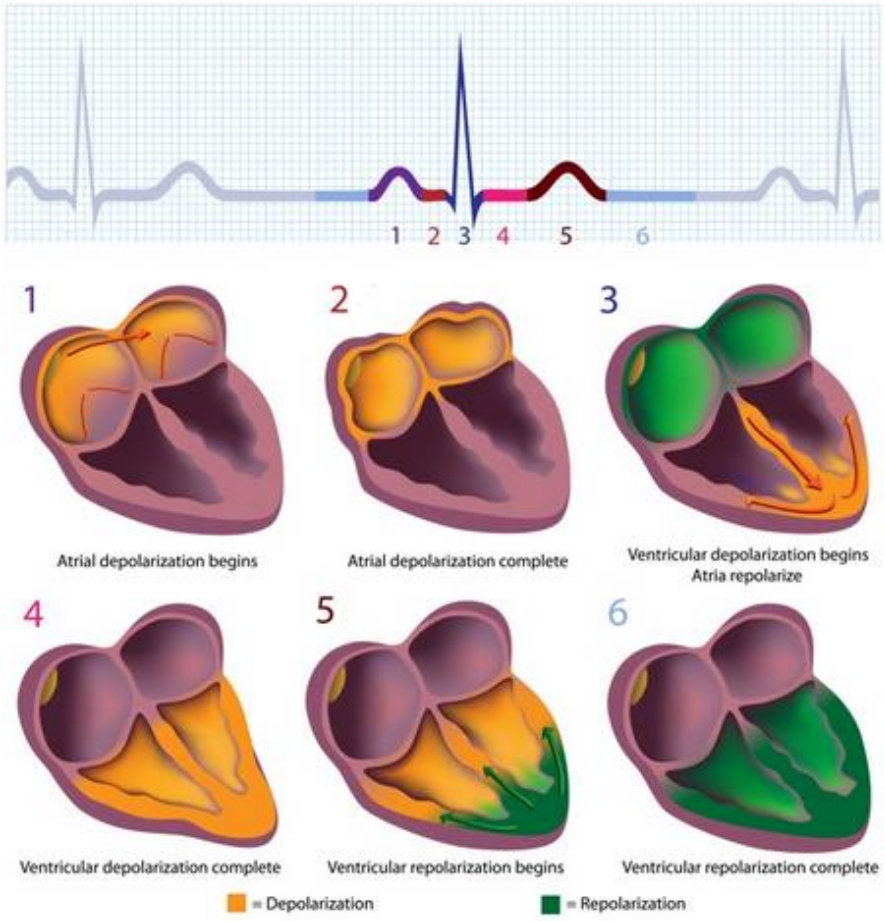


Figure 2.5 – ECG and electrical activity propagation

The ECG during a normal sinus rhythm has three main components.

The P wave indicates atrial depolarization. P wave is the first deflection and is normally a positive (upward) waveform.

QRS complex follows the P wave. It normally begins with a downward deflection, Q; a larger upward deflection, R; and then a downward S wave. The QRS complex consists of a Q wave, R wave and S wave and represents ventricular depolarization. The time period between QRS complexes is known as the RR interval. The measurement of QRS duration requires the detection of the beginning (onset) and the end (offset) of the QRS complex

The T wave comes after the QRS complex. T wave is normally a modest upward waveform, representing ventricular repolarization.

The waveforms can vary in size, timing, and morphology, depending on the rate, rhythm, heart condition, and electrode locations.

Illustration of the standard intervals and segments that are measured from the ECG is shown in Figure 2.6. Normal values of ECG components are summarized in Table 2.1.

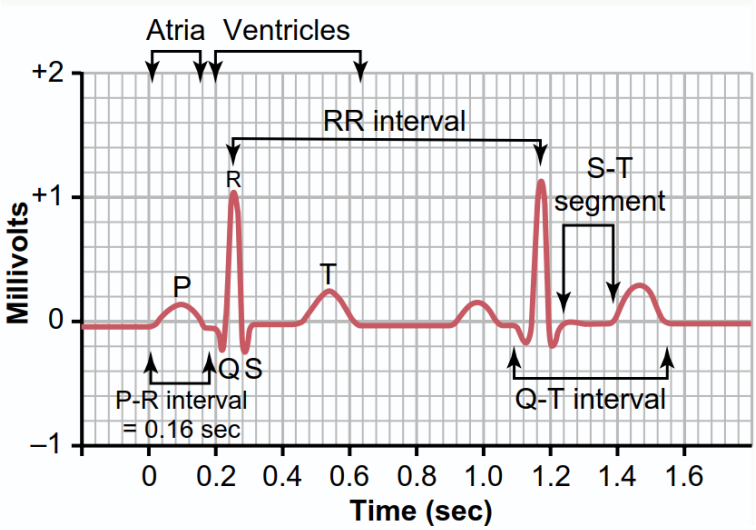


Figure 2.5 – Illustration of the standard intervals and segments that are measured from the ECG.

Table 2.1 – Normal values of ECG components.

Heart rate	60 – 100 bpm
PR interval	0.12 – 0.20 s
QRS interval	≤ 0.12 s
QT interval	< half RR interval (males < 0.40 s; females < 0.44 s)
P wave amplitude (in lead II)	≤ 3 mV (mm)
P wave terminal negative deflection (in lead V1)	≤ 1 mV (mm)
Q wave	< 0.04 s (1 mm) and < 1/3 of R wave amplitude in the same lead

2.2 Operating procedure

Assignment 1. Record the electrocardiogram from the three standard bipolar limb leads.

For standard limb leads, connect the red terminal to the right arm, the yellow terminal to the left arm, the green terminal to the left leg, and the black terminal to the right leg (ground).

Assignment 2. Analyze the ECG, compare it with the normal values:

1. Rhythm

For ventricular rhythms, examine the R to R intervals on the ECG strip. Paper marks

can be used to fix the distance for one R-R interval and then this distance can be compared to other R-R pairs. Use small boxes. Count the number of small boxes for a 5 R-R intervals. Multiply it to 0.04 seconds to determine R-R interval. Put the data into the Table 2.2. Calculate the mean value and deviations from it.

Table 2.2 – R-R Duration.

R-R interval No.	1	2	3	4	5	Mean value
R-R interval, small boxes						
R-R interval, sec						
Deviation from the mean, %						

Answer the questions:

- Are R-R intervals regular, meaning that each heart beat's R-R interval is equal? Small variations of up to 10% are considered equal.

- Is the rhythm regularly irregular? For example is there a pattern, such as increasing R-R durations? Or perhaps groups of similar intervals as illustrated on the right? Or are R-R intervals completely irregular?

- For atrial rhythm, observe the P-P intervals. Are they regular (minor variations can be caused by the breath cycle)? If P-P intervals are irregular, is there a pattern?

2. Determine heart rate. Use the R-R interval mean value.

Heart rate = 60/ R-R interval (in sec)

3. P wave

P wave represents atrial depolarization. In a normal ECG, the P-wave precedes the QRS complex. It looks like a small bump upwards from the baseline. The amplitude is normally 0.05 to 0.25mV (0.5 to 2.5 small boxes). Normal duration is 0.06-0.11 seconds (1.5 to 2.75 small boxes). The shape of a P-wave is usually smooth and rounded.

Answer the questions:

- Are P-waves present?
- Do they occur regularly?
- Is there one P-wave for each QRS complex?

- Are the P-waves smooth, rounded, and upright?
- Do all P-waves have similar shapes?
- What is the P wave amplitude in lead II (in mV)?

4. PR Interval.

The PR interval indicates AV conduction time.

The PR interval is the time period between the P-wave onset and the QRS onset. Counting small boxes methods can be used to determine PR Intervals. Normally this interval is 0.12 to 0.20 seconds (3 to 5 small boxes) in adults, longer in elderly people. This interval shortens with increased heart rate.

Also evaluate if PR intervals are constant or varying across the ECG strip. If they vary, determine if the variations are a steady lengthening until the point where an expected QRS does not appear.

Answer the questions:

- Measure the QRS interval. Does the QRS interval fall within the range of 0.08-0.10 seconds?
- Are the QRS complexes similar in appearance across the ECG tracing?
- R wave amplitude (in mV): in lead I? in lead II? in lead III?

5. QRS Complex.

The QRS complex indicates ventricular depolarization. Depolarization triggers contraction of the ventricles.

Because of the larger tissue mass, the QRS complex is larger than the P wave. While the prototypical QRS complex consists of three wave components, one or two of these components may be missing.

In this step, measure the QRS interval from the end of the PR interval to the end of the S wave. Use counting small boxes. Normally this interval is 0.08 to 0.10 seconds.

Answer the questions:

- Measure the QRS interval. Does the QRS interval fall within the range of 0.08-0.10 seconds?
- Are the QRS complexes similar in appearance across the ECG tracing?
- R wave amplitude (in mV): in lead I? in lead II? in lead III?

6. T wave.

The T wave indicates the repolarization of the ventricles. It is a slightly asymmetrical waveform that follows (after a pause), the QRS complex. Take note of T waves that have a downward (negative) deflection or of T waves with tall, pointed peaks.

The U-wave is a small upright, rounded bump. When observed, it follows the T-wave.

Answer the questions:

- Are T-waves present?
- Are the T-waves positive /negative: in the lead I? in the he lead II? in the lead III?

7. QT Interval.

The QT interval represents the time of ventricular activity including both depolarization and repolarization.

The QT interval is a measure of the time period from the QRS onset to the end of the T wave. QT intervals provide an estimate of the time required for the ventricles to repolarize following activation. Normally, the QT interval is 0.36 to 0.44 seconds (9-11 boxes). The QT interval will vary with patient gender, age and heart rate. Another guideline is that normal QT interval is less than half of the R-R interval for heart rates below 100 bpm. Because the time to repolarize is dependent on the heart rate, a heart-rate-corrected QT interval (QTc) is often used. QTc is calculated by the following formula (Bazett's formula):

$$QT_c = \frac{QT}{\sqrt{RR}}$$

where

QT is in seconds

RR is in seconds

A QTc interval of greater than 0.450 s for males and 0.470 s for females is an indicator for vulnerability to ventricular arrhythmias. Other rate correction formulae have also been used:

For men

$$[\mathrm{QT}] = 0.37 \times \sqrt{[RR]}$$

For women

$$[\mathrm{QT}] = 0.40 \times \sqrt{[RR]}$$

where RR is in seconds

Answer the questions:

– Calculate QT interval according to the sex of the patient.

– Measure QT interval and compare it with the normal value.

8. ST Segment.

The ST segment represents the early part of ventricular repolarization.

The ST segment is the line that from the end of the QRS complex to beginning of the T wave. Normally the ST segment is flat relative to the baseline.

Answer the questions:

- Is the ST segment flat relative to the baseline?

Control Questions.

1. Explain spontaneous rhythmicity in cardiac tissue.
2. Define what is meant by ECG and describe its utility in medicine.
3. Explain why Kirchhoff's law results in lead I + lead III = lead II.
4. Describe why Einthoven's triangle is an idealized abstraction that is useful but not strictly valid.
5. What amplification is required to record an ECG? What factors can cause noise during ECG registration?
6. Describe the events in the heart that produce the P wave, QRS complex, T wave.

3 BLOOD PRESSURE DETERMINATION BY AUSCULTATORY METHOD.

3.1 Purpose and basic theoretical information

Purpose: Get acquainted with the principles and methods of a blood pressure measurement.

Instruments and materials. Manometer, stethoscope, timer.

Work task.

1. Get acquainted with the principles auscultative method of blood pressure measurement.
2. Measure the blood pressure and heart rate.
3. Calculate the related indices and compare them with normal values.

Theoretical information

Methods for an indirect blood pressure measurement:

- Pressure Cuff Methods
  - Riva-Rocci Method
  - Oscillometry
  - Continuous Vascular Unloading
- Tonometry

Korotkoff method for a blood pressure determination.

A stethoscope is placed over the antecubital artery and a blood pressure cuff is inflated around the upper arm (Figure 3.1). As long as the cuff continues to compress the arm with too little pressure to close the brachial artery, no sounds are heard from the antecubital artery with the stethoscope. However, when the cuff pressure is great enough to close the artery during part of the arterial pressure cycle, a sound then is heard with each pulsation. These sounds are called Korotkoff sounds.

The exact cause of Korotkoff sounds is still debated, but they are believed to be caused mainly by blood jetting through the partly occluded vessel. The jet causes turbulence in the vessel beyond the cuff, and this sets up the vibrations heard through the stethoscope.

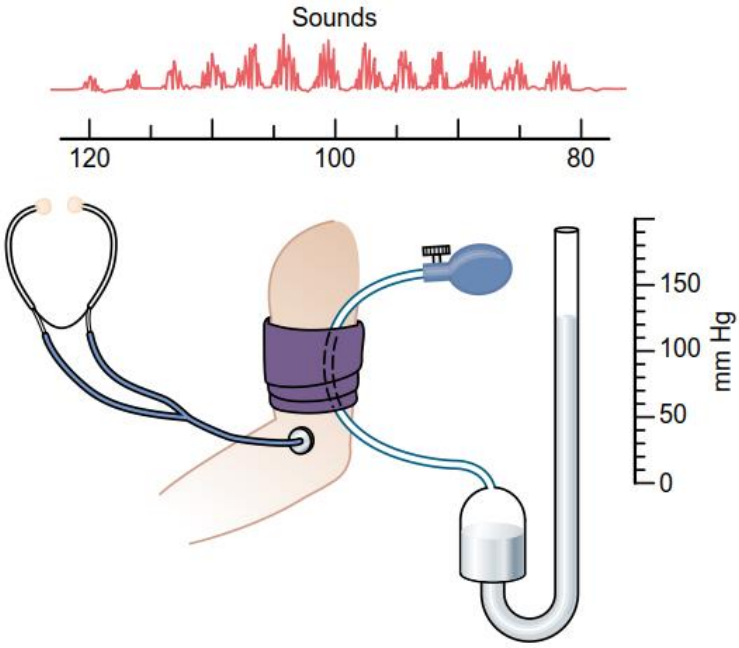


Figure 3.1 – Korotkoff method for a blood pressure determination.

In determining blood pressure by the auscultatory method, the pressure in the cuff is first elevated well above arterial systolic pressure. As long as this cuff pressure is higher than systolic pressure, the brachial artery remains collapsed so that no blood jets into the lower artery during any part of the pressure cycle. Therefore, no Korotkoff sounds are heard in the lower artery. But then the cuff pressure gradually is reduced. Just as soon as the pressure in the cuff falls below systolic pressure, blood begins to slip through the artery beneath the cuff during the peak of systolic pressure, and one begins to hear tapping sounds from the antecubital artery in synchrony with the heartbeat. As soon as these sounds begin to be heard, the pressure level indicated by the manometer connected to the cuff is about equal to the systolic pressure.

As the pressure in the cuff is lowered still more, the Korotkoff sounds change in quality, having less of the tapping quality and more of a rhythmical and harsher quality. Then, finally, when the pressure in the cuff falls to equal diastolic pressure, the artery no longer closes during diastole, which means that the basic factor causing the sounds (the jetting of blood through a squeezed artery) is no longer present. Therefore, the sounds suddenly change to a muffled quality, then disappear entirely after another 5- to 10-millimeter drop in cuff pressure. One notes the manometer pressure when the Korotkoff sounds change to the muffled quality; this pressure is about equal to the diastolic pressure.

The auscultatory method for determining systolic and diastolic pressures is not entirely accurate, but it usually gives values within 10 per cent of those determined by direct catheter measurement from inside the arteries.

Arterial pressure indices to be measured:

– systolic (maximal) arterial pressure ($AP_{diastol}$);

– diastolic (minimal) arterial pressure (AP_{systol}).

Arterial pressure optimal values for adults who are 18 years and older, adopted by the European Society of Cardiology (ESC) and the European Society of Hypertension (ESH) are < 120 mmHg – systolic BP, and < 80 mmHg – diastolic BP.

Arterial pressure indices to be calculated.

– **Pulse pressure (**AP_{pulse}**).** The pulse pressure is the difference between systolic and diastolic pressures. It depends on CO, stiffness of large arteries (where it is measured), and wave reflection:

$$\text{Pulse pressure} = \text{systolic arterial pressure} - \text{diastolic arterial pressure}$$

– **Mean arterial pressure (**MAP**).** The mean arterial pressure is the average of the arterial pressures measured millisecond by millisecond over a period of time (Figure 3.2).

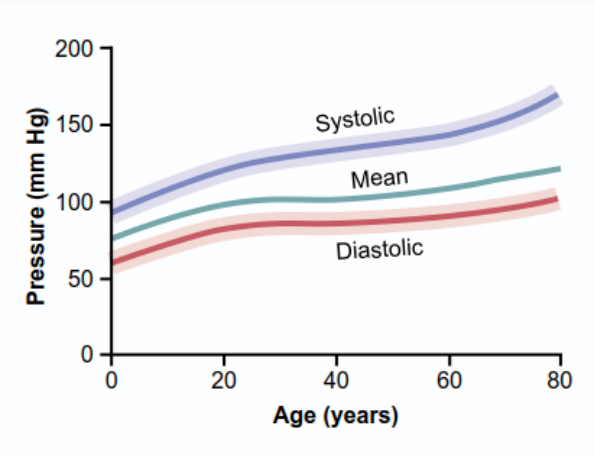


Figure 3.2 – Mean arterial pressure.

It is not equal to the average of systolic and diastolic pressure because the arterial pressure remains nearer to diastolic pressure than to systolic pressure during the greater part of the cardiac cycle. Therefore, the mean arterial pressure is determined about 60 per cent by the diastolic pressure and 40 per cent by the systolic pressure.

$$\mathrm{MAP} = \mathrm{AP}_{diastol} + \frac{1}{3}\mathrm{AP}_{pulse}$$

3.2 Operating procedure

1. Get acquainted with the principles and methods of auscultative method of blood pressure measurement (according to Korotkoff 's method), measure heart rate and blood pressure 3 times in the sitting position. Have a rest of 2 minutes or more before each measurement. Put results in a table 3.1.

Table 3.1 – Heart rate and blood pressure measurements.

	Arterial pressure, mm Hg.				HR min^{-1}
	Systolic pressure	Diastolic pressure	Pulse pressure	Mean arterial pressure	
1					
2					
3					
mean					

2. Calculate the pulse pressure and mean arterial pressure.
3. Compare the measured and calculated indices with a normal values.

Control Questions.

1. What methods for an indirect blood pressure measurement do you know?
2. Origin of Korotkoff sounds.
3. Identify the parts of the aortic pressure trace including systolic pressure, diastolic pressure, dicrotic notch, pulse pressure, and mean arterial pressure?
4. What factors determine blood pressure?
5. What are the optimal values of arterial pressure for a healthy adults?
6. What arterial pressure regulation mechanisms do you know?

4 HEMODYNAMICS CALCULATIONS.

4.1 Purpose and basic theoretical information

Purpose: Determine the parameters of systemic hemodynamics and pumping function of the heart by blood pressure and heart rate.

Instruments and materials. Manometer, stethoscope, timer.

Work task.

1. Get acquainted with the principles and methods of hemodynamics calculations.
2. Measure the heart rate and blood pressure.
3. Calculate the parameters of systemic hemodynamics and pumping function of the heart by blood pressure and heart rate.

Theoretical information

Stroke volume (SV), cardiac output (CO), cardiac index (CI), stroke index (SI).

These parameters directly characterize the pumping function of the heart and determine the delivery of oxygen to the tissues. Knowing the SV, it is possible to calculate other hemodynamic parameters (cardiac output, systemic vascular resistance, pulmonary vascular resistance, etc.), which provides a more complete picture of the functioning of the circulatory system.

Stroke volume (SV).

Each beat of the heart ejects a volume of blood, the **stroke volume**, equal to the difference between the end-diastolic volume of the left ventricle (EDV) and the end-systolic volume (ESV):

$$SV = EDV - ESV$$

The ejected volume then travels through the systemic circulation that perfuses all of the tissues except the lungs. The **cardiac output** is the average flow into the aorta, calculated as:

$$CO = SV \times HR$$

where CO is the **cardiac output**, SV is the **stroke volume**, and HR is the **heart rate**, in beats per minute. The units of CO are $L \cdot min^{-1}$. Typical values at rest are 4–6 $L \cdot min^{-1}$, but this can increase as much as 5-fold during strenuous exercise. CO can be varied by changing SV or changing HR or both

Cardiac output determination.

a. **Calculation** method for determining the stroke volume based on the Starr formula. First, stroke volume is calculated by the Starr formula:

$$SV = 93 + 0.62 \times \mathrm{AP}_{pulse} - 0.45 \times \mathrm{AP}_{diastol} - 0.61 \times A$$

where SV – stroke volume (ml), AP_{pulse} .– pulse pressure (mm Hg), $\mathrm{AP}_{diastol.}$ – diastolic pressure (mm Hg) , A – age (years).

Starr's formula gives reliable results when examining healthy people at rest.

Next, the cardiac output is calculated as

$$CO = SV \times HR$$

where CO is the **cardiac output** (ml),

SV is the **stroke volume**,

HR is the **heart rate**

b. **Fick principle:**

c. **Indicator dilution method**

d. **Thermodilution**.

The **cardiac index** is the value of the minute volume of blood circulation divided by the body surface area in m^2. The **cardiac index** is to neutralize the effect of individual anthropometric differences on the CO value.

$$\mathrm{CI} = \mathrm{CO}/\mathrm{BSA}$$

where CI is the cardiac index in is L / (min × m^2),

CO is the cardiac output in L/min

BSA is the body surface area in m^2

The a values of the cardiac index normally vary from 2 to 4 L / (min × m^2) (a range 3–3.5 L / (min × m^2) is given more often).

The **stroke index** is the ratio of the **stroke volume** of blood circulation to the body surface area in m^2:

$$\mathrm{SI} = \mathrm{SV}/\mathrm{BSA}$$

where SI is the stroke index in is ml / m^2

SV is the stroke volume in ml

BSA is the body surface area in m^2

Work output of the heart.

Total work of the heart includes pressure, kinetic, and gravitational terms.

The stroke work output of the heart is the amount of energy that the heart converts to work during each heartbeat while pumping blood into the arteries. Minute work output is the total amount of energy converted to work in 1 minute; this is equal to the stroke work output times the heart rate per minute.

Work output of the heart is in two forms. First, by far the major proportion is used to move the blood from the low-pressure veins to the high-pressure arteries. This is called **volume-pressure work** or **external work.**

$$\mathrm{W}_{v-p} = \mathrm{P} \times \mathrm{V} = \mathrm{MAP} \times \mathrm{SV}$$

where $W_{v\text{-}p}$ is the volume-pressure work,

MAP is the mean arterial pressure,

SV is the stroke volume.

Second, a minor proportion of the energy is used to accelerate the blood to its velocity of ejection through the aortic and pulmonary valves. This is the **kinetic energy** of blood flow component of the work output.

$$\mathrm{W}_{kinetic} = ½\ \mathrm{m}v^2 = ½\ \mathrm{SV} \times \rho \times v^2$$

where $W_{kinetic}$ is the kinetic energy of blood flow component,

SV is the stroke volume,

ρ is a mass density of blood,

v is the velocity of ejection.

Right ventricular external work output is normally about one sixth the work output of the left ventricle because of the sixfold difference in systolic pressures that the two ventricles pump. The additional work output of each ventricle required to create kinetic energy of blood flow is proportional to the mass of blood ejected times the square of velocity of ejection.

Ordinarily, the work output of the left ventricle required to create kinetic energy of blood flow is only about 1 percent of the total work output of the ventricle and therefore is ignored in the calculation of the total stroke work output. But in certain abnormal conditions, such as aortic stenosis, in which blood flows with great velocity through the stenosed valve, more than 50 percent of the total work output may be required to create kinetic energy of blood flow.

Work output of the heart is a sum of its **volume-pressure work** and **kinetic energy** of blood flow component:

$$W = W_{v-p} + W_{kinetic}$$

Systemic vascular resistance.

In the systemic circulation, the most resistance to blood flow is small arteriolar resistance.

Resistance cannot be measured by any direct means, instead, resistance must be calculated from measurements of blood flow and pressure difference between two points in the vessel, according to the hydrodynamic analogue of Ohm's law:

$$Q = \frac{P_a - P_v}{R}$$

where Q is the Flow,

P_a – P_v is a difference between the mean arterial and venous pressures,

R is the resistance.

Systemic vascular resistance (SVR) can be calculated as

$$SVR = \frac{MAP - CVP}{CO}$$

where SVR is the systemic vascular resistance,

MAP is the mean arterial pressure,

CVP is a central venous pressure, which normally is equal to 0,

CO is the cardiac output.

4.2 Operating procedure

Assignment 1. Measurement of blood pressure and a heart rate.

1. Measure the heart rate and blood pressure 3 times in a sitting position. Have a rest of 2 minutes or more before each measurement. Put results in a table 4.1.

Table 4.1 – Heart rate and blood pressure measurements.

	Arterial pressure, mm Hg.				HR min^{-1}	Cardiac cycle duration, s	Systole duration, s
	Systolic pressure	Diastolic pressure	Pulse pressure	Mean arterial pressure			
1							
2							
3							
mean							

2. Calculate the cardiac cycle duration from the heart rate and the systole duration by Bazett's formula:

$$t = k \times \sqrt{\text{cardiac cycle duration } (in\ seconds)},$$

where k – coefficient, which is 0.37 for men, and is 0.40 for women.

Assignment 2. Calculation of a parameters of systemic hemodynamics and pumping function of the heart.

1. Calculate your body surface area (BSA), stroke volume (SV), cardiac output (CO), cardiac index (CI), stroke index (SI):
    a) body surface area (BSA) by Mosteller:

$$BSA = \sqrt{\frac{H \times W}{3600}}$$

where *BSA* is the body surface area in m^2

H is the height in cm,

W is the weight in kg.

b) Stroke volume SV in ml by Starr equation;

c) Cardiac output CO in ml/min;

d) Cardiac index CI in L / (min × m^2);

e) Stroke index in ml / m^2

2. Compare the values of your own cardiac index with the norm.

3. Assuming the blood velocity in the aorta is 0.4 m/s and mass density of blood is 1.05 g/cm^3, calculate for your heart:

   a) Volume-pressure work of the left ventricle per one beat;

   b) Stroke work output of the heart;

   c) Minute work output;

   d) Power of the heart during a systole. Take the systole duration that you have calculated in the Table 4.1.

   e) Systemic vascular resistance in MPa × sec × m^{-3}

Control Questions.

1. Give the definition of the stroke volume and ejection fraction.
2. Starr's formula reliability.
3. What is the cardiac output? How does it depend on a heart rate and stroke volume?
4. Identify components of cardiac work.
5. Why is the output of the right heart the same as the output of the left heart?
6. How can you determine cardiac output from oxygen consumption?
7. How can you determine cardiac output from indicator or thermal dilution?
8. Describe the basic hemodynamic parameters. How are they related? Write an equation.
9. Define vascular resistance. Where is most of the resistance in the circulatory system?
10. What cardiovascular function parameters are controlled in the living organism?

5 BLOOD CELLS

5.1 Purpose and basic theoretical information

Purpose: Investigate the quantitative characteristics of the formed elements of human blood.

Instruments and materials: Microscope equipped with digital camera; immersion oil; blood smear, stained by Romanovsky-Gimza; personal computer; AxioVision software.

Work task

1. Identify blood cells on a stained blood smears.
2. Take digital photos of blood cells.
3. Measure the parameters of blood cells in digital photos with AxioVision software.

Theoretical information

Blood is a complex, heterogeneous suspension of formed elements, the blood cells, or hematocytes, suspended in a continuous fluid called plasma. It is accounting for about 8 ± 1% of total body weight and averaging in volume 5200 mL. The volume percentage of blood cells in blood (hematocrit) is normally 40.7–50.3% for males and 36.1–44.3% for females.

Normal red blood cells are biconcave discs having a mean diameter of about 7.8 micrometers and a thickness of 2.5 micrometers at the thickest point and 1 micrometer or less in the center. The average volume of the red blood cell is 90 to 95 cubic micrometers. In healthy men, the average number of red blood cells per cubic millimeter is 5,200,000 (±300,000); in women, it is 4,700,000 (±300,000).

The leukocytes, also called *white blood cells,* are the *mobile units* of the body's protective system. They are formed partially in the bone marrow (*granulocytes* and *monocytes* and a few *lymphocytes*) and partially in the lymph tissue (*lymphocytes* and *plasma cells*). After formation, they are transported in the blood to different parts of the body where they are needed.

Six types of white blood cells are normally present in the blood. They are *polymorphonuclear neutrophils, polymorphonuclear eosinophils, polymorphonuclear basophils, monocytes, lymphocytes,* and, occasionally, *plasma cells.* In addition, there are large numbers of *platelets,* which are fragments of another type of cell similar to the white blood cells found in the bone marrow, the *megakaryocyte.* The first three types of cells, the polymorphonuclear cells, all have a granular appearance, as shown in Figure 5.1, and for this reason are called *granulocytes,* or, in clinical terminology, "polys," because of the multiple nuclei.

The adult human being has about 7000 white blood cells per *microliter* of blood (in comparison with 5 million red blood cells). Of the total white blood cells, the normal percentages of the different types are approximately the following: Polymorphonuclear neutrophils – 62.0%, Polymorphonuclear eosinophils – 2.3%, Polymorphonuclear basophils – 0.4%, Monocytes – 5.3%, Lymphocytes - 30.0%.

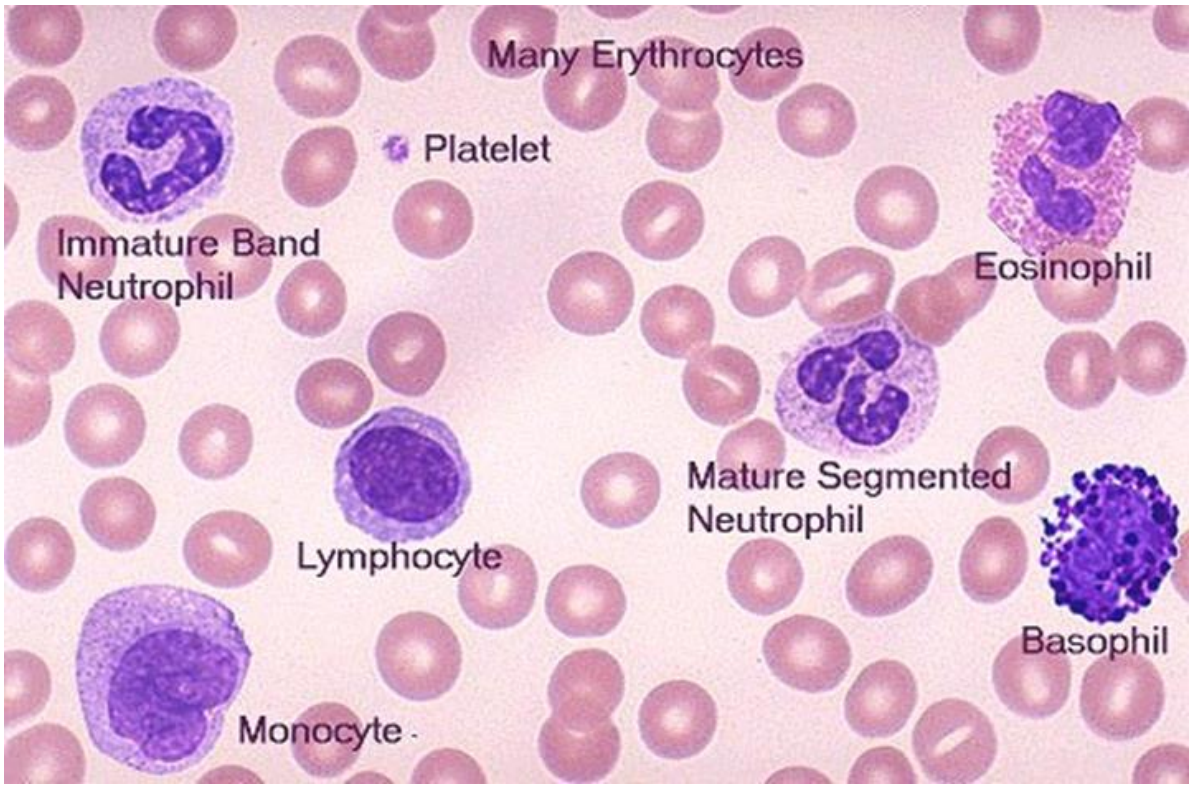


Figure 5.1 – Blood smear

5.2 Operating procedure

Assignment 1. Identify blood cells on a stained blood smear.

1. Turn on the microscope.

2. Start the AxioVision.

3. Swing the eyepiece tubes symmetrically slightly toward or away from one another to adjust the distance between the tubes to your individual interpupillary distance (The adjustment of the interpupillary distance is correct when you see only one round image while looking through the two eyepieces). Swivel the eyepiece tubes fully up or down to adjust the viewing height to your individual requirements.

4. Put the specimen onto the mechanical stage. Look at the specimen through the eyepiece with the eyepiece pointer and focus on the microscopic image by turning on the focusing drive. When in the above-mentioned eyepiece both microscopic image and eyepiece pointer appear sharply, focus the image for the second eye by turning the focusing ring of the second eyepiece.

5. Adjust an illumination intensity.

6. Find image with the use of control knobs for X travel and for Y travel of mechanical stage.

7. In the **AxioVision** main menu, click **Live** to transfer the image to the monitor screen.

8. Install the 100× objective, apply a drop of immersion oil on the specimen. Focus on the specimen with the use of the coarse focusing drive and the fine focusing drive.

9. Find red blood cells, pay attention to their color, shape, structure. Find leukocytes of different species and consider their color, granules in the cytoplasm, shape of the nucleus; identify them. Take digital photos of selected items. To do this, click **Snap** in the AxioVision main menu. Save cell images as *.zvi file to your own folder.

Assignment 2. Measure the parameters of blood cells in digital photos with AxioVision software.

Measure linear dimensions and area of 3 red blood cells and of 3 leucocytes.

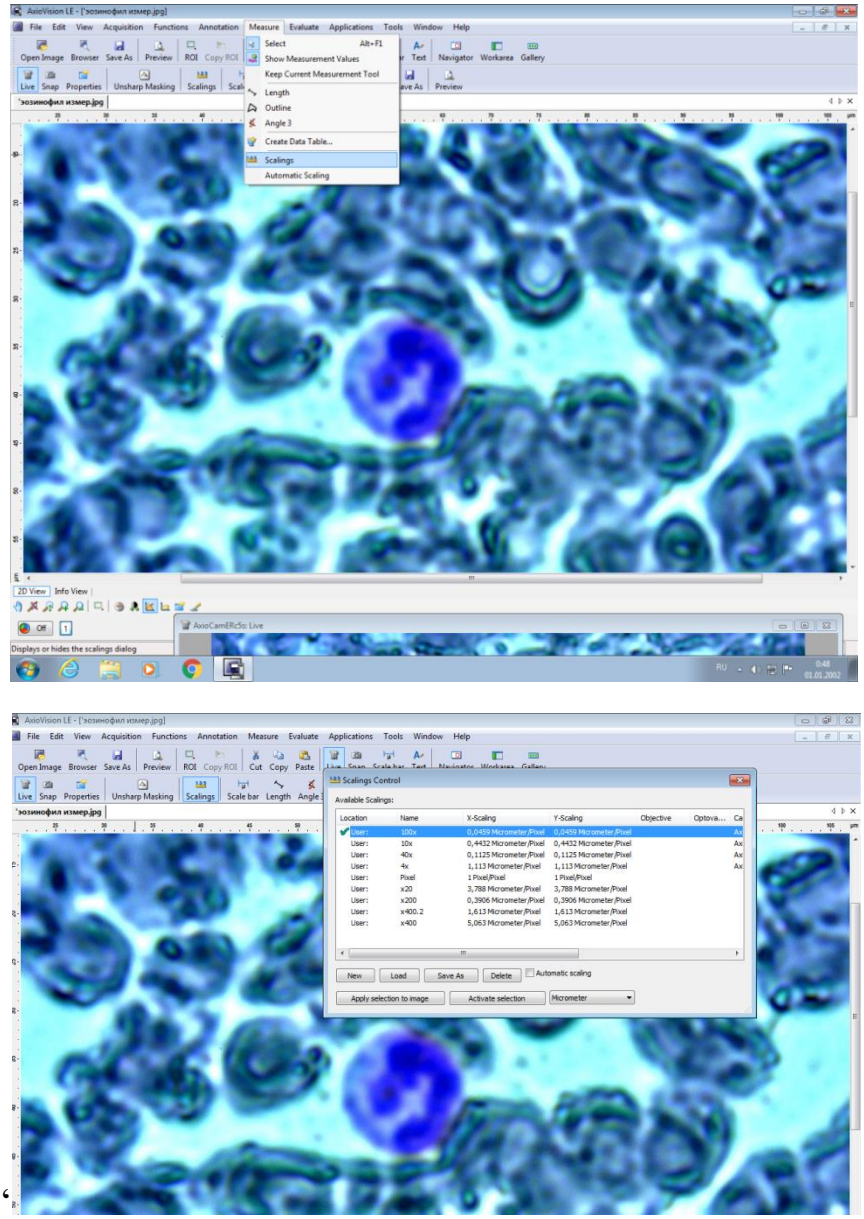
1. Open the image (**Open Image**) and zoom it to the size of the window.

2. Select the image scaling options that match the magnification of the objective at which the image was taken (Figure 5.3, *a, b*). For this:

– Select **Scalings** in the **Measure** menu.

– Select **100**× in the **Scaling control** panel.

– Click **Apply selection to image** & **Activate selection**.



a ***b***

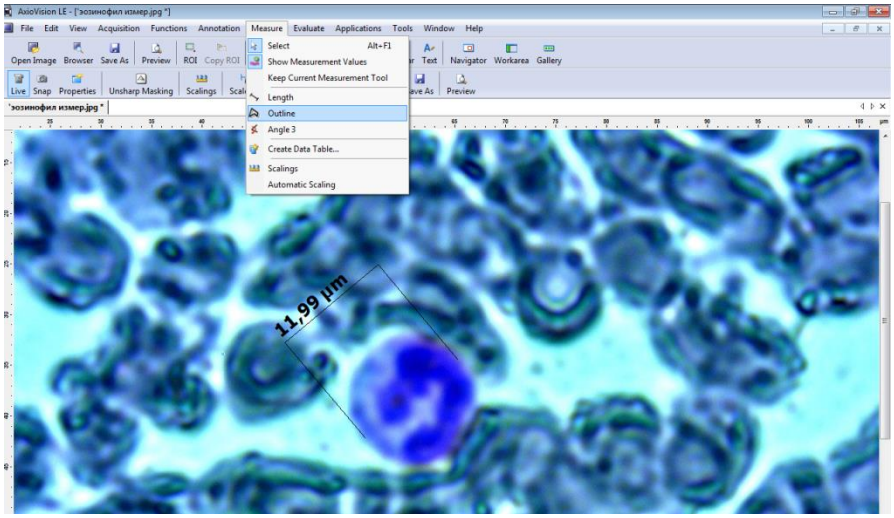
Figure 5.3 – Image scaling selection

3. Select one or more cells in the image. Select the instruments in the **Measure** drop-down box and measure the necessary parameters (Figure 5.4, *a*, *b*):

– cell diameter (**Length**),

– cell area (**Outline**)

а



б

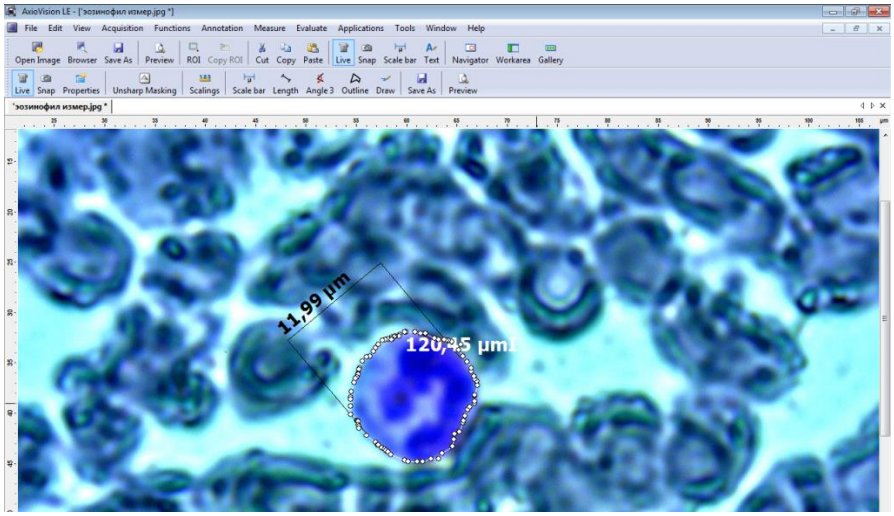


Figure 5.4 – Measurement of quantitative characteristics of a blood cells.

4. Draw a scale bar in the image. For this select **Scale bar** in **Annotations** drop-down box (Figure 5.5, 5.6):

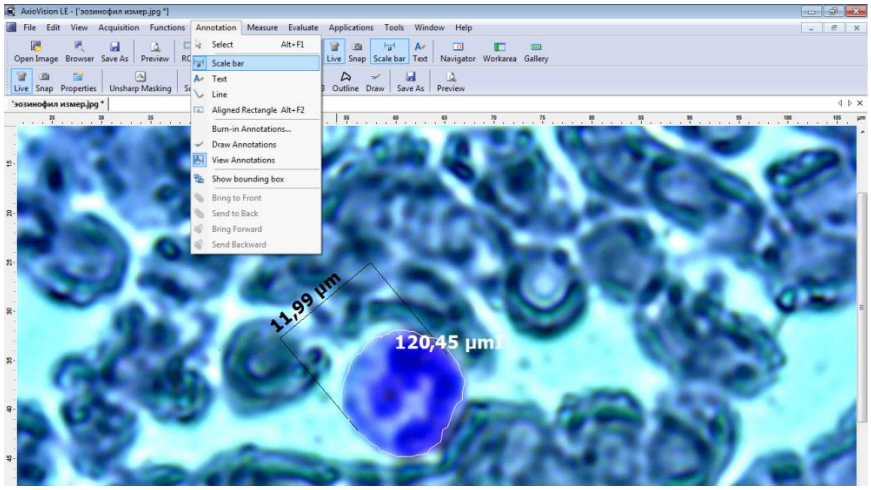


Figure 5.5 – Drawing a scale bar in the image.

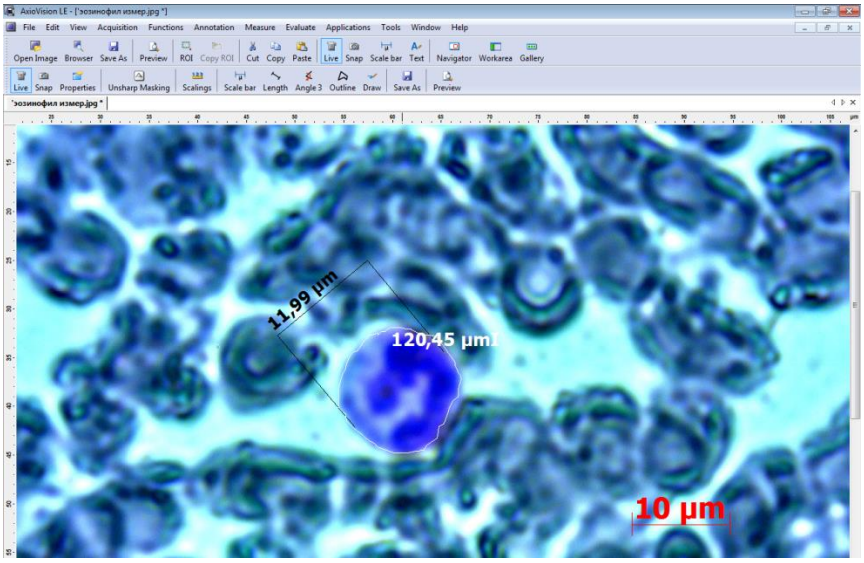
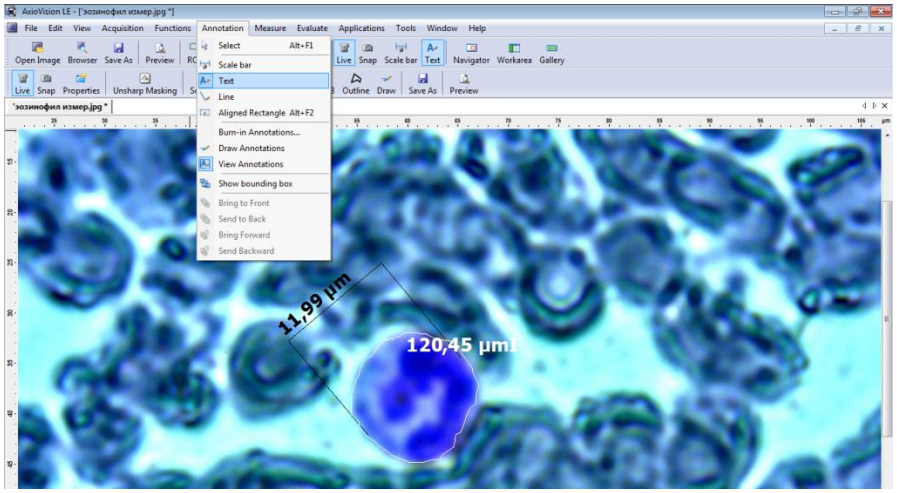


Figure 5.6 – Image with a scale bar.

5. Sign the cell:

– Select **Text** in **Annotations** drop-down box (Figure 5.7, *a*). Select the area in the image for the caption (Figure 5.7, *b*).

а



б

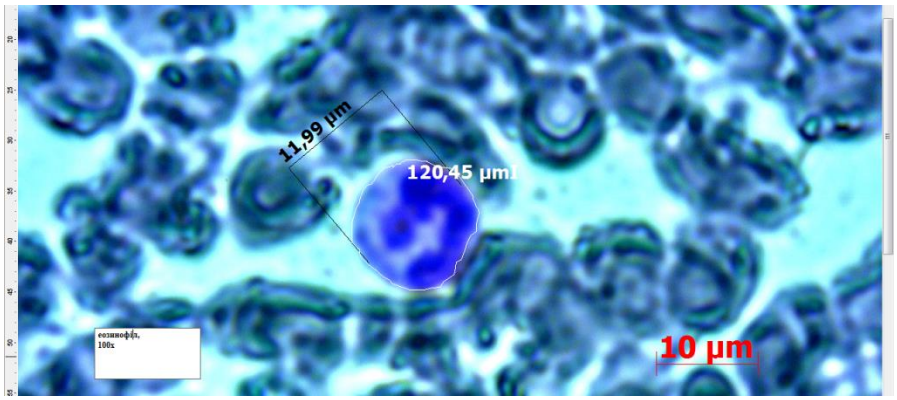


Figure 5.7 – Creating an annotation.

– Change formatting. To do this, right-click on the desired label, go to settings (**Properties**), and go to **Attributes** (Figure 5.8). Choose font, size and color of the text.

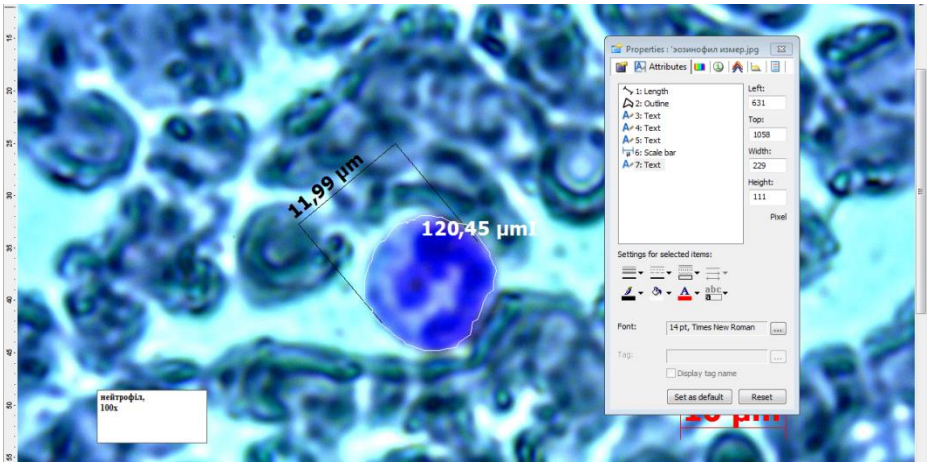


Figure 5.8 – Text formatting.

6. Save image as***.jpg** (Figure 5.9).

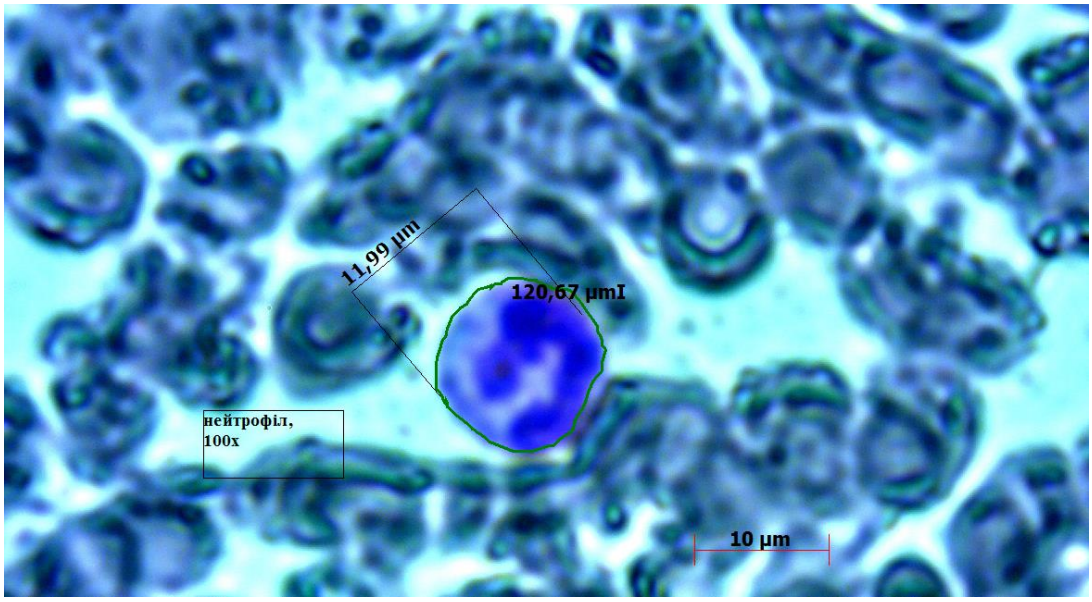


Figure 5.9 – Sample image

Assignment 3. Comparative characteristics of blood cells based on the results of the images obtained.

Write the results of measurements and observations into the table 5.1. Based on our own data, make conclusion about the differences in a number and size of blood cells.

Table 5.1 – Comparative characteristics of blood cells.

Cells	Cell shape	The presence of the nucleus and its shape	Linear dimensions	Cell area
Red blood cells				
Neutrophils				
Lymphocytes				
Basophils				
Eosinophils				
Monocytes				

Control Questions.

1. Explain the differences between blood, plasma and serum. How does this affect their hemodynamic characteristics?
2. What is a hematocrit? What is its normal value?
3. What blood cells do you know? Give their quantitative characteristics.
4. What is the physiological significance of the shape of erythrocytes?

The laboratory work report should contain:

- name and purpose of work;
- images of blood cells with a scale bar, linear dimensions and cell area measured, and annotation (cell name and magnification);
- table with a comparative characteristics of blood cells;
- conclusion;
- answers to control questions.

6 PULSE CO-OXIMETRY

6.1 Purpose and basic theoretical information

Purpose: Experimentally establish the parameters of oxygen transport function of blood.

Instruments and materials: Pulse CO-oximeter RAD-57c, timer.

Work task

1. Get acquainted with the design of the pulse CO-oximeter RAD-57s and the method of measuring the oxygen transport function of the blood.
2. Measure %SpO_2, SpCO, Perfusion Index.
3. Data analysis.

Theoretical information.

The principle of the method of pulse oximetry. Pulse oximetry is a continuous and non-invasive method of measuring the level of arterial oxygen saturation in blood. Pulse CO-oximetry is a continuous and non-invasive method of measuring the level of arterial carbon monoxide saturation (SpCO) in arterial blood.

Pulse oximetry is governed by the following principles:

1. Oxyhemoglobin (oxygenated blood), deoxyhemoglobin (non-oxygenated blood), carboxyhemoglobin (blood with carbon monoxide content), methemoglobin (blood with oxidized hemoglobin) and blood plasma constituents differ in their absorption of visible and infrared light (using spectrophotometry) (Figure 6.1).

2. The amount of arterial blood in tissue changes with your pulse. Therefore, the amount of light absorbed by the varying quantities of arterial blood changes as well.

The Rad-57 Pulse CO-Oximeter uses a multi-wavelength sensor to distinguish between oxygenated blood, deoxygenated blood, blood with carbon monoxide, oxidized blood and blood plasma. The Rad-57 utilizes a sensor with various light-emitting diodes (LEDs) that pass light through the site to a photodiode (detector).

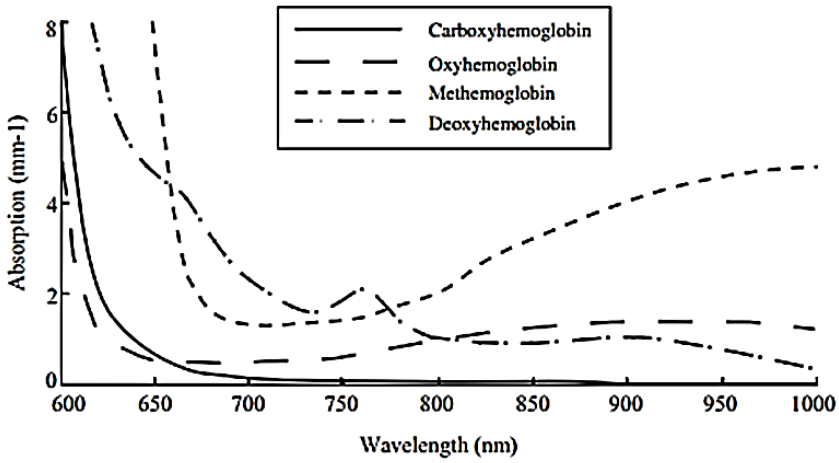


Figure 6.1 – Oxyhemoglobin, deoxyhemoglobin, carboxyhemoglobin and methemoglobin spectra.

Signal data is obtained by passing various visible and infrared lights (LED's, 500 to 1000 nm) through a capillary bed (for example, a fingertip, a hand, a foot) and measuring changes in light absorption during the blood pulsatile cycle. This information may be useful to clinicians. The detector receives the light, converts it into an electronic signal and sends it to the Rad-57 for calculation.

Once the Rad-57 receives the signal from the sensor, it utilizes Masimo Rainbow SET signal extraction technology to calculate the patient's functional oxygen saturation (%SpO_2), total hemoglobin concentration (SpHb [g/dl]), blood levels of carboxyhemoglobin (SpCO), and pulse rate.

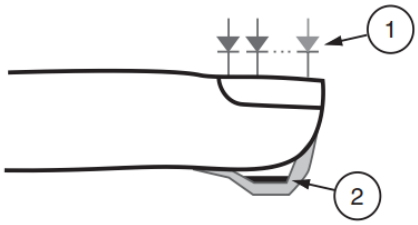


Figure 6.2 – Arrangement of optical elements in the sensor: 1 – light emitting diodes (LEDs), (7 + wavelengths), 2 – detector

The perfusion index (PI).

The perfusion index (PI) is the ratio of the pulsatile blood flow (AC) to the nonpulsatile or static blood (DC) in peripheral tissue (Figure 6.3a). Perfusion Index thus represents a noninvasive measure of peripheral perfusion that can be continuously and noninvasively obtained from a pulse oximeter. It is a calculated percentage between the

pulsatile signal and nonpulsatile signal of arterial blood moving through the site:

$$\mathrm{PI} = \frac{\mathrm{AC}}{\mathrm{DC}} \times 100\%$$

When the patient moves, another component is taken into account, which appears due to the shift of the moving venous blood (Figure 6.3b).

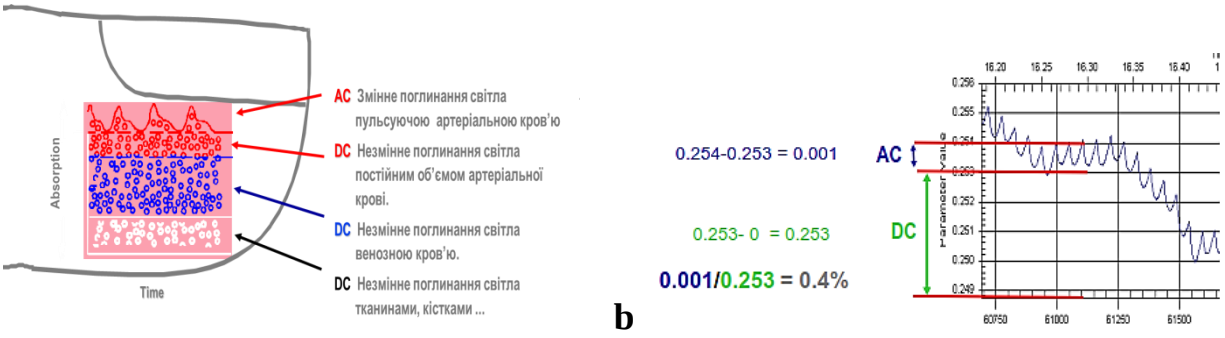


Figure 6.3 – Model of light absorption in the absence of movement of the patient (a) and the calculation of the perfusion index by the infrared signal (b)

PI may be used to find the best perfused site and to monitor physiological changes in the patient. It displays a range of < .1% to > 5% on the bar graph display and a numerical range of 0.03% to 20.0% on the display screen. A percentage greater than 1.00% is desired. Extreme changes in the display number are due to changes in physiology and blood flow (Figure 6.4).

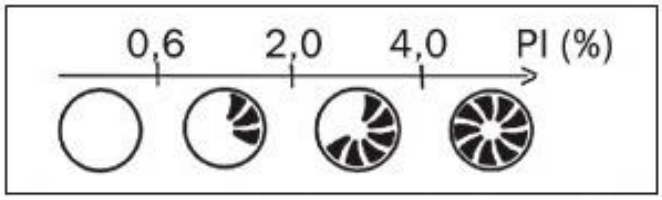


Figure 6.4 – The filling of the capillary bed at different values of the perfusion index

The measurement.

The measurement is taken by placing a sensor on a patient, usually on the fingertip for adults, and the hand or foot for neonates (Figure 6.5). The sensor connects to the pulse oximetry instrument with a patient cable. The sensor collects signal data from the patient and sends it to the instrument. The instrument displays the calculated data:

1) Percent value for arterial oxygen saturation (SpO_2)

2) Pulse rate (PR)

3) Percent value for carboxyhemoglobin (SpCO)

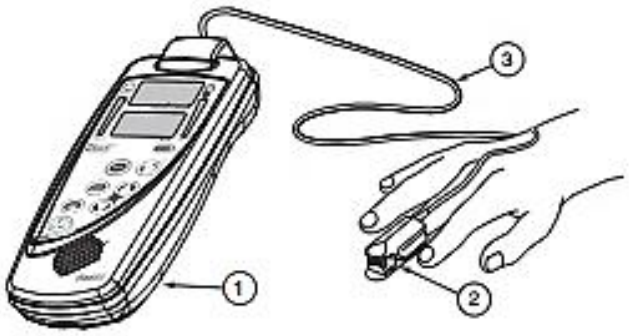


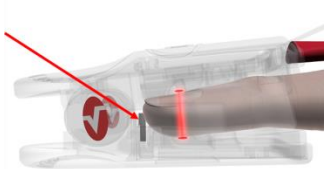
Figure 6.5 – General monitoring setup: 1 – instrument, 2 – sensor, 3 – patient cable

For the successful monitoring:

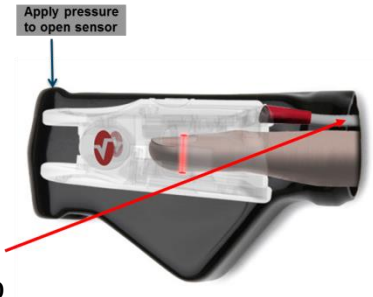
- Place the sensor on a site that is not too thick, has sufficient perfusion and provides proper alignment of the LED's and photodetector;
- Place the sensor on a site that has unrestricted blood flow;
- Do not secure a sensor with tape;
- Do not select a site near potential electrical interference (electrosurgical unit, for example).

Optimal sensor placement:

- When possible, use ring finger, non-dominant hand.
- Insert finger until the tip of finger hits the STOP Block (Figure 6.6a). Sensor should not rotate or move freely on finger.
- There is a top and bottom, cable should be on top (nail side) as shown in the Figure 6.6b.
- LED's (red light) should pass through mid-nail, not cuticle (Figure 6.7).



a



b

Figure 6.6 – Optimal sensor placement and use of a Masimo Light Shield

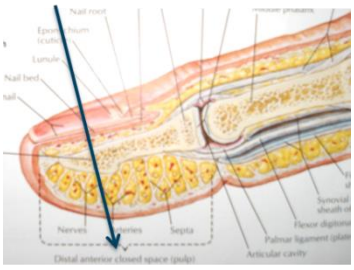


Figure 6.7 – Optimal LED path.

Stability of the SpO_2 readings may be a good indicator of signal validity. Although stability is a relative term, experience will provide a good feeling for changes that are artifactual or physiological and the speed, timing, and behavior of each. The stability of the readings over time is affected by the averaging mode being used. The longer the averaging time, the more stable the readings tend to become. This is due to a dampened response as the signal is averaged over a longer period of time than during shorter averaging times. However, longer averaging times delay the response of the oximeter and reduce the measured variations of SpO2 and PR. Inaccurate measurements may be caused by

- Elevated levels of carboxyhemoglobin
- Elevated levels of methemoglobin
- Intravascular dyes such as indocyanine green or methylene blue
- Externally applied coloring (such as nail polish)

A stable SpCO reading is associated with correct sensor placement, small physiological changes during the measurement and acceptable levels of arterial perfusion at the measurement site. Physiological changes at the measurement site are mainly caused by fluctuations in the oxygen saturation, blood concentration and perfusion. Inaccurate measurements may be caused by:

- Elevated levels of bilirubin
- Motion artifact
- Low arterial oxygen saturation levels including altitude induced hypoxemia

6.2 Operating procedure

Assignment 1. Checking the performance of the pulse CO-oximeter RAD-57 and the correctness of its settings.

1. Get acquainted with the principles of operation.
2. Make sure it is a firm connection of the patient cable with a sensor and the cable is not twisted, sliced or frayed.
3. Place sensor on finger (relatively clean & dry). Shield sensor from flashing lights, strobes or high ambient light with Masimo Light Shield*
4. Turn the Instrument on by pressing the “Power” button. The LEDs will scroll in the display window as the sensor calibrates, verify all indicators illuminate and speaker sounds a brief tone.
5. Verify the front-panel display is free of alarm and system failure messages and the battery indicator shows sufficient charge.
6. Check the Instrument configuration for the regional power line frequency (60 Hz) to allow for the cancellation of noise introduced by fluorescent lights and other sources.

Assignment 2. %SpO_2, SpCO, Perfusion Index Measurement.

NB! Measurement should be done in the sitting position.

1. Place sensor on finger (relatively clean & dry). Insert finger until the tip of finger hits the STOP Block. Shield sensor from flashing lights, strobes or high ambient light with Masimo Light Shield* as shown in Figures 6.6, 6.7.
2. Press “Power” button.
3. Verify All LED’s light up and a 1 second tone is heard
   - Startup mode begins
   - All preset configurations are displayed
   - Scrolling zeroes 0 – 0 – 0 and flashes dashed lines
   - May take up to 25 seconds
   - Do not move sensor during startup until complete reading is displayed
4. Begin patient monitoring.
   - Defaults to pulse rate and oxygen saturation reading

- "PI" bar graph displays strength of arterial perfusion

5. Display after Startup:
   - Oxygen Saturation on top in Red
   - Pulse Rate on bottom in Green
   - Green PI scale, indicates strength of arterial pulse
   - Low SIQ LED indicates poor SpO2 signal quality
   - Press "Display" to display % carboxyhemoglobin
   - Press "Bell" to silence alarms
6. Press "**Display**" button for SpCO reading.
7. Write the data into the table 6.1.
8. Press "Power" button to turn the Instrument off.
9. Repeat the measurements three times on other fingers

Table 6.1 – %SpO2, SpCO and Perfusion Index Measurements

	SpO_2 (%)	SpCO (%)	PI (%)	Pulse rate
1				
2				
3				
Mean				

Assignment 3. Data analysis.

1. Indicate your value of SpO_2 in the Oxyhemoglobin dissociation curve (Figure 6.8).
2. Compare the data obtained with the norm..

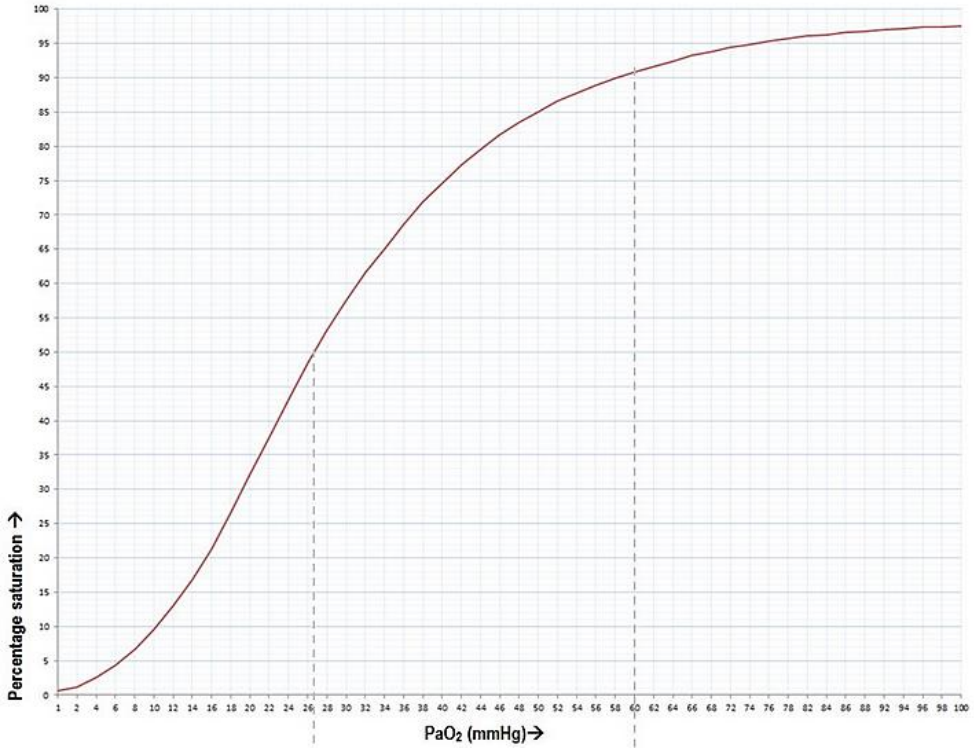


Figure 6.8 – Oxyhemoglobin dissociation curve

Control Questions.

1. The principle of the method of pulse oximetry.

2. What is the utility of an S-shaped oxygen dissociation curve?

3. What parameter of the oxyhemoglobin dissociation curve indicates cooperativity? What does cooperativity mean?

4. Describe the consequence of positive cooperativity on the oxygen saturation curve of hemoglobin.

5. What causes a rightward shift in the oxygen dissociation curve? Does a rightward shift mean oxygen dissociates more or less easily from Hb?

6. In what forms is CO_2 carried in the blood?

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Appendix I. Body surface area calculation

	Authers	Year	Formula	Designation
1	DuBois		$S = 0.107 \times \sqrt[3]{P^2}$	S is the body surface area in m^2; P is the body mass in kg
2	Costeff		$S = \frac{4P + 7}{P + 90}$	
3	DuBois and DuBois	1916	BSA = 0.007184 × H $^{0.725}$ × W $^{0.425}$	BSA is the body surface area in m^2; W is the body mass in kg, H is the height in cm
4	Takahira	1925	BSA = 0.007241×H$^{0.725}$×W $^{0.425}$	
5	Boyd	1935	BSA = 0.017827 × H $^{0.5}$ × W $^{0.4838}$	
6	Gehan and George	1970	BSA = 0.0235 × H $^{0.42246}$ × W $^{0.51456}$	
7	Haycock et al.	1978	BSA = 0.02465 × H $^{0.39646}$ × W $^{0.5378}$	
8	Mosteller (modified by Gehan and George)	1987	$BSA = \sqrt{\frac{H \times W}{3600}}$	
9	Fujimoto	1968	BSA = 0.008883 × H $^{0.663}$ × W $^{0.444}$	

Appendix II. Primo Star microscope description and adjustment

The Primo Star is a transmitted-light microscope of compact design with a small footprint. Beside the high-resolution, infinity-corrected objectives and the important microscopy techniques, the microscope is available with a camera AxioCam for photo and video documentation. The optical heart of the microscope are the objectives. Disposal objective set includes the objectives with a magnification of 4×, 10×, 40×, 100×. Since an eyepiece magnification is 10×, the maximum total magnification of is 1000×.

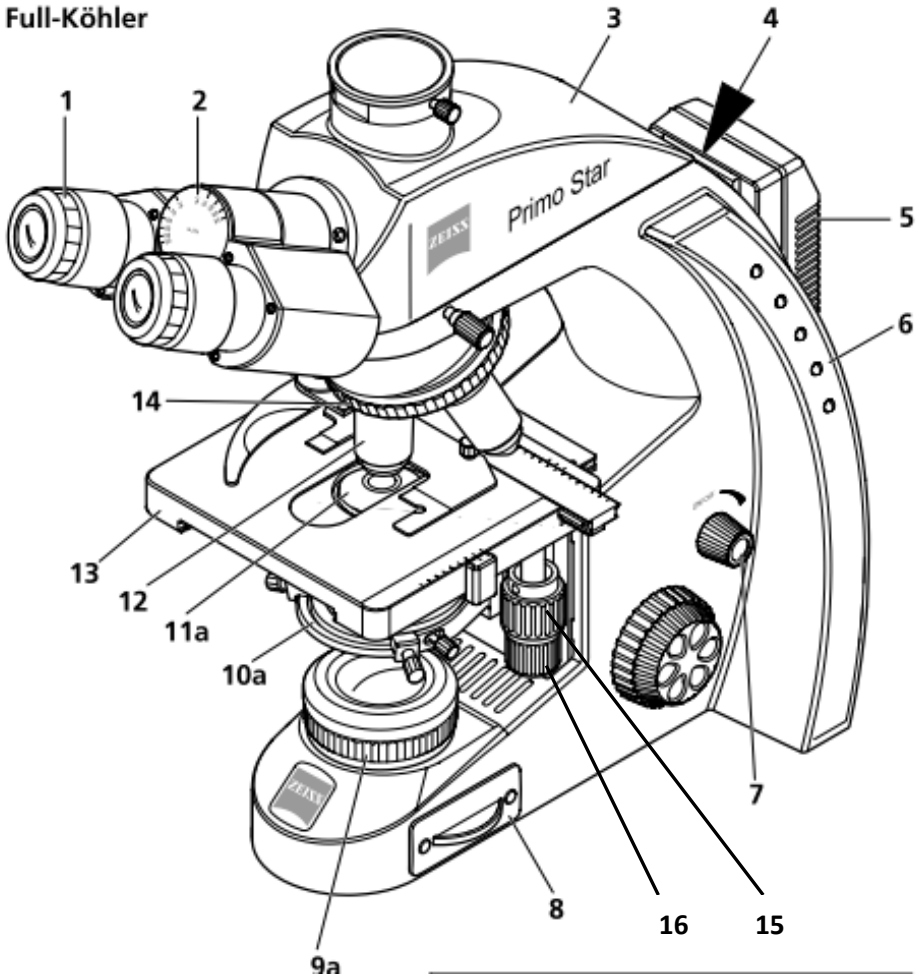

Figure II.1 – Total view and control elements of a Primo Star microscope («Full Köhler»)

1 – Eyepiece

2 – Binocular head

3 – Tube

4 – Carrying handle

5 – Plug-in power unit

6 – Stand

7 – ON/OFF switch; illumination intensity

8 – Illumination module

9a – Luminous-field diaphragm, adjustable

10a – Condenser carrier, Full Köhler

11a – Abbe condenser, Full Köhler

12 – Objective

13 – Microscope stage

14 – Objective nosepiece

15 – Control knob for X travel of mechanical stage

16 – Control knob for Y travel of mechanical stage

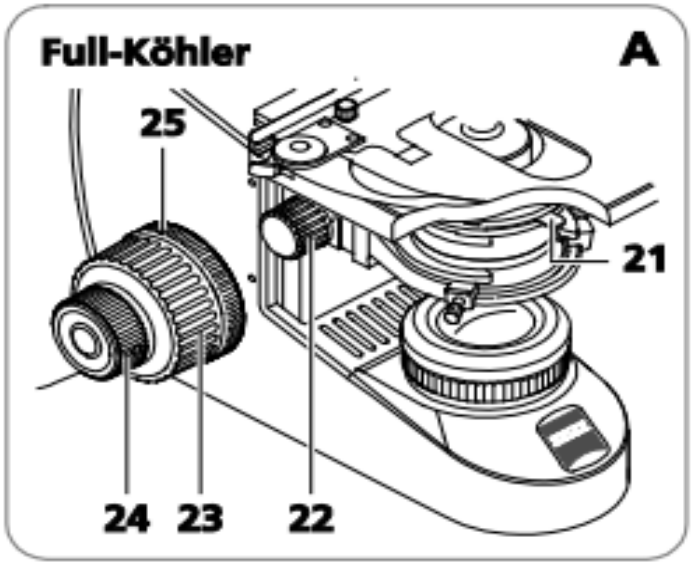

Figure II.2 – Controls of Primo Star microscope («Full Köhler»):

21 – Lever for the adjustment of the aperture diaphragm of the condenser

22 – Knurled knob for vertical adjustment of condenser

23 – Coarse focusing drive (left side)

24 – Fine focusing drive (left side)

25 – Knurled ring for adjusting the smoothness of the coarse focusing drive

Adjustment of a Primo Star microscope

Setting the interpupillary distance. Swing the eyepiece tubes symmetrically slightly toward or away from one another to adjust the distance between the tubes to your individual interpupillary distance (The adjustment of the interpupillary distance is correct when you see only one round image while looking through the two eyepieces). (Figure II.3). The adjustment of the interpupillary distance is correct when you see only one round image while looking through the two eyepieces.

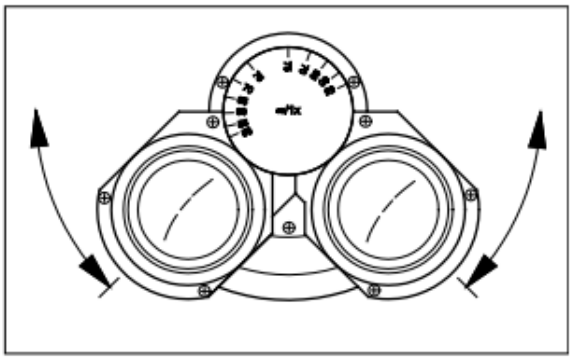

Figure II.3 – Setting interpupillary distance

Adjusting the viewing height. Swivel the eyepiece tubes fully up (Figure II.4/A) or down (Figure II.4/B) to adjust the viewing height to your individual requirements.

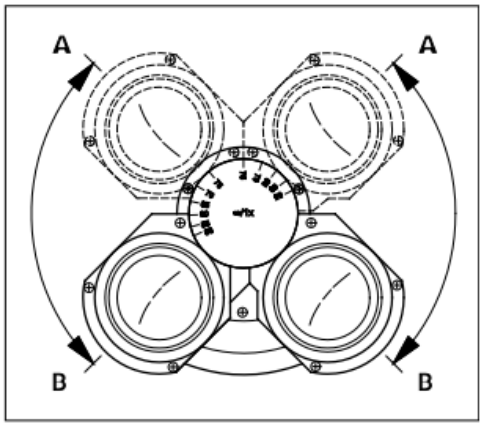

Figure II.4 – Adjusting the viewing height

Adjusting transmitted-light brightfield on the Full Köhler microscope.

Specimen field size and objective aperture change after every objective change. Therefore, repeat the adjustment of luminous-field diaphragm and aperture diaphragm to obtain optimum results.