Test material sampling and handling

Content

0. General issues

E. Gulbis laboratory takes all possible measures to ensure proper test material sampling, storage, dispatching (transporting), reception, handling, protection, disposal of testing materials, including everything that is necessary to protect the integrity of the testing materials, as well as the interests of the laboratory and customers.

Information on possible examinations (VD-10) at E. Gulbis laboratory and special rules for material collection for each test (NO-02, VD-38) is available at each material sampling point and the laboratory website www.egl.lv and it can be obtained by phone if necessary.

The request for laboratory examinations (tests) helps a testing material user to choose the correct method of material sampling. The following information is indicated on the examination request: adequate data for the patient`s correct identification and the necessary information on the payer, which promotes the appointed examination and other activities, but does not require unnecessary personal information. The patient receives an explanation as to why information is requested.
If a client (payer) prefers to transfer material anonymously then the required information is: an assumed name, sex and a year of birth.

Laboratory testing material identification system has been computerized and designed to ensure that materials are not mixed either when transmitting or documenting them.

Test samples are taken by specially trained laboratory staff. Customers who take samples themselves are informed of the correct procedure for taking, processing and delivery to the laboratory. Customers can order special sampling tools, containers and request forms for laboratory examination. They are free of charge by filling out the order form (VL-34).

If the whole serum is not utilized, it is stored frozen for one month to allow the performance of additional or repeated examinations, there is no need for repeated blood sampling.

0.1. Processing of requests for a laboratory examination

The request for a laboratory examination should contain the following information:

- ✓ Patient identification – surname, first name (in block letters)
- ✓ Personal code, address, gender
- ✓ Address, gender (preferably phone, e-mail if patient pays personally)

- ✓ Number of disease history or card number
- ✓ Invoice payer for analyses
- ✓ For patients of the sickness insurance fund – the area code and the code of the sickness insurance fund
- ✓ For insurance patients – insurance institution, policy number, policy expiry date
- ✓ Diagnostic code or details of the patient's condition
- ✓ If several copies of the Test report are required, a note indicating to whom and how to send – by e-mail, FAX or by post
- ✓ The name, surname, institution stamp, signature, date of issue of the referral by the doctor or other person who is legally authorized to request examinations, or use medical information,
- ✓ Signature of a blood collector
- ✓ Blood group determined in the presence of a patient (if the laboratory is required to determine RH, blood group), signature
- ✓ The date and time of the primary sample, urine collection and blood sampling.

0.2 Tests for which blood should be drawn on an empty stomach.

High–density cholesterol, low-density cholesterol, total cholesterol, betaCTx, free fatty acids, C peptides, folic acid, gastrin, glucose, triglycerides, vitamin B12.

1. Blood

1.1. Venous blood sampling.

Venous blood is drawn using a closed blood sampling system. The choice of vacutainer® BD tubes used in our laboratory is indicated in the request for laboratory examinations of "E. Gulbis Laboratory". The tubes should be kept at room temperature so that the vacuum does not disappear.

Sequence of blood sampling in tubes:

Cap colour Vacutainer BD	Notation	Additive
1. BacT/ALERT bottle – aerobic FA, anaerobic FN, children PF (bioMerieux)	F	A medium for bacteria, mushroom cultivation
2. Blue	Zi	Na citrate
3. Red	S	Without additive
4. Yellow	Dz	Serum separating gel
5. Green	Za	Heparin
6. Violet	V	EDTA
7. Grey	P	Na fluoride

<u>It is prohibited:</u>

To open the bottle or tube by removing the cap before blood sampling.

To draw the blood with a syringe and fill the tube with a puncture through the cap.

Tubes with separating gel, which when being centrifuged moves and stays on the serum and clot boundary and forms an impenetrable barrier between the serum and erythrocytes, are used instead of serum tubes (red cap) to ensure stable serum storage.

1.2. Venous puncture technique and blood sampling in Vacutainer BD tubes and BacT/ALERT bottles.

Vacutainer® BD system consists of 3 parts:
1. Vacutainer tubes with a precisely defined vacuum and/or the BacT/ALERT bottles of blood inoculants, where the vacuum is not dosed, and the collected blood level should correspond to the appropriate mark on the label.
2. The needle holder, which is intended to connect the tube and needle, should be new for each patient. The special BacT/ALERT needle holder for blood sampling inoculations is used repeatedly, but the Luer adapter is changed for each patient. The adapter is screwed into the BacT/ALERT needle holder and attached to the needle in the vein.
3. Sterile disposable needle or "butterfly system" is for filling one or more tubes.
* Luer adapter – the Luer syringe is required to be added to the needle in the vein if, after blood sampling, intravenous medication is necessary or the BacT/ALERT needle holder should be attached for blood sampling inoculations. There should be a new one for each patient.
* BacT/ALERT adapter is inserted in the BacT/ALERT needle holder to collect blood in Vacutainer tubes after blood sampling for inoculation has been performed. It is used repeatedly.
The patient's identification should be checked before vein puncture.
Peripheral venous blood is drawn from the elbow vein, whenever possible. Blood circulation in the veins of the palm is slower, the veins are more mobile and the needle is likely to cause painful sensations, therefore, the blood is drawn from the palm in cases when elbow vein puncture is not possible. Prepare the necessary tools (Vacutainer system tubes in sampling sequence, tourniquet, disinfectant, cotton wool, gloves).
1. Screw the protective cover with the needle into the holder.
2. In order to better see or find the vein, place the arm in an extended position and apply a tourniquet.
3. Put on gloves – new ones for each patient.
4. Select the puncture location and disinfect it with 70° alcohol or isopropanol. Allow the disinfected area to dry.
5. Keep the patient's arm down.
Do not let the patient tighten his/her fist and fingers.
6. Remove the protective cover of the needle and make a puncture. During this time, the patient's arm is still down, but the tube is inserted into the needle holder by pushing the needle membrane and tube cap upwards.
Do not take samples from a vein in which medications are administered.

7. As soon as the blood appears in the tube, release the tourniquet – do not hold for more than 2 minutes.
8. If the blood does not enter the tube or the blood flow stops before the sufficient blood sample volume is reached up to the mark, the following measures are recommended:
    8.1. Adjust the needle position in the vein.
    8.2. Remove the tube and insert another tube.
    8.3. Take a new needle, a new tube and start everything from the beginning.
9. If the first tube is full and the blood flow is over, remove the tube from the holder.
10. Then, inserting the next tube in the holder, the needle membrane and the tube cap are again pierced, for the blood stream to enter the tube.
11. While the next tube is filling up, the previous one needs to be stirred, inverting it 8 times (including serum tubes – with red, yellow caps) but EDTA and heparin tubes 10 times. Do not shake.
12. As soon as the last tube is filled, the needle is removed from the vein, and a dry swab is applied to the puncture place. The swab should be pressed and held for 3 – 5 minutes until the bleeding has stopped.
13. The needle is removed from the holder and placed in an unbreakable container for destruction, but the gloves and needle holder are placed into a plastic bag for burning.

1.3. Blood sampling for inoculations in Bact/ALERT bottles.

It is advisable to draw blood inoculations before initiating antibiotic therapy. If this is not possible, then it is done before the next antibiotic dose. When taking blood inoculants it is prohibited to palpate the vein after skin disinfection.
1. Check the Bact/ALERT bottle label.
2. Make sure the fluid in the bottle is clear (there may be opalescence, precipitates).
3. Check the bottle sensor. It must be without any damage and bluish-green.
    Do not use bottles with a yellow sensor.
4. Remove the Bact/ALERT bottle protective cap and disinfect the rubber stopper with 70% ethyl alcohol for ~ 1 minute, allow the cap to dry for ~ 1 minute.
5. Perform the blood sampling as stated above.
    Blood inoculants may also be drawn with a syringe and, by piercing the disinfected bottle stopper, injected into the Bact/ALERT bottle.
6. Draw up to 10 ml blood for inoculants (for children, depending on the age, 0.5 – 4 ml in a Paediatric bottle. Before drawing blood mark the bottle label to ensure accuracy because the vacuum is not calibrated in bottles. First draw the blood in an aerobic bottle, and then if necessary, in an anaerobic inoculant bottle.
7. If the bottles of blood inoculants cannot be immediately placed in the Bact/ALERT analyzer, store them at +15 to +30 °C (room) temperature and within 48 hours at the latest put them in the analyzer. Keep away from direct sunlight.

8. The following information should be indicated on a tube and Bact/ALERT bottles:
A patient`s name, surname (do not write on bar codes, serial numbers).
Date of sampling for examinations – repeated examinations during the day.

It is prohibited:
To open the bottle by removing the cap before drawing blood.
To keep bottles in a refrigerator or thermostat.

1.4. Blood sampling and dispatch for alcohol detection.

For this purpose all materials must be collected, stored and sent for examination in accordance with the existing regulations of the Cabinet of Ministers – the procedure for checking the effects of alcohol, narcotic psychotropic or toxic substances if the examinations are carried out for expert examination.
In the place of venipuncture, the skin is treated with a non-alcoholic disinfectant, a furacilin solution (1:5000), rivanol solution (1:500).
Blood is drawn in a vacutainer tube with an anticoagulant – heparin (green cap).
Special referral sheets for drug and alcohol detection are in accordance with the existing regulations of the Cabinet of Ministers – the procedure for checking the effect of alcohol, narcotic, psychotropic or toxic substances if examinations are carried out for expert examination.
Store the tube in a refrigerator where the temperature is not higher than 4 °C.
Hermetically sealed tube (containing the patient's name, surname, personal identification number, blood sampling time, the blood collector's name and surname) is dispatched to the laboratory in a sealed envelope with all fields of the referral form filled, indicating the disinfection method used.
A laboratory assistant signs for the material received.

1.5. Blood sampling and storage for unstable test parameters in the blood.

If such tests are required, where parameters are not stable in the blood, then after blood sampling:
Vacutainers with a red cap are kept in a vertical position (in a stand) at room temperature until the blood coagulates (60 min), then it is centrifuged at 1300 – 2000 g for 10 minutes. The serum is siphoned into a clean tube (vacutainer with a red cap or Eppendorf tube. It is marked according to the primary tube.
Vacutainers with gel are held in a vertical position (in a stand) at room temperature until the blood coagulates (minimum 30 min), then it is centrifuged at 1300 – 2000 g for 10 minutes. The serum should not be poured out. Tubes are dispatched and stored at t° 0 as it is indicated below and in VD-20, VD-38, LIS until the performance of examinations.
Vacutainers containing Na citrate for the detection of VIIIF other factors are centrifuged at 2500 g for 15 minutes. Plasma is separated immediately from blood cells. The tube

is labelled with a label that is identical to primary tube label, , note "plasma" is added and the tube is send to the laboratory.
Homocysteine (HOMCIS), ECP (coagulated in a standardized way for 30 minutes and then centrifuged), zinc, potassium, lithium, C3, C4, LDH, iron, renin, BNP, gastrin, haptoglobin, acid phosphate, P, transferrin, soluble transferrin receptors - dispatch the tube for examination within one hour or separate serum (renin, BNP – plasma) from the clot as soon as possible.

Before drawing blood for STH analysis, the patient should have an empty stomach and should remain at rest for 30 minutes. The patient should have an empty stomach when drawing blood for HOMCIS, CPEPT and PTH analyses.

Growth hormones (IGF1, STH), parathyroid hormone (PTH), NH3 (ammonia), ACTH, calcitonin, homocysteine (only homocysteine can stay in ice water for 6 hours until centrifugation) – after blood sampling the tube is , placed in a bowl of cold water with ice cubes and dispatched to the laboratory at 366 Brivibas Avenue within one hour. Or, after the coagulation of blood – serum and plasma should be separated. The tube is placed in the "Cold transportation system" and dispatched to the laboratory within one hour, or is frozen and then dispatched on the examination day in the "Cold Transportation system".
5.6 If the above-mentioned types of serum, plasma preparation and transportation are not possible, the patient is sent for blood sampling to EGL, 366 Brivibas Avenue, Linezers, ARS or other branches.
5.7 Lactate should be withdrawn in a cold tube with NaF, then it is centrifuged for 15 minutes and sent to the laboratory within one hour, or frozen in the "Cold transportation system".
5.8 Renin. Blood should be drawn at room temperature in a tube with EDTA vacutainer with a violet cap). Then the tube should be dispatched at room temperature within one hour to the laboratory or it is centrifuged immediately and the plasma is separated. The separated plasma is frozen and sent to the laboratory in the "Cold transportation system".

1.6. Parameter stability and blood storage temperature and time.

The maximum permissible storage time is a time period that maintains 95% of the original content of the parameter to be determined. In the case of pathology, its stability can be greatly reduced.
The main causes of material deterioration are:

- ✓ Blood cell metabolism.
- ✓ Evaporation/sublimation.
- ✓ Chemical reactions.
- ✓ Impact of microorganisms.

- ✓ Osmotic processes.
- ✓ Light effect.
- ✓ Gas diffusion.

1.6.1. Stability of parameters for clinical chemistry and hormonal examinations.

Detectable parameter	Stability in the blood	Stability in serum/plasma		
	Room temperature	-20 °C	4 – 8 °C	20 – 25 °C
HBL cholesterol	2 days	3 months	7 days	2 days
ACTH	unstable (put in ice water immediately, freeze plasma immediately)	4 weeks	unstable	unstable (2 hours)
AMH	1 day	6 months	5 days	3 days
AFP	3 days	3 months	3 days	3 days
ALT	4 days	7 days	7 days	3 days
Albumin	7 days	3 months	1 month	7 days
Aldosterone	?	4 weeks	5 days	?
Androstendion	1 day	2 months	24 hours	1 day
Antistreptolysin	2 days	6 months	2 days	2 days
Apolipoprotein A1	1 day	2 months	3 days	1 day
Apolipoprotein B1	1 day	2 months	3 days	1 day
Bilirubin	2 days (related) 1 day (total)	6 months	7 days	19 days (related) 1 day (total)
Beta 2 microglobulin	3 days	2 weeks	7 days	3 days
BNP	1 day	9 months	24 hours	1 day
Anti CCP	2 hours	1 month	3 days	-
Cyfra 21-1CA 72-4	2 hours	6 months	4 weeks	-
CA 72-4	2 hours	3 months	30 days	-
Cyclosporine	6 hours	1 month	7 days	6 hours
CEA	7 days	6 months	7 days	7 days
Cyclosporine	6 hours	1 month	7 days	6 hours
C peptide	2 -3 hours	7 days	2 – 3 hours	2 -3 hours
Ceruloplasmin	11 days	3 months	2 months	11 days
Zinc	1 hour	1 year	2 weeks	1 week
CRP	3 days	3 years	8 days	3 days
High sensitive CRP	1 day	2 months	3 days	1 day
DHEA – SO4	2 days	2 months	2 days	2 days
Iron	2 hours	years	3 weeks	7 days
Erythropoietin	24 hours	2 months	7 days	2 days
Estradiol	20 hours	6 months	2 days	20 hours
Estriol	1 day	6 months	7 days	1 day
Ethanol (alcohole)	2 weeks	?	6 months	2 weeks
ECP	2 hours	3 months	7 days	8 hours
Ferritin	1 day	2 months	7 days	1 day
Folic acid (not light)	2 hours	1 month	2 days	2 hours
Phosphorus	1 hour		4 days	24 hours
Fructosamine	12 hours	2 months	2 weeks	3 days

Phosphorus	1 hour		4 days	24 hours
Fructosamine	12 hours	2 months	2 weeks	3 days
FSH	2 days	2 months	7 days	2 days
FT3	1 day	2 months	2 days	1 day
FT4	1 day	3 months	2 days	2 days
Gastrin	2 hours	30 days	4 hours	2 hours
GGT	1 day	years	7 days	7 days
Glucose (in venous blood plasma)	(with inhibitor for 7 days)	1 day (stabilised)	7 days (stabilised)	1 day (stabilised)
HbA_{1c}	3 days (EDTA in the blood)	6 months	7 days	3 days
Haptoglobulin	1 hour		3 months	3 months
HER-2		1 month	24 days	8 hours
HE4	5 hours	12 weeks	48 hours	5 hours
Free β HCG	8 hours	10 months	7 days	8 hours
HCG	1 day	2 months	7 days	1 day
Cholesterol	7 days	3 months	7 days	7 days
Homocysteine	1 hour	6 months	14 days	unstable
HIV	1 day	3 months	4 weeks	7 days
IGF1	unstable	12 months	24 hours	unstable
Immunoglobuline A	7 days	6 months	3 months	7 days
Immunoglobuline E	3 days	6 months	3 days	3 days
Immunoglobuline G	11 days	6 months	3 months	3 months
Immunoglobuline M	2 months	8 months	8 months	2 months
Insulin	8 hours	3 months	7 days	8 hours
Interleukin 6		6 months	1 day	
Calcium (total)	2 days	8 months	3 weeks	7 days
Calcitonin	unstable	15 days		unstable
Potassium	1 hour	1 year	1 week	1 week
Complex C3	1 hour	8 days	8 days	4 days
Complex C4	1 hour	?	2 days	2 days
Cortisol	2 hours	3 months	5 days	-
Creatinine	2 -3 days	3 months	7 days	7 days
Creatine kinase Creatine kinase MB	1 day	4 weeks (in the dark)	7 days	2 days
Lactate dehydrogenase	1 hour	6 weeks	4 days	7 days
LH	2 days	2 months	2 weeks	3 days
Lipase	7 days	1 year	7 days	7 days
Lithium	1 hour	6 months	7 days	1 day
Magnus	1 day	1 year	7 days	7 days
Sodium	4 days	1 year	2 weeks	2 weeks
Protein electrophoresis		3 weeks	7 days	1 day
Osteocalcine	8 hours	3 months	3 days	8 hours (in serum), 2 days EDTA in plasma
BCTX (EDTA in plasma)		3 months	5 days	1 day
PAPP-A	8 hours	3 months	3 days	8 hours
Parathormons	unstable	2 months	8 hours	unstable
Progesterone	2 days	3 months	7 days	2 days

Procalcitonin		6 months	2 days	
Prolactin	2 days	1 year	3 days	1 day
PSA	1 day	6 months	5 days	1 day
Renin	1 hour	1 month	unstable	?
Alkaline phosphatase	4 days	2 months	7 days	7 days
Alkaline phosphatase bone fraction	?	?	3 days	?
SHBG	2 days	2 months	7 days	2 days
Acid phosphate	1 hour		8 hours	2 hours
SCC	1 day	3 months	14 days	5 days
Selenium	2 days	1 year	2 weeks	1 week
Lead	7 days	?	?	7 days
STH (HGH)	unstable	2 months	8 hours	unstable
Testosterone	1 day	6 months	7 days	-
TSH receptor antibodies	8 hours	1 month	3 days	1 day
Tiroglobulin	2 days	2 months	3 days	2 days
T3	2 days	3 months	8 days	2 days
T4	7 days	1 month	7 days	2 days
Triglyceride	7 days	3 months	7 days	2 days
TNF	unstable	6 months	2 hours	
Troponin I	4 hours	1 month	24 hours	4 hours
Troponin T	8 hours	12 months	24 hours	8 hours
Urea	1 day	1 year	7 days	7 days
Transferrin-soluble receptors	1 hour	6 months	8 days	8 days
Uric acid	5 days	6 months	7 days	5 days
Valproic acid		1 month	2 days	8 hours
Vancomycin		1 month	2 days	8 hours
Vancomin		6 months	2 days	8 hours
Vitamin B12	unstable	2 months	2 days	2 hours
Vitamin B12 active	16 hours	3 months	3 days	16 hours
Vitamin D3 (25 OH)	no data	4 months	4 days	8 hours
LDL cholesterol	1 day	1 month	7 days	1 day

1.6.2. Stability of blood gases and electrolytes in the blood.

Detectable parameter	Stability in the blood at room temperature and tendency to change	Stability in the blood at 4 °C to 8 °C
pH	< 15 minutes ↓	2 hours
pCO_2	<15 minutes ↑	2 hours
Bicarbonates Base excess	<15 minutes ↓	2 hours
pO_2	<15 minutes ↑ or ↓	2 hours
Ionised calcium	15 minutes ↑	2 hours
Sodium	1 hour	1 hours
Potassium	1 hour ↑↑	1 hours
Chlorides	1 hour	1 hours

1.6.3. Stability of haematological parameters detectable in EDTA blood in EGL analyzers

Detectable parameter	Stability in the blood	
	20 – 25 °C	4 – 8 °C
Leukocyte formula	1.5 days	3 days
✓ Stabilized crude neutrophils	1.5 days	3 days
✓ Segment dipole neutrophils	1.5 days	3 days
✓ Eosinophilic	1.5 days	3 days
✓ Basophiles	1.5 days	3 days
✓ Monocytes	1.5 days	3 days
✓ Lymphocytes	1.5 days	3 days
RBC (Red blood cell)	1.5 days	3 days
Haemoglobin	1.5 days	3 days
Leukocytes	1.5 days	3 days
Reticulocytes	1 day	1 day
Platelets	2 days	2 days
Lymphocyte subpopulations	48 hours	not allowed
HLA–B27	24 hours	not allowed
CD 38	6 hours	
Anti-HLA-DR	6 hours	

1.6.4. Coagulometric parameter stability in the blood with Na citrate EGL methods.

Detectable parameter	Stability in the blood	Stability in plasma		
	Room temperature	– 20 °C	4 – 8 °C	20 – 25 °C
Antithrombin III	8 hours	1 month	2 days	8 hours
D-Dimer	4 hours	1 month	8 hours	4 hours
Factor II	24 hours	1 month		24 hours
Factor V	4 hours	1 month		4 hours
Factor VII	24 hours	1 month		24 hours
Factor VIII	4 hours	1 month		4 hours
Factor IX	4 hours	1 month		4 hours
Factor X	24 hours	1 month		24 hours
Factor XI	2 hours	1 month		2 hours
Factor XII	2 hours	1 month		2 hours
Factor XIII	4 hours	1 month		4 hours
Fibre monomers	24 hours	3 months		24 hours
Fibrinogen	8 hours			8 hours
APTL	4 hours	1 month		4 hours
Lupus anticoagulant	4 hours	1 month		4 hours
Protein C	8 hours	1 month		8 hours
Protein S	4 hours	1 month		4 hours
Prothrombin time	24 hours	1 month		24 hours
Thrombin time	4 hours			4 hours
Vwf Ag	8 hours	1 month		8 hours
Vwf activity	24 hours	1 month		24 hours

1.7. Capillary blood sampling

In some cases a capillary is used for the detection of haematological parameters and for the detection of certain biochemical parameters. Blood is obtained by making a puncture with a blood pin in the heel of a newborn or the fingertip of older children and adults.

Capillary blood sampling system with the appropriate color labels (EDTA – violet, glucose – yellow, grey, Na citrate – black) is used for capillary blood collection.

Blood sampling:

1. Prepare the necessary tools (disposable sterile scarifier, capillary blood sampling system, cotton wool, disinfectant).
2. Disinfect the planned puncture site, wipe with a tissue (remains of disinfectant may cause haemolysis).
3. Make a puncture with a sterile lancet.
4. The first drop of blood should be wiped off because it is diluted with tissue fluid.
5. Blood drops are collected with a haematocrit tube. The blood should flow freely – the tissues around the puncture should not be squeezed or massaged. To make it easier to get free blood flow, capillary blood should be drawn from warm tissue.
6. When a full capillary is collected, the collection system is turned round vertically until the blood flows out of the capillary.
7. When the required amount of blood is drawn, capillaries are removed from the system and placed in a disinfectant solution.
8. Close the container and immediately invert 5 times in order to mix the blood with an anticoagulant. Do not shake!
9. The scarifier is placed in an unbreakable container for disposal.
10. Wipe the puncture site with sterile cotton wool and cover with a plaster.
11. The container of the capillary blood sampling system is labelled in the same way as vacutainers.

1.8. Blood sampling for glucose test. Patient preparation.

1. To maintain normal eating habits (at least 150 – 200 g carbohydrates per day) for at least 3 days before the test.
2. At least 3 days before the test, to stop taking medications that may affect blood glucose (if this does not endanger the patient's state of health).
3. To continue the habitual physical activities, to avoid intense physical load or prolonged bed rest.
4. Women should not take the test during menstruation.

Test procedure.

5. The first blood sampling is performed in the morning when the patient has an empty stomach. The blood is drawn in a tube with a stabiliser NaF (grey cap, label – 1st tube).
6. After that, within 5 minutes, drink glucose dissolved in water.

7. During the test (2 hours after the glucose solution is taken until the next sampling) the patient should be in a peaceful atmosphere in the medical institution, without physical activity, eating and refraining from smoking.
8. The next blood sampling is performed after 120 minutes (label 2nd tube).
Blood sampling for pregnant women is performed after 60 minutes (label 2nd tube) and after 120 minutes (label 3rd tube).

Glucose solution preparation:
1. For adults 75 g of glucose is dissolved in 250 – 300 ml of water.
2. For children 1.75 g of glucose per 1 kg body weight is dissolved, but not more than 75 g in total.

1.9. Blood sampling for immunohaematological examinations.

1. There is no need for patients to be specially prepared.
2. Blood sample is required for IMH examination:
   - for the detection of EDTA blood group, Rh(D) affiliation, Rh-phenotype, DAT:
   - EDTA or without anti-coagulant for anti-human RBC antibody screening, antibody titre;
   - without anti-coagulant for Rh-phenotype, recipient's and donor's compatibility tests (ST);
   - EDTA and without anti-coagulant for the detection of antibodies VADC.
3. All blood samples should be labelled after blood sampling in the presence of the patient.

For stationary patients:
person's name, surname, date of birth, blood group (blood group, Rh(D) affinity, anti-epithelial antibody screening, ST, Rh-phenotype);
person's name, surname – DAT.
For ambulatory patients whose blood sampling has been performed outside the EGL laboratory:
person's name and surname;
information on pre-defined blood group can be found by Nr. LIS.
For ambulatory patients whose blood sampling has been performed at EGL:
EGL label with a bar code and number;
Information can be found on pre-defined blood group by Nr. LIS.
3.1. In some cases the blood sample may be incompletely labelled, for example, the patient is unconscious and without documents.
4. The accompanying document (laboratory examination request or ST application) is attached to the blood sample. The accompanying document must contain the blood sampling time, the blood group, the nurse's and doctor's signatures and the doctor's stamp. Blood samples for examination in VADC according to requirements.

5. Blood samples must be dispatched to the laboratory in isothermal containers (permissible temp. from +2 to +25 °C) in a vertical position as soon as possible, not later than 24 hours after blood sampling.
6. Blood sample is valid for testing for 48 hours, compatibility tests and anti-thyroid antibody detection VADC is valid for 24 hours.

1.10. Blood sampling for immunological examinations – HLA B27, Ly immunophenotyping, CD38, activated T lymphocytes (CD3+HLA-DR).

Blood is drawn in a separate EDTA tube.
Store and dispatch at +20 °C – 25 °C.
Blood samples should not be stored in the refrigerator or dispatched in cold weather.
These tests have a limited material storage time. HLA-DR, CD38 samples should be tested within 6 hours. Consequently, Anti-HLADR and CD38 can only be drawn at Riga reception points from 7.30 until 12.00. Samples should be dispatched to the laboratory by 13.00 (at the latest). HLA-B27 and Ly immunophenotype samples should be dispatched to the laboratory at 233 Brivibas Avenue within 24 hours. These tests can also be drawn outside Riga on Mondays, Wednesdays, Thursdays until the driver or post courier arrives. So the material cannot be drawn on Thursday afternoon, Friday, Saturday and HLA-B27 and Ly can be drawn at Riga reception points on Mondays, Tuesdays, Wednesdays, Thursdays all day, but in the morning on Friday (on Fridays the material should be at 366 Brivibas Avenue by 13.00).

1.11. Blood sampling for molecular biological DNA/RNA examinations.

1.11.1. CMV DNS, FBV DNS, Aspergillus DNS, HIV RNS, HSV DNS, VZV DNS.

Blood is drawn in the EDTA tube and dispatched to the laboratory within 6 hours, or in sterile conditions, plasma is separated in a sterile tube with a screw cap.
Separated plasma can be stored for up to 24 hours in a refrigerator at +2 °C – +8 °C but for longer storage, it is frozen at -20 °C to -80 °C. It is only allowed to unfreeze plasma once.

1.11.2. For HCV RNA, HBV DNA detection.

Blood is drawn in the tube without an anticoagulant (a red cap). The blood is dispatched to the laboratory within 2 hours. It should be stored at +2 °C – +25 °C.
For longer storage, serum is separated as in 1.11.1.

1.11.3. Borrelian for DNS storage.

Blood is drawn in the EDTA tube. It is delivered to the laboratory at +4 °C within 24 hours. It is frozen for longer storage at -20 °C to -80 °C.

1.11.4. BCR/ABL gene transcript, venous thrombotic risk alleles (II and V) for genetic predisposition analysis and celiac genetic preposition detection (DQ2DQ8).

The blood is drawn in the EDTA tube or PAXgene Blood RNA tube and delivered to the laboratory within 24 hours. Blood is valid for up to a week for the risk detection of venous thrombosis, or for BCR/ABL detection, 48 hours for PQ2DQ8 detection.

1.12. Material sampling for chromosomal examinations.

1.12.1. Bone marrow aspirates sampling.

VL-157

The bone marrow is drawn in a tube of Li-heparin, in which a medium is added in advance in sterile conditions.

The specially prepared tubes with a green cap are to be pre-ordered at the EGL cytogenetic department. 0.5 – 1 ml of the bone marrow is aspirated and added to the medium in each tube. If the number of cells in bone marrow is small, 3 tubes are required.

It is recommended so that the aspirate does not have a final exudate containing a large number of erythrocytes.

It should be immediately delivered to the laboratory at 366 Brivibas Avenue and stored at room temperature. It cannot be frozen.

1.12.2. Peripheral blood sampling.

VL-158

The peripheral blood is drawn in a Li-heparin tube (green cap). For an adult 2 ml is drawn, for newborns – 1 ml.

At least 5 ml of blood should be drawn for onco-haematological examinations, when the number of leukocytes is >15x 10*9/L and blasts >10%. In other cases the bone marrow should be sampled.

It should be immediately delivered to the laboratory at 366 Brivibas Avenue. It is stored at room temperature and cannot be frozen.

1.13. Blood sampling for parasitological examinations.

Blood sampling for the diagnostics of malaria and filariasis is performed in the EDTA tube at any time of day.

The tube must be delivered immediately to the laboratory at 366 Brivibas Avenue, as it should be examined within one hour.

Cytological analysis.

1.14. Blood sampling for Aspirin and Plavix detection with the VerifyNowSystem.

Each test requires blood sampling in to two Greiner Vacuette tubes with 3.2% Na Citrate. The material from the second consecutive tube will be used for examination. (Compulsory labelling).
Blood can be drawn at three EGL sampling points: 1 Zemitana Square, 5 Slokas Street and 366 Brivibas Avenue, on Tuesdays and Thursdays from 11.00 – 14.00.
It should be delivered (dispatched) to 366 Brivibas Avenue immediately, as it should be examined within one hour.
It is necessary for the patient to take Aspirin at least 2 – 5 days before blood sampling, and Plavix 75 mg for 5 – 7 days.

2. Urine.

2.1. Urine sampling for urine tests with spike test and for microscopy.

The morning urine portion is used for clinical urinalysis.
If necessary, any portion of the urine may be used during the day (with proper urine collection).

Urine is collected in a clean plastic container. It is desirable to use disposable plastic containers (may be obtained in the laboratory). Patients should wash themselves before urinating (see picture, information for patients), women should use a tampon to prevent adding vaginal discharge to urine before urinating. In order to exclude impurities, the first stream of the urine portion is poured out and the next one is taken for analysis.
Urine is poured into a special tube with a preservative – BD VACUTAINER Urinalysis, (a tube with a yellow – red cap which can be obtained at the laboratory free-of-charge) and can be delivered for examination within 24 – 28 hours. The urine tube is labelled (collecting urine outside the laboratory), indicating:

- ✓ Patient's name;
- ✓ Date and time of urine collection, when it is important for examination (e.g. repeated examinations during the day).

2.2. Urine sampling for clinical chemistry.

Urine for certain chemical reactions (see information in the Patient Manual VD-38, VD-20 or www.egl.lv) is collected in a clear tube (a tube with a yellow or light brown cap, obtained in the laboratory free-of-charge) or in a container without preservative.

2.3. Urine sampling for microbiological investigations.

Before starting treatment with antibiotics, collect the average urine portion in a special BD VACUTAINER bottle with boric acid (VACUTAINER C&S – a light green cap, obtained

in the laboratory free-of-charge) as indicated in section 2.1, for microbiological examination. It should be inverted 8 – 10 times and delivered to the laboratory within 48 hours. It can also be collected in a sterile container and delivered to the laboratory at 366 Brivibas Avenue within 2 hours.

2.4. 24-hour urine collection for clinical chemistry examinations.

1. In the morning the patient urinates but does not collect urine (see picture, patient information).
2. All further urine portions are collected in one container. They should be stored at 2 to 8 °C.
3. The next morning the patient urinates and urine is added to the previous portions.
4. The amount of 24-hour urine is measured and marked on the examination assignment.
5. Urine is mixed up in a container.
6. Urine is poured into a smaller container ~ 200 ml.
7. Name, surname, date, amount of 24-hour urine is written on the container and then taken to the laboratory.

!!! For the following examinations:

- ✓ adrenaline;
- ✓ dopamine;
- ✓ noradrenaline;
- ✓ vanilla manganic acid;
- ✓ methanephrine;

before urine collection, a preservative of 30 ml of 6N hydrochloric acid, which can be obtained in the laboratory, should be poured into the container.

2.4.1. Creatinine clearance.

Urine collected for 24 hours without a preservative (see 24-hour urine collection, patient information) is used for creatinine clearance detection.

The patient should ensure normal urine output during urine collection while taking an adequate amount of liquid. The patient should refrain from using intense physical exercise and urine release agents.

A blood sample should also be drawn from the vein to determine creatinine clearance. Blood should be drawn in a tube without anticoagulants (Vacutainer® with a red cap).

To be delivered to the laboratory:

1. 5 to 10 ml of collected urine, indicating the collection period and the amount of urine collected. If it is impossible to measure the amount of urine, all the urine collected should be delivered to the laboratory.
2. Blood tube.
3. Information on the patient's height and weight.

2.5. Urine collection and dispatch for alcohol and drug detection.

All materials must be collected, stored and sent for examination in accordance with the existing regulations of the Cabinet of Ministers – the procedure for checking the effects of alcohol, narcotics, psychotropic or toxic substances if the examinations are carried out for expertise.

15 – 20 ml urine is collected without preservatives in a clean glass or plastic container (leaving as little air as possible above the surface of the sample) under the supervision of a medical practitioner. The container is immediately covered up and marked indicating the patient's name, surname, personal identification number, sampling time and the medical practitioner's name and surname.

The container is placed in a sealed envelope with a referral, in which all the columns are filled in.

Special referral sheets for drug and alcohol detection are in accordance with the existing regulations of the Cabinet of Ministers – the procedure for checking the effects of alcohol, narcotic, psychotropic or toxic substances if examinations are carried out for expertise.

Urine is stored in a refrigerator where the temperature is not higher than 4 °C or frozen and stored in the refrigerator freezer.

A laboratory assistant signs for the material received.

2.6. Urine collection for DPD detection.

DPD (deoxypyridoline): urine is collected without preservatives, the first or the second urine portion is collected before 10.00 o'clock.

2.7. Urine collection for Beta2 macroglobulin detection.

First of all, empty the bladder, then drink ~ 200 ml of water, collect the urine sample without preservatives within an hour. ~ 10 ml of urine should be delivered to the laboratory.

2.8. Urine collection for mercury, chromium, iodine detection.

20 ml of morning urine portion in a closed container with a screw cap is necessary for the examination.

2.9. Urine sampling for molecular biological Chlamydia trachomatis and Gonorrhoea RNS detection.

The first urine blast is required for an examination which is collected in a clean container without additives (about 20 – 30 ml). It is advisable to take the container to the laboratory immediately after collection. If it is not possible, store at 2 – 8 °C for no longer than 24 hours or freeze at -20 to -80 °C, or pour into Gen probe Aptma Urine Collection tube (2 ml of urine is filled in a transport tube by using the pipette included in the set. The urine level in the tube should be between the black arrows).

Urine sampling for the performance of molecular and biological detection of several sexually transmitted pathogens (STIP)

The initial stream urine is required for examination, which is collected in a clean container without additives (about 20 – 30 ml, or at least 8 ml). If it is not possible, store it at 2 – 8 °C for no longer than 24 hours or freeze at -20 to -80 °C.

2.10. Stability of parameters in urine.

Parameters	Stability in urine			Affecting factors
	-20 °C	4 – 8 °C	20 – 25 °C	
Albumin	6 months	1 month	7 days	Physical load
Alpha amylase	3 weeks	10 days	2 days	Renal function
Ethanol	?	30 days	?	
Glucose	2 days	2 hours	2 hours	Pregnancy, diet, age
B2 microglobulin	2 months	2 days		
DPD	for a long time	7 days	1 day	
IgG	7 days	1 month	Unstable	
Calcium	3 weeks	4 days	2 days	Nutrition, bad rest
Cocaine	4 months	3 weeks	?	
Creatine	6 months	6 days	2 days	Age, food (meat), muscle mass
LSD	2 months	1 month	1 month	
Morphine	1 year	?	?	
pH	unstable			Nutrition
Protein	1 month	7 days	1 day	Physical load, pregnancy
Urea	1 month	7 days	2 days	Protein–rich nutrition, infusion (amino acids)
Uric acid	unstable		4 days	Physical load, nutrition
Vanillylmandelic acid	years	7 days (if pH is 3 – 5)		

2.11. Urine collection for parasitic examinations – schistosomiasis diagnostics.

Urine should be collected in a clear container during the day or within 24 hours and ~ 20 ml of the urine sediment (clear urine layer should be drained off – precipitate) should be brought to the laboratory.

2.12. Urine collection for cytological examinations.

After collection urine should be dispatched immediately to the laboratory or centrifuged for 10 minutes at a speed of 400 – 600 g. The sediment should be plated in a thin layer on the slide, dried in a reclined position, labelled and then dispatched to the laboratory.

3. Sputum

3.1. Sputum sampling for sputum analysis, for acid-resistant-bacteria analysis.

The material that is released only by expectorating should be collected in a clean container with a lid.
To avoid an oral cavity content admixture, it is necessary to rinse the oral cavity with clean water before the collection of sputum.
The first sputum portion is usually sufficient for sputum analysis – it is lung secretion accumulated all night long. Salivary and nasal secretions should be avoided.
Sputum is collected in a clear, dry container with a wide opening and a cap that closes the container tightly.
Sputum should not be on the outside of the container.
Sputum is stored in the refrigerator at 2 – 8 °C until examination.

3.2. Sputum collection for microbiological and cytological examination.

Only freshly collected sputum, as indicated in 3.1, is used for microbiological and cytological examination. It should be dispatched to the laboratory within 1 – 2 hours.

3.3. Sputum collection for pneumocyst diagnostics.

Sputum is collected, as indicated in 3.1, or the material is obtained by performing a bronchoscopy. The material for examination should be dispatched immediately.

4. Nasal secretion.

Material for examination is taken by a doctor. The material is collected on the slides, dried at room temperature. The slides are labelled with the patient's identification data, corresponding to the data on the request for laboratory examinations. The slides, together with the request, are placed in a container and delivered to the laboratory.

4.1. Collection of nasopharyngeal smears, aspirates, bronchoalveolar lavage for molecular biology tests (RIPV, RIPVB) and the flu express test (GRIPAGT).

The material is collected in a special container with preservative solution in COPAN culturette.

5. Stool

5.1. Stool sampling for the co-programme.

1. Stool should be fresh (no later than 12 hours after collecting). It should be stored in the refrigerator until dispatching to the laboratory.

2. About 10 g of stool is placed in a glass or plastic container taking up 1/3 of the volume.
3. Stool for testing is taken from different places of one portion (from the top and deep). Correct collection of all factions of heterogeneous material from stool containing large amounts of mucus is especially important!
4. The stool sample should be without urine admixture, should not be collected after an enema, after taking medication that changes the stool composition (barium, iron, bismuth preparations, diarrhoea agents).
5. To judge the functional abilities of the digestive tract following the results of the co-programme, the patient should consume varied food and fats.

5.2. Stool sampling for occult blood detection.

Stool is collected as indicated in the co-programme.
Sampling should not be performed during menstruation (also 3 days after), haemorrhoids and other known or apparent bleeding. There are no restrictions on the use of diet and medication. However, it is advisable to abstain from alcohol, aspirin and other irritating and bleeding agents before occult blood detection.

5.3. Stool sampling to examine parasitic eggs, protozoon vegetative forms and cysts.

Stool is collected, as indicated in 5.1.
Three re-examinations are desirable to examine parasitic eggs, protozoon vegetative forms and cysts, sampling stool special container, which can be obtained in the laboratory.

5.4. Stool sampling for calprotectin detection.

Stool is collected in a clean, dry container, avoiding contamination with toilet disinfectants and dispatched immediately to the laboratory.

5.5. Perianal scrape for enterobiasis detection.

A tube with a cotton wool applicator for material removal can be obtained in the laboratory. The material should be taken before washing oneself in the morning.
1. In the morning, after the child's wakes up, moisten the cotton wool at the tip of the applicator with warm water.
2. Anal verge should be cleaned up with moistened cotton wool by rolling the applicator.
3. The applicator is placed in a tube by closing the tube with clean cotton wool, and dispatched to the laboratory on the same day or the next morning.

5.6. Stool sampling for microbiological examination.

The material for examination should be taken before antibacterial therapy.
1. Stool sampling is performed immediately after defecation, as indicated in 5.1, and dispatched to the laboratory within one hour.

2. A special tube with the AMIES transport medium can be obtained in the laboratory, allowing the examination time to be extended by up to 24 hours:
As much stool as possible should be taken from the portion in different places and the tampon with stool is placed in the transport medium.
3. To detect Salmonella, Shigella, the stool should be dispatched immediately to the laboratory or a Cary Blair transport medium can be used.

5.6.1. Stool sampling to detect Adenovirus Ag, Rotavirus Ag, Giardo lamblia Ag, Yesinia, Aeromonas, Clostridium difficile toxin.

Fresh stool is necessary.
About 10 g of stool is placed in a glass or plastic container and delivered to the laboratory within one hour.

5.6.2. Stool sampling for Campylobacter detection.

Stool should be collected immediately after defecation in a special Port A Cul medium that can be obtained in the laboratory.

5.7. Stool sampling for molecular biology tests – ZTP, GEP, GEPB.

There is a special container for stool sampling, which can be obtained in the laboratory (stool container). Stool should be stored at +2 to+ 8 °C or frozen until being dispatched to the laboratory.

6. Material dispatch for histology.

Histologically examined samples (surgery and biopsy) should be fixed with 10% formalin solution.
Drying or storage in saline solution is inadmissible.

7. Sampling and dispatching of serous cavity (transudates, exudates) for examination.

1. Puncture is performed by the doctor.
2. Serous cavity liquid is collected in dry (to prevent lysing of the blood cells) clean containers and is delivered immediately to the laboratory.
3. To prevent coagulation, an anticoagulant, heparin or Na citrate (1 drop per 5 ml of material) is added to the test material.
4. If there is a lot of material, then the last portions are dispatched to the laboratory.
5. The patient's name, surname and the type of the material is labelled on the container.
6. If serious cavity liquid after puncture for cytological examination cannot be dispatched immediately to the laboratory, it should be centrifuged for 10 minutes at a speed of 400 – 600 g. The sediments should be plated in a thin layer on the slide and should be dried and labelled in a horizontal position.

7.1. Serous cavity collection for Borrelia burgdorferi DNA examination.

1. Puncture is done by the doctor.
2. Serous cavity liquid is collected in dry (to prevent lysing of the blood cells) containers.
3. The patient's name, surname and the type of material is labelled on the container.
4. It should be dispatched to the laboratory at 4 °C within 24 hours. It is frozen for longer storage at -20 °C to -80 °C.

8. Cerebrospinal fluid sampling for examination and storage.

1. Puncture to get cerebrospinal fluid is performed by the doctor.
2. Cerebrospinal fluid is collected in a dry and clean tube. Collection of larger quantities of cerebrospinal fluid can cause severe complications. Wet tubes can cause lysing of liquor formelements.
3. The sealed tube is delivered to the laboratory at 366 Brivibas Avenue immediately after the puncture, because the collapse of formelements begins after 3 minutes. There is always a cytological examination.
4. The cerebrospinal fluid should be analyzed immediately after receiving it.
5. Cerebrospinal fluid is delivered to the laboratory together with a request for laboratory examination indicating the date, the time of puncture and the sender's name.

6. It is also necessary to analyze the cerebrospinal fluid on the BC, then it is collected in 2 tubes of 5 ml each.
7. Tubes are labelled with the patient's name, surname and the type of material.

8. The microbiological examination sample should be taken in a sterile container of at least 2 ml in the second tube and dispatched immediately to the laboratory at 633 Brivibas Avenue.
9. Cytology examination should be carried out in the same way as for dispatching serous cavity liquid.
10. Stability of parameters detected in the cerebrospinal fluid.

Parameter	Stability		
	– 20 °C	4 – 8 °C	20 – 25 °C
Albumin	1 year	2 months	1 day
Glucose		months	3 days
LgG	7 days	1 day	unstable
Lactate	months	24 hours	3 days
Leukocytes		3 – 5 hours	1 – 2 hours
Protein total	1 year	6 days	1 day

8.1. Cerebrospinal fluid collection for Borrelia burgdorferi DNA examination.

1. Puncture is performed by the doctor.
2. Serous cavity liquid is collected in dry (to prevent lysing of the formelements) containers.
3. The patient's name, surname and the type of material is labelled on the container.
4. It should be dispatched to the laboratory at 4 °C within 24 hours. It is frozen for longer storage at -20 °C to -80 °C.

9. Sampling and dispatching for fungal bacterioscopy and microbiological diagnostics.

It is recommended for the patient, before sampling for microscopic and bacteriological examination, not to wash the skin with soap, not to use cosmetics and not to use local therapies for 2-3 days.
The sample should be taken from fresh, but already typically developed damage foci, where fungal elements are found in greater concentration.

- The material is taken from the hairy part of the scalp with sterile tweezers by pulling out 3 - 4 damaged hairs (split or with a changed colour and shape) then, with a disposable scalpel, skin scales are scraped from the damaged area. In the case of infiltrative - purulent forms, hair and skin scales should be taken in the periphery of the damage foci.

- The material from the smooth skin is taken from the periphery of fresh, foci of damage with clear-cut contours by using a disposable scalpel, scraping off the skin scales and pulling out the fluffy hair with sterile tweezers.
- The material for superficial nail lesions is scraped with a disposable scalpel; thickened nails are cut with scissors or pliers. Material for Demodex folliculorum bacterioscopic diagnostics is taken from the characteristic places – the nose, lip folds, chin, between the eyebrows, as well as from other places where there is a rash, from the outer ear passage and eyelashes.
- The material from the skin is taken with a disposable scalpel by scratching the skin scales, squeezing out the content of sebaceous glands and rash and pulling out the skin hair with sterile tweezers.
- Eyelashes are pulled out with sterile tweezers (4 – 6 eyelashes from each eyelid).
- The material from the outer ear passage is taken with a sharp spoon by scraping the skin scales.

The material is placed in disposable small petri containers or in closed containers (can be obtained at the laboratory) labelled and dispatched to the laboratory with a request for laboratory examination, indicating the type of material and the place of sampling.

Smears on slides should be prepared for materials of mucous consistency.
At the request of the laboratory, the patient's identification data and the number corresponding to the date and the number should be written with a pencil on the polished part of each slide.
Several slides with smears are dispatched to the laboratory in a closed box with the institution's label. One patient's smear slides are placed in a separate container, or envelope.
Material should not be placed between slides, as it is potentially infectious.
The transport system (AMIES) is used for material smears.

10. Sperm sampling for clinical and microbiological examination.

There is no special room for sperm sampling at E. Gulbis Laboratory.
Sperm sampling can be performed:

a) At home. **The sample is delivered to the sample reception point at 366 Brivibas Avenue** on working days from 8.00 to 12.00, one hour after collection, keeping the sample at 20 °C to 40 °C.
b) At the premises of "Embrions" Ltd at the Clinic "Linezers" calling 67543350 in advance.

1. It is necessary to refrain from sexual intercourse for a period of 3 days before sperm sampling for clinical examinations.

2. Before sperm sampling it is advisable to urinate and wash the genitals so that the bacteria from the external genitalia do not interfere with the sperm.
3. The sperm is collected by masturbation. It cannot be collected in a condom.
4. The sperm (all ejaculate) is collected in a container that can be obtained at a laboratory or purchased at a pharmacy.

11. Smears.

11.1. Smear sampling for the detection of trichomonosis, gonorrhoea, gardnerellosis and fungi.

For men

1. Discharge or mucous membrane scrapes from the following are examined:
   - ✓ The urethra;
   - ✓ Other localizations (anal opening, etc.)
2. Before sampling material from the urethra, it is recommended that the patient abstain from urination for 4 – 5 hours.
3. The area of the urethra should be washed with a sterile swab soaked in physiological saline. The first free-flowing drops are washed away, but the next are applied to the slide and smears are prepared.
4. If there is little or no discharge, urethra massage is performed and mucous membrane scrapes are taken.
5. After placing the material on the slide, it should dry at room temperature. The patient`s identification data and number, corresponding to the number and data on the request, are written with a pencil on the faceted surface of each slide.
   Several slides with smears are dispatched to the laboratory in a closed box with the institution's label. One patient's smear slides are placed in a separate container, or envelope.

For women

1. The material is taken from:
   - ✓ Urethra;
   - ✓ Cervical canal;
   - ✓ Vagina.
2. Before sampling material from the urethra canal the orifice is wiped with a dry, sterile cotton swab. The material is taken at a depth of 1.5 – 2 cm.
3. Material sampling from the vagina is performed after inserting the gynaecological mirror. If there is a lot of discharge it is wiped off with a dry cotton swab. Scrapes are carried out with a material sampling brush or a spatula.
4. The cervix is wiped with a dry cotton swab. A material sampling brush is inserted in the cervical canal at a depth of 1 cm and the sample is taken from the walls.

5. After placing the material on the slide, it should be dried at room temperature.
6. Two smears are necessary for examination.
7. The patient`s identification data and number, corresponding to the number and data on the request, are written with a pencil on the faceted surface of each slide or a special label from the special sampling and labelling set is attached.
   One patient's smear slides are placed in a separate container, or bag.

Special material sampling and labelling set is provided upon request for **gynaecologists.**
Material labelling and delivering procedure can be found in 11.3 - Sampling for cytological examination of gynaecological material.

11.2. Prostatic secrete sampling.

Sampling for examination is taken by the doctor. The material is collected on slides and dried at room temperature. The slides are labelled with the patient's identification data, corresponding to the data on the request for laboratory examinations, are placed in a container and together with the request, dispatched to the laboratory.

11.3. Blood sampling for cytological examination of gynaecological material

Each doctor is provided with the material necessary for material sampling and labelling at a customer's request (VL-34):

- a separate bag for each patient for dispatching the sample and request,
- dispatch,
- 8-label set for each patient with a unique, unrepeatable identification number and barcode,
- Rovers Cervix Brush for material sampling,
- slides,
- containers for dispatching slides,
- sample collection container with preservative (for papillomavirus detection and/or cytological testing of the material – "liquid" cytology),
- dispatch.

I BEFORE SAMPLING:

- The referral should be filled in accurately.
- Labelling: one 8-label set is provided for one patient.
- Separate labels are marked with: referral, each slide, container with a preservative.

The label should be pasted on the faceted slide edge.
If several samples are taken and there is a lack of labels then the unlabeled slides should be marked with a pencil, with the same number as on the label.

- ✓ Unused 1st set labels are not applicable to the labelling of another patient's material. They should be thrown out!

II SAMPLING:
Gynaecological preparations for cytological examination are prepared as T-shaped smears, where the perpendicular smear is from the posterior vault of the vagina but the parallel smear is taken from the outer opening of the cervix.

100045 (label No.)	Posterior vaginal vault	Cervix

- The smear material is plated on the faceted surface where the label is attached.
- If the material is taken from the cervix and put in a container with a preservative to detect papillomavirus, then cytological testing from the same material can also be carried out. It is only necessary to write in the request – liquid cytology.

Slides should be placed in special containers.

It is advisable to take materials for cytological examination:

- ✓ During endovaginal treatment;
- ✓ At IUD evacuation;
- ✓ Earlier than 3 weeks after electrocoagulation;
- ✓ Earlier than 6 weeks after electrical excision of the cervix;
- ✓ Pre and post the first menstruation, and after abortion;
- ✓ Sooner than 2 months after childbirth.

III AFTER SAMPLING:
The request for examination, containers with slides and a container with a preservative should be placed in a bag, closed and dispatched to the laboratory.
One patient – one set of labels, one bag.

11.4. Smear sampling for papilloma viruses and/or fluid cytology.

Gynaecological material for liquid cytology (including papilloma viruses) is collected with a Rovers Cervex Brush. After collection of the sample, the head of the Rovers Cervex Brush is removed and placed in a sample collection bottle Sure Path with a preservative. When using a brush with a non-removable head, it should be vigorously stirred in a preservative liquid 10 – 15 times to remove the collected cells.

11.5. Sampling to determine Chlamydia trachomatis/Neisseria gonorrhoeae by DNA/RNA hybridisation test (GEN PROBE).

Sampling for both men and women is performed using the Unisex Swab specimen collection kit (**Gen-Probe-APTIMA**).

For men:
Patients are advised to refrain from urinating for at least 1 hour before material collection.

1. Before material sampling mucous is removed with the white swab from the collection kit.
2. The blue swab is entered in the urethral canal at a depth 3 to 4 cm and the material is collected by rotating the swab.
3. The blue swab is pulled out and placed in the Gen/Probe tube, it is broken off at the mark.
4. The tube should be properly fixed.
5. The tube can be stored at room temperature for up to 60 days.
6. The tube is labelled, indicating the patient's identification data and the request for laboratory examination.

For women:

1. Before material sampling with the white swab in the collection kit, mucous from the cervical canal is cleaned.
2. The second blue swab is inserted in the cervical canal at a depth of 1 – 1.5 cm and the material is collected by rotating the swab.
3. The blue swab is removed without touching the vaginal mucous.
4. The blue swab is inserted in the Gen/Probe tube and broken off at the mark.
5. The tube is properly fixed.
6. The tube can be stored at room temperature for up to 60 days.
7. The tube and the request for laboratory examination, indicating the patient`s personal data, are labelled.

11.6. Smear sampling to determine Ureaplasma urealyticum and Mycoplasma hominis.

The material is taken in the AMIES transportation system before starting antibiotic therapy.
Vaginal material.

- ✓ Mucous is removed from the cervix canal;
- ✓ The material is taken from the interval cervical canal (for pregnant women only from the outside) with a swab.

Urethral canal.

- ✓ The orifice is cleaned;
- ✓ The material is taken from the mucous membrane with a swab by rotating it (at least 3 hours after the last urination).

Other materials.

- ✓ Some ml of sperm is collected in a clean tube or container or in a tube with a preservative (Vacutainer C&S – a light green cap) and dispatched to the laboratory.
- ✓ Some ml of the first urine portion is collected into a clean tube or container, or in a tube with a preservative (Vacutainer C&S – a light green cap) and dispatched to the laboratory.

The transport system (is labelled UROTUBO, CULTURETTE) is dispatched to the laboratory within 24 hours.

11.7. Smear sampling for microbiological examinations.

The transport system with the AMIES medium is used.

1. Take out a swab from the kit.
2. The material is taken from the area to be examined (skin, external ear canal, throat, wounds, urethra, vagina, etc.).To determine C. diphtheriae, the smear is taken in a separate AMIES transportation medium.
3. The swab is placed in a tube with a medium.
4. The tube and the request for laboratory examinations are labelled indicating the patient's personal data, the type of material and the place of sampling (skin, external ear canal, throat, wounds, urethral canal, vagina, etc.).
5. The tube is stored at room temperature (15 to 25 °C) for up to 24 hours until dispatched to the laboratory.

11.8. Smear sampling for molecular biological examinations of A/B influenza virus.

The material should be taken from the nasal passages, or the throat or the urethral canal (for SST-Strip) panel detection) with a special Copan Swab removal transport system that can be obtained at the laboratory:

1. Open the sterile system.
2. Take the material with a cotton swab, trying to collect epithelial cells as much as possible.
3. Place the cotton swab with material in the transport system.
4. Label Copan Swab and referral according to the patient.
5. The sample should be dispatched to the laboratory in the transport system within 5 days and kept at 2° to 25 °C.

When collecting materials, the usual precautions should be taken when dealing with potentially infectious materials, such as disposable gloves, mask and work clothes.

12. Dispatch of various materials for cytological examinations.

The reference to the cytological examination should include the patient's personal data, the medical institution, the doctor who prepared the material, where it was taken from, in what way the material was removed – as a smear or with a preservative and the date, as well as corresponding anamnesis, a diagnosis, a doctor's signature, a stamp.

12.1. Dispatch of aspirates from the cervix, uterine cavity and the pouch of Douglas.

Aspirates from the cervix, uterine cavity and the pouch of Douglas and others are placed on several slides in a thin layer, completely dried, and placed horizontally.

12.2. Dispatch of mammary gland excretions for cytological examinations.

Pathological mammary gland excretions which are not related to location, pregnancy or recent abortion of normal duration are placed in a thin layer on degreased slides and completely dried. R or L is written on the label, according to the side the material is taken from, and the same is written on the request for laboratory examination.

12.3. Dispatch of prints and scrapings from the skin and mucous membrane.

The material is removed and smear preparation is performed by a doctor-clinician. The material is taken after the removal of necrotic masses by a blunt instrument mildly scraping or imprinting. The obtained material is evenly distributed on slides in a thin layer. A clean, degreased slide is pressed firmly against the damaged surface for imprints. The preparations are dried at room temperature, and are labelled with a barcode number.

12.4. Dispatch of fine needle aspiration material for cytological examinations.

Preparation of fine needle aspiration material is performed during the procedure. The aspiration material is prepared both on the slide and preservative liquid, to obtain as many cells as possible. When preparing the smear, one drop of the material from the needle is slowly dripped on one side of the slide and by applying light, well-balanced, even pressure to the smooth surface of the second slide, the material is evened over the entire slide, achieving a thin and levelled smear. Smears are dried in the air. The remaining content of needle aspiration material is carefully injected in the bottle with

a preservative, rinsing the syringe several times in preservative liquid, by drawing the liquid into the syringe and releasing it into a collection container 5 or more times. The reference to the cytological examination should include the patient's personal data, the medical institution, the doctor who prepared the material, when it was taken, in what way the material was removed – in the form of a smear or with a preservative and the date, as well as corresponding anamnesis, a diagnosis, a doctor's signature, a stamp.

13. Collection of amniotic fluid, chorion, skin biopsy for chromosomal examinations.

13.1. Collection of amniotic fluid.

1. Material for FISH and/or standard cytogenetics is collected in a sterile syringe with a plastic plunger).
2. Amniotic liquid is recommended for standard cytogenic examination and FISH analysis:

Pregnancy weeks	Total amount (ml)	Quantity for standard cytogenetic analysis (ml)	Minimum amount for FISH analysis (ml)
11 - 15	14	12	2
16	16	13	3
17	16	13	3
18	18	14	4
19	18	14	4
20 - 27	20+	15+	5+

Brownish or bloody amniotic liquid with FISH cannot be analysed because it can contain maternal blood.

13.2. Collection of chorionic villi and skin biopsy.

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A little piece of chorionic villi tissue or skin should be placed in a centrifuge tube, containing:
5 ml of RPMI-1640 medium supplemented with antibiotics (penicillin 10,000 units/ml, streptomycin 10,000 µg/ml and heparin (5,000 units/ml). Tubes can be ordered and received at the E. Gulbis laboratory.
Chronic villi material should not be contaminated with maternal tissue.
The material is dispatched to the laboratory within 24 hours and stored in a refrigerator at 4° to 8 °C. It cannot be frozen.

14. Sampling of tumour material for the detection of BRAF, KRAS and EGFR gene mutations.

KRAS/BRAF (KRBR test) requires a tissue sample of colorectal tumour that is fixed in formaldehyde and embedded in a paraffin block A BRAF (BRAF test) requires a melanoma tumour or a metastasis tissue sample fixed in formaldehyde and embedded in paraffin block

15. Transportation of testing material to the laboratory.

1. Until dispatching, the samples should be stored in conditions required for the testing material and the type of test: in the refrigerator or at room temperature (see in the laboratory Patient Guide NO-02, VD-38 or www.egl.lv).
2. Samples are transported in such a way that guarantees the safety of the courier, all of the public and the recipient's laboratory – special thermal insulation bags are used that protect samples from qualitative changes (due to chemical, physical and mechanical exposure).
3. Requests for laboratory tests are placed in folders, separate from the material to be examined.
4. In the laboratory test material transportation has been developed and agreed with customers according to the schedule. The test material is accepted in urgent cases round-the–clock (see admission times in the Patient Guide or at www.egl.lv).

16. Refusal criteria for examinations.

1. Insufficient or incorrect patient identification (for example, the patient's name and surname do not match the name on the request and the testing material label).
2. Sample is taken improperly: smear material is placed in a non-sterile container, in the wrong medium, the blood is drawn in a non-matching colour labelled tube, material for histological examination is without formalin, etc.
3. The sample is not labelled.
4. Inaccurately taken material: incorrect blood/anticoagulant ratio, insufficient amount.
5. The data of sampling is unknown, or the time, if two samples are taken per patient per day.
6. There is evident sample contamination or changes in quality – haemolysis, hyperlipidaemia.
7. Inappropriate sample storage, transportation (The serum for unstable parameters should be separated from blood cells within an hour).

17. References.

11.1. Samples: From the Patient to the Laboratory, W.G. Guder, 1996, ISBN 3-928862-22-6;
11.2. Urinalysis and Collection, Transportation and Preservation of Urine Specimens; Approved Guideline, NCCLS, 1995, Vol. 15 No. 15;
11.3. Collection, Transport, and Processing of Blood Specimens for coagulation Testing and General performance of Coagulation Assays; Approved Guideline - Third Edition, NCCLS, 1998, ISBN 1-56238-363-9;
11.4. Methods for the Erythrocyte Sedimentation Rate (ESR) Test- Third Edition; Approved Standard, NCCLS, 1993, ISBN 1-56238-198-9;
11.5. Procedures for the Collection of Diagnostic Blood Specimens by Venipuncture; Approved Standard - Fourth Edition, NCCLS, 1998, ISBN 1-56238-350-7;
11.6. Manufacturer's methodologies.