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Петрашенко Вікторія Олександрівна
Зміст

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**Topic 14: “INVASIVE METHODS OF PREGNATAL DIAGNOSIS”**

1. **The general aim** - to know the main invasive methods for prenatal diagnosis of inherited pathology.
2. **Student must know:**
   - The tasks and possibilities of invasive methods of prenatal diagnosis
   - Principles of methods, indications and contraindications for different types of invasive methods
3. **Student must be able:**
   - To choose invasive methods for prenatal diagnosis of inherited pathology.
   - To treat the results of different invasive tests in prenatal diagnosis.

4. Plan of conducting of studies

| Introduction | Classroom | 5 min |
| Control and correction of initial level of knowledges | Computer class | 10 min |
| The tasks and possibilities of invasive methods of prenatal diagnosis | Classroom | 20 min |
| Principles of methods, indications and contraindications for different types of invasive methods | Classroom | 25min |
| Demonstration of invasive methods in prenatal diagnosis | Computer class | 10 min |
| Educational control and correction of level of knowledges | Classroom | 5 min |
| Conclusion | Classroom | 5 min |
Invasive methods of prenatal diagnosis
There are a variety of invasive techniques available for prenatal diagnosis. Each of them can be applied only during specific time periods during the pregnancy for greatest utility. The invasive techniques employed for prenatal diagnosis include:

- Amniocentesis
- Chorionic villus sampling
- Cordocentesis
- Fetoscopy

CHORIONIC VILLUS SAMPLING (CVS)
In this procedure, a catheter is passed via the vagina through the cervix and into the uterus to the developing placenta under ultrasound guidance. Alternative approaches are transvaginal and transabdominal. The introduction of the catheter allows sampling of cells from the placental chorionic villi. These cells can then be analyzed by a variety of techniques. The most common test employed on cells
obtained by CVS is chromosome analysis to determine the karyotype of the fetus. The cells can also be grown in culture for biochemical or molecular biologic analysis. CVS can be safely performed between 9.5 and 12.5 weeks gestation.

**Indications**

Possible reasons for having a CVS can include:

- Abnormal first trimester screen results
- Increased nuchal translucency or other abnormal ultrasound findings
- Family history of a chromosomal abnormality or other genetic disorder
- Parents are known carriers for a genetic disorder
- Advanced maternal age (maternal age above 35). AMA is associated with increase risk of Down's syndrome and at age 35, risk is 1:400. Screening test are usually carried out first before deciding if CVS should be done.

![Fig. 1. A diagram of the technique of transvaginal chorionic villus sampling.](image)

CVS has the disadvantage of being an invasive procedure, and it has a small but significant rate of morbidity for the fetus; this loss rate is about 0.5 to 1% higher than for women undergoing amniocentesis. Rarely, CVS can be associated with limb defects in the fetus. The possibility of maternal Rh sensitization is present. There is also the possibility that maternal blood cells in the developing placenta will be sampled instead of fetal cells and confound chromosome analysis.

**Amniocentesis**

Amniocentesis is a procedure used to diagnose fetal defects in the early second trimester of pregnancy. A sample of the amniotic fluid, which surrounds a fetus in the womb, is collected through a pregnant woman's abdomen using a needle and syringe. Tests performed on fetal cells found in the sample can reveal the presence of many types of genetic disorders, thus allowing doctors and prospective parents to make important decisions about early treatment and intervention.

Since the mid-1970s, amniocentesis has been used routinely to test for Down syndrome, by far the most common, nonhereditary, genetic birth defect, affecting about one in every 1,000 babies. By 1997, approximately 800 different diagnostic tests were available, most of them for hereditary genetic disorders such as Tay-Sachs disease, sickle cell anemia, hemophilia, muscular dystrophy and cystic fibrosis.

Amniocentesis, often called amnio, is recommended for women who will be older than 35 on their due-date. It is also recommended for women who have already borne children with birth defects, or when either of the parents has a family history of a birth defect for which a diagnostic test is available. Another reason for the procedure is to confirm indications of Down syndrome and certain other defects which may have shown up previously during routine maternal blood screening.

One of the most common reasons for performing amniocentesis is an abnormal alpha-fetoprotein (AFP) test. Alpha-fetoprotein is a protein produced by the fetus and present in the mother's blood. A simple blood screening, usually conducted around the 15th week of pregnancy, can determine the AFP
levels in the mother’s blood. Levels that are too high or too low may signal possible fetal defects. Because this test has a high false-positive rate, another test such as amnio is recommended whenever the AFP levels fall outside the normal range.

Amniocentesis is generally performed during the 16th week of pregnancy, with results usually available within three weeks. It is possible to perform an amnio as early as the 11th week but this is not usually recommended because there appears to be an increased risk of miscarriage when done at this time. The advantage of early amnio and speedy results lies in the extra time for decision making if a problem is detected. Potential treatment of the fetus can begin earlier. Important, also, is the fact that elective abortions are safer and less controversial the earlier they are performed.

As an invasive surgical procedure, amnio poses a real, although small, risk to the health of a fetus. Parents must weigh the potential value of the knowledge gained, or indeed the reassurance that all is well, against the small risk of damaging what is in all probability a normal fetus. The serious emotional and ethical dilemmas that adverse test results can bring must also be considered. The decision to undergo amnio is always a matter of personal choice.

**Procedure**

Before the start of the procedure, a local anesthetic can be given to the mother in order to relieve the pain felt during the insertion of the needle used to withdraw the fluid. After the local anesthetic is in effect, a needle is usually inserted through the mother's abdominal wall, then through the wall of the uterus, and finally into the amniotic sac. With the aid of ultrasound-guidance, a physician punctures the sac in an area away from the fetus and extracts approximately 20ml of amniotic fluid. If used for prenatal genetic diagnosis, fetal cells are separated from the extracted sample. The cells are grown in a culture medium, then fixed and stained. Under a microscope the chromosomes are examined for abnormalities. The most common abnormalities detected are Down syndrome (trisomy 21), Edwards syndrome (trisomy 18), and Turner syndrome (monosomy X). In regard to the fetus, the puncture heals and the amniotic sac replenishes the liquid over the next 24–48 hours.

![Amniocentesis Diagram](image)

**Fig. 2. A diagram of the technique of amniocentesis.**

**Indications and results**

1. Early in pregnancy, amniocentesis is used for diagnosis of chromosomal and other fetal problems such as:
   - Down syndrome (trisomy 21)
   - Trisomy 13
   - Trisomy 18
   - Fragile X
   - Rare, inherited metabolic disorders
   - Neural tube defects (anencephaly and spina bifida) by alpha-fetoprotein levels.

2. Lung maturity. Amniocentesis can predict fetal lung maturity, which is inversely correlated to the
Main material

The umbilical cord is closest to the placenta. However, there is a risk of maternal blood contamination at a relatively stable segment of the umbilical cord. A typical sampling site would be where the segment of which helps to reduce the risk of clot formation. During the procedure, the first step is to locate a paralyzed using a fetal paralytic drug.

If movement of the fetus is a risk to the success of the procedure, the fetus may be paralyzed using a fetal paralytic drug. If movement of the fetus is a risk to the success of the procedure, the fetus may be paralyzed using a fetal paralytic drug. During the procedure, the first step is to locate a paralyzed using a fetal paralytic drug. If movement of the fetus is a risk to the success of the procedure, the fetus may be paralyzed using a fetal paralytic drug. During the procedure, the first step is to locate a paralyzed using a fetal paralytic drug. If movement of the fetus is a risk to the success of the procedure, the fetus may be paralyzed using a fetal paralytic drug.

Infection, in which amniocentesis can detect a decreased glucose level, a Gram stain showing bacteria or an abnormal differential count of white blood cells.

Rh incompatibility

Decompression of polyhydramnios

An emerging indication for amniocentesis is in the management of preterm rupture of membranes where measurement of certain amniotic fluid inflammatory markers may be helpful. If amniotic fluid IL-6, a marker of inflammation, is elevated, the fetus is at high risk and delivery should be considered.

**Risks and drawbacks**

Amniocentesis is performed between the 15th and 20th week of pregnancy; performing this test earlier may result in fetal injury. The term "early amniocentesis" is sometimes used to describe use of the process between weeks 11 and 13.

**Complications of amniocentesis** include preterm labor and delivery, respiratory distress, postural deformities, chorioamnionitis, fetal trauma and alloimmunisation of the mother (rhesus disease). Studies from the 1970s originally estimated the risk of amniocentesis-related miscarriage at around 1 in 200 (0.5%).

**Social implications**

The prenatal diagnosis of chromosomal abnormalities can have social drawbacks as technology changes the way people think about disability and kinship. There is potential for intensification of attitudes of discrimination towards those with a disability, whose births could have been prevented through technology such as amniocentesis. When reproduction becomes stratified, groups of people become dis-empowered to reproduce and the standard of entry into human community is questioned. In one sense, amniocentesis offers a window of control and in another, an anxiety-provoking responsibility to make rational decisions about complex, emotional and culturally contingent issues.

**Cordocentesis**

Percutaneous umbilical cord blood sampling (PUBS), also called cordocentesis, fetal blood sampling, or umbilical vein sampling is a diagnostic genetic test that examines blood from the fetal umbilical cord to detect fetal abnormalities. Fetal and maternal blood supply are typically connected in utero with one vein and two arteries to the fetus. The umbilical vein is responsible for delivering oxygen rich blood to the fetus from the mother; the umbilical arteries are responsible for removing oxygen poor blood from the fetus. This allows for the fetus’ tissues to properly perfuse. PUBS provides a means of rapid chromosome analysis and is useful when information cannot be obtained through amniocentesis, chorionic villus sampling, or ultrasound (or if the results of these tests were inconclusive); this test carries a significant risk of complication and is typically reserved for pregnancies determined to be at high risk for genetic defect. It has been used with mothers with immune thrombocytopenic purpura.

**Procedure**

If the fetus is viable, the procedure is performed close to an operating room in case an emergency cesarean section is necessary due to complications caused by the procedure. Currently, there is no definite age of viability because this depends on the fetus’ ability to survive outside the womb, which in cases of premature births, can depend on access to medical care and technology needed to keep the fetus alive through the neonatal stage. Fetal viability typically occurs at about 24 to 25 weeks of gestation. When the fetus is in between the ages of 24–34 weeks, a glucocorticoid is given to the patient about 24 hours before the procedure to stimulate lung maturity. An ultrasound is performed before the procedure to view the position of the fetus and may be used during the procedure to help guide the needle. The mother’s blood is drawn for comparison against fetal blood, and intravenous access is established in the mother in order to supply medications as needed. To reduce the risk of intraamniotic infection, antibiotics are supplied through the intravenous access about 30–60 minutes before the procedure. If movement of the fetus is a risk to the success of the procedure, the fetus may be paralyzed using a fetal paralytic drug.

A 20 or 22 gauge spinal needle is typically used in PUBS and may be prepared with an anticoagulant, which helps to reduce the risk of clot formation. During the procedure, the first step is to locate a relatively stable segment of the umbilical cord. A typical sampling site would be where the segment of the umbilical cord is closest to the placenta. However, there is a risk of maternal blood contamination at...
this site. Blood sampling may be achieved with more ease if the placenta is in the anterior position. However, if the placenta is in the posterior position, the fetus might block direct access to the umbilical cord. Once the umbilical cord is reached and the correct position of the needle is confirmed, the fetal blood is drawn. The needle is removed after all necessary samples are taken. The site of puncture is monitored after the procedure for bleeding. Also, if the fetus is viable, fetal heart rate is monitored post-procedure for one to two hours.

After the blood samples are obtained, they are placed into tubes containing anticoagulants in order to stop the blood from clotting. If the blood sample was obtained at the site close to the placenta, a fetal blood confirmation test should be done to ensure no mixing of fetal and maternal blood occurred before the diagnostic tests are done on the blood. Fetal red blood cells (RBC) are usually bigger than maternal RBCs, and the average volume of RBCs, the mean corpuscular volume (MCV), is one of the methods used to determine whether or not the fetal blood has been contaminated. Another method, human chorionic gonadotropin (hCG) determination, can detect maternal blood because maternal blood has high levels of hCG. The hemoglobin alkaline denaturation test (Apt test) can detect the presence of maternal blood, which is indicated by a color change from red to brown when the sample is added to alkali reagent. Blood typing would also detect maternal blood, as the I antigen only occurs in adults. The Kleihauer–Betke test can detect very small amounts of maternal blood before the third trimester of pregnancy by monitoring hemoglobin elution in acid because adult and fetal hemoglobin elute differently in acid. Finally, a white blood cell count can detect maternal blood in the sample, as fetal white blood cells are primarily leukocytes, while maternal white blood cells are mostly neutrophils. If amniotic fluid infiltrated the sample, then there would be a reduction in the volume of RBCs, white blood cells, and platelets in the sample. Also, patterns consistent with amniotic fluid would be visible in the sample.

**Fig. 3. Percutaneous umbilical cord sampling, also known as cordocentesis.**

**Associated risks**

The most common complication is a hemorrhage, or bleeding, of the puncture site and can be especially dangerous when the fetus is younger than 21 weeks. The risk of hemorrhage is greater if the fetus has a defect that affects its platelets. A transfusion of donor platelets is usually done in such cases to reduce the risk of bleeding. If the bleeding is severe, immediate delivery is an option as long as the fetus is old enough to survive, or fetal blood volume restoration may be considered. Another possible complication is cord hematoma, which doesn’t have any characteristic symptoms but can be indicated by sudden bradycardia. If the hematoma is under control, the fetus is monitored until stabilized. If the fetus remains unstable, a delivery may be done. Fetomaternal hemorrhage is another complication that occurs when the fetal blood mixes into the maternal blood. A small fetomaternal hemorrhage could cause an increase in maternal antigens, while a large fetomaternal hemorrhage could cause fetal anemia and death. Fetal bradycardia, low heart rate, is another complication that may occur. Most cases of fetal bradycardia are self-resolved within five minutes. The complication of infection has a low incidence rate, and preventative measures are implemented against the risk of infection, such as antibiotic usage and the aseptic technique. However, vertical transmission of a virus such as HIV may occur. Fetal loss may also occur, especially in the presence of several risk factors, including fetal abnormalities, operator
errors, placental penetration, and viability of the fetus.

Intrahepatic vein fetal blood sampling may be done as an alternative to PUBS. It involves the needle being inserted into the intrahepatic part of the umbilical cord in the fetal abdomen. The benefits of this alternative, compared to PUBS, are that chances of contamination of the fetal blood are very low, the risk of fetomaternal hemorrhage are reduced, the risk of bleeding from the sampling site is reduced, and access to the sampling site is easy regardless of the position of the placenta. In pregnancies with high risk of fetal thrombocytopenia, this is the preferred method of blood samples due to the very low risk of site bleeding.

Indications and contraindications

PUBS is not a diagnostic test that is indicated in every pregnancy. It is, however, suggested in pregnancy cases in which the blood gas levels and pH would aid in diagnosis of a condition, such as anemia, or delivery plan, if termination of the pregnancy is being considered or special plans must be made. Severe fetal growth issues in conjunction with low oxygen in the fetus’ blood and high levels in the mother’s blood also indicate the use of PUBS. With more detailed observations and information on fetal tissue perfusion and metabolism, better predictions on development can be made. For pregnancies in which genetic abnormalities may be present, PUBS can be used to construct a karyotype, usually within 48 hours, and detect irregular chromosomal patterns. Karyotypes are able to confirm or detect monosomies, trisomies, or missing portions of chromosomes to give a detailed picture of the severity of the genetic defect as well as predicting developmental future. PUBS is also indicated in the cases of twins with accumulation of amniotic fluid and substantially different growth rates (at least 10%), if the fetus is expected to be breaking down red blood cells improperly, and in the alleviation of hydrops fetalis, a build-up of fluid in at least 2 parts of the fetus. Suspicion of fetal infections, such as rubella and toxoplasmosis, as well as the need to supply medicine or blood transfusions to the fetus are indications for the use of PUBS.

Due to its invasive nature, the contraindications of PUBS, reasons to not undergo the procedure, must be taken into account in order to ensure the safety of the fetus and the mother. During the first 18 weeks of pregnancy, the umbilical vein from which the blood sample is taken is not very stable, which could lead to excessive bleeding; therefore, PUBS is contraindicated in any fetus under the age of 18 weeks old. While blood gas levels and pH values are able to give parents and medical professionals a snapshot of fetal status, these fetuses can be monitored with less invasive procedures and equipment, such as ultrasounds, cardiotocography, or maternal blood tests. Mothers affected by hepatitis B are not advised to undergo PUBS. In these cases, the fetus would be put at an increased risk of contracting the hepatitis virus from the mother. However, the necessity of the procedure should be considered along with this risk. PUBS should not be performed in mothers testing positive for the human immunodeficiency virus (HIV) due to increased risk of fetal contraction. If PUBS is being used to determine if the fetus has been infected with HIV it may not be contraindicated.

Fetoscopy

Fetoscopy is a procedure that utilizes an instrument called a fetoscope to evaluate or treat the fetus during pregnancy.

There are two different types of fetoscopy: external and endoscopic.

External fetoscopy

An external fetoscope resembles a stethoscope, but with a headpiece. It is used externally on the mother’s abdomen to auscultate (listen to) the fetal heart tones after about 18 weeks gestation. It also allows a birth attendant to monitor the fetus intermittently and ensure that the baby is tolerating labor without the mother having to be attached to a continuous fetal monitor.

Endoscopic fetoscopy

The second type of fetoscope is a fiber-optic endoscope. It is inserted into the uterus either transabdominally (through the abdomen) or transcervically (through the cervix) to visualize the fetus, to obtain fetal tissue samples, or to perform fetal surgery.

Approximately 3% of babies born each year have a complex birth defect. Certain birth defects are complicated by the labor and delivery process, while others may progress quickly after birth to cause significant disability or death. Fetal surgical techniques utilizing the endoscopic fetoscope offer early intervention in order to treat such defects before they become serious.

Some of the fetal abnormalities that may be treated by endoscopic fetoscopy are:

- Congenital diaphragmatic hernia (CDH). In babies with CDH, the diaphragm (the thin muscle that separates the chest from the abdomen) doesn’t develop properly. The abdominal organs may enter the chest cavity through a hole (hernia) and cause pulmonary hyperplasia (underdeveloped lungs). CDH occurs in about one out of every 2,000 births.
- Urinary tract obstruction. The urethra (the tube that carries urine from the bladder to the outside of the body) may become obstructed in utero or fail to develop normally. When this happens, urine can back up into the kidneys and destroy tissue or cause the bladder to become enlarged. The amount of amniotic fluid also decreases because fetal urine is its major component.
Pulmonary hypoplasia usually results because the lungs rely on amniotic fluid in their development.

Twin/twin transfusion syndrome (TTTS). In some twin pregnancies, the two fetuses will share a placenta (called a monochorionic pregnancy). TTTS occurs in approximately 15% of these twins when blood volume between the fetuses is unequal, causing abnormally low blood volume in the donor twin and abnormally high blood volume in the recipient twin. There is often a large difference in size between the twins. Approximately 70–80% of fetuses suffering from TTTS will die without intervention.

Acardiac twin. This condition also occurs in monochorionic pregnancies, but one twin develops normally while the other develops without a heart. The acardiac twin receives its blood supply from the normal twin, whose heart must now work harder to pump blood through both fetuses. Approximately 50–75% of acardiac twins will die as a result. An acardiac twin occurs in 1% of monochorionic pregnancies and one out of 35,000 overall pregnancies.

The external fetoscope is used to listen to fetal heart tones for rate and rhythm. The earpieces and the headpiece allow auscultation (listening) via both air and bone conduction. External fetoscopy is inexpensive, noninvasive, and does not require electricity. It is difficult, however, to clearly hear the fetal heart tones prior to 18 to 20 weeks gestation. Doppler ultrasound can detect fetal heart tones around weeks 10 to 12.

Endoscopic fetoscopy uses a thin (1 mm) fiberoptic scope. Developed in the 1970s, the endoscope was originally inserted transabdominally to visualize the fetus for gross abnormalities suspected by ultrasound or to obtain tissue and blood samples. It was performed after about 18 weeks gestation. Even with practitioner expertise, associated fetal loss was 3–7%. During the 1980s, ultra-sound-guided needle sampling of cord blood replaced fetoscopy when samples of fetal blood were required.

As laparoscopic and microsurgical techniques have become more common and the instrumentation has become more advanced technologically, fetoscopy has improved for fetal diagnostic and therapeutic purposes. Fetal surgery performed through an open maternal abdomen has a higher risk of such complications as infection, premature rupture of membranes, preterm labor, or fetal death. If surgery is performed via fetoscopy, which requires a very small transabdominal incision, the risks are much smaller. Techniques have advanced enough to allow some fetoscopy to be performed in the first trimester via the mother’s cervix. The term “obstetrical endoscopy” may be used for surgery on the placenta, umbilical cord, or on the fetal membranes. The term “endoscopic fetal surgery” is used for such procedures as the repair of a fetal congenital diaphragmatic hernia or obstructed bladder.

**Diagnosis/Preparation**

The use of external fetoscopy requires access to the maternal abdomen, with the mother lying supine or in a semi-seated position. Afterwards, the mother is able to get up and resume a normal activity level.

Preparation for endoscopic fetoscopy will depend on the extent of the procedure, and whether it is performed transcervically or transabdominally. Obtaining a small fetal tissue sample is a smaller procedure by comparison to fetal surgery. Other factors include outpatient versus inpatient stay and anesthesia (both maternal and fetal). For some procedures medication may be administered to temporarily decrease fetal movement to lower the risk of fetal injury. Maternal anesthesia may be local, regional, or general.

**Aftercare**

External fetoscopy does not require aftercare. The care following fetal endoscopic use will depend on the extent of the procedure and the type of anesthesia used. If the procedure is done on an outpatient basis, the mother and fetus will be monitored for a period of time prior to discharge. More extensive surgery will require inpatient hospital postoperative care.

**Risks**

The only potential complication with external fetoscopy is the possibility of missing an abnormal heart rate or rhythm. Its usefulness and accuracy depend on the skill of the practitioner.

Endoscopic fetoscopy has the potential for causing infection in the fetus and/or mother; premature rupture of the amniotic membranes; preterm labor; and fetal death. When endoscopic fetal surgery is done instead of open-uterus fetal surgery, the risks to the mother and fetus are decreased. The risks are
because the incision is significantly smaller, with less potential blood loss, decreased uterine irritability, and decreased risk of early miscarriage.

**Normal results**

The normal fetal heart rate is 120 to 160 beats per minute, regardless of the method used for auscultation (external fetoscopy or Doppler ultrasound). Some variability of fetal heart rate is expected, as the heart rate increases with fetal activity and slows with fetal rest.

Results expected using endoscopic fetoscopy will vary depending on the procedure undertaken. The goal is for the maximum benefit with the minimum of risk or complication to both the mother and fetus.