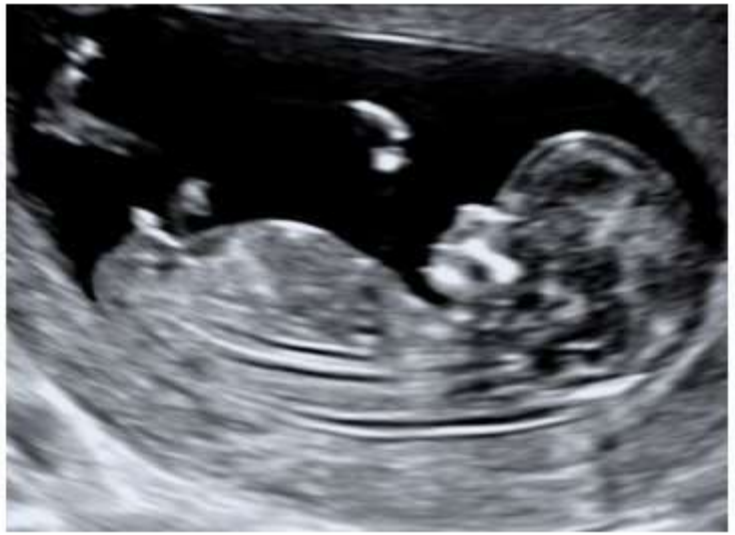


# PRENATAL DIAGNOSIS

Dr Valens NKUBITO GATERA

# Prenatal Diagnosis

- Over the last 4 decades, the genetic basis of an increasing number of diseases is becoming understood.

- At the same time, safe and effective fetal diagnostic techniques are being developed.

# Prenatal Diagnosis of fetal Abnormalities

**Benefits:**

**1.Malformation incompatible with life may be terminated.**

**2.Certain abnormalities may be correctible in-utero.**

**3.Provides opportunity to arrange corrective measures before hand.**

**Offer a chance to be delivered at a place where the required facilities are available.**

**4. Parents decision to continue pregnancy/ mentally prepare to have a handicapped child.**

# Classification of Congenital Abnormalities

- 1 Chromosomal Abnormalities: - Trisomy 21 (D.S) - Trisomy 18 (E.S) - Trisomy 13 (P.S)
- 2 Structural Abnormalities: - CNS - CVS - GIT - Bone - Renal system
- 3 Genetic Disorders: Inborn error of metabolism - Haemoglobinopathies

# What Should We Do?

- **Every pregnancy should be evaluated with the most definite test.**
- **Practically & economically not feasible because**
  - ✓ **Expensive**
  - ✓ **Invasive**

**Worldwide practice is to carry out**

**-Screening procedures**

**-Definite (diagnostic)tests for screening positive cases**

# Screening Procedures

**These are:**

- **Simple**
- **Cheap**
- **Least invasive**
- **Safe**
- **Easily repeatable**

# Screening Procedures --- Cont

. 1. History:

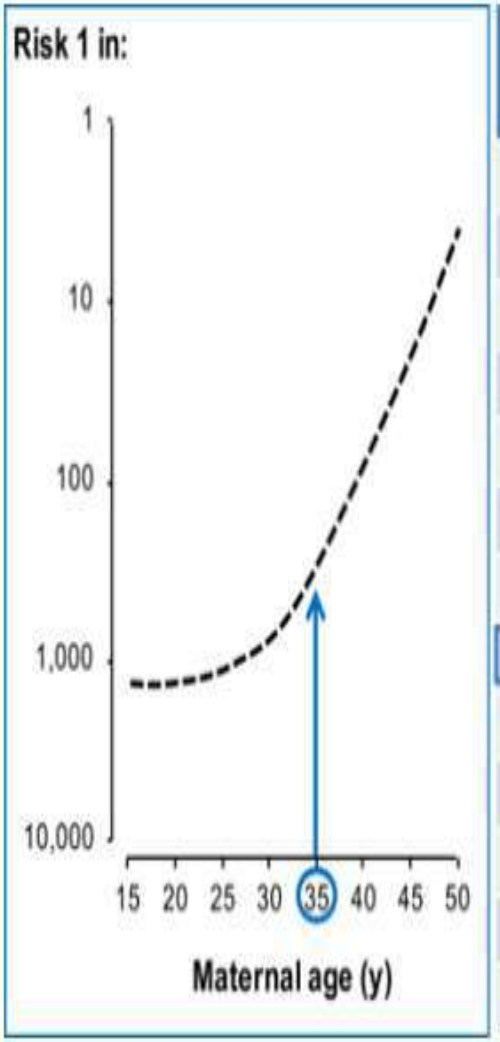
- Increasing maternal age
- Congenital anomalies in previous children
- F/Hx of Stillbirth, Recurrent 1st trimester abortion, relative marriage,...

# Maternal age

## Risk for trisomy 21

- Increases with maternal age.
- Decreases with gestational age because about 30% of affected fetuses die between the 12th and 40th week of pregnancy.

**For example**, in a woman who is 35 years old the risk for trisomy 21 at the 12th week of pregnancy is 1 in 250, but the chance that she will deliver an affected baby at 40 weeks is 1 in 350.




| Age (y) | Gestational age | | |
|---|---|---|---|
| | 12 w | 20 w | 40 w |
| 20 | 1100 | 1300 | 1500 |
| 25 | 1000 | 1100 | 1400 |
| 30 | 650 | 750 | 900 |
| 31 | 550 | 650 | 800 |
| 32 | 450 | 550 | 650 |
| 33 | 400 | 450 | 550 |
| 34 | 300 | 400 | 450 |
| 35 | 250 | 300 | 350 |
| 36 | 200 | 250 | 300 |
| 37 | 150 | 185 | 220 |
| 38 | 120 | 140 | 160 |
| 39 | 90 | 110 | 130 |
| 40 | 70 | 80 | 100 |

# Screening Procedures ---Cont

## 2. Features of current pregnancy:

- Drug intake(antiepileptics e.g. warfarin, alcohol, smoking)
- Radiation exposure
- Maternal diseases e.g.DM, cardiac, renal
- Uterine fundas large/ small for date
- Decrease fetal movements
- Fetal malpresentation
- Viral infection in early pregnancy

# Screening Procedures --- Cont.

**3. Ultrasonography:**

- **Screening tool in all trimesters**
- **At 10-14 weeks if fetal nuchal translucency > 2.5 mm- chromosomal anomalies association**
- **At 18-20 weeks 75% fetal abnormalities can be diagnosed**

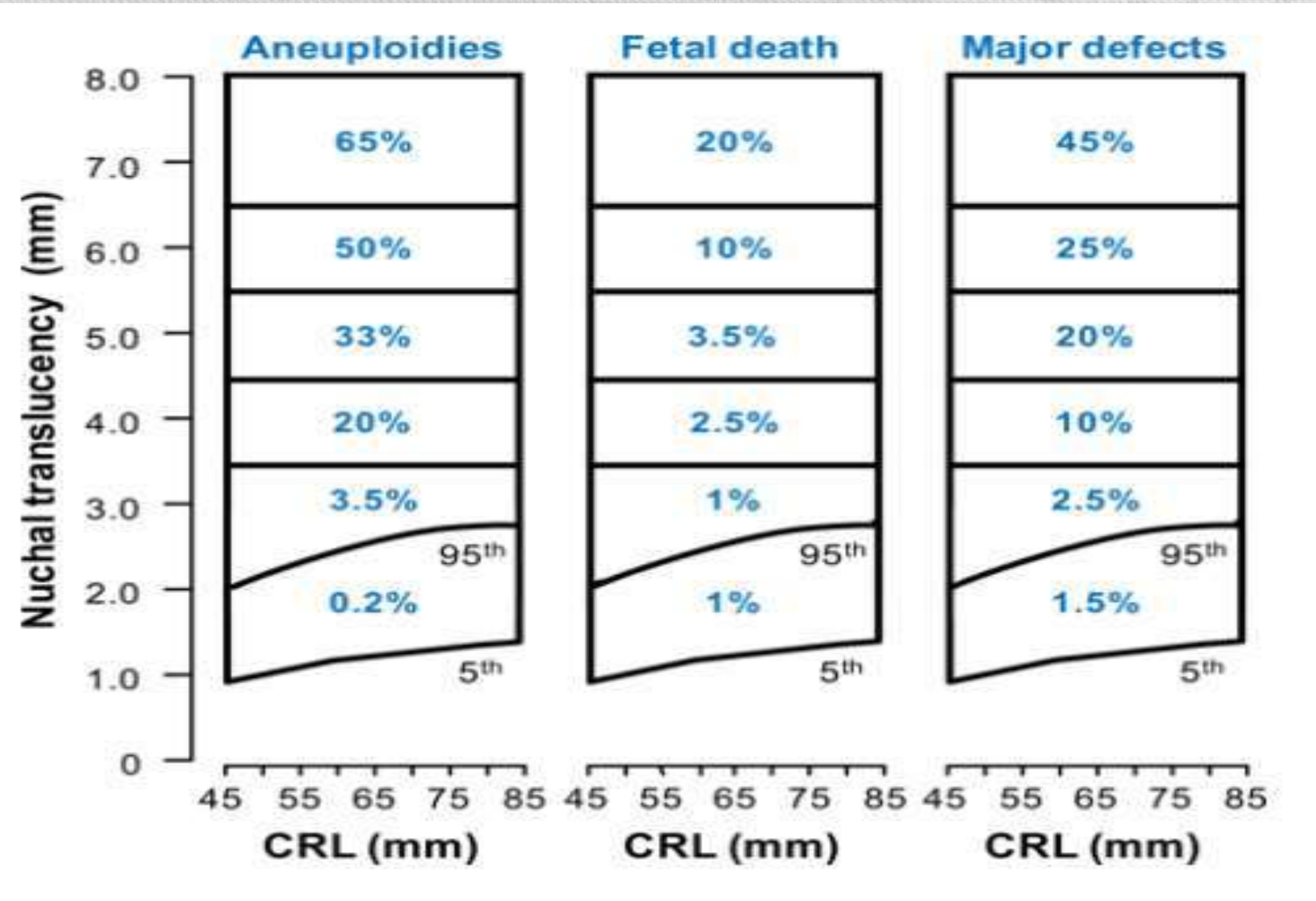
# Screening Procedures --- Cont.

**An ultrasound scan at 11-13 weeks:**

- to measure the collection of fluid behind the fetal neck (nuchal translucency)
- to examine the fetal nose and palate
- to measure the fetal heart rate
- to assess the flow of blood across the tricuspid valve of the fetal heart and the ductus venosus

**Detection of major chromosomal abnormalities of 95%**

# Why measuring NT?



# Why fetal heart rate?

**Euploid and trisomic fetuses**

- **In euploid fetuses** the heart rate increases from about 110 bpm at 5 weeks of gestation to 170 bpm at 10 weeks and then gradually decreases to 150 bpm by 14 weeks.
- **In trisomy 21** the FHR is mildly increased and is above the 95th centile in about 15% of cases.
- **In trisomy 18** the FHR is mildly decreased and is below the 5th centile in about 15% of cases.
- **In trisomy 13** the FHR is substantially increased and is above the 95th centile in 85% of cases.

# 4. Serum biochemistry

- **Trisomic pregnancies** are associated with altered maternal serum concentrations of various feto-placental products.
- **Screening in the second trimester** by maternal age and various combinations of total or free ß-hCG, AFP, uE3 and Inhibin A can identify 56-71% of trisomy 21 pregnancies for a false positive rate of 5%.
- **Screening in the first trimester between 11-14 weeks** by a combination of maternal age, fetal NT, FHR and serum free ß-hCG and PAPP-A identifies about 90% of trisomy 21 pregnancies for a false positive rate of 3%

# Serum biochemistry

## Screening for other aneuploides

**In euploid pregnancies** the average free ß-hCG is 1.0 MoM and PAPP-A is 1.0 MoM.

**In aneuploid pregnancies** the average MoM values of free ß-hCG and PAPP-A are:

| | free ß-hCG | PAPP-A |
|---|---|---|
| **Trisomy 21** | 2.0 | 0.5 |
| **Trisomy 18** | 0.2 | 0.2 |
| **Trisomy 13** | 0.3 | 0.3 |
| **Turner** | 1.2 | 0.5 |
| **Triploidy** | | |
| » Digynic | 0.2 | 0.1 |
| » Diandric | 9.0 | 0.7 |

# Screening Procedures --- Cont.

Other benefits of the 11–13 weeks scan include:

- Accurate dating of the pregnancy
- Early diagnosis of many major fetal abnormalities
- The detection of multiple pregnancies with reliable diagnosis of **chorionicity**, which is the main determinant of the outcome in multiple pregnancies
- Another recent development is that the 11-13 weeks scan can be used to identify **women at increased risk for the development of preeclampsia in pregnancy.**

# Screening Procedures --- Cont.

These are some of the structural abnormalities which can be detected by the first trimester ultrasound:

**CNS:** Acrania/exencephaly, Alobar holoprosencephaly, Encephalocele, Spina bifida, Sacral agenesis

**Face**: Facial clefts, micrognatia

**Thorax:** Diaphragmatic hernia

**Cardiac**: TOF/TA, AoCo/HLH, Right aortic arch, Atrioventricular septal defect, Right atrial isomerism, Double-outlet right ventricle.

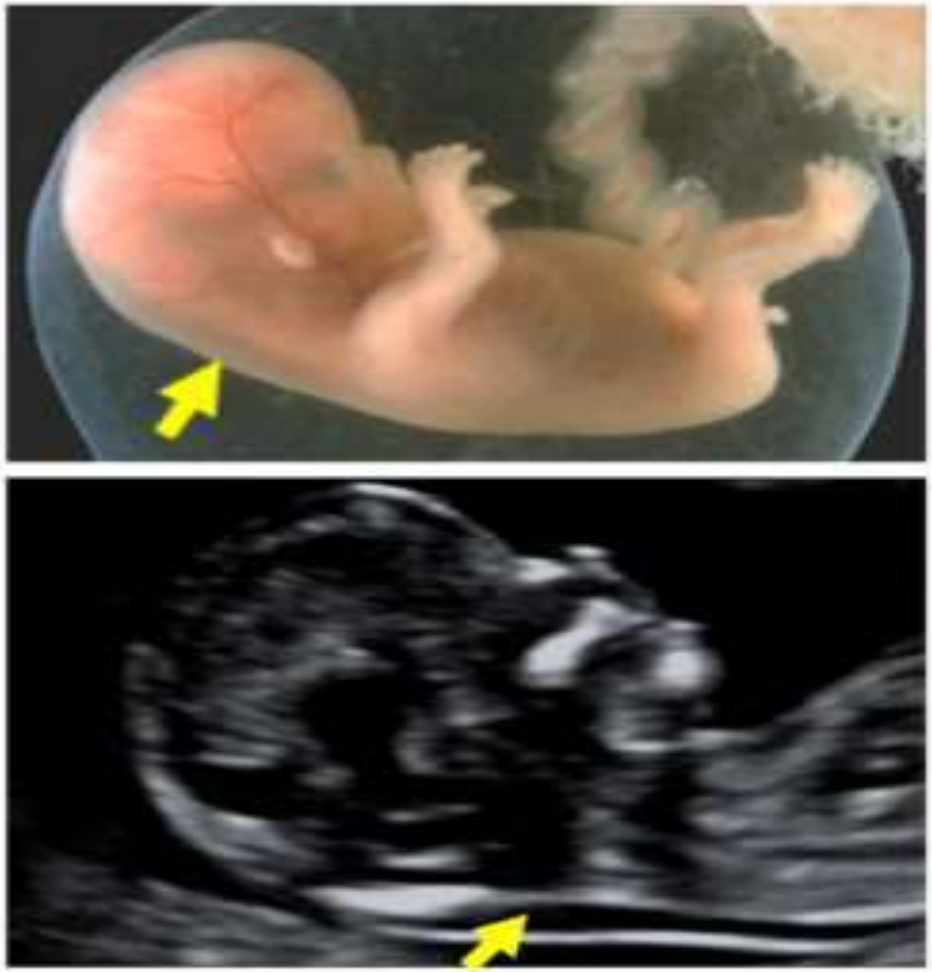
**Abdominal wall/GI defect**: Exomphalos, Gastrischisis, Body stalk anomaly

**GU**: Megacystis,

**Skeletal**: Skeletal dysplasia, Aplasia/hypoplasia of the radius/ulna (club hand), Limb reduction defects.

**Many of these abnormalities can be associated.**

# 1st Trimester Ultrasound • NT Ultrasound



# Combined test or Double Test

- Pregnancy associated plasma proteins-A (PAPP-A) level and serum free Beta-hCG during 1st trimester(11-14 weeks)
- Measurement of NT
- Maternal age
- And other biomarkers

High risk--------CVS or Amnio

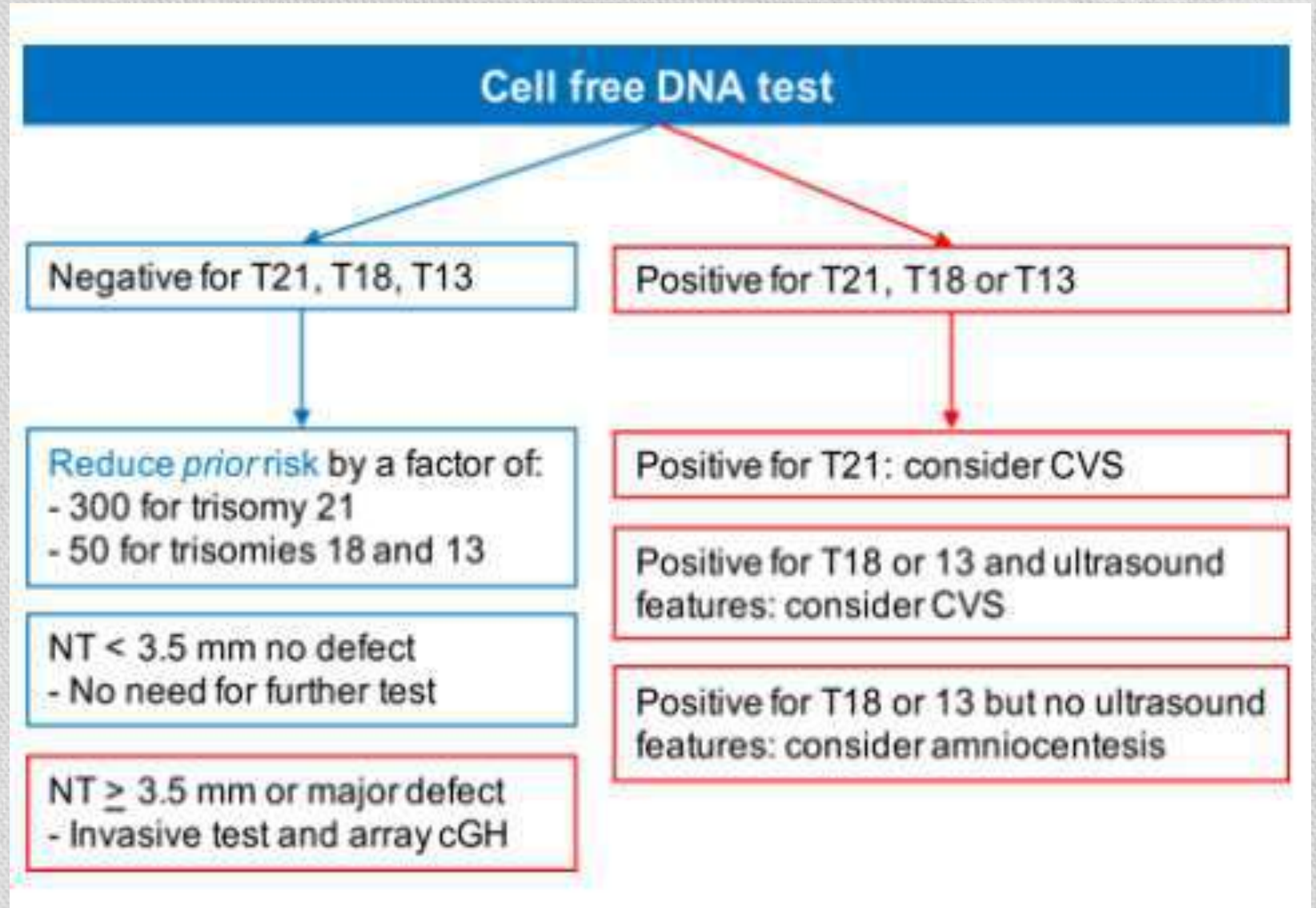
Low risk--------Ftal assessment at 20 weeks

## Cell free DNA/ NIPT

- Analysis of cfDNA in maternal blood
- detect about 99% of fetuses with trisomy 21 and 98% of fetuses with trisomies 18 or 13.
- false positive rate (FPR) of 0.1-0.2%

| Method of screening | Proportion of total population | Proportion of all cases of T21 |
|---|---|---|
| Maternal age | 5% / 20% | 30% / 50% |
| Second trimester serum biochemistry | 5% | 50-70% |
| Age, NT, FHR, ß-hCG, PAPP-A (combined) | 3-5% | 90% |
| Combined plus additional ultrasound markers | 2.5% | 95% |
| Cell free DNA in maternal blood | <0.1% | >99% |

# Limitations(cfDNA)



# Limitations(cfDNA)

- **Failure to obtain result in 1-5% of singleton pregnancies**
- Repeat testing provides a result in 60-70% of cases
- **Main reason for failure is low fetal fraction** (low proportion of fetal to total cfDNA in maternal blood).
  - Obesity
  - Low placental mass
- In trisomies 18 and 13, but not in trisomy 21, the fetal fraction is lower.
- Cost (200-1000 USD)

# DIAGNOSTIC TESTS

- For high risk women on basis of screening tests
- An ideal test should be :
  - Least invasive
  - Diagnose c. abnormality in early pregnancy.
  - Minimally interfering developing pregnancy
  - Diagnostic tests are also not risk free.

# Counseling

**Organize an appointment**
**Couple should be present**
**Explain:**

- **Risk of occurance of c. abnormality**
- **All tests available, their procedure, cost, diagnostic ability and benefits, possible risks**
- **Possible management plan**
- **If termination of pregnancy is unacceptable diagnostic tests would be fruitless.**

# NON INVASIVE TESTS

**Ultrasonography:**

- **Diagnostic USG is different from screening USG,**
  - **It takes longer time**
  - **Dx. Wide range of c. anomalies**
  - **Non invasive and diagnosis at spot possible**
  - **Requires special training**
  - **Colour doppler further enhance the capability especially for cardiac malformations and renal agenesis.**

# INVASIVE TESTS

## AMNIOCENTESIS:

- Aspiration of amniotic fluid which contain fetal cells
- Fluid can be used for estimation of bilirubin level (for fetal haemolytic disease),AFP, Acetyl cholinesterase
- Cells used for karyotyping (Chromosomal dis.) and array CGH
- Fetal cells cultured for 3 weeks for karyotyping.
- New technique like PCR, FISH give result in 48 h.

# AMNIOCENTESIS---Cont.

## Procedure:

- Preliminary USG to confirm the gestation age, placental site, adequacy of liqour
- Sterilize the abdomen
- 22 G spinal needle is used.
- About 20 cc amniotic fluid is withdrawn.
- Give Anti- D to all Rh-ve mothers.
- Ask rest for 30 min.& restrict movements for 48h

# AMNIOCENTESIS---Cont.

- Limitations (difficulties)of procedure:
  - Anteriorly placed placenta
  - Multiple pregnancy.
  - Maternal obesity
  - Oligohydramnios

- Risks:

- Pregnancy loss 1 % , Bleeding , Infection, ROM, PTL/IUD, Leaking of Amniotic fluid, Increase risk of RDS in newborn.

# CHORIONIC VILLUS SAMPLING

- **Collection of fragments of placental tissue (chorionic villi)- cells are examined for Dx. of C.Anomalies.**

- **Cytotrophoblastic (rapidly dividing) cells are used for direct karyotyping, result available within 24-48 h.**

- **Chorionic villi are best source of DNA**

- **CVS can be performed at 10 weeks gestation.**

# CVS

**Indications:**

**1-DNA analysis for SCD, thallasemias, CF, hemophillias**

**2-Chromosomal abnormalities**

**3-Inborn error of metabolism**

# CHORIONIC VILLUS SAMPLING

- **Procedure:**
- **Transabdominal approach preferred under USG guidance in supine position**
- **Transcervical approach is easy.**
- **In lithotomy position, sterilize area & Aspiration catheter and biopsy forceps.**
- **Introduce through Cx. under USG into placental tissue avoiding membrane rupture**

- **Risks:**
  - **Pregnancy loss 2% or less**
  - **Before 10 weeks- associated with limb deformities, micrognathia, microglossia**

# FETAL BLOOD SAMPLING (FBS)

- **Fetal blood**
- **Lymphocytes are rapidly cultured, results within 48-72 hours.**

**Indications:**

**1. Prenatal Dx. DNA available for Cytogenetic studies In failed amniocentesis, and mosaicism in chorion or amniotic fluid.**

**2. Fetal assessment: for red cell alloimmunization, (Hb;Hc,TrF) Hydrops fetalis, viral infection, platelets alloimmunization**

**• Unfortunately Associated with highest rate of fetal loss.**

**• Currently used for blood transfusion in utero in fetal anemia.**

# FETAL BLOOD SAMPLING (FBS)

•**Procedure : (cordocentesis):**

• **The sites for FBS are placental insertion of umbilical cord, abdominal insertion of cord, intrahepatic fetal vein and fetal heart.**

• **Suitable time is 20-28 weeks •**

**Risks:**

- **Bleeding from site of puncture**
- **Cord haematoma**
- **Fetal bradycardia**
- **Fetal death**

# EMBRYOSCOPY & FETOSCOPY

- Direct visualization of embryo and fetus.
- Limited field of vision.
- Provide information only about external fetal structures .

# NEW MOLECULAR ANALYTIC TECHNIQUES

- **Fetal cell obtained by CVS and Amniocentesis can be used for prenatal Dx.**

**For congenital anomalies by following new techniques**

**1- Southern blotting**

**2- PCR**

**3- FISH**

**4- Array CGH**

# Polymerase chain reaction (PCR)

- **Amplify specific DNA and RNA fragments**
- **Once nucleotide sequence of a region of DNA strand is known, complimentary oligonucleotides & polymerase are added to single strand DNA**
- **Repeat process 30 times to get adequate DNA**
- **PCR identify specific DNA sequence for gene mutation & prenatal Dx. at an earlier stage before an embryo transfer in IVF cycle.**

# FLOURESCENT IN SITU HYBRIDIZATION

- **FISH allows detection & localization of specific DNA sequence in interphase or metaphase.**
- **Advantage – results available in 24-48 h.**
- **Disadvantage – fail to detect big structural rearrangements**
- **Identify 80% clinically relevant abnormalities, helpful for early decision about further management of affected pregnancies.**

# MANAGEMENT OF FETAL C. ANOMALIES

- It is a tedious task, requires skillful, sympathetic & professional approach.
- Management options:
  - Termination of pregnancy
  - In utero management if possible
  - Conservative management
  - Involvement of pediatricians is very crucial
  - Need to involve surgeons, geneticists, psychologists,

**Multidisciplinary team is needed!!!**

# POSTPARTUM MANAGEMENT OF C.A.

- **For better understanding of congenital anomalies and its impact on future reproductive performance of couple, following procedures should be carried out on affected babies/ abortuses whenever possible after consent:**

**1.Physical examination /postmortem**

**2.Fetal tissue(blood, skin, placenta) for genetic analysis**

**3.Placenta and membrane for histopathology**

**4.Placental & baby swab for microbiology & virology.**

**5.Baby gram (x-rays of whole baby)**

**6.Baby photograph**

# Take home message

- Fetal abnormalities are frequent;
- First trimester scan and screening for chromosomal abnormalities between 11 and 14 weeks should be our standard practice;
- Combined test can be performed in our settings and can detect more 95% of major abnormalities in the first trimester and quadruple test in the second trimester to detect around 60%;

# Take home message

- NIPT/ cfDNA is a screening tool with more accuracy the the combined but its still expensive;
- A 20 weeks scan is important for fetal anatomical assessment;
- Only perform a diagnostic invasive test if screening tests are positive
- Counseling of the couple with expalining risks and benefits
- No need to carry out a diagnostic test if termination is not an option

**In case of fetal abnormalities, a multidisciplinary team is need for assessment and management.**

# THANKS