

# Population genetic screening

## Principles and applications

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## **Definition: Population Screening**

**Presumptive identification of unrecognized disease by application of tests, examination, and other procedures.**

**The procedures can be applied to rapidly sort out “apparently” well individuals who probably have a disease from those who probably do not.**

**The procedures are not intended to provide a definitive diagnosis.**

## Definition: Genetic screening

Identification of individuals possessing certain *genotypes* that either are (A) associated with disease or predisposition, or (B) which lead to disease in their descendants.

### Examples:

(A) Newborn screening for metabolic disease

(B) Heterozygote detection of Tay-Sachs or Gaucher disease

## Objectives of genetic screening

- Early recognition of disorder:  
for intervention that prevents or reverses the disease process,  
  
or to ensure optimal management of the patient, eg. appropriate referrals to specialists when symptoms are anticipated
- Informed reproductive decisions/ disease management, eg. trisomies 13 and 18, which are generally lethal in the newborn period.

# Principles of screening

## **Disease**

1. Disease is serious and relatively common - cost/benefit
2. Natural history well defined
3. Acceptable and effective treatment
4. Prenatal diagnosis available for some conditions

## Test

1. Inexpensive
2. Valid and reliable
3. Easy to perform
4. “Socially acceptable”

## Principles of screening (continued)

### System

1. Resources for diagnosis/treatment accessible.
2. Results communicated efficiently and effectively.
3. At-risk population can then be targeted with more accurate, often more expensive subsequent tests.

## Box 13-1

# Genetic Screening and Prenatal Diagnosis

- I. Population screening of genetic disorders
  - A. Newborn Screening
    - 1. Blood
      - a. PKU, all 50 states in the United States
      - b. Galactosemia, all 50 states in the United States
      - c. Hypothyroidism, all 50 states in the United States
      - d. Other: hemoglobinopathies, cystic fibrosis
    - 2. Urine: aminoacidopathies
  - B. Heterozygote Screening
    - 1. Tay Sachs disease, Ashkenazi Jewish population
    - 2. Sickle cell disease, African American population
    - 3. Thalassemias, at risk ethnic groups
    - 4. Cystic fibrosis, pilot programs only

- II. Prenatal diagnosis of genetic disorders
  - A. Diagnostic testing (invasive prenatal diagnosis)
    - 1. Amniocentesis
    - 2. Chorionic villus sampling
    - 3. Percutaneous umbilical cord sampling (PUBS)
  - B. Fetal visualization techniques
    - 1. Ultrasonography
    - 2. Radiography
    - 3. MRI
  - C. Population screening
    - 1. Maternal age > 35 years
    - 2. Family history of condition diagnosed by prenatal techniques
    - 3. Abnormal maternal serum  $\alpha$  fetoprotein estriol, human chorionic gonadotropin
- III. Family screening of genetic disorders
  - A. Family history of chromosomal rearrangement (e.g. translocation)
  - B. Screening female relatives in an X-linked pedigree (e.g. Duchenne muscular dystrophy, Fragile X syndrome)
  - C. Heterozygote screening within at-risk families (e.g. cystic fibrosis)
  - D. Presymptomatic screening (e.g. Huntington disease, breast cancer, colon cancer)

# How useful is a screening test?

Table 13-1 Definitions of Sensitivity and Specificity\*

Screening test	<u>D i s e a s e</u> <u>S t a t e</u>	
	Affected	Unaffected
Results positive (+)	a (true positive)	b (false negative)
Results negative (-)	c (false positive)	d (true negative)

\*Sensitivity =  $a / (a + c)$ ; specificity =  $d / (b + d)$

Proportion of true positives    Proportion of true negatives

Accuracy: what is the proportion of persons with a positive test who truly have the disease in question?  
Termed the positive predictive value.

*Benefit* of a screening test is also related to its positive predictive value.

TABLE 13-2 Hypothetical Results of Screening for Congenital Adrenal Hyperplasia in a Low-Prevalence Caucasian Population and a High Prevalence Yupik Population\*

Screening Test	C A H P R E S E N T		C A H A B S E N T	
	Positive	Negative	Positive	Negative
Caucasian	47	3	5,000	494,950
Yupik	24	1	100	9,875

\*Caucasian positive predictive value =  $47 / (47 + 5,000) = 1\%$

Yupik positive predictive value =  $24 / (24 + 100) = 19\%$

The *positive predictive value* is different from *sensitivity*.

# Prenatal testing

## Benefits include:

- Reassurance to at-risk families when results are normal
- Providing risk information to couples who without the info might not choose to begin a pregnancy
- Allowing couple to prepare for birth of an affected baby
- Help health care professional plan delivery, management of an affected infant prior to birth.
- Providing risk information to couples for whom pregnancy termination is an option.

**98% of prenatal diagnoses yield a normal test result.**

**The majority of families receive reassurance; only a small minority face the issues of delivery of an affected child or termination.**

# Ultrasound: noninvasive screening procedure to visualize the fetus

Selected Disorders Diagnosed By Ultrasound in the Second Trimester\*

## System Complex

Hydrops  
Oligohydramnios  
Polyhydramnios  
Intrauterine growth  
retardation

## Central Nervous System

Anencephaly (85-90%)  
Encephalocele  
Holoprosencephaly  
Hydrocephalus

## Craniofacial

Cleft lip

## Chest

Congenital heart disease  
Diaphragmatic hernia

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## Abdomen/ Pelvis

Gastrointestinal atresias  
Gastroschisis  
Omphalocele  
Renal agenesis  
Cystic Kidneys  
Hydronephrosis

## Skeletal System

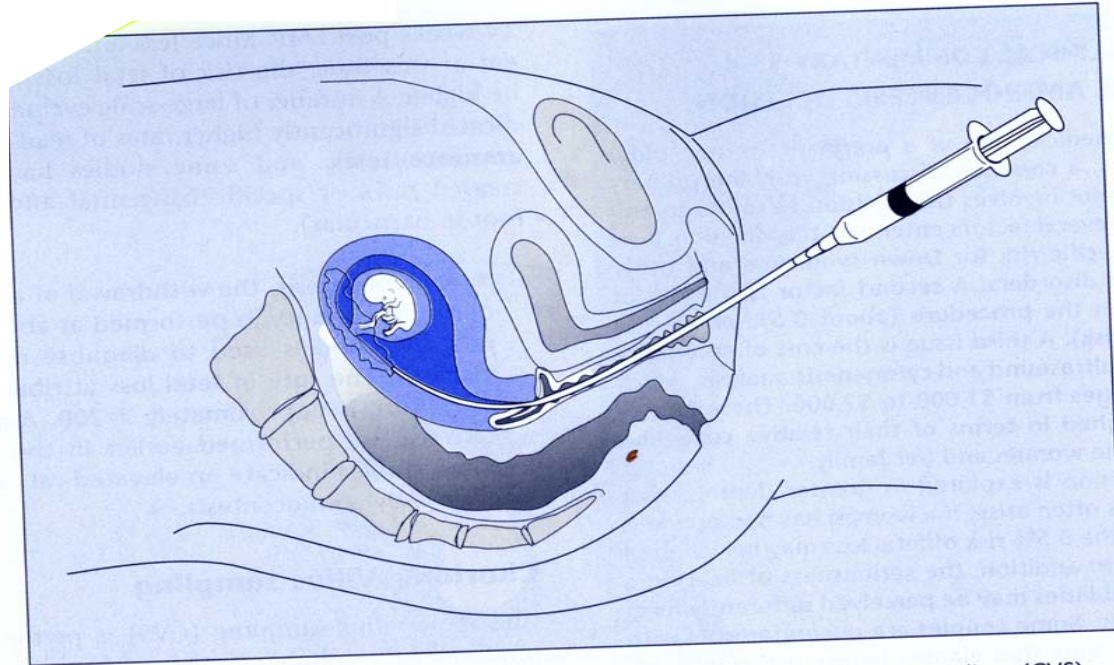
Limb reduction defects  
Many chondrodystrophies,  
including thanatophoric  
dysplasia and osteo-  
genesis imperfecta

Overall: 30-50% sensitivity  
99% specificity

\*Detection rate varies by condition

## Chorionic villus sampling (CVS)-

aspirating fetal trophoblasts either trans-cervically or -abdominally



**FIG. 13-5** A schematic illustration of a transcervical chorionic villus sampling (CVS) procedure. With ultrasound guidance, a catheter is inserted, and several milligrams of villus tissue are aspirated.

- CVS performed at 10-11 weeks vs 15-17 weeks for amniocentesis after last menstrual period. More rapid results generally.
- Risk of fetal loss is higher than amnio procedure: 1-1.5%
- Confined placental mosaicism (in extraembryonic tissue only): 1-2%
- Possible limb shortening when CVS performed early (<10 weeks).

## **Amniocentesis - a diagnostic test:**

At 15-17 weeks after last menstrual period, a needle is inserted into the abdominal wall into the amniotic sac guided by real-time ultrasound. Amniocytes are cultured and cytogenetic (chromosomal studies) are performed.

Results usually available in 10-12 days.

Risk of fetal loss due to procedure 0.5%. Women <35 yrs of age: probability of loss due to amnio > probability of detecting a diagnosable condition.

### **Box 13-3      Indications for Prenatal Diagnosis by Amniocentesis**

Maternal age > 35 years

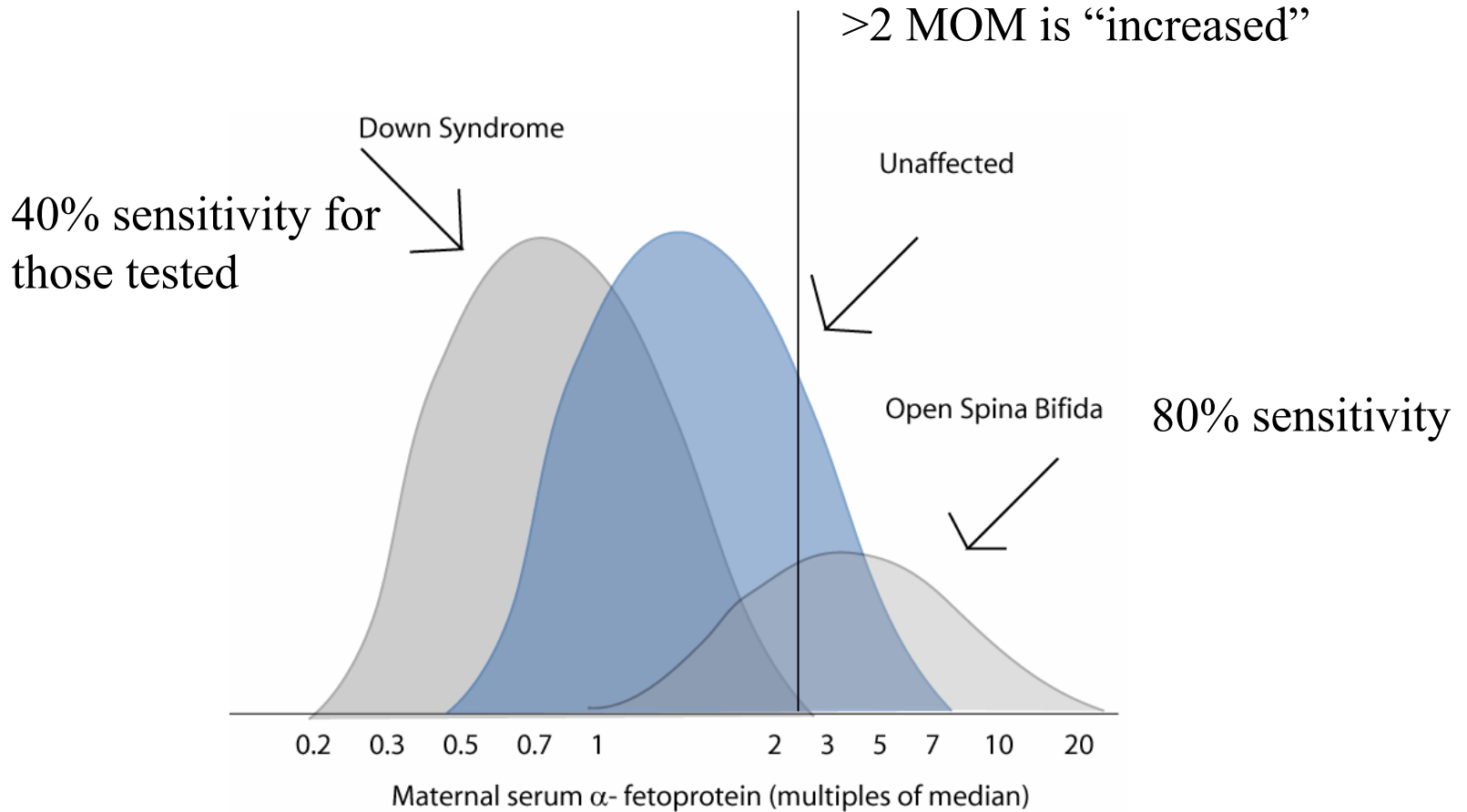
Previous child with chromosome abnormality

History of structural chromosome abnormality in one of the parents

Family history of genetic defect that is diagnosable by biochemical or DNA analysis

Risk of neural tube defect (NTD)

# Alpha fetoprotein screening



MSAFP levels in mothers carrying normal fetuses vs. fetuses with Down syndrome or open spina bifida. It is lowered in Down syndrome somewhat, and substantially increased in spina bifida.

Low pos. predictive value (6%), but justified because 90-95% NTD have no family history.

Better sensitivity for Down syndrome screening is achieved with triple test of:

MSAFP,  
serum unconjugated estriol,  
and  
chorionic gonadotropin.

MSAFP alone = 40% of Down syndrome pregnancies.

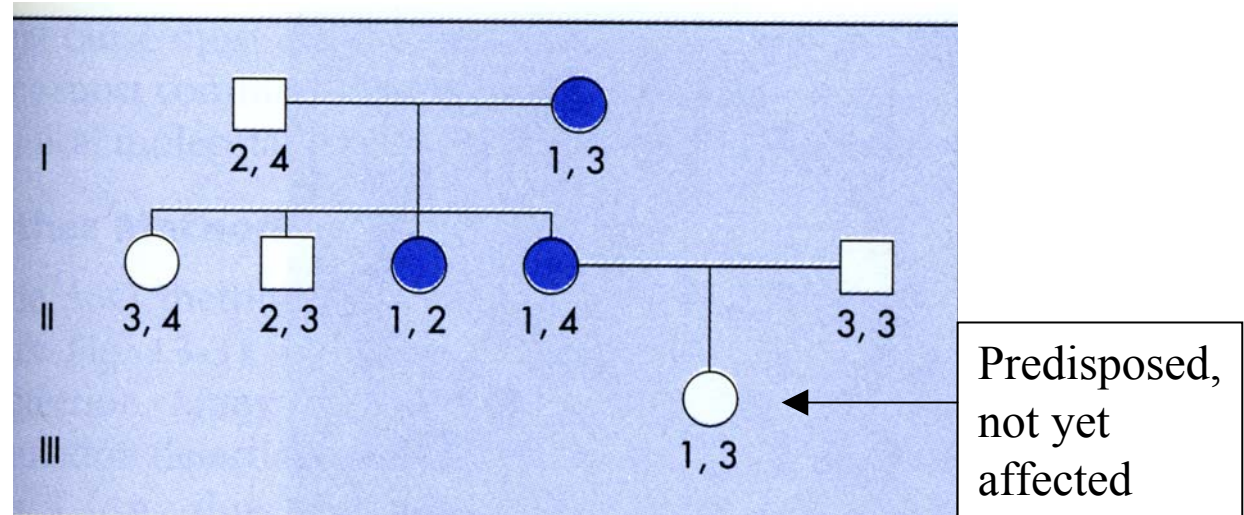
All three = 70% (false positive rate of 5%).

**Table 13-5 Selected Single-Gene Disorders for Which Family Screening and Prenatal Diagnosis by DNA are Available**

<b>Disease</b>	<b>DNA Analysis</b>
Neurofibromatosis type 1	Linkage analysis/ direct mutation analysis
Myotonic dystrophy	Direct mutation analysis
Cystic fibrosis	Direct mutation analysis
Sickle-cell disease	Direct mutation analysis
Fragile X syndrome	Direct mutation analysis
Hemophilia A	Direct mutation analysis/ linkage analysis
Huntington disease	Direct mutation analysis
Duchenne muscular dystrophy	Direct mutation analysis/ linkage
Familial breast cancer	Direct sequencing/ linkage
Hemochromatosis	Direct mutation analysis

# Screening for disease predisposition by genetic linkage analysis

Age-related expression, eg. breast cancer



Test direct descendants and relatives of affected individual using genetic marker closely linked to disease locus. Grandmaternal allele 1 is linked to the disease allele in this family. If III-1 were tested, this would be a *presymptomatic* test.

Table 13-6 Selected Inborn Errors of Metabolism That are Diagnosable Through Amniocentesis and/ or CVS

Disease	Measurable Enzyme
<b>Disorders of Amino Acid/ Organic Acid Metabolism</b>	
Maple syrup urine disease	Branched-chain ketoacid decarboxylase
Methylmalonic academia	Methylmalonic CoA mutase
Multiple caboxylase deficiency	Biotin responsive carboxylase
<b>Disorders of Carbohydrate Metabolism</b>	
Glycogen storage disease, type 2	$\alpha$ -Glucosidase
Galactosemia	Galactose-1-uridyl transferase
<b>Disorders of Lysosomal Enzymes</b>	
Gangliosidosis (all types)	$\beta$ -Galactosidase
Mucopolysaccharidosis (all types)	Disease-specific enzyme
Tay-Sachs disease	Hexosaminidase
<b>Disorders of Purine and Pyrimidine Metabolism</b>	
Lesch-Nyhan syndrome	Hypoxanthine-guanine phosphoribosyl transferase
<b>Disorders of Peroxisomal Metabolism</b>	
Zellweger syndrome	Long-chain fatty acids

# Newborn genetic screening

- Opportunity for presymptomatic detection and intervention
- All states screen newborns for  
PKU,  
galactosemia and  
hypothyroidism  
Mental retardation in these conditions  
can be prevented by early detection and effective intervention.
- Other screening programs include hemoglobinopathies (up to 15% with HbS die of infections before age 5), muscular dystrophy, and other rare biochemical disorders that are prevalent in local groups (Glycogen storage disease type II, MSUD, methylmalonic acidemia in Amish).

Table 13-3 Characteristics of Selected Newborn Screening Programs

Disease	Inheritance	Prevalence	Screening Test	Cost	Treatment
Phenylketonuria	Autosomal recessive	1/10,000-1/15,000	Guthrie test	\$1.25	Dietary restriction of phenylalanine
Galactosemia	Autosomal recessive	1/50,000-1/100,000	Transferase assay	\$1.00	Dietary restriction of galactose
Congenital Hypothyroid	Usually sporadic	1/5,000	Measurement of T4 or TSH	\$1.50	Hormone replacement
Sickle cell disease	Autosomal recessive	1/400-1/600 African Americans	Isoelectric focusing or DNA diagnosis	\$1.50	Prophylactic penicillin

# Heterozygote screening

- Target population: group known to be at risk - often an ethnic background
- Intervention: presentation of risk figures, option of prenatal diagnosis
- Candidate disorders are generally:
  - autosomal recessive,
  - prenatal diagnosis and counseling are available, feasible and accurate

## Phenylketonuria

1 in 10-15K births

Untreated, 95% become moderately/severely MR.

Not identified clinically in 1st year of life

Dietary treatment usually (except those with biotin synthesis defect) results in normal intelligence.

Guthrie test: blood incubated with Phe-deficient *B. subtilis* strain. Growth is related to PHE levels.

Test must be confirmed by repetition, since sensitivity depends on diet.

## Recommended Guidelines:

1. Screening should be voluntary, and confidentiality must be ensured.
  2. Screening requires informed consent.
  3. Providers of screening services have an obligation to ensure that adequate education and counseling are included in the program.
  4. Quality control of all aspects of the laboratory testing, including systematic proficiency testing, is required and should be implemented as soon as possible.
  5. There should be equal access to testing.
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From Elias S, Annas G, Simpson JL (1991) Carrier screening for cystic fibrosis: implications for obstetric and gynecologic practice. *Am J Obstet. Gynecol.* 164: 1077-1083.

**Table 13-4 Selected Examples of Heterozygote Screening Programs in Specific Ethnic Groups**

Disease	Ethnic Group	Carrier Frequency	At-risk couple frequency	Disease incidence in newborns
Sickle-cell disease	African-Americans	1/12	1/150	1/ 600
Tay-Sachs disease	Ashkenazi Jews	1/30	1/900	1/ 3,600
$\beta$ - Thalassemia	Greeks, Italians	1/30	1/900	1/ 3,600
$\alpha$ - Thalassemia	Southeast Asians and Chinese	1/25	1/625	1/ 2,500
Cystic Fibrosis	Northern Europeans	1/25	1/625	1/ 2,500

Accurate carrier testing for **Tay-Sachs** (HEXA) has reduced number of newborns 90% since 1970. Tay Sachs results in accumulation of GM2 ganglioside which leads to blindness, seizures, hypotonia, and death by age 5.

**CF** screening is about to be recommended for Caucasians by Am. College of Ob/Gyn, but has not by American College of Med Genetics. Cost/benefit is less clear-cut. (1/600 couples would be heterozygotes, 1/2400 affected ). Counseling requirements would be impractical.

# Summary

- Genetic screening and testing are well accepted by health care professionals and patients for certain conditions.
- Criteria for screening and/or testing include (a) benefits to the patient, (b) test accuracy, sensitivity and specificity, and (c) availability of resources for diagnosis and treatment and a system for communicating results efficiently and effectively.
- Prenatal, newborn and heterozygote screening and testing are available











