Genetic Testing for Prenatal Diagnosis of Chromosomal Abnormalities

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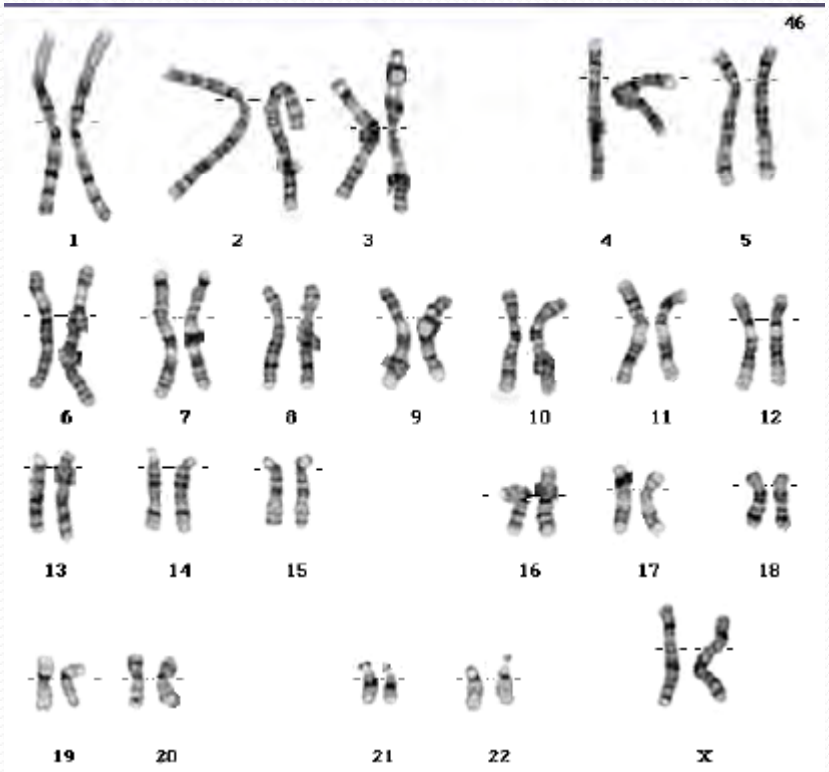
Outline

- Standard Cytogenetics Testing
- Molecular cytogenetics
- Diagnostic Yield of screening methods

Issues

- Time: 2-3 weeks for results
- Small deletion and duplications will be missed (<5MB)
- Living tissue required



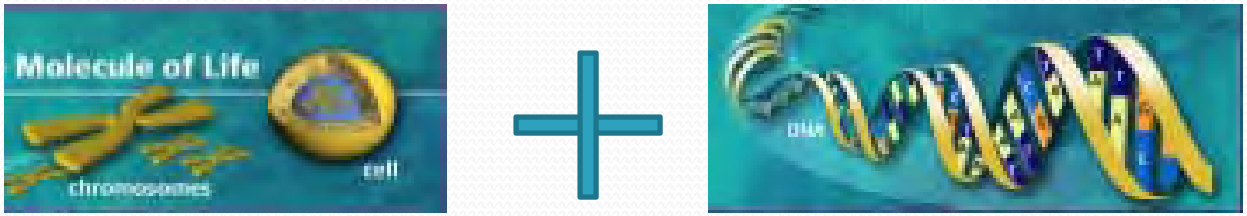
Data

- Total prenatal diagnosis: 1170
- Abnormal karyotypes: 69

Data

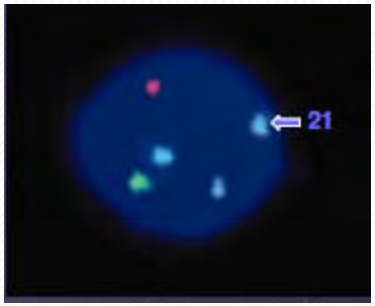
Reason for referral	Total	Abnormal karyotype	Positive predictive value
Screen positive	974	42	4.3%
Abnormal ultrasound findings	66	7	10.6%
Advanced maternal age	56	4	7.1%
Positive family history	45	7	15.5%
Parental anxiety	23	0	0
others	4	0	0

Molecular Cytogenetics

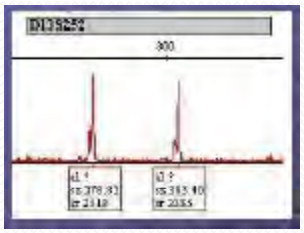


Molecular Cytogenetics

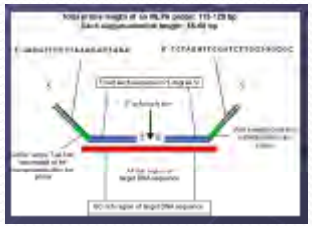
Molecular cytogenetic techniques



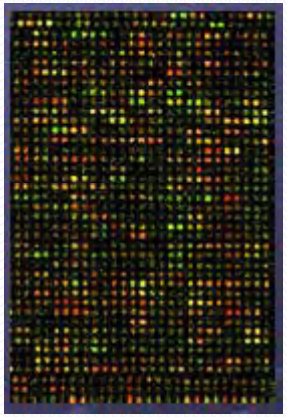
FISH



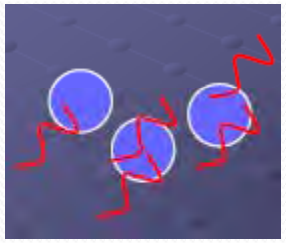
QF-PCR



MLPA



Array-CGH



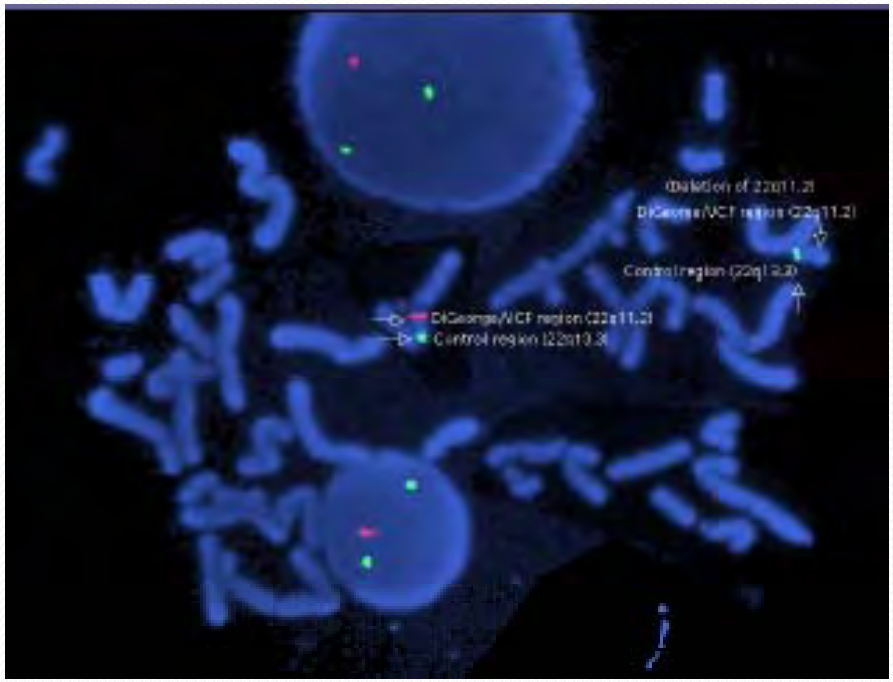
BACs on Beads

FISH

- Quick answers (24-48 hours)

But:

- Labor intensive
- **You must know what you are looking for**



QF-PCR

- Quick answers
- Limited to common chromosomal aneuploidies involving chromosomes 21, 18, 13, X and Y

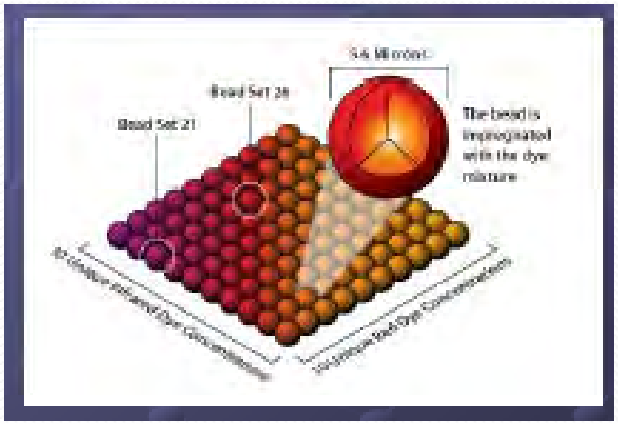
Array-CGH

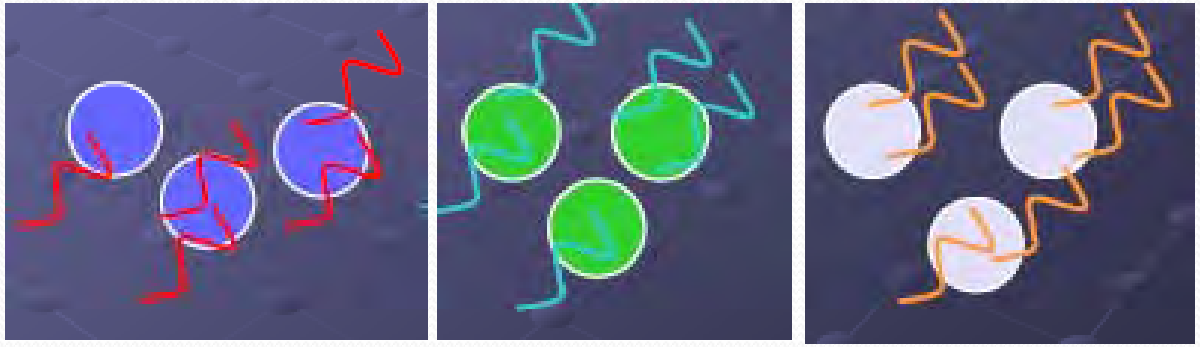
- Advantages:
  - High throughput
  - High resolution
- Disadvantages:
  - Costly
  - Detecting changes with unknown clinical significance

ACOG committee opinion November 2009

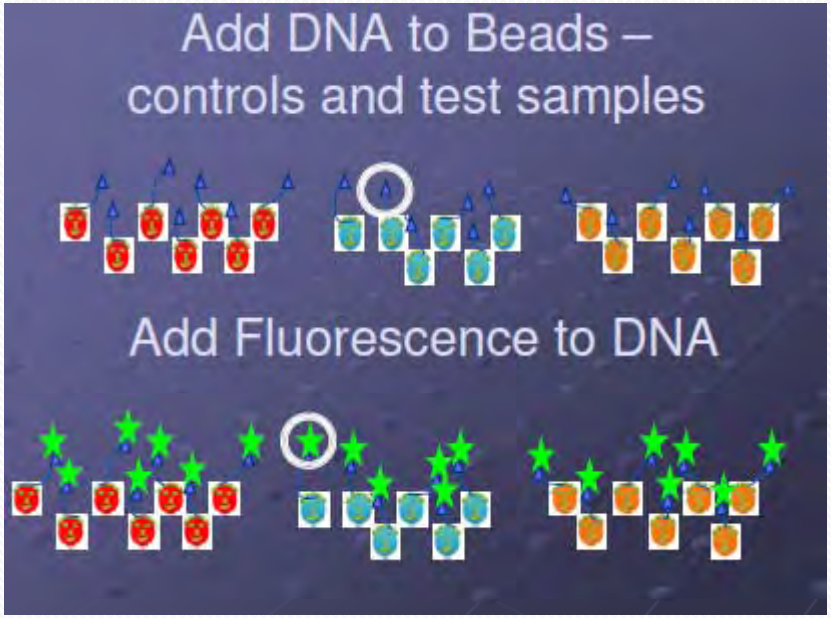
- Array-CGH in prenatal diagnosis
  - Karyotyping remains the principal cytogenetic tool in prenatal diagnosis
  - Targeted array-CGH in concert with genetic counseling, can be offered as an adjunct in prenatal cases with abnormal anatomic findings and a normal karyotype, as well as in case of fetal demise with congenital anomalies
  - Targeted array-CGH may be useful as a screening tool; however, further studies are necessary and are underway to fully determine its utility and its limitations

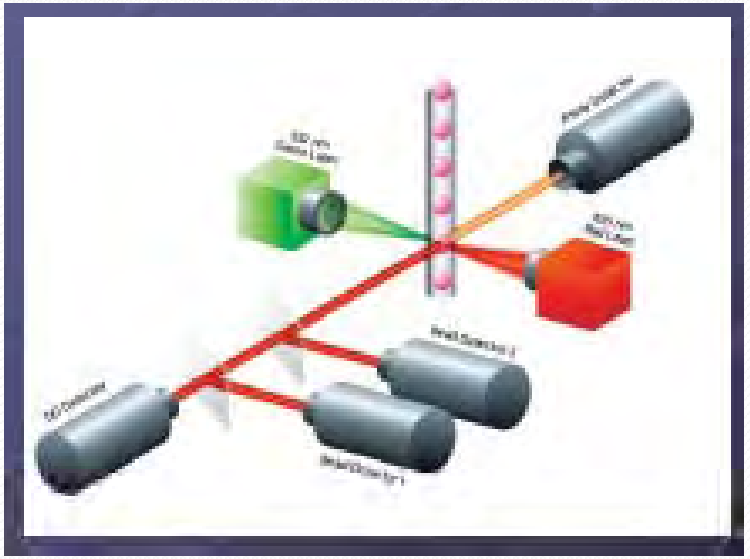
BACs on Beads



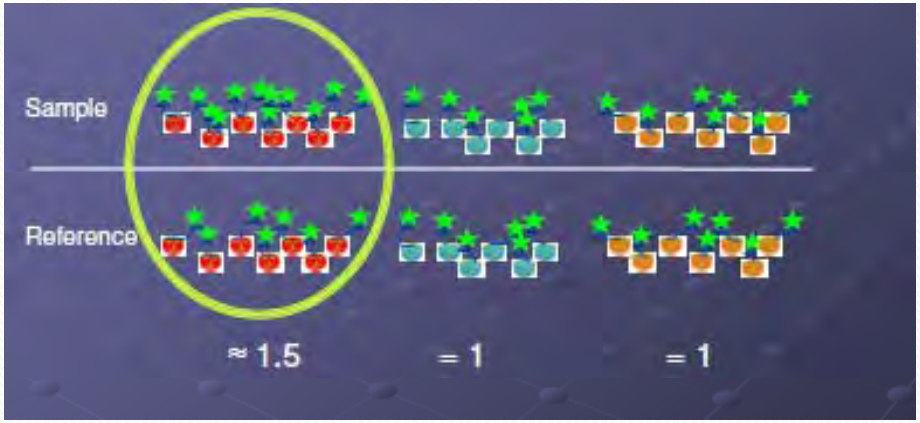


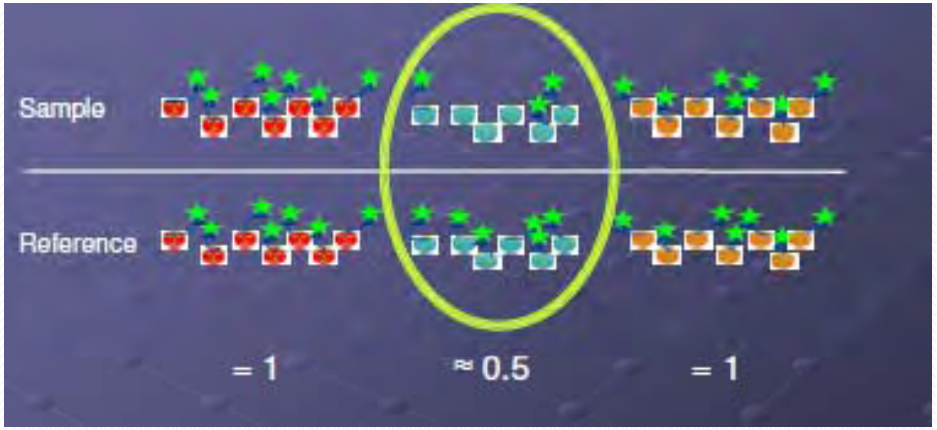
BACs on Beads



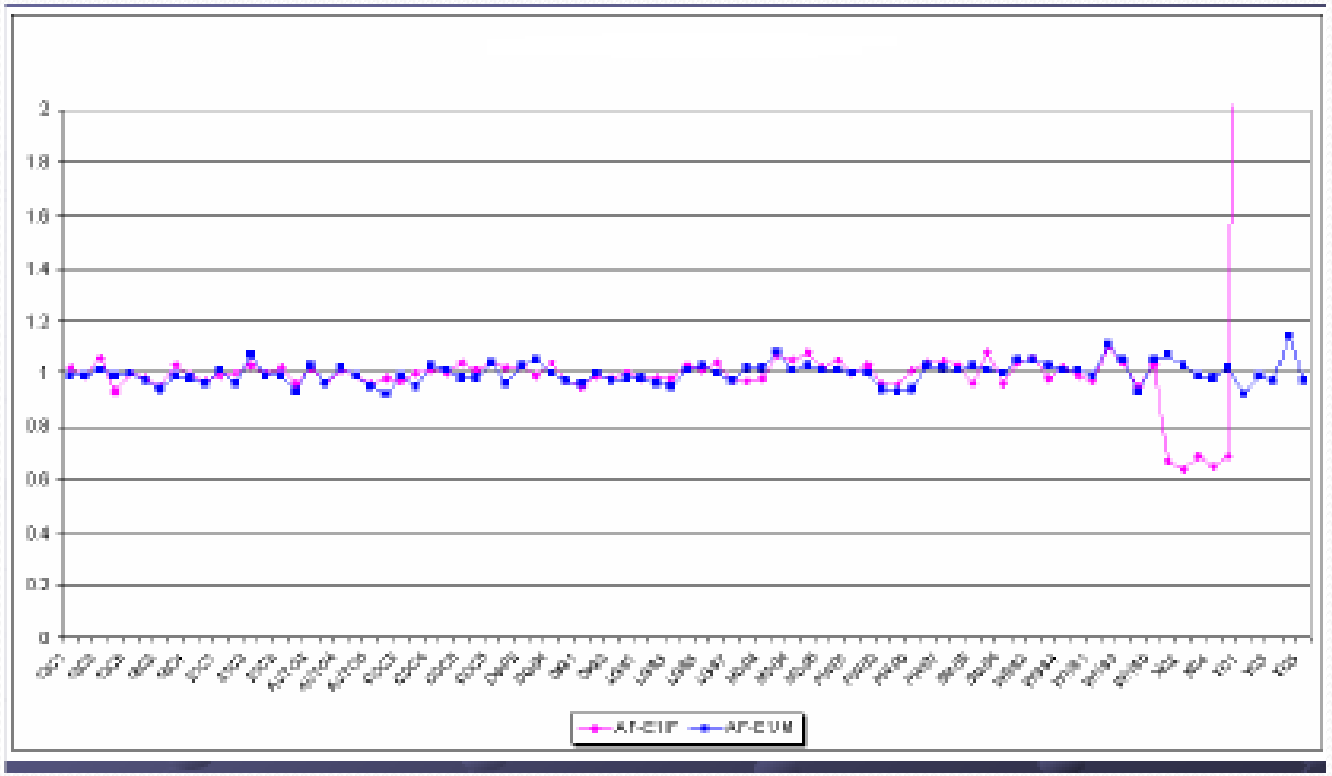


BACs on Beads

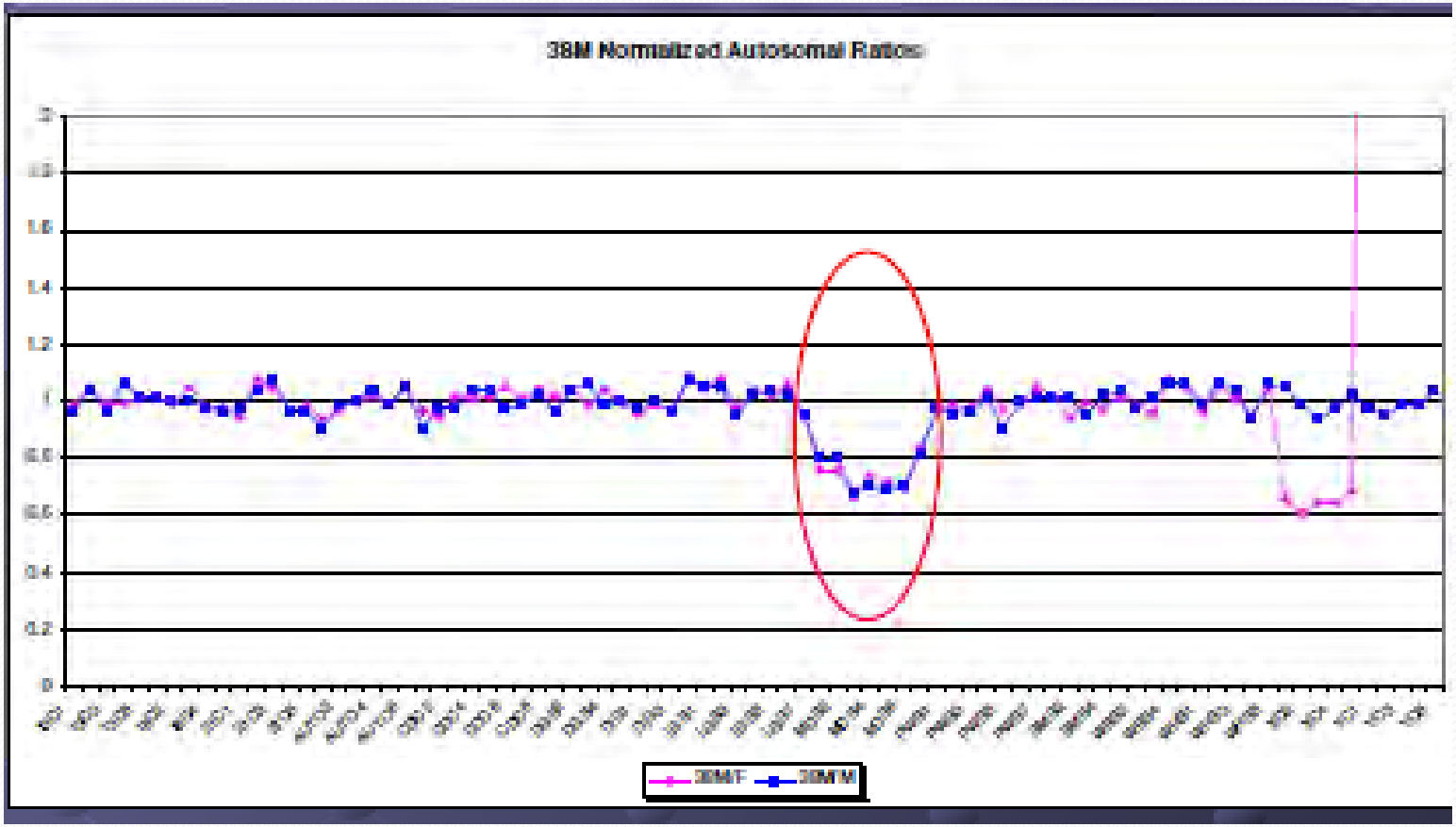




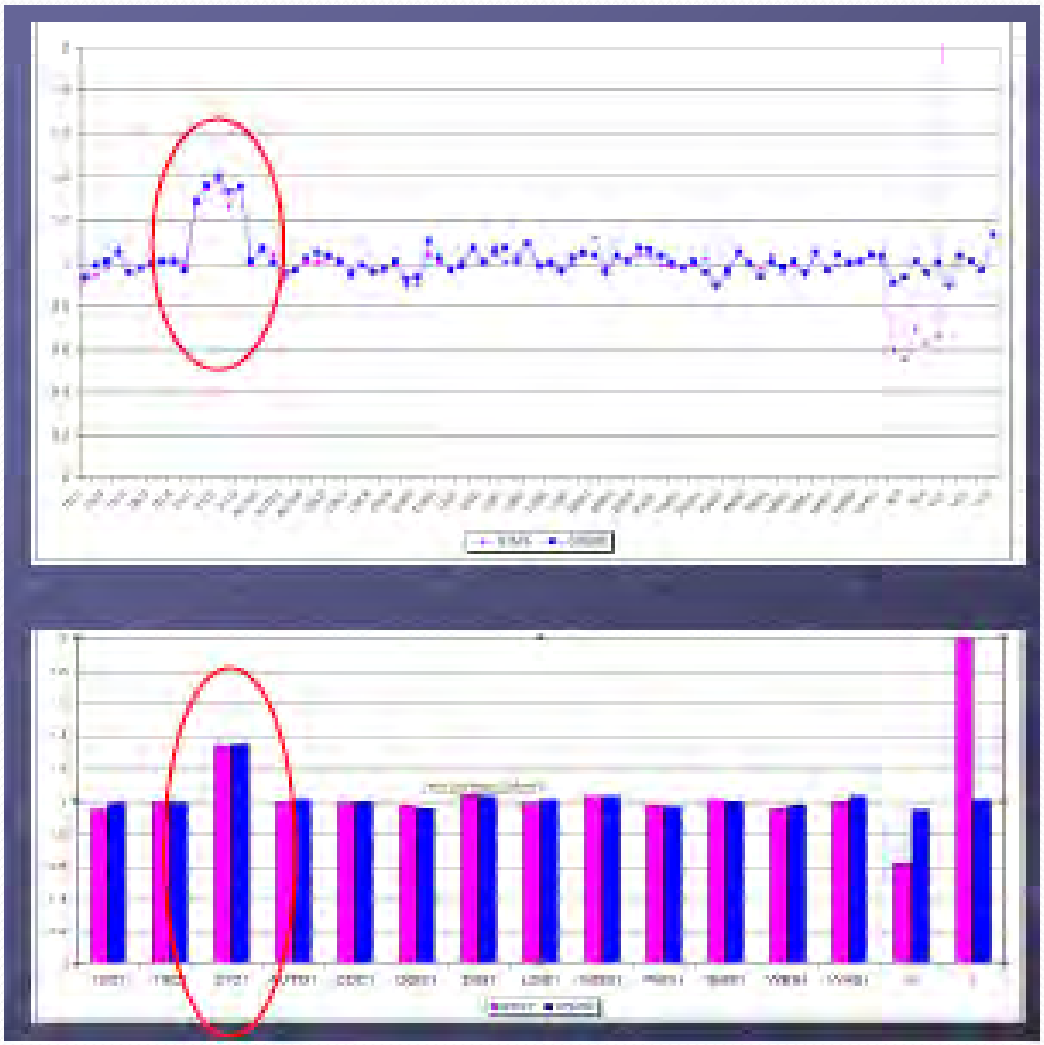
Analysis, normal



Analysis, Miller-Dieker syndrome



Down Syndrome



Screening of Microdeletion Syndromes

- Indications:
  - Significant morbidity and mortality
  - Relatively common
  - Majority of cases are caused by deletion
  - May be missed on detailed sonography
  - Not detectable by karyotyping

Microdeletion syndromes

Microdeletion Syndromes

- Prevalence: **1/1600** deliveries
- Prevalence of Down syndromes: **1/800** deliveries

- With 4 million deliveries in the US/year
  - Approximately 5700 Down syndrome births per year
  - Approximately 2500 microdeletion syndromes per year

Screening of Microdeletion Syndromes

- ***Would increase detection of serious disorders associated with mental retardation or birth defects by approximately 40%***

Detection rates

Conditions	Fraction	Decimals
Numeric chromosome abnormalities only	**1 in 326**	0.003
Deletion syndromes only	**1 in 1426**	0.0007
Deletion syndromes plus numeric abnormalities	**1 in 265**	0.003
Karyotype only	**1 in 154**	0.006
Karyotype plus microdeletion syndromes	**1 in 139**	0.007

Recommendation

- In otherwise **normal pregnancies** undergoing invasive tests because of advanced maternal age or parental anxiety:

  aneuploidies and microdeletion syndromes

- In pregnancies with **abnormal ultrasound findings**: **targeted array-CGH analysis**

Our study

Table 1. Exclusion criteria.

Factors	Variables	n
Parental	• Pedigree analysis	
	○ Previous MR/DD child	1
	○ Family history♦ of MR/DD	8
	○ Recurrent abortion/ repeated IUFD	5
	○ History of fertility problems	5
	○ Family history of congenital anomaly	2
	• Parental preference	10
Fetal	• Abnormal ultrasound findings	3
Technical	• Blood contamination of the sample	7
Total		41*

♦ first-degree relatives of the parents, *2 women opted for MLPA despite the initial exclusion.

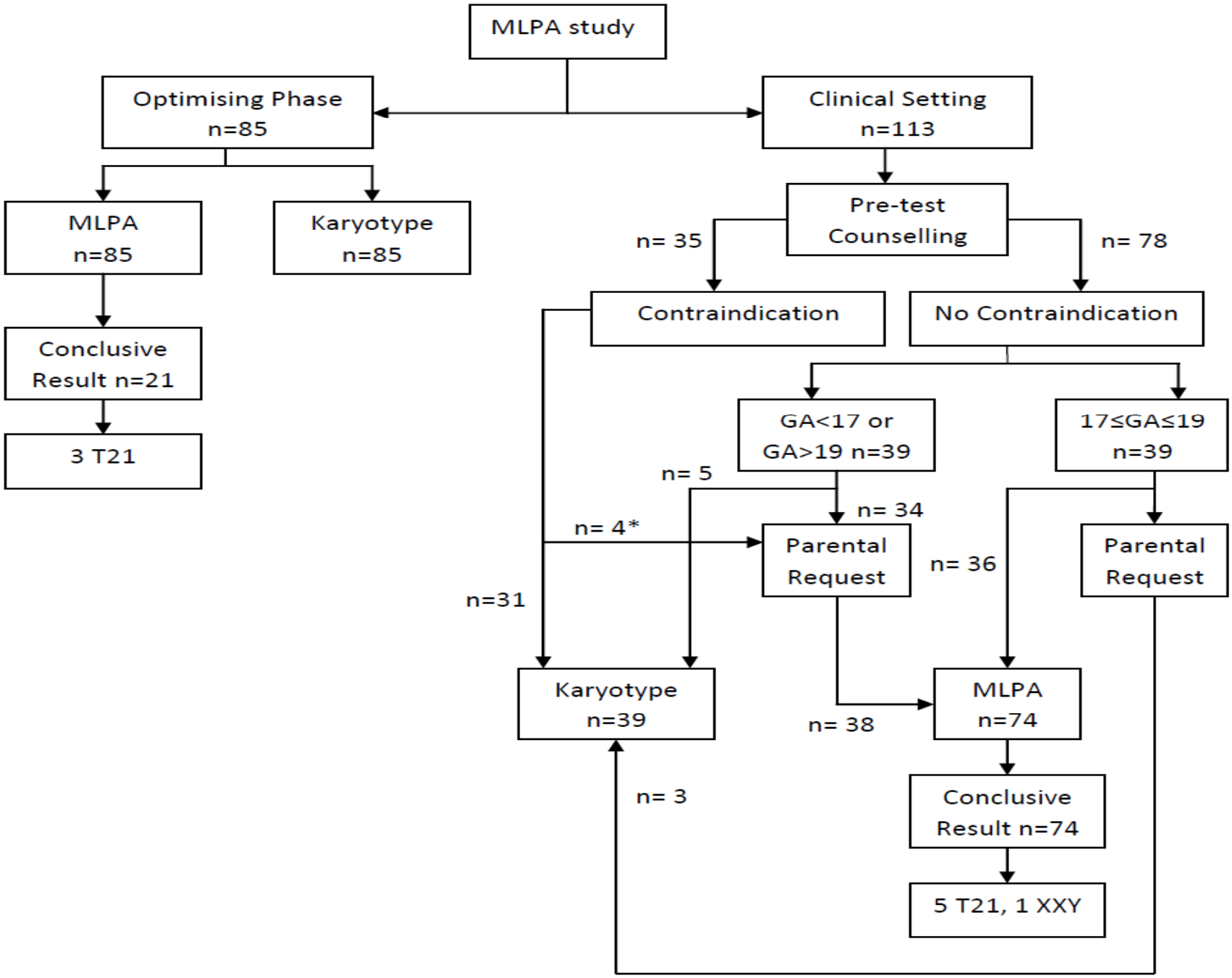

MLPA study
Optimising Phase n=85
Clinical Setting n=113
MLPA n=85
Karyotype n=85
Conclusive Result n=21
3 T21
Pre-test Counselling
n= 35
n= 78
Contraindication
No Contraindication
GA<17 or GA>19 n=39
17≤GA≤19 n=39
n= 5
n= 34
n= 4*
Parental Request
Parental Request
n= 36
n=31
Karyotype n=39
MLPA n=74
n= 38
n= 3
Conclusive Result n=74
5 T21, 1 XXY

Table 2. Chromosomal abnormalities missed by rapid tests in published studies.

	Cain 2005	Kooper 2008	Boormans 2010	Lim 2010	
Rapid test	FISH/ PCR	MLPA	MLPA	FISH	
Total samples	119528	1000	4585	5883	1
Total abnormal	3081	78	124	647	
Abnormal missed by rapid test					
Structural balanced	415	16	18	30	479 (
Structural unbalanced	85	6	2	34	127
Marker chromosome	113	0	4	0	117
Tetraploidy	3	2	0	0	5 (
Sex chromosome abnormality	366	0	0	0	366

Abnormal karyotypes

Category	No	subgroups	No	Pedigree analysis	Detectable by RAD
Common chromosomal aneuploidies	41	T21	34		Yes
		T18	4		
		XXY	1		
		XXX	1		
		XO	1		
Balanced translocations	5	Confirmed hereditary	3	Recurrent abortion: 1 Family history of balanced translocation: 1 Abnormal sonography: 1 Screen positive: 2	Not indicated, no clinically significant
		Not assessed	2		

Abnormal karyotypes

Category	No	Subgroup	No	Pedigree analysis	Detectable by RAD
Structural rearrangements	2	Ring chromosome, deletion	1	Abnormal sonography	Not indicated
		deletion	1	Recurrent abortion	Not indicted
Triploidy	1				
Double trisomy	1	48,XX,+18, +mar	1	Screen positive	Yes

Abnormal karyotypes

Category	Subgroup	No	Karyotype	Pedigree analysis	Detectable by RAD
Mosaicism	Sex chromosomes	2	mos, 45,X[19[/47,XXX[11]	Screen positive: 1 Positive family history: 1	No
			mos, 47,XXX[26]/46,XX[6]		
	Autosomal chromosomes	2	mos, 46,XY[22]/47,XY,+18[8]	Screen positive: 2	No
			mos, 46,XY[36]/47,XY,+13[2]		
	Marker chromosomes	2	mos, 46,XY[36]/47,XY,+mar[4]	Screen positive: 1 Positive family history: 1	No
			mos, 47,XX,+mar[17]/46,XX[13]		