



neoBona®
Certitude for you



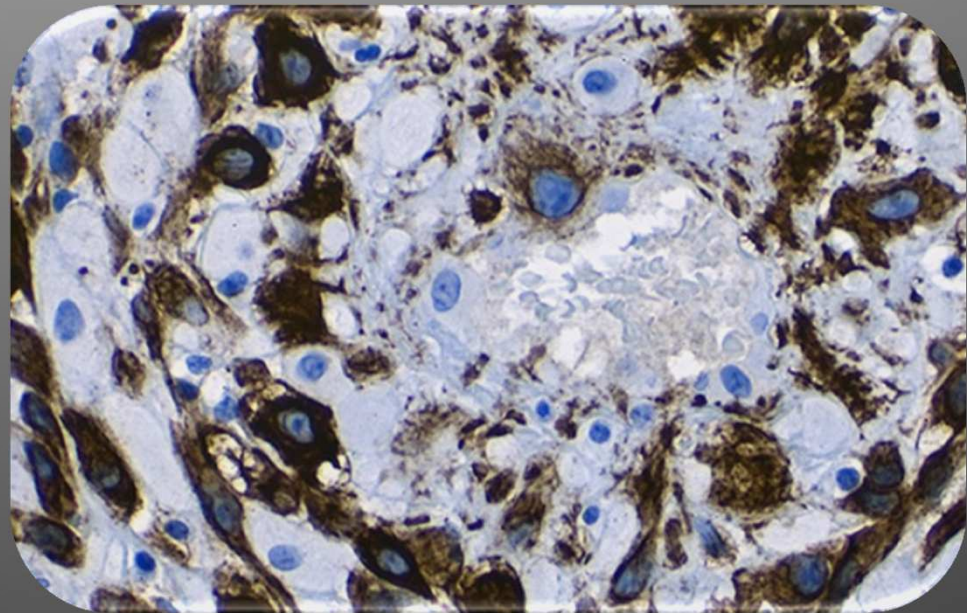
NIPT *onfield*

*DNA fetale circolante e
Test Prenatali non invasivi*

Lamberto Camurri PhD

*RDI Rete Diagnostica Italiana
Padova*

*Università Tor Vergata Roma
Roma*



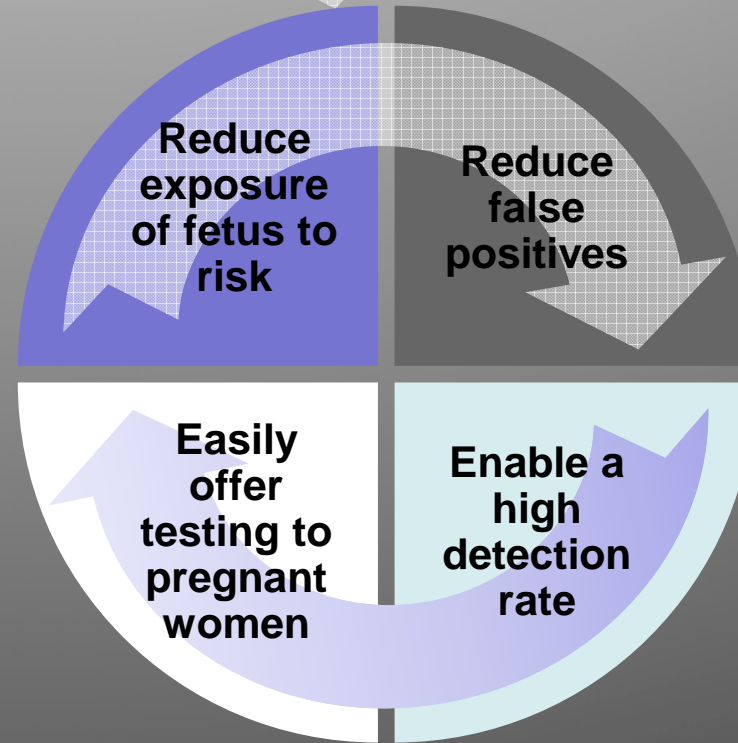
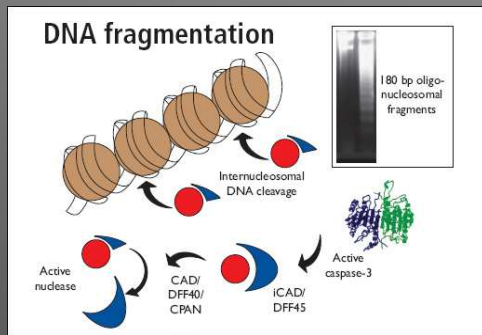
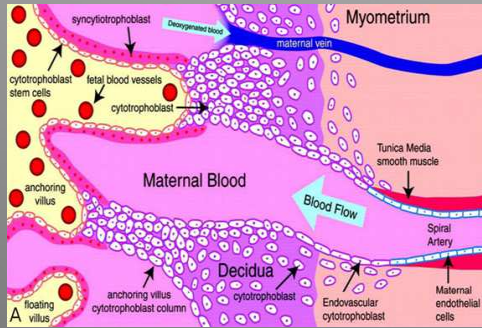
NIPT

NON

INVASIVE

PRENATAL

TEST




NIPT

NON

INVASIVE

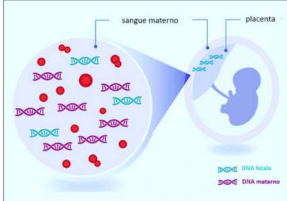
PRENATA
L

TEST


Ministero della Salute
Consiglio Superiore di Sanità
Sezione I

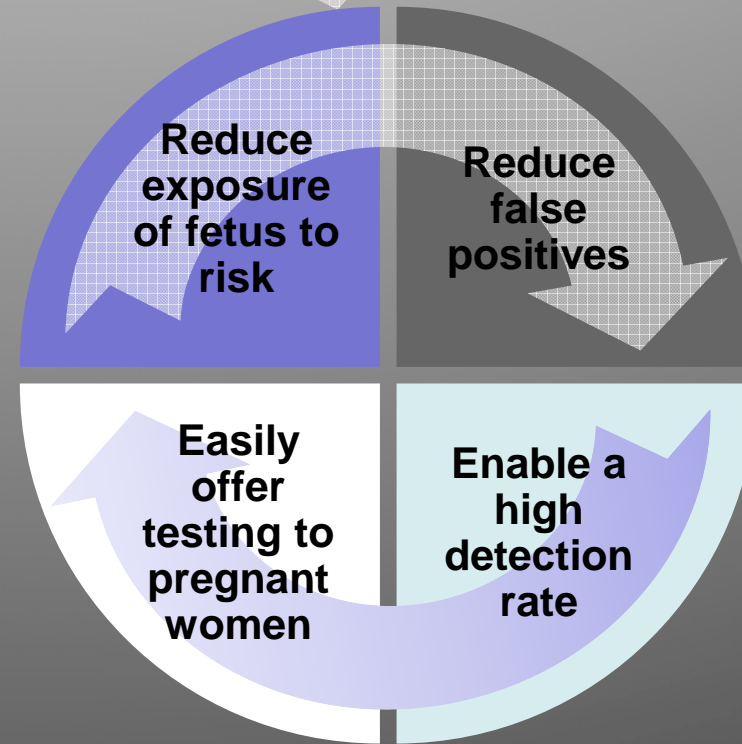
Linee-Guida

**Screening prenatale non invasivo basato sul DNA
(Non Invasive Prenatal Testing – NIPT)**



Nel plasma materno in gravidanza sono presenti cellule fetali nucleate e DNA libero (cffDNA) non-cellulare proveniente dalle cellule della placenta

Maggio 2015



NIPT

NON

INVASIVE

PRENATA
L

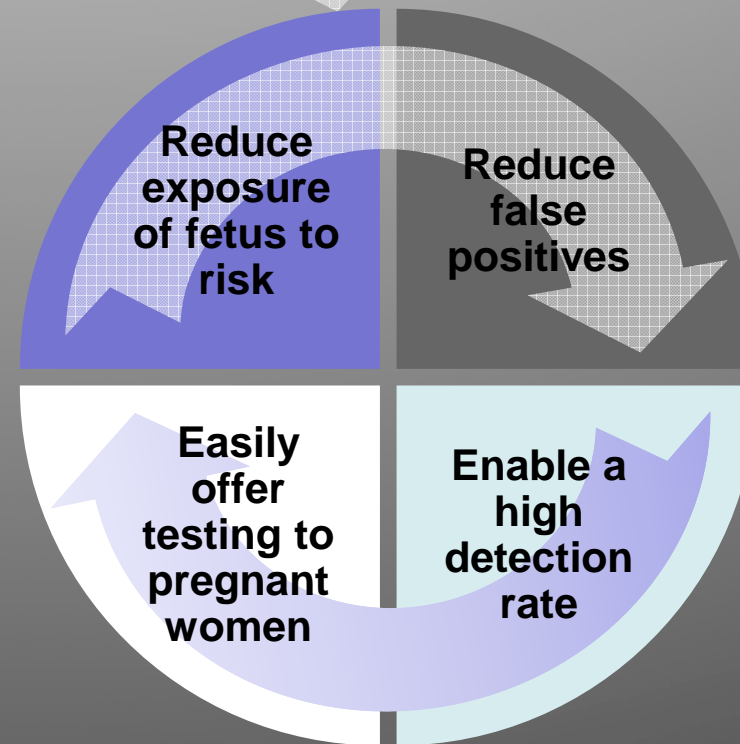
TEST

CONSULENZA PRE TEST

La disponibilità di varie tecniche che utilizzano il DNA fetale per la ricerca di anomalie genetiche nel corso della gravidanza rende tassativa la consulenza pre-test, che rappresenta lo strumento di elezione per informare la gestante/coppia sulle diverse opzioni disponibili.

Infatti, è stato dimostrato che la comprensione delle potenzialità e dei limiti del test cfDNA/NIPT è fortemente compromessa, in assenza della consulenza.

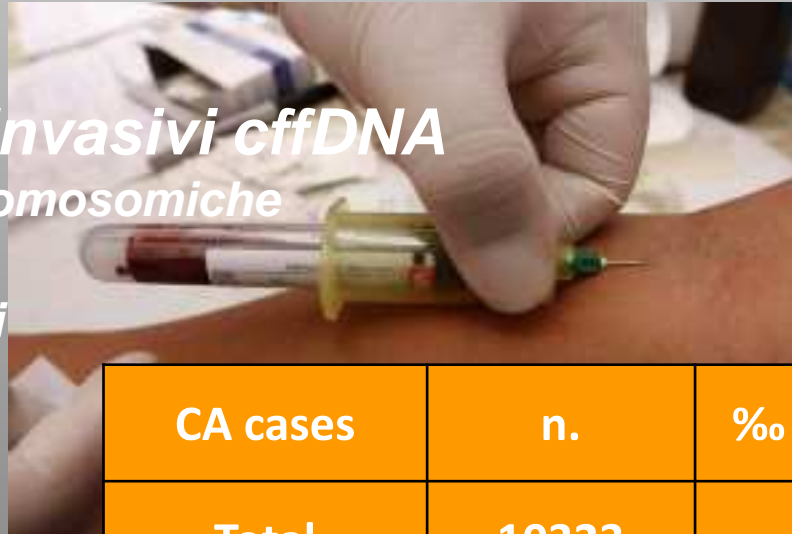
La consulenza pre-test deve essere effettuata da uno specialista esperto di medicina fetale.





Screening non invasivi cffDNA

Rischio Anomalia Cromosomiche



T21 1/700 nati

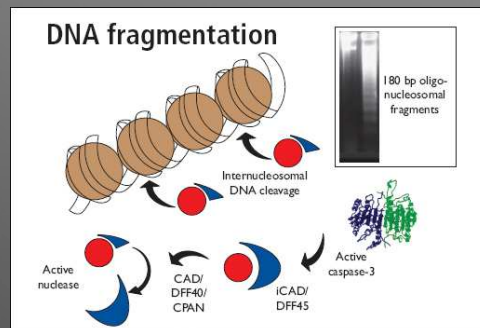
T18 1/2000

T13 1/5000

X/Yaneupl.

RCA

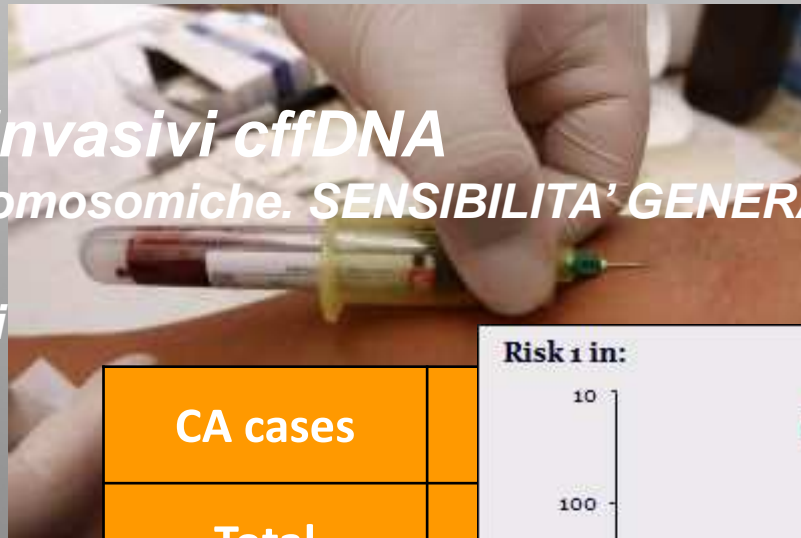
CA cases	n.	%o prevalence	% CA
Total	10323	4,4	
T21 T18 T13	7335	3,1	71 53 - 13 - 5
X-Y trisomies	473	0,2	5
X0	778	0,33	8
RCA	1737	0,7	17



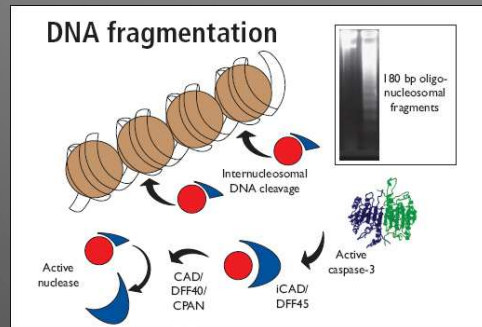


Screening non invasivi cffDNA

Rischio Anomalia Cromosomica. SENSIBILITA' GENERALE



T21 1/700 nati
 T18 1/2000
 T13 1/5000
 X/Yaneupl.
 RCA



CA cases	Risk 1 in:	% CA
Total	10	
T21 T18 T13	100	48 < 77
X-Y trisomies	1000	5
X0	10,000	8
RCA	100,000	10 > 40
	1,000,000	



Screening non invasivi cffDNA

Rischio Anomalie Cromosomiche. SENSIBILITA' GENERALE

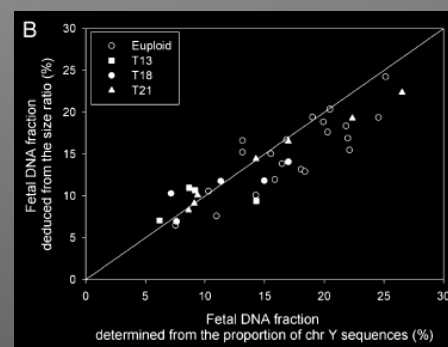
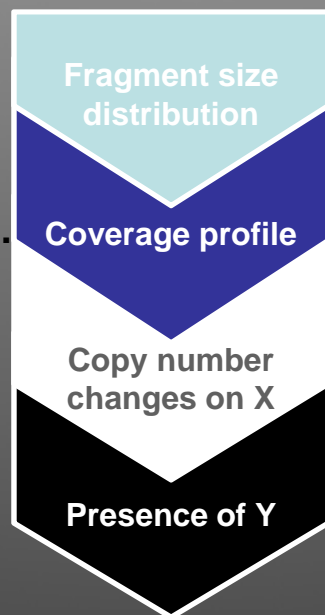
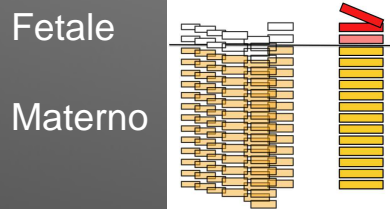
Fetal fraction and Expected ratio for trisomy

Fetal Fraction	Expected ratio for Trisomy
4%	1.02
10%	1.05
20%	1.10
40%	1.20

SNP

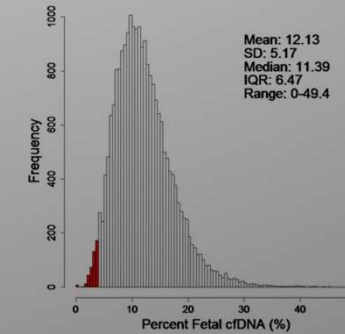
ABRSJA5517	Maternal (buffy coat)	A/C	G/G	C/T	A/T	A/A	A/G	C/T	C/C	A/C	A/G
	Fetal (cfDNA)					A/G			C/C		

Fragment size, #X CNV & #Y



Fetal fraction and Expected ratio for trisomy

Fetal Fraction	Expected ratio for Trisomy
4%	1.02
10%	1.05
20%	1.10
40%	1.20



NIP Tonfield 1000

Fetal fraction and test failure

Cases	1200
Successful 1 st tier	1187
Successful 2nd tier	9 (0,7%)
Low DNA	7
High variance/1 obese	2
Double fail	4 (0,3%)
Low DNA-HV / FIV ovod.	2
High Variance / obese	2

Fetal fraction and gestation

weeks

Mean weeks	12,34
Mean fetal fraction	11,5%



Screening non invasivi cffDNA

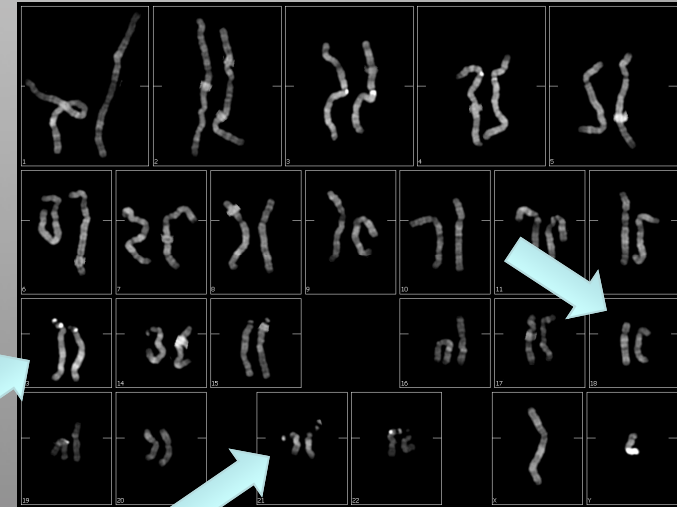
Rischio Anomalie Cromosomiche. SENSIBILITA' SPECIFICITA'

Validation and aneuploidy risk prediction: **high**

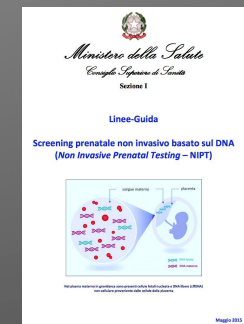


Frequenza Anomalie Cromosomiche Autosomi

T21 1/700 nati+ivg
T18 1/2000
T13 1/5000



		Trisomy 21	Trisomy 18	Trisomy 13
CVS trophoblast	False positive rate/specificity	0.08%	0.06%	0.2%
62000 cases*	False negative Population rate	0.02%	0.01%	NS
	False negative/sensitivity	0.74%	1.59%	0.74%
NIPT total 2013-2015	False positive rate/specificity	0.09%	0.13%	0.13%
	False negative Population rate	0.08%	0.06-0.12%	0.18-0.36%
	False neg. /detection rate	0.8%	3.7%	9%



Screening non invasivi cffDNA

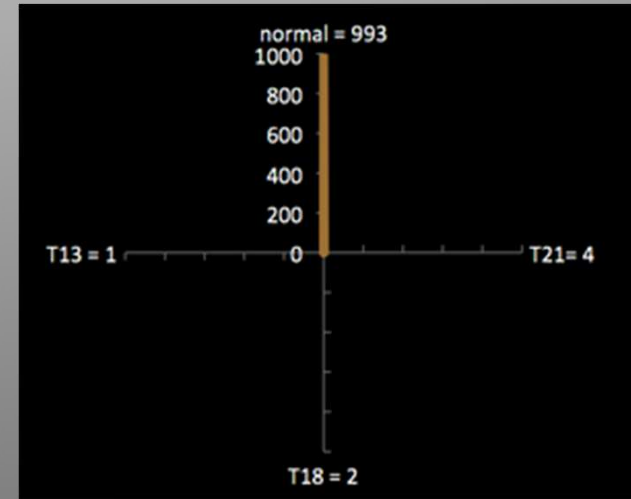
Rischio Anomalie Cromosomiche. SENSIBILITA' SPECIFICITA'

Validation and aneuploidy risk prediction: **high**

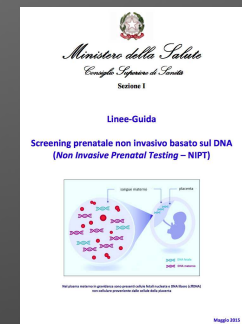


Frequenza Anomalie Cromosomiche Autosomi

T21 1/700 nati+ivg
T18 1/2000 nati
T13 1/5000 nati



		Trisomy 21	Trisomy 18	Trisomy 13
CVS trophoblast	False positive rate/specificity	0.08%	0.06%	0.2%
62000 cases*	False negative Population rate	0.02%	0.01%	NS
	False negative/sensitivity	0.74%	1.59%	0.74%
NIPT total 2013-2015	False positive rate/specificity	0.09%	0.13%	0.13%
	False negative Population rate	0.08%	0.06-0.12%	0.18-0.36%
	False neg. /detection rate	0.8%	3.7%	9%



Screening non invasivi cffDNA

Rischio Anomalie Cromosomiche. SENSIBILITA' SPECIFICITA'

Validation and Aneuploidy risk prediction: **medium**

Frequenza Anomalie Cromosomiche SEX

45X

47XXX,47XXY,47XYY

65-90%

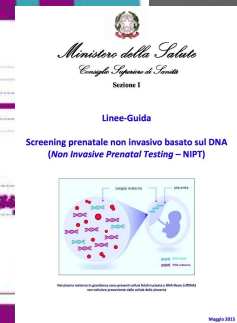
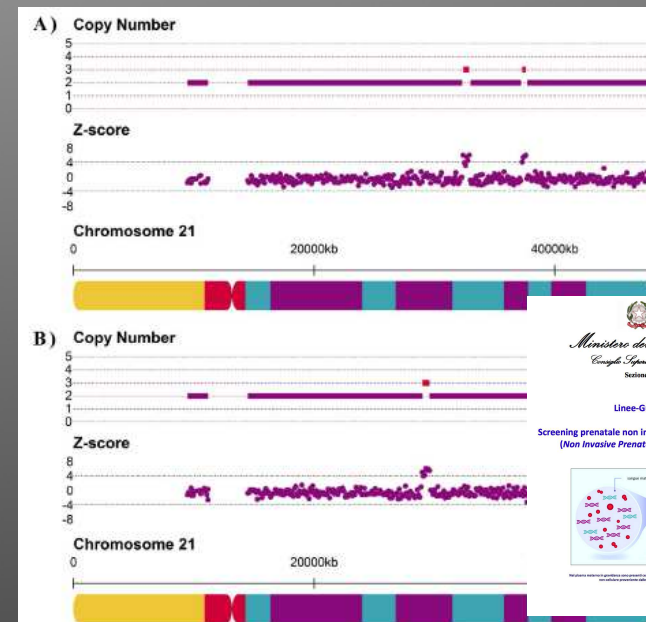
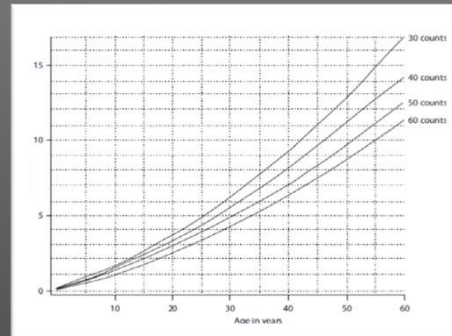


Figure 3. Detection of maternal copy number variations (CNVs)

Screening non invasivi cffDNA

Rischio Anomalie Cromosomiche. SENSIBILITA' SPECIFICITA'

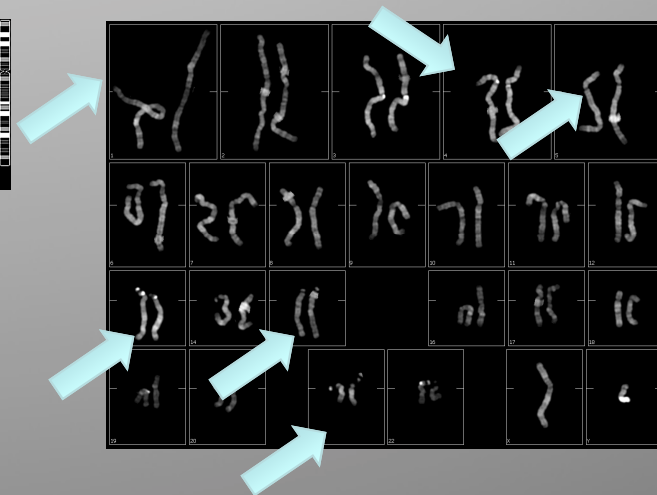
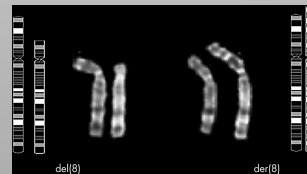
Validation and Aneuploidy risk prediction: **low/not**

validated....

Frequenza Anomalie Cromosomiche

RCA Altre (15-50%) >1/1800

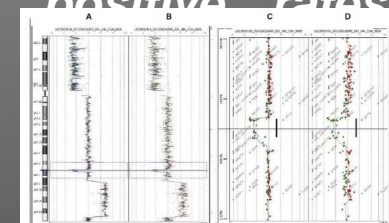
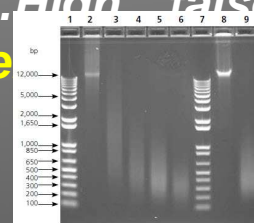
DELEZIONI > 10 Mb



.....why?

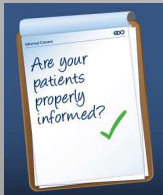
Because tested with simulated fragmentation by.... DNA sonication
..... than NGS and sequencing...High false positive rates

Sonicated DNA vs.....cffDNA fragme



Screening non invasivi cffDNA

Rischio Anomalie Cromosomiche. SENSIBILITA' SPECIFICITA'

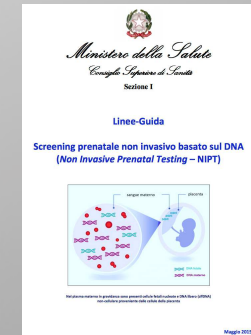


Appropriate Options for Patient Management

	Clinically relevant	Adequately validated	Accurate	Impacts clinical mgmt
Core NIPT (T21, T18, T13)				
Invasive testing				
Expanded NIPT (microdelets)				



Twin pregnancies



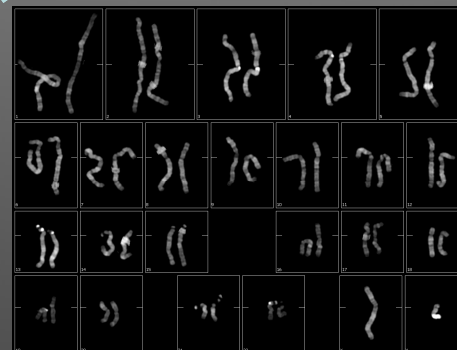
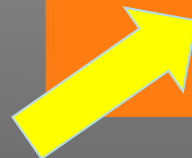
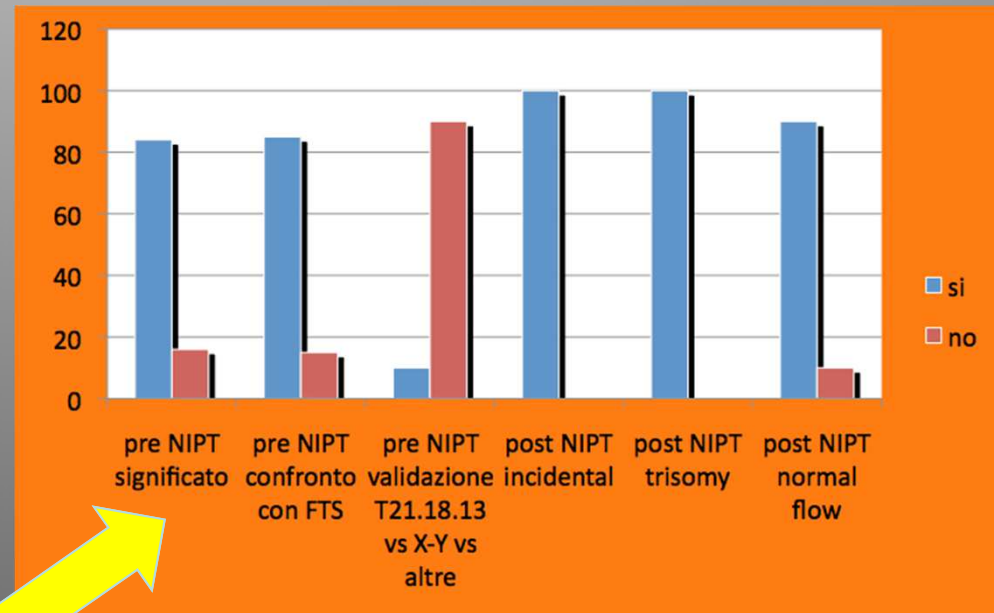
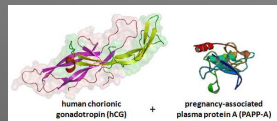
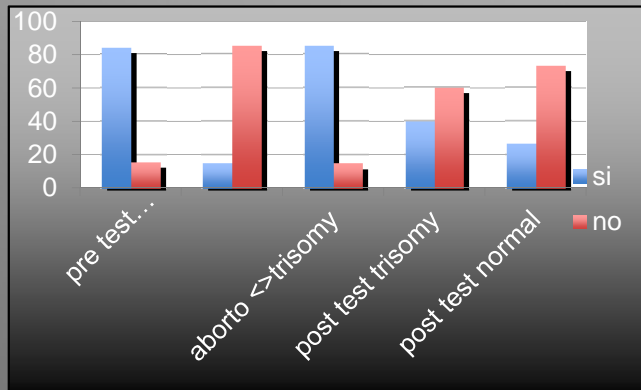
Twins/multiple	
2 max	Valore di rischio distribuito
regular	No sex, no identificazione feto affetto
IVF	No sex, no identificazione feto affetto
Oocytes etero	No sex, no identificazione feto affetto
Sensitivity T21*	93.7% (FN/tested popul. 0.6%)
Specificity T21*	99.8% (FPR 0.23%)
DNA fetal fraction	m 7,4% (12% single)
*Gil M et al 2015	Ultrasound Obstet Gynecol 45, 249



Screening non invasivi cffDNA

Rischio Anomalie Cromosomiche. SENSIBILITA' SPECIFICITA'

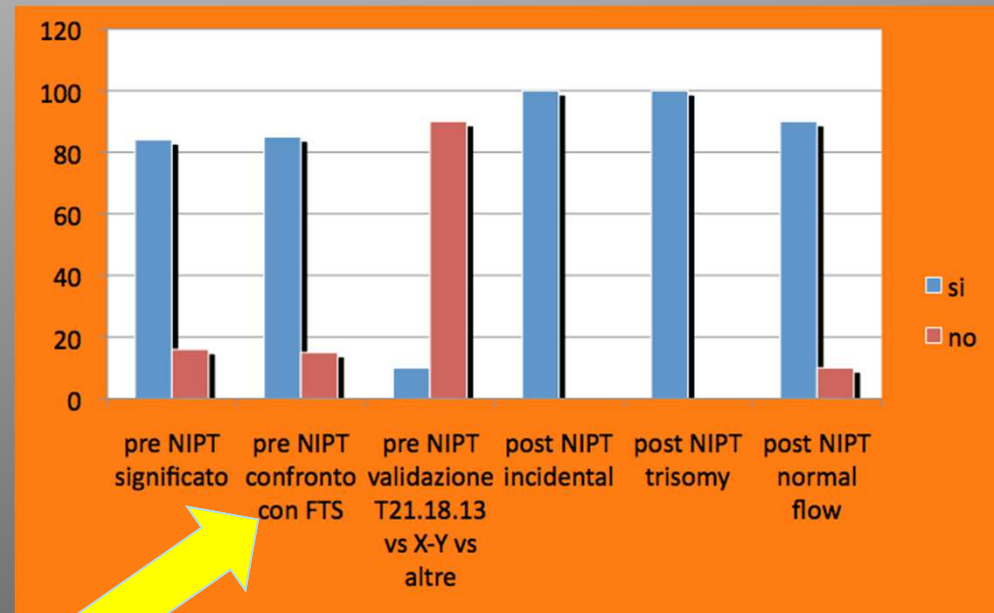
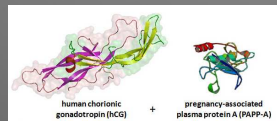
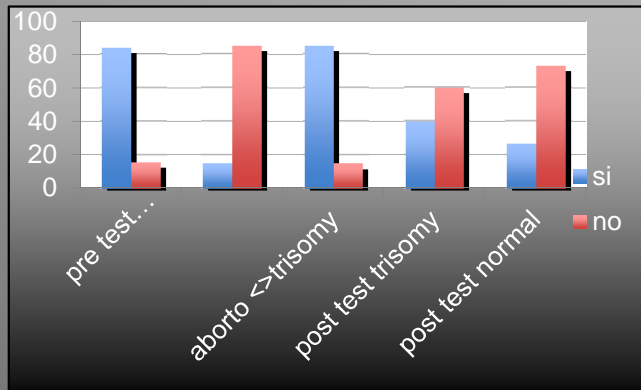
Patient information and understanding **NIPTonfield 1000**



Screening non invasivi cffDNA

Rischio Anomalie Cromosomiche. SENSIBILITA' SPECIFICITA'

Patient information and understanding **NIPTonfield 1000**



2015 NEXT STUDY 16000 parallel cases: data trisomy 21

cffDNA

vs

FTS

DETECTION RATE

36/36 (100%)

28/36 (77.8%)

FALSE POSITIVE RATE

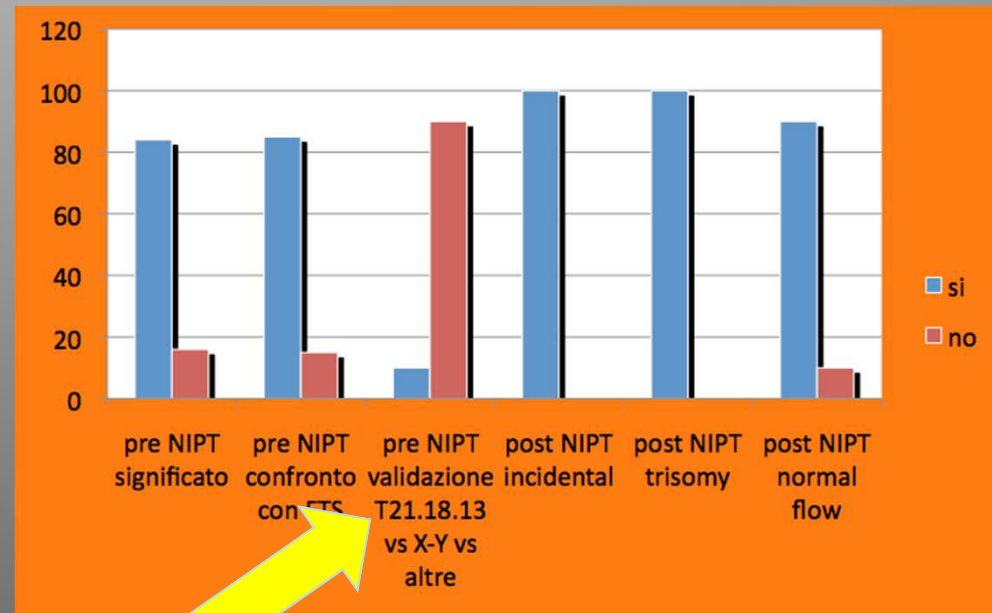
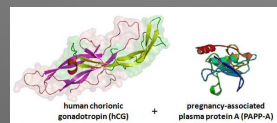
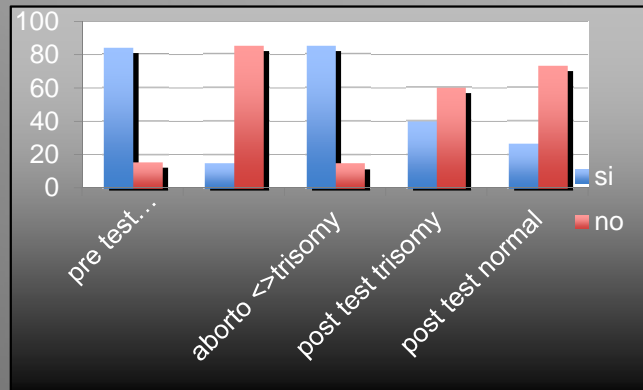
9/15050 (0.06%)

818/15050 (5.4%)

Screening non invasivi cffDNA

Rischio Anomalie Cromosomiche. SENSIBILITA' SPECIFICITA'

Patient information and understanding **NIPTonfield 1000**



Anomalie Cromosomiche NIPT validazione

T21 T18 T13

>99

Validated

XXX XXY XYY X0

60-90

Validated

Rare Chromosome Anomalies

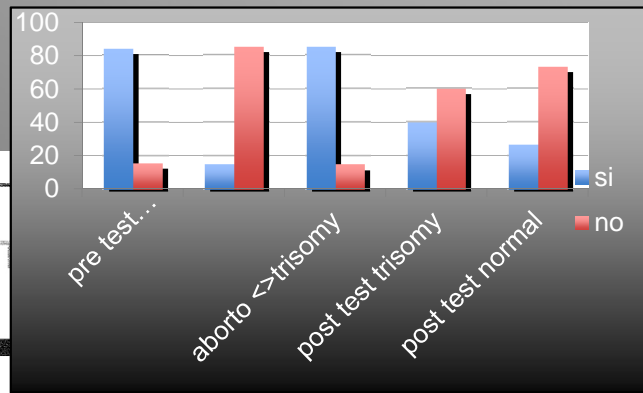
no on-field clinical data

Not validated

Screening non invasivi cffDNA

Rischio Anomalie Cromosomiche. SENSIBILITA' SPECIFICITA'

Patient information and understanding **NIPTonfield 1000**



Geneva - Switzerland

Medical Record/Patient ID:
Gestational Age at Draw (weeks): 12
Indication:
Counselor:
Client Sample ID:



PRENATAL ANEUPLOIDY TEST RESULTS -- Singleton Pregnancy	
RESULT	INTERPRETATION
No aneuploidy detected	Results consistent with two copies of chromosome 21
No aneuploidy detected	Results consistent with two copies of chromosome 18



SAMPLE ID			
Order ID:	Sample ID:	Draw Date: 02.Mar.2015	Receipt Date: 06.Mar.2015

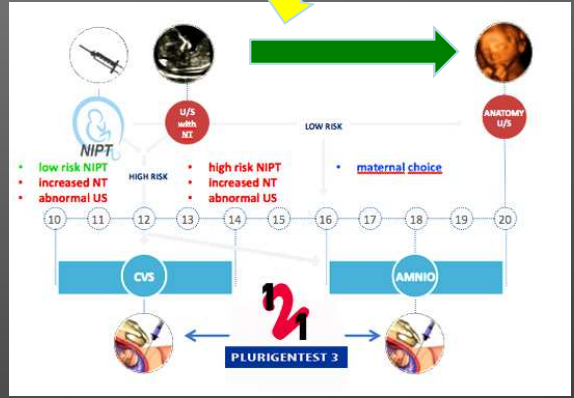
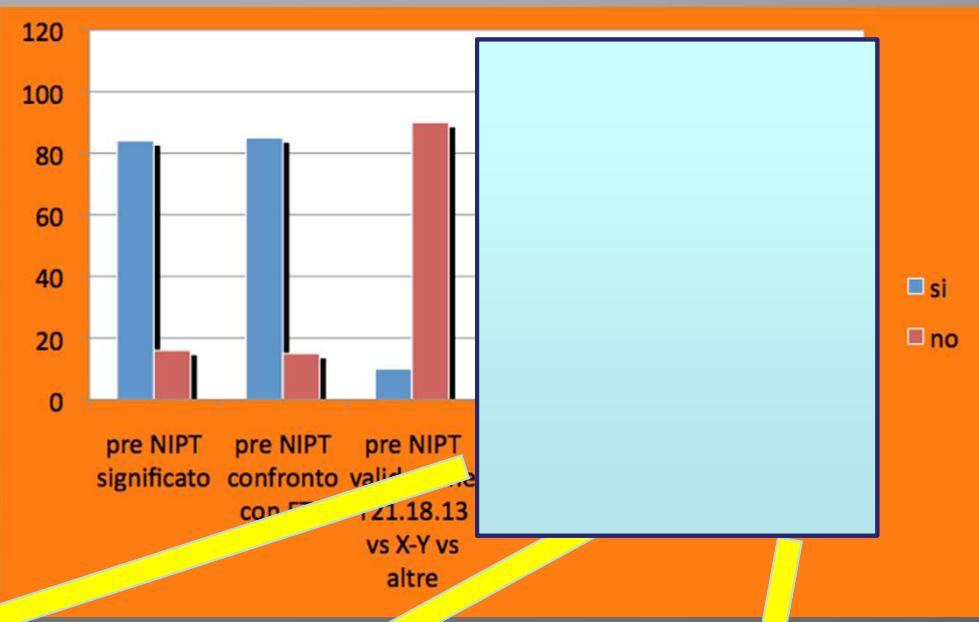
Test Method:
Non Invasive Prenatal Test: nucleic Acid detection, DNA sequencing using NGS technology, and analysis of sequencing results to determine fetal aneuploidy.

Limitations of Test:
This test is designed to detect chromosome aneuploidies and test is intended for singleton and twin pregnancies with one before and after testing is necessary. These results do not associated with other chromosomal or subchromosomal abnormalities. This test is not intended to identify or diagnose a risk for copy number variations in a twin pregnancy. The status of each individual chromosome of Y chromosome material can be reported in a aneuploidy such as 45,XYY, XYY, and YYY cannot be evaluated. The absence of chromosomal abnormalities such as X and Y chromosomes. There is a small possibility that in fetus, but may not detect chromosomal changes of the placenta (chromosomal mosaicism). Results of 'aneuploidy detected' patients should perform invasive prenatal procedures for confirm, in order to obtain a definitive diagnosis.

Disclaimer:
The manner in which the information is used in a patient care is the responsibility of the healthcare provider, including advising for the need for genetic counseling or additional diagnostic testing like amniocentesis or chorionic villus sampling. Any diagnosis mentioned is intended in the context of all available clinical findings. This test has not been created or approved by regulatory agencies as a diagnostic test. This test represents the newest service currently available for prenatal testing; however, as with any emerging genetic test, there is always a chance of failure or error in sample analysis. Cautious measures are taken to avoid these errors.

This prenatal test is sold for Geneva SA, 17, rue de la Saboterie, 1206 Plan-les-Coteaux, Switzerland, tel: +41 98 1000 100, a wholly owned subsidiary of Esperite NV group. Esperite NV, P.O. Box 11, 2284 JH Zoetermeer, The Netherlands, tel: +31 (0) 20 520 100.
Genoma is a company of esperite

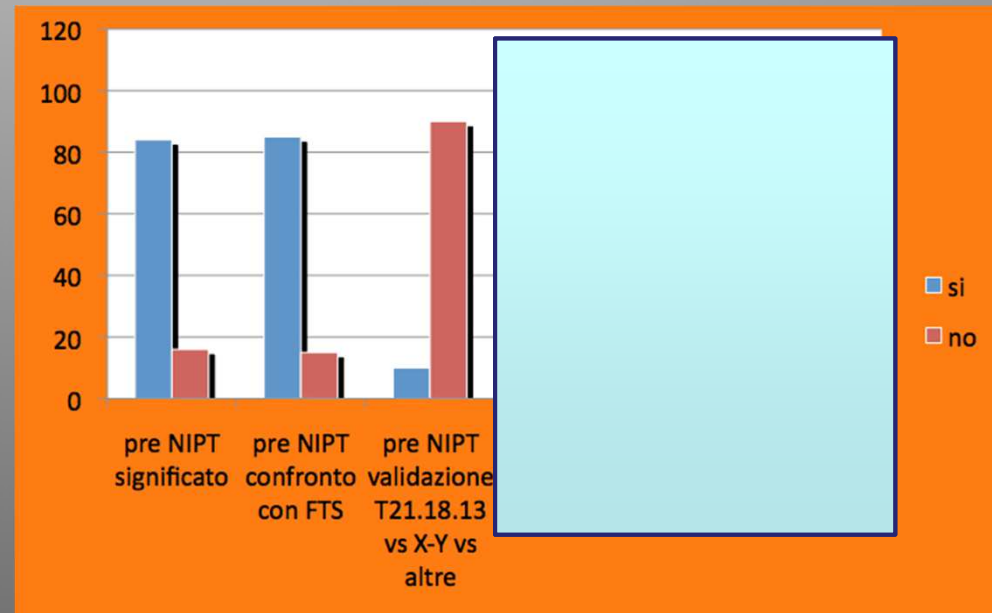
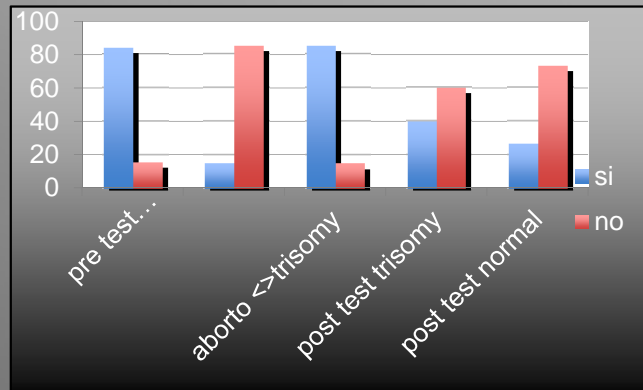
False Positive Population rate Case to case (PPV)	T21	T18	T13	45X
0.09%	0.13%	0.13%	0.23%	
7/100	23/100	66/100	62/100	



Screening non invasivi cffDNA

Rischio Anomalie Cromosomiche. SENSIBILITA' SPECIFICITA'

Patient information and understanding **NIPTonfield 1000**



**indice di comprensione dopo
consulenza genetica pre test passa
dal 25-40% al 85-100%**



LLGG.NIPT Raccomandazioni generali

I Centri che offrono il test devono avere competenze nella diagnosi ecografica, nella diagnosi prenatale; devono avere competenze qualificate per la consulenza pre-test e post-test; devono garantire la tracciabilità del campione ed essere collegati con il laboratorio che esegue il test. Nel caso il laboratorio sia all'estero o a distanza è necessario un efficiente contatto a sostegno della interpretazione dei risultati.



NIPTonfield1000

Poliambulatori con competenze integrate. Visita ostetrica/ecografia. Consulenza genetica pre test. NIPT e/o altre indagini. Consulenza e/o Supporto post test. Decorso ecografico.

NIPT non è per "il venditore di medicine".....(C.Santamaria)

ricerca indagate caratteristiche disponibili
prendendo fallimento tassativa
considerando materna possibilità
varie fetale termini
disponibilità materno
genetica
illustrare DNA opzioni
informare
nel villi del liquido
analisi rende
test limiti
quelli
sui all origine come
circolo
pre-test
dei sulle quando
stimata
delle definire
per relazione
sugli placentare
embriologica
compresi amniotico
consequenti dimostrato
necessario elezione
anomalie

Consulenza
patologie post-test diverse amniociti affrontare corso
referto
senza
coralli
cliniche
quelle infatti alcuni risultati indagini presente assenza riferimento strumento
implicazioni