

Advancing the Frontier of DNA Testing

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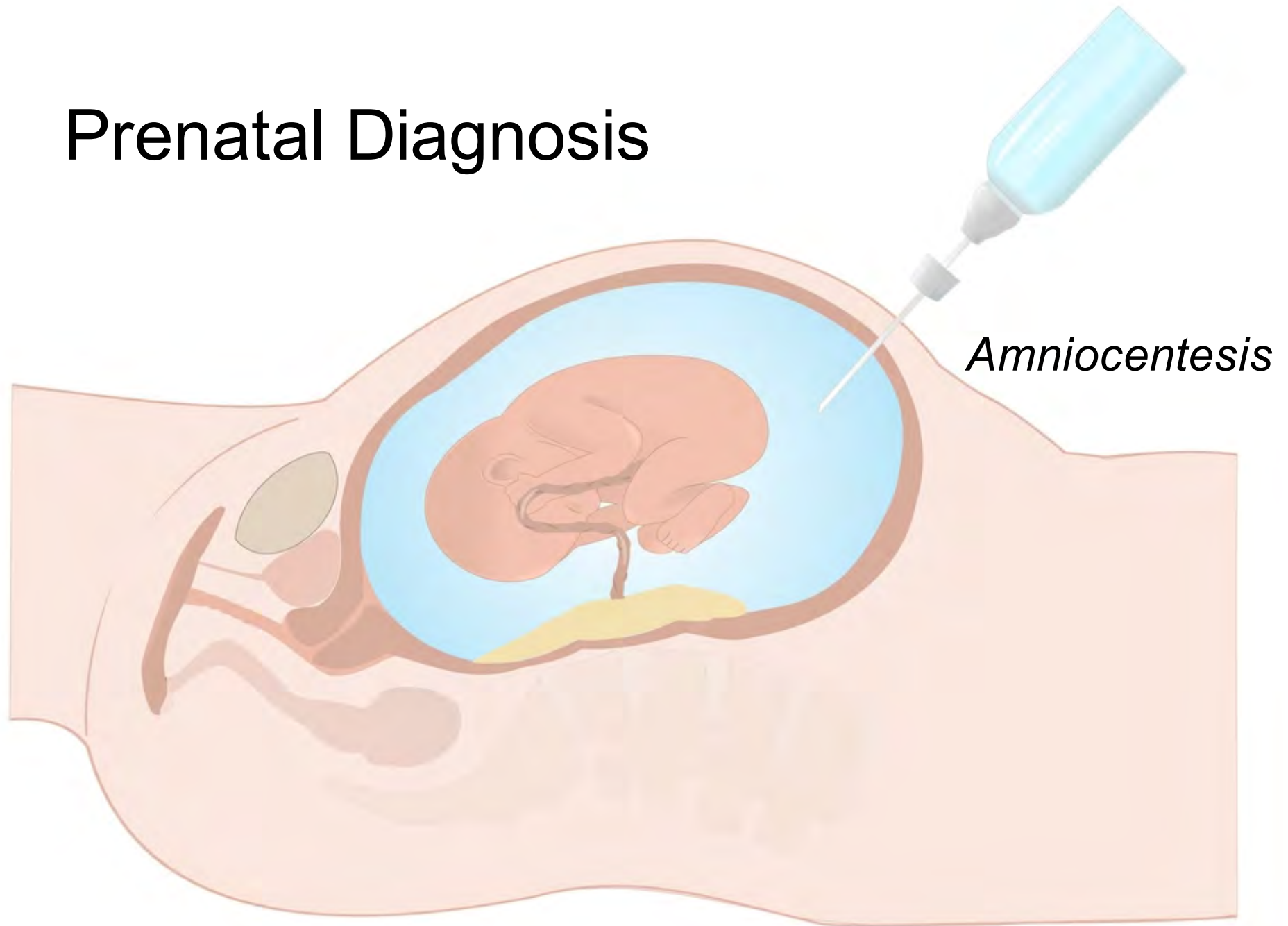
The Chinese University of Hong Kong

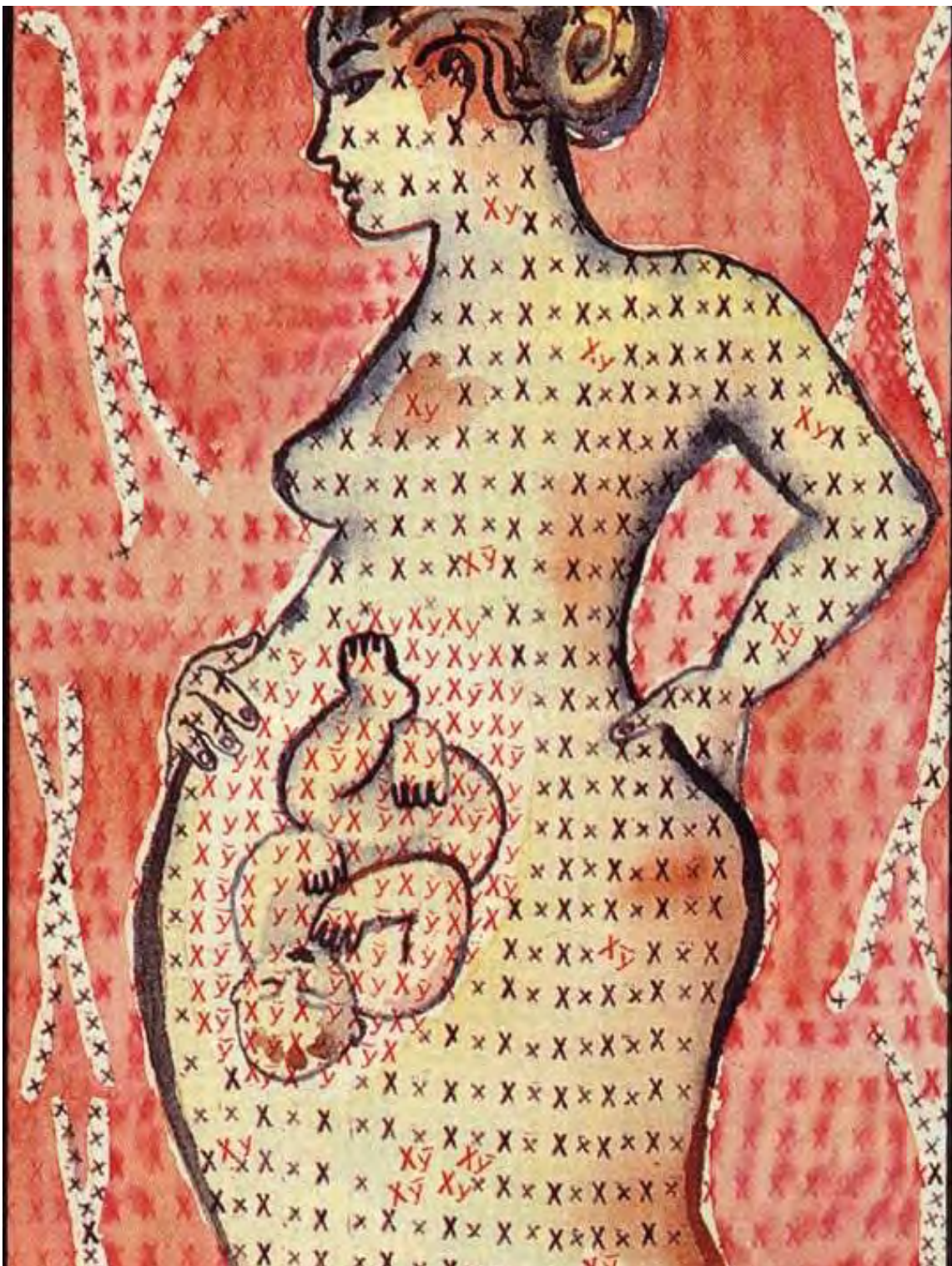
香港中文大學醫學院

Faculty of Medicine

The Chinese University of Hong Kong

Prenatal Diagnosis





Lo et al. *Lancet* 1997; 350:485

Non-invasive prenatal testing (NIPT)



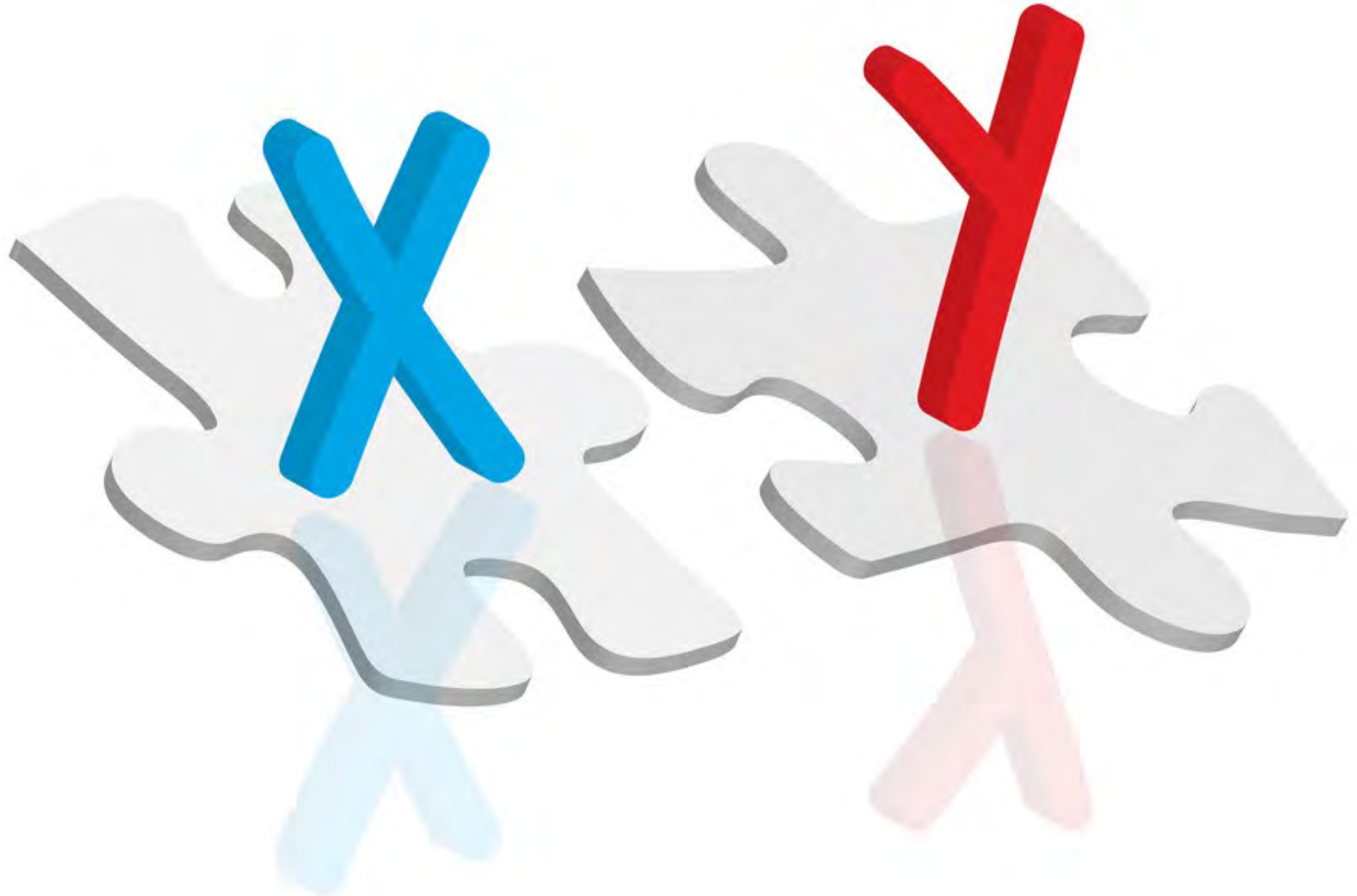
Fetal DNA:

Present from 1st trimester

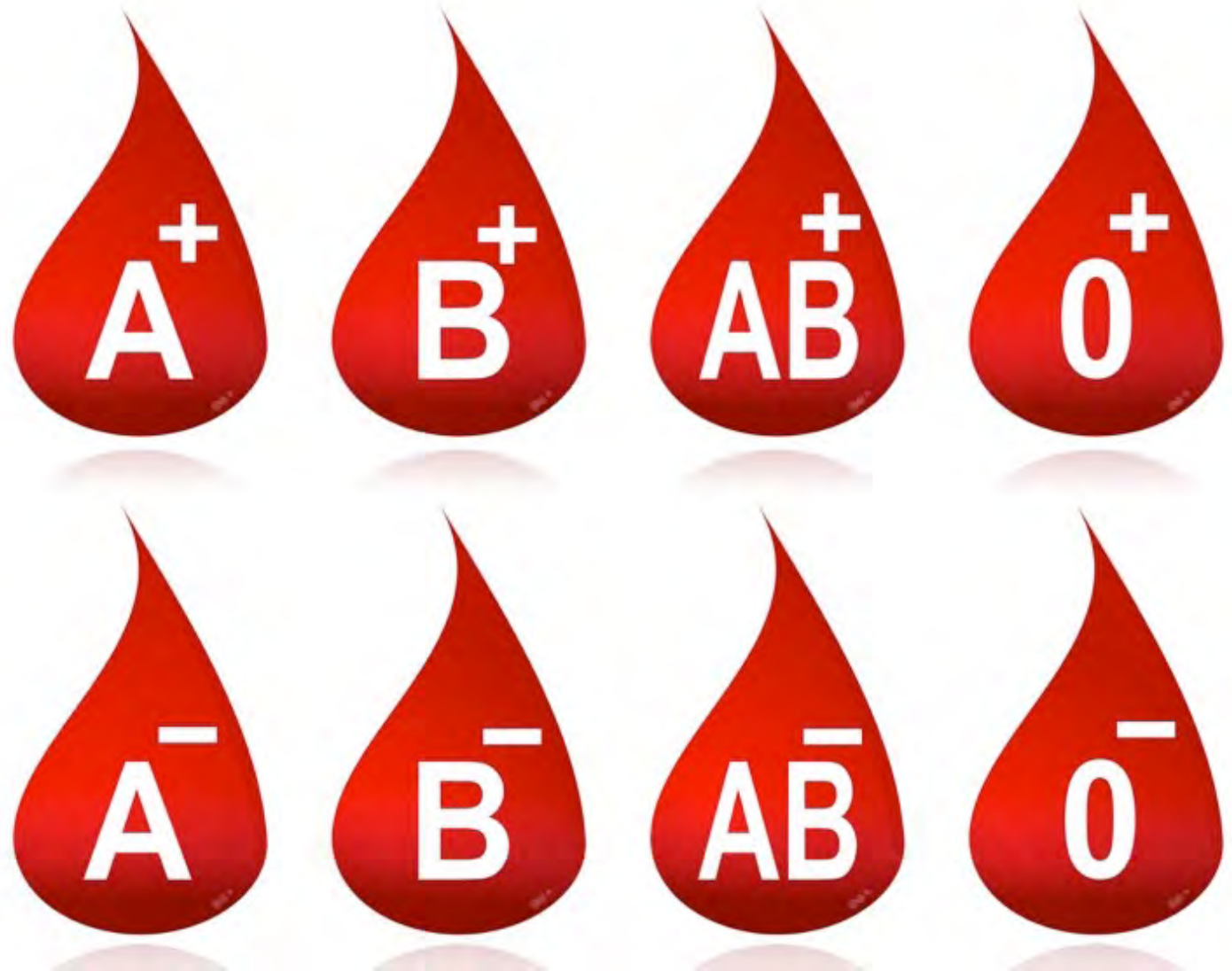
Mean conc.: 13%

Cleared rapidly after delivery

Sex-linked disorders



Fetal
Blood Group
Genotyping



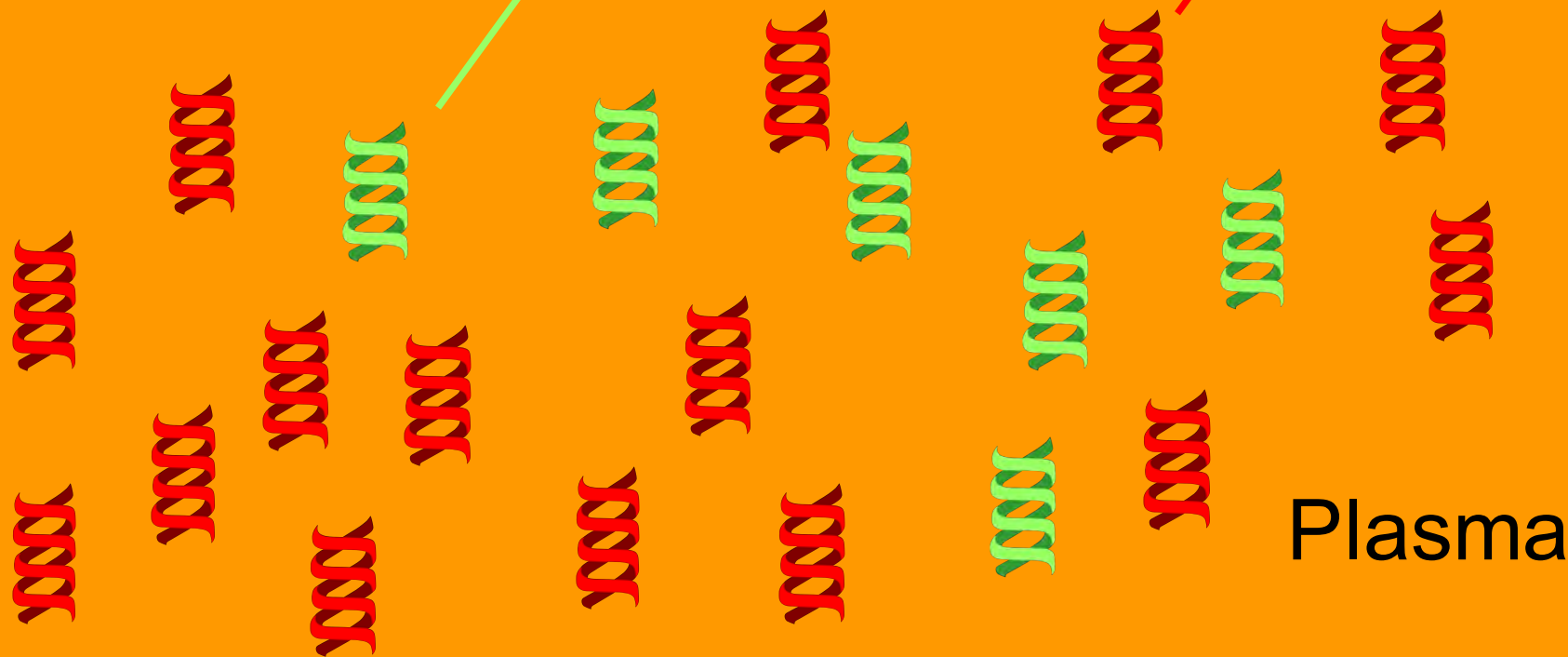
Down syndrome detection



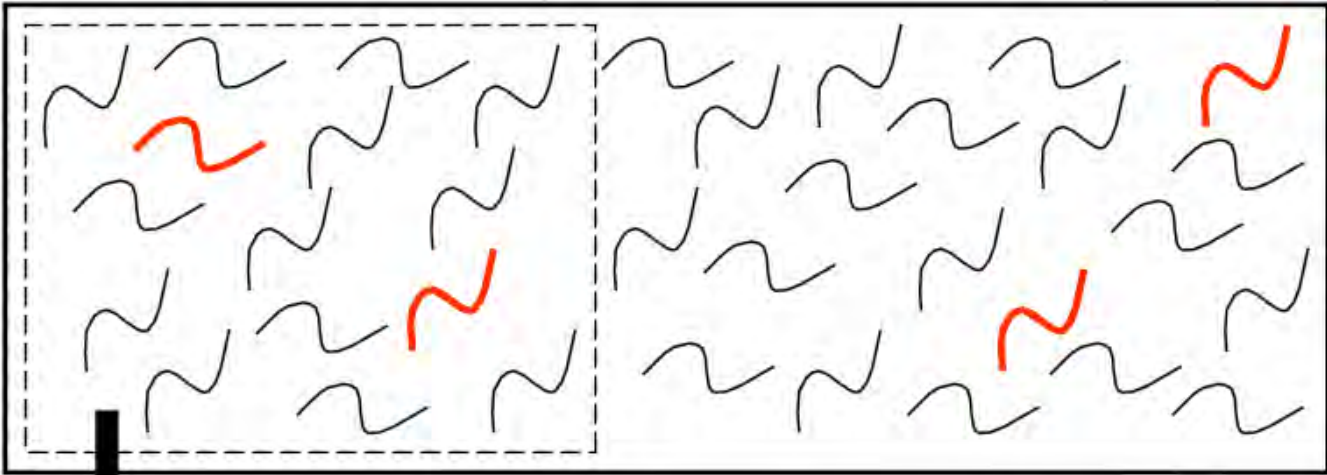
A challenging problem

Fetal DNA

Maternal DNA



DNA fragments in maternal plasma



Sequence and align

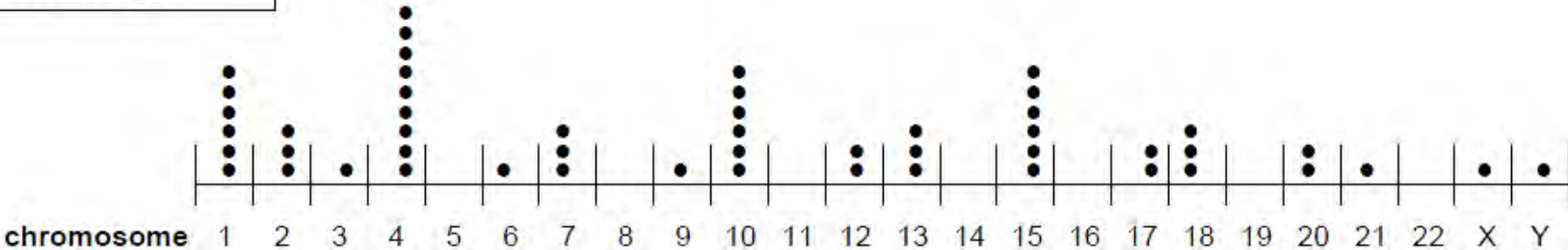
36 bp

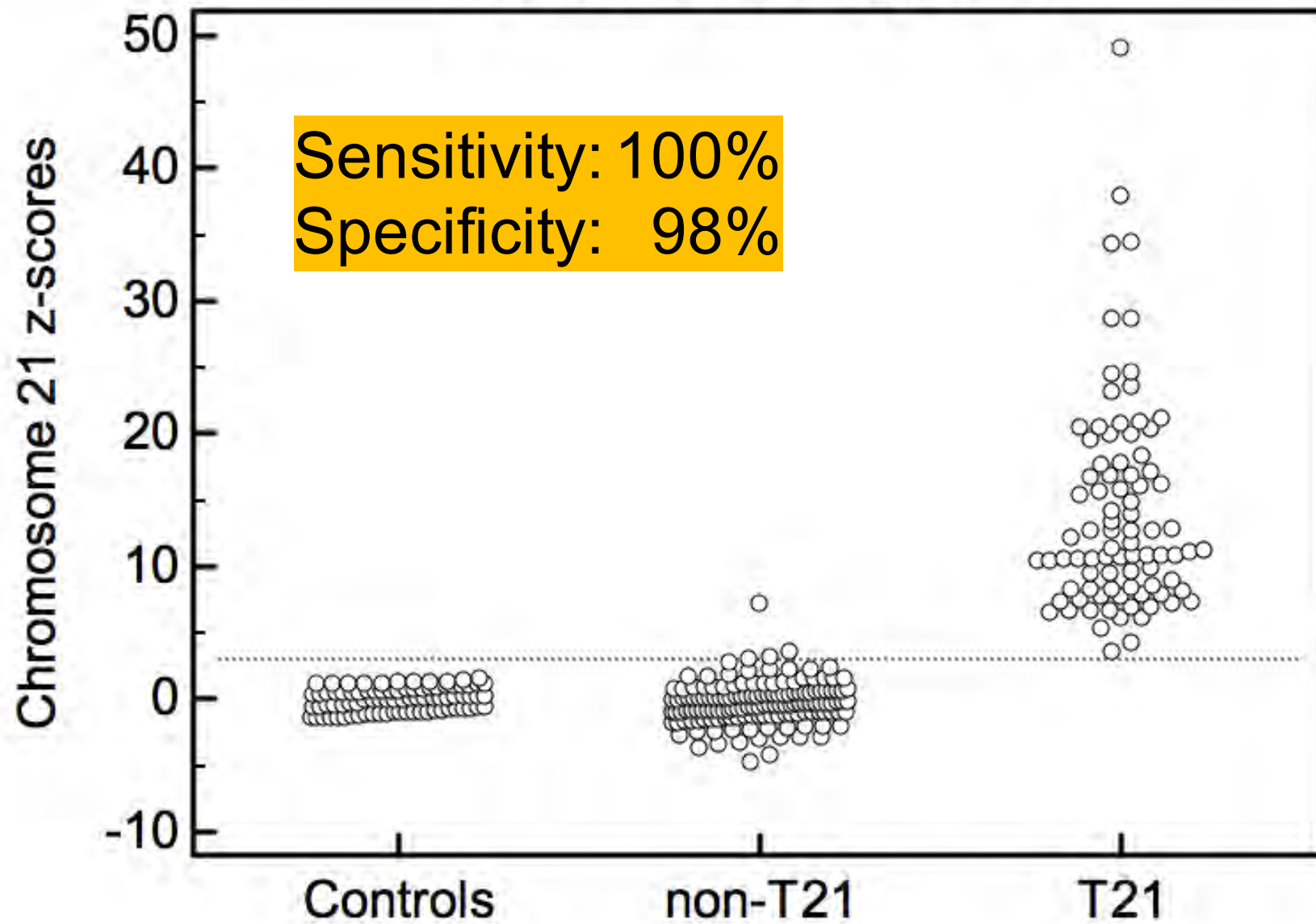
AAGCT...
CTAGT...
TAGGC...
GCATG...
⋮
nth sequence

Bioinformatics alignment

Chr1
Chr7
ChrX
Chr13
Chr1
Chr21
Chr18
ChrY and so on...

Sequence counting





GENETICS

Fetal gene screening comes to market

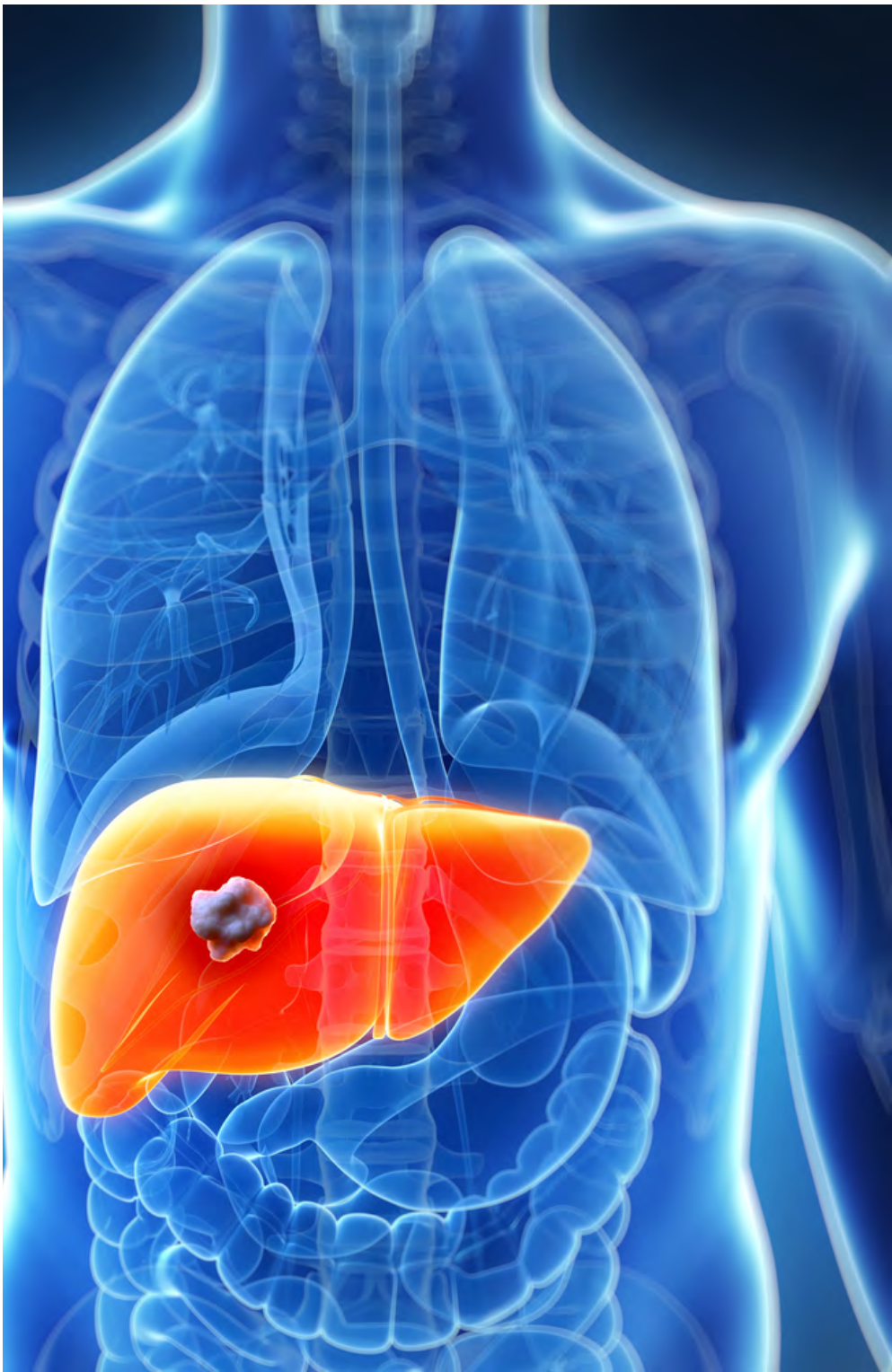
Non-invasive procedure could make prenatal testing easier, but it comes with ethical problems.

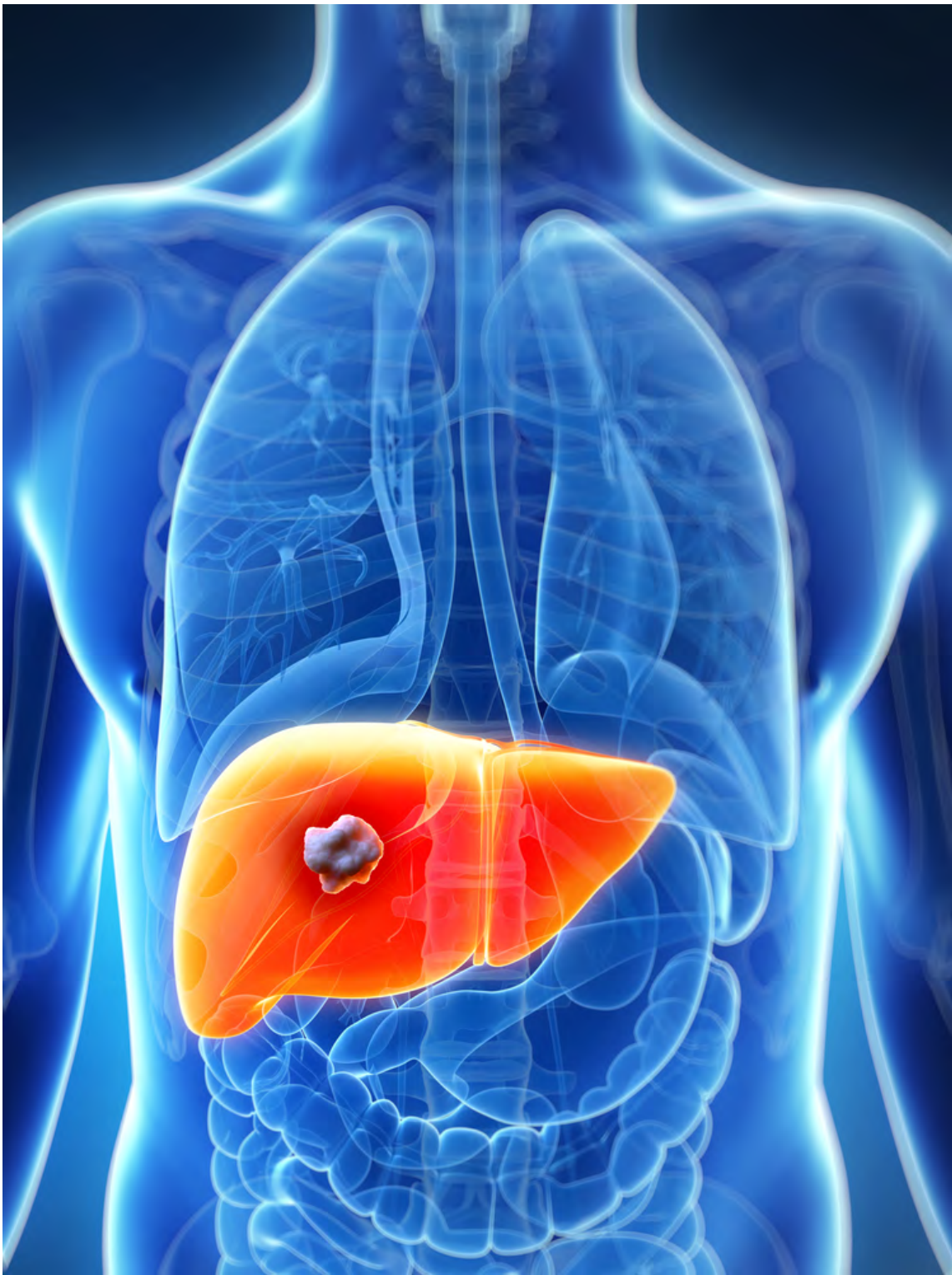
Nature (27 Oct 2011)

Clinical Service

- ◆ China
- ◆ US
- ◆ Israel
- ◆ Germany
- ◆ UK
- ◆ Canada
- ◆ The Netherlands
- ◆ Austria
- ◆ Switzerland
- ◆ Czech Republic
- ◆ Slovakia
- ◆ Japan
- ◆ Singapore
- ◆ Malaysia
- ◆ Australia, etc

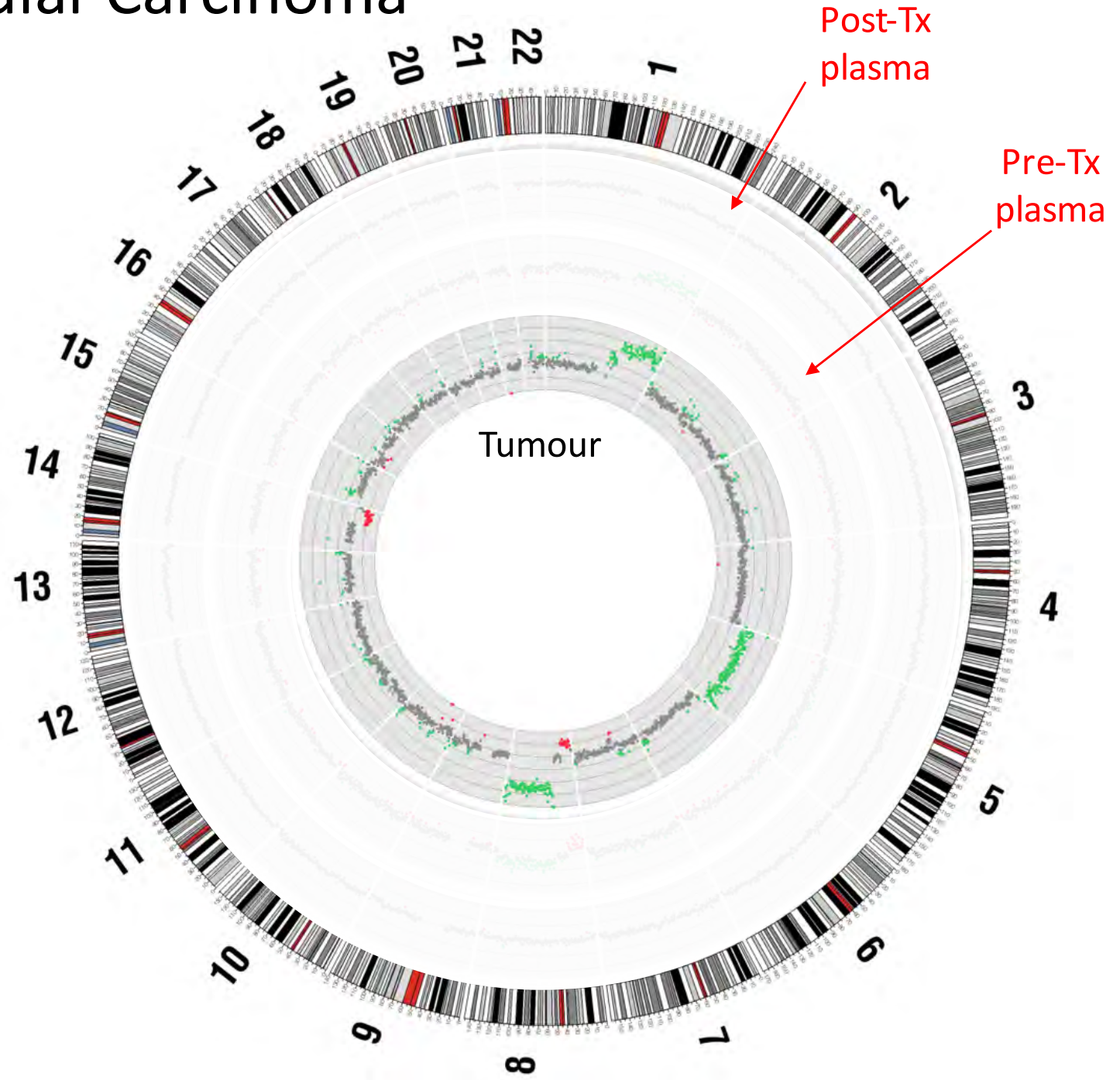




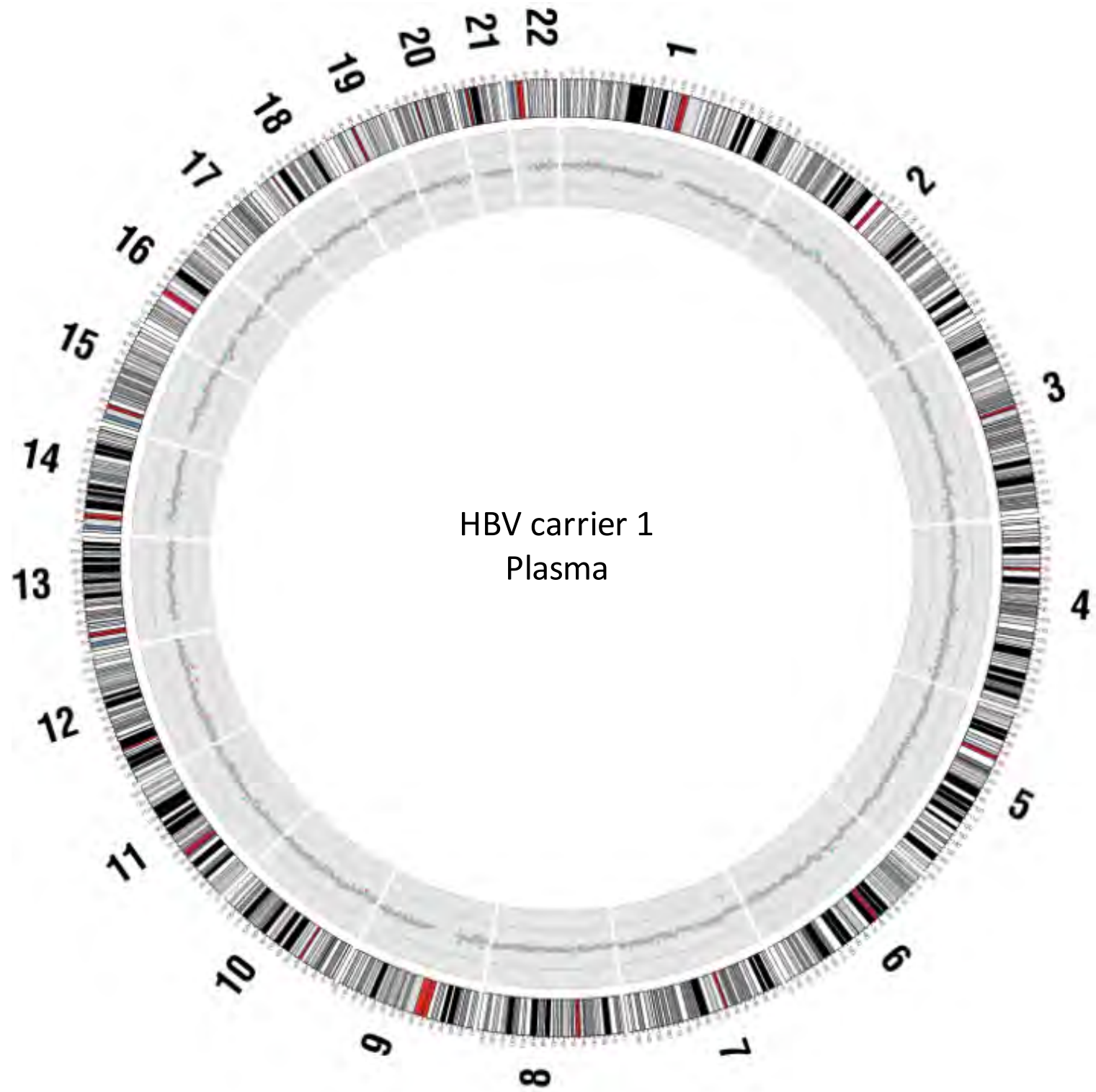


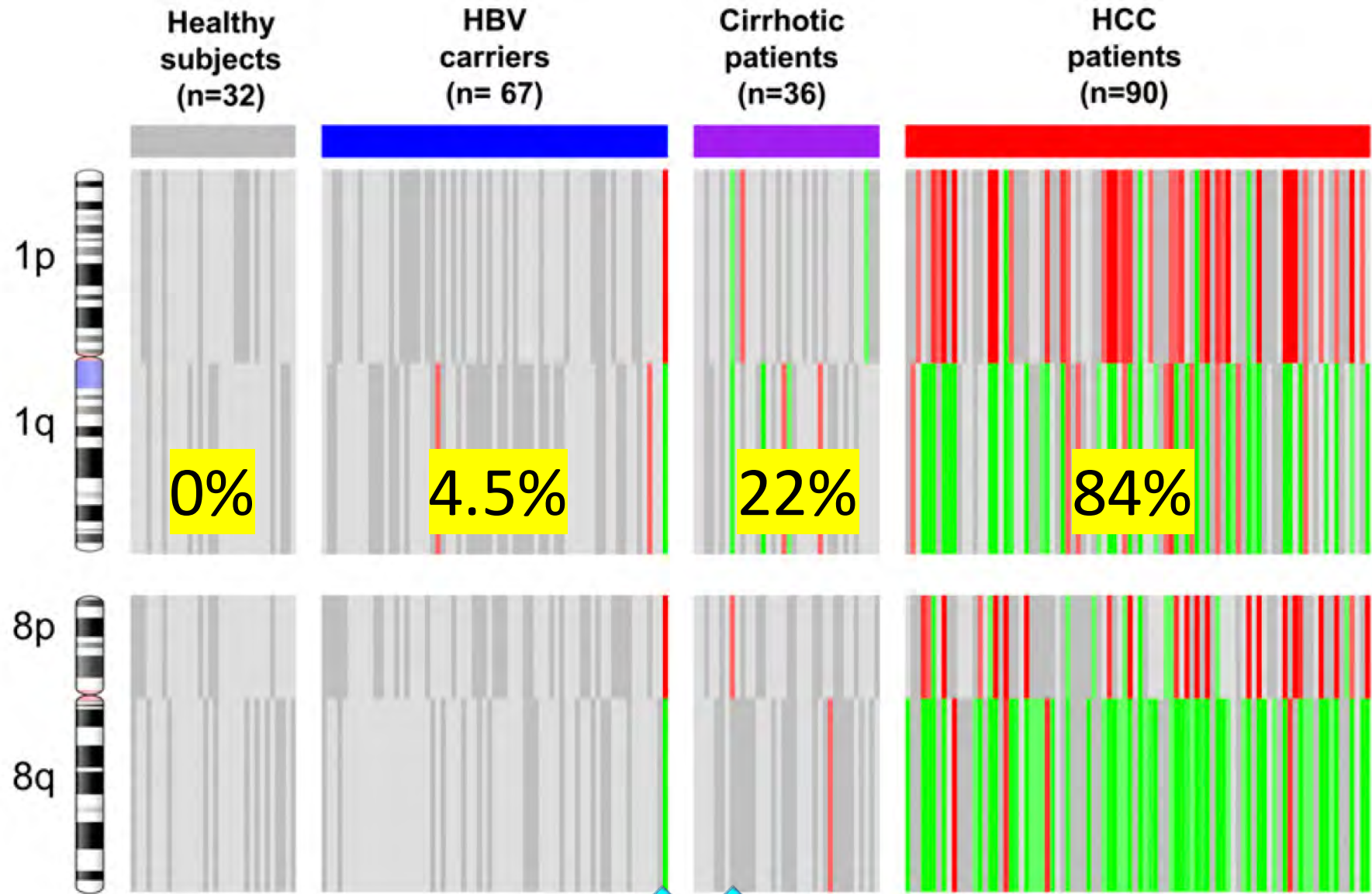
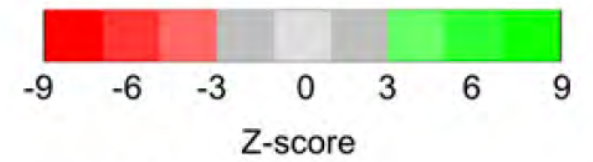
Liver Cancer

Hepatocellular Carcinoma

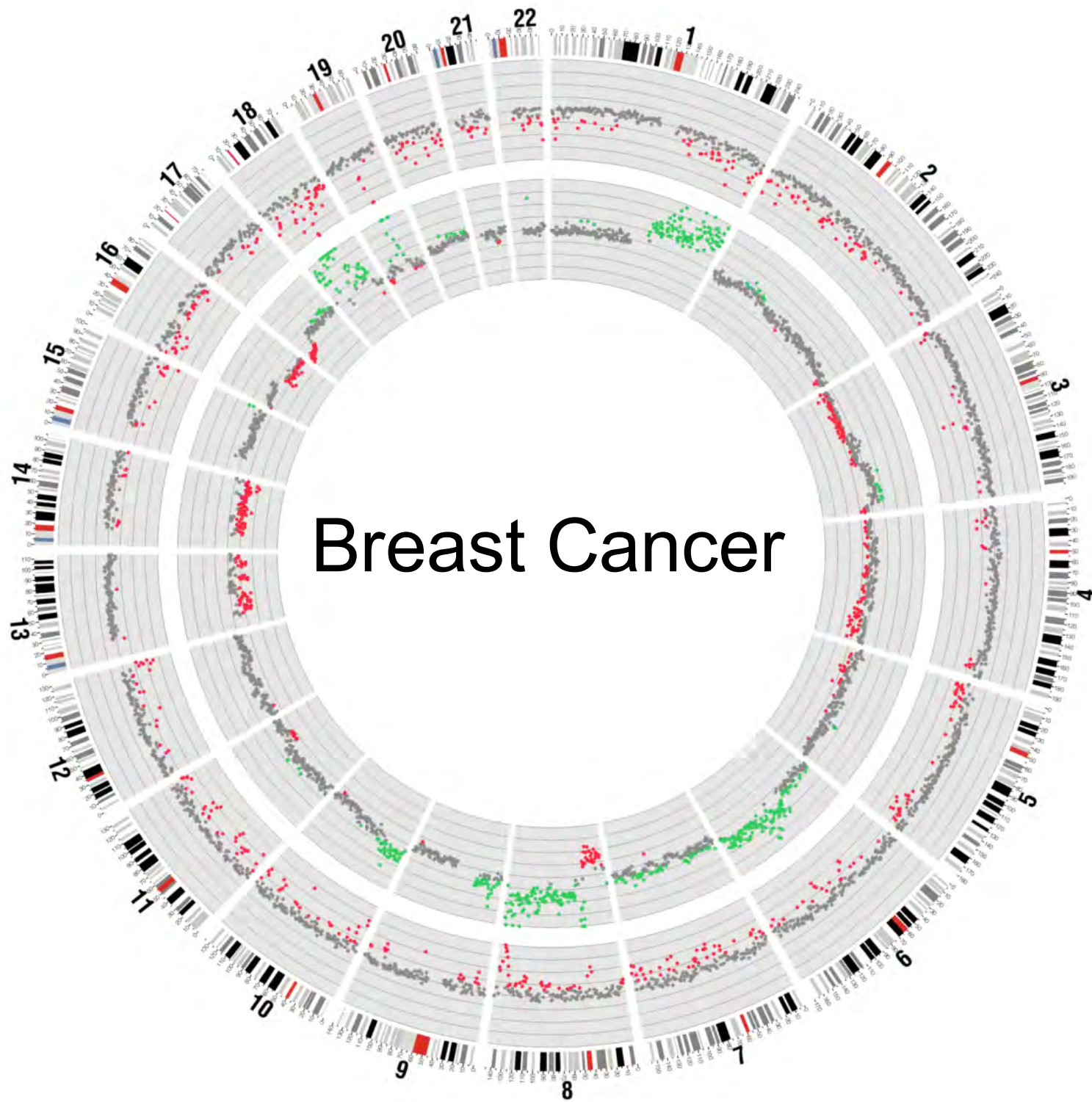


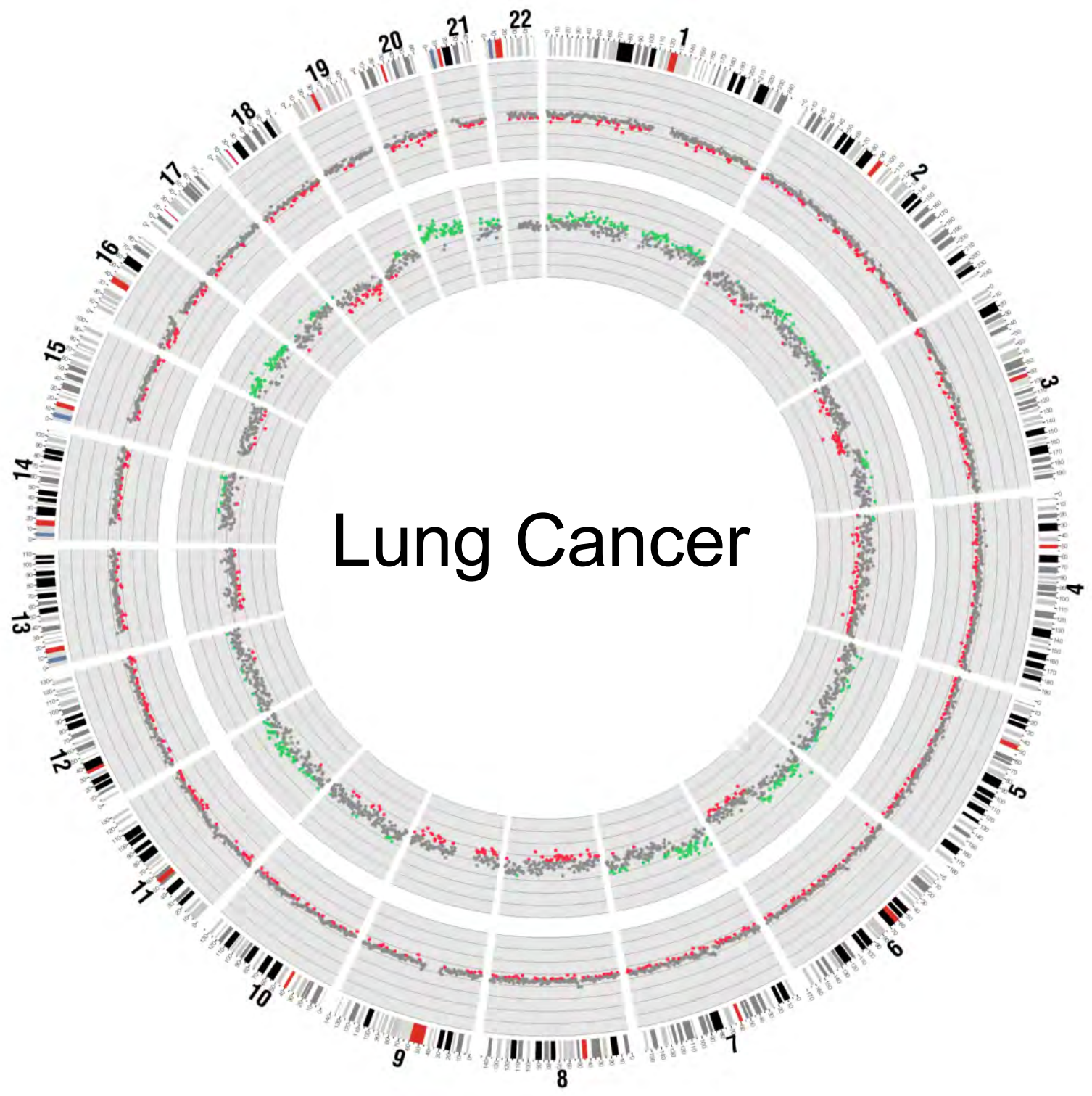
Chronic hepatitis B carriers
without liver cancer





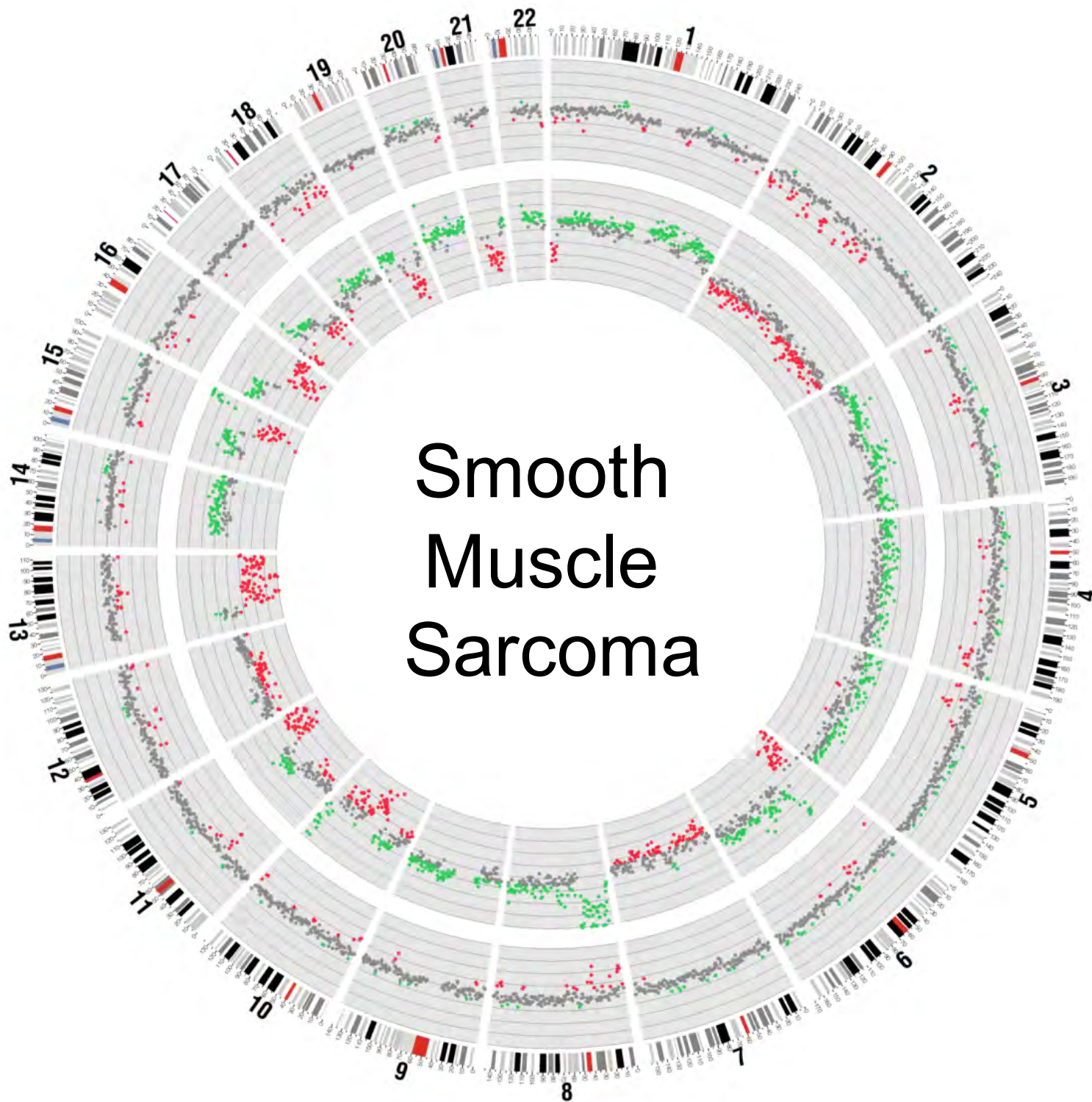
Jiang et al. PNAS 2015; 112: E1317

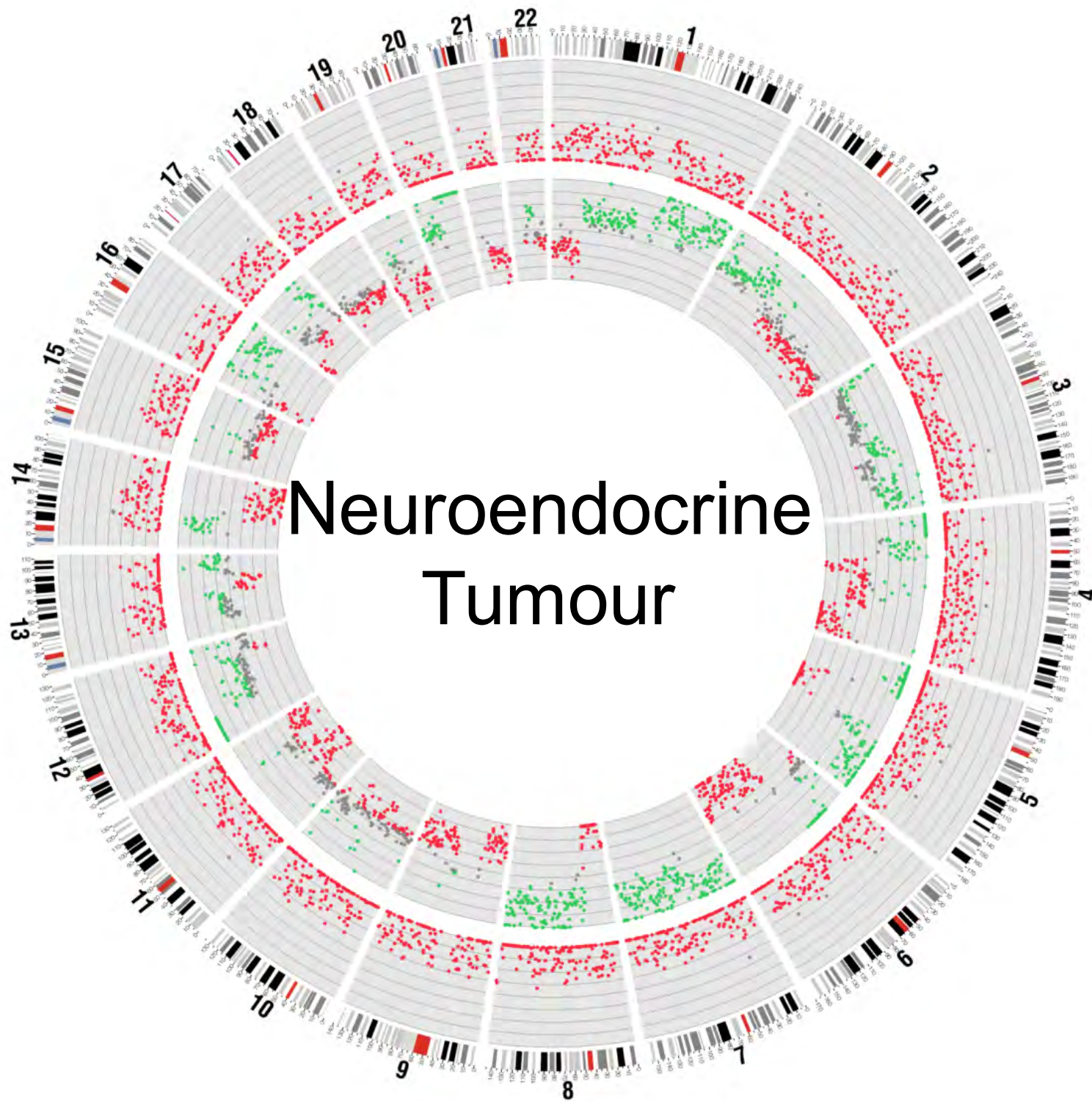




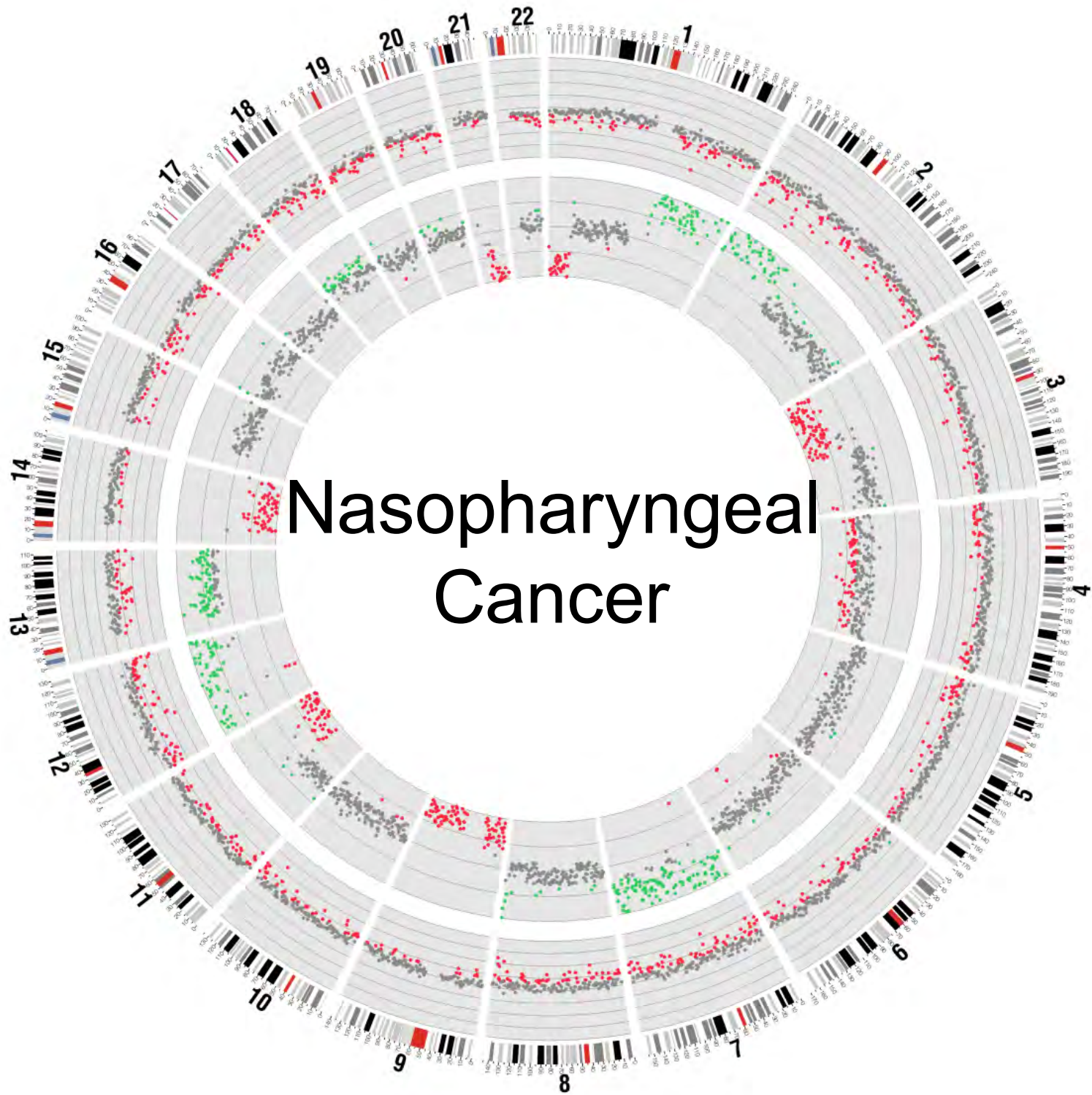
Lung Cancer

Smooth Muscle Sarcoma

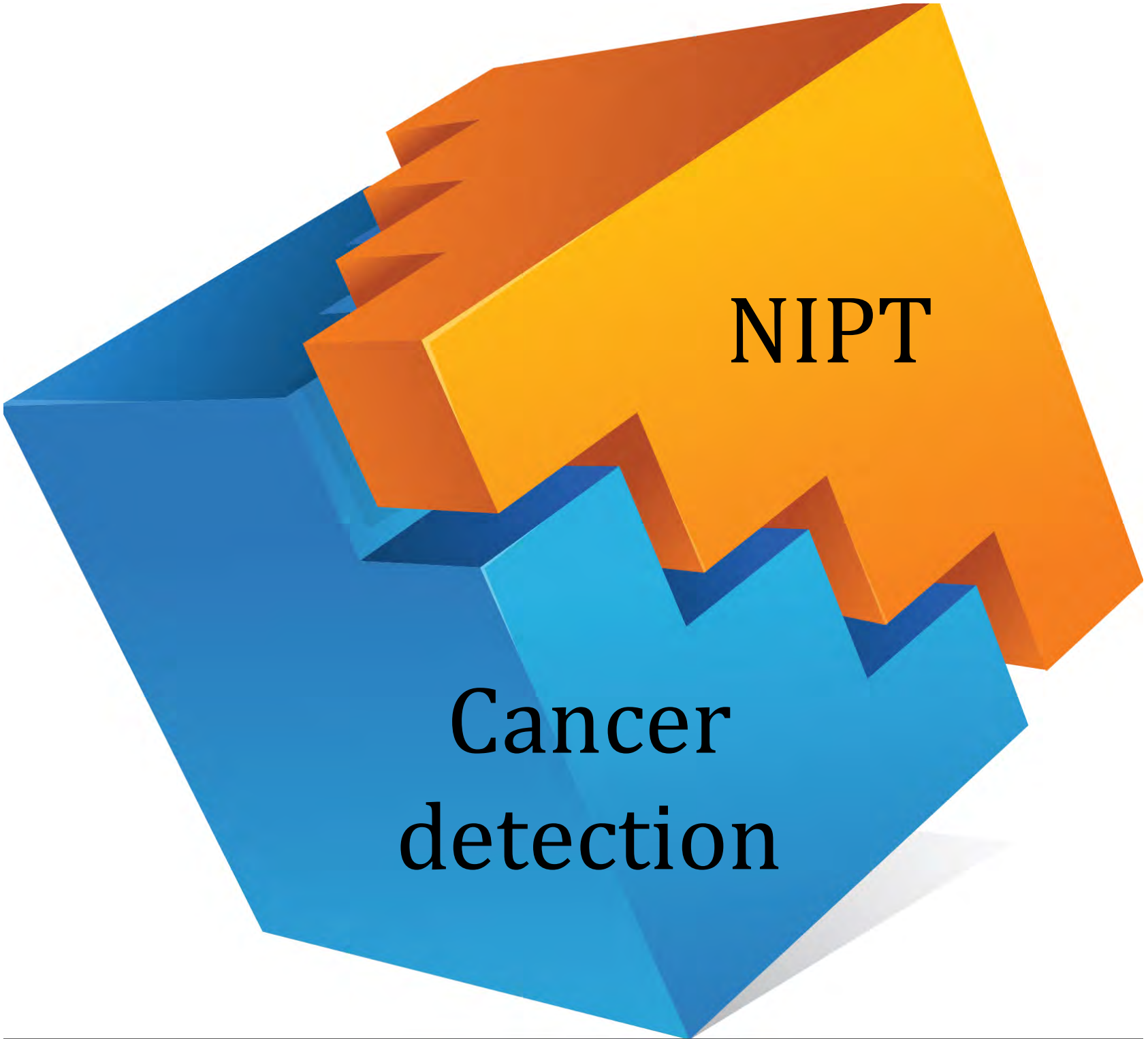




Neuroendocrine Tumour



Nasopharyngeal Cancer



Cancer
detection

NIPT



Noninvasive Prenatal Testing and Incidental Detection of Occult Maternal Malignancies

Bianchi et al.

JAMA 2015; 314: 162-9

Presymptomatic Identification of Cancers in Pregnant Women During Noninvasive Prenatal Testing

Amant et al.

JAMA Oncol 2015

Tracing the Origin of Plasma DNA Aberrations

What tissues contribute DNA into plasma?

Plasma DNA Tissue Mapping



Sun et al PNAS 2015

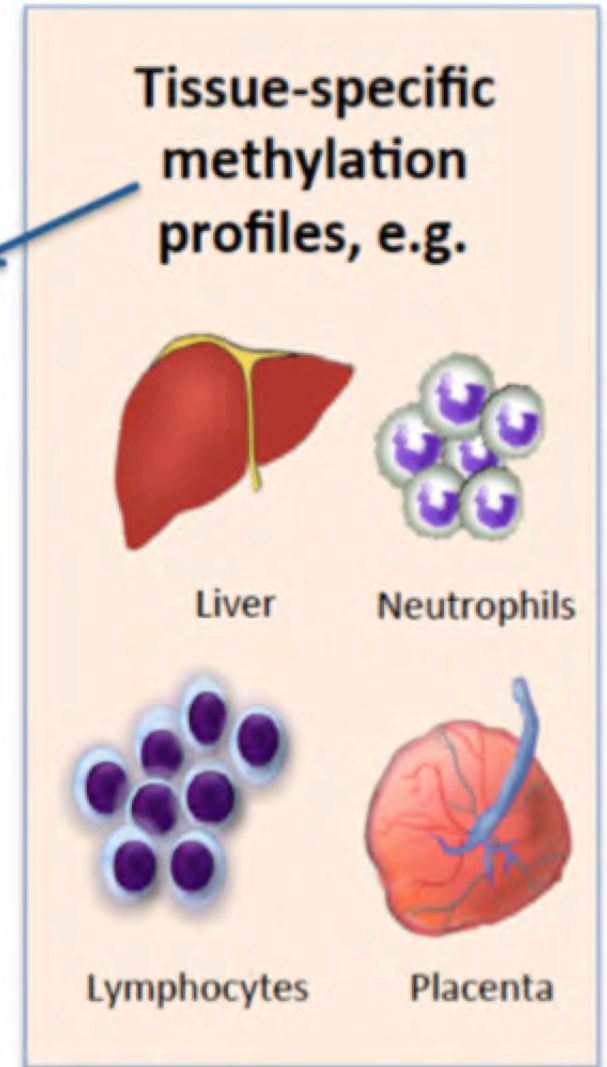
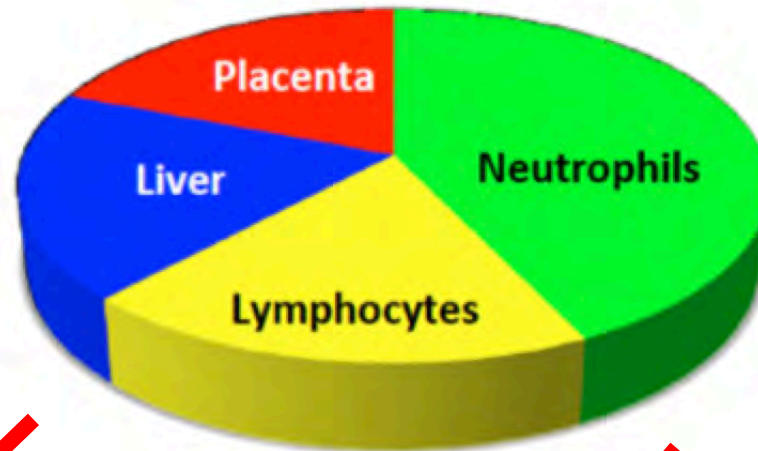


Genomewide
bisulfite
sequencing



Plasma DNA
tissue mapping

Tissue contribution %



Prenatal testing

Cancer detection

Transplantation monitoring

Organ damage, e.g. trauma, autoimmune

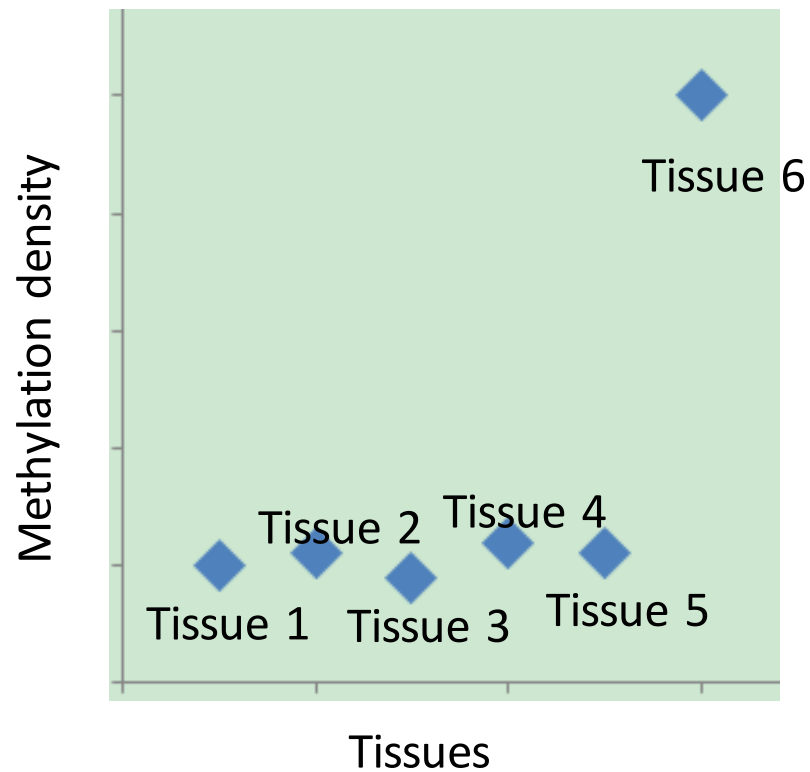
Marker Identification

Marker Identification

Total: 5820 markers

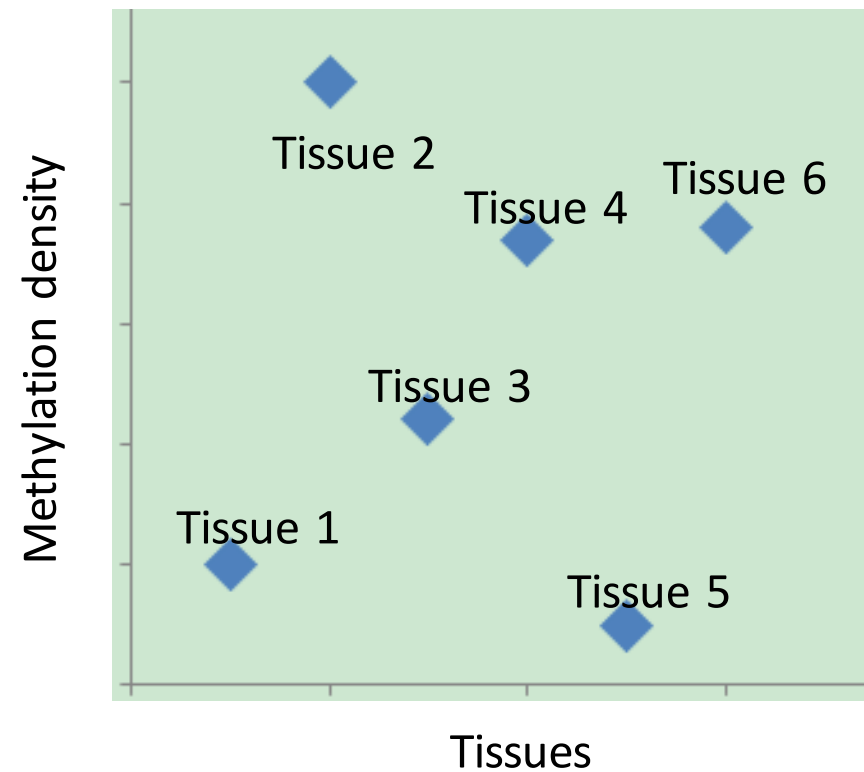
1013 markers

Type I




4807 markers

Type II



- DNA mixture experiments
- Clinical scenarios





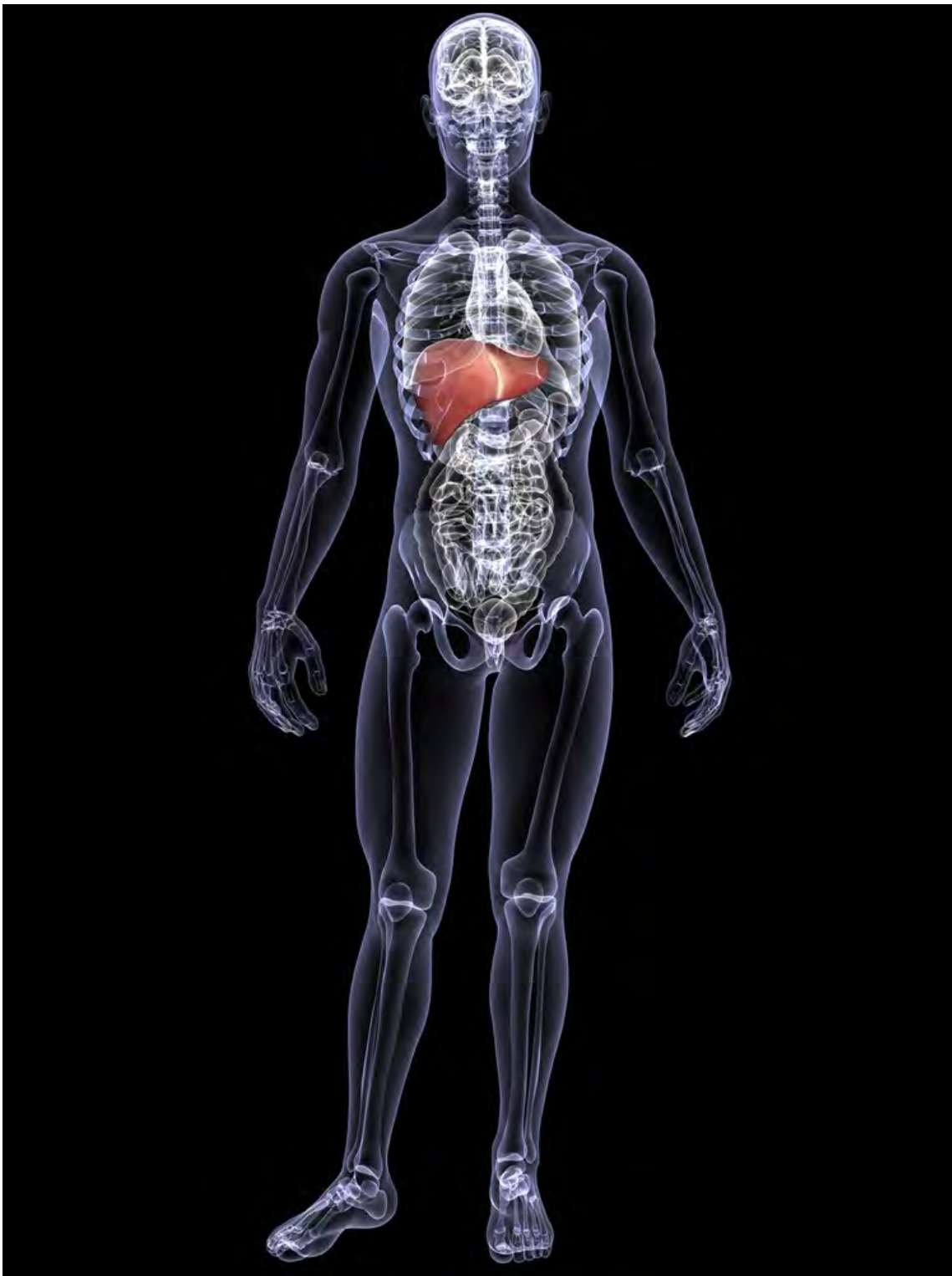
Fetal (placental) DNA in Maternal Plasma

Chim et al. PNAS 2005



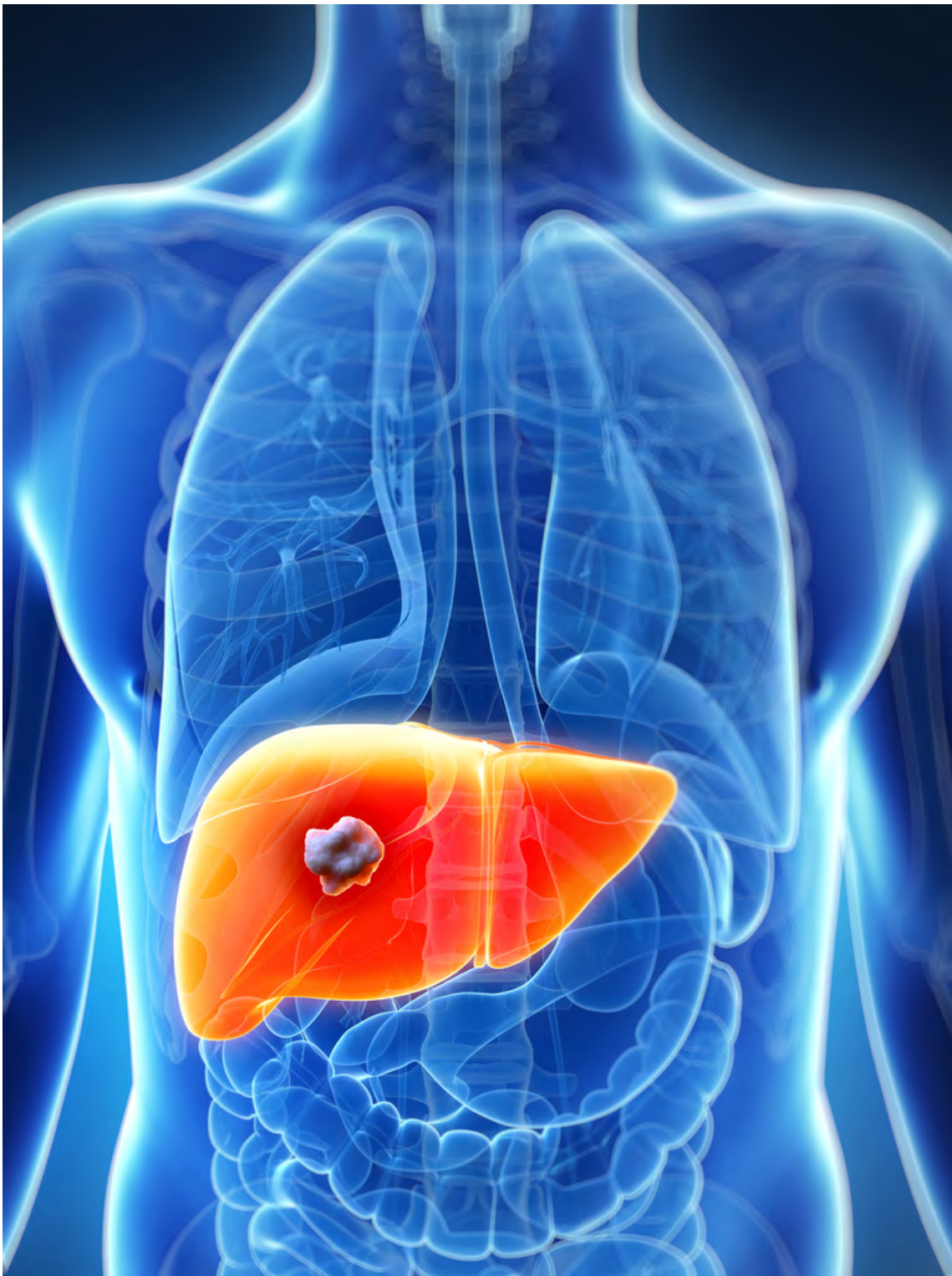
Bone Marrow Transplantation

Lui et al Clin Chem 2002



Liver Transplantation

Lo et al Lancet 1998

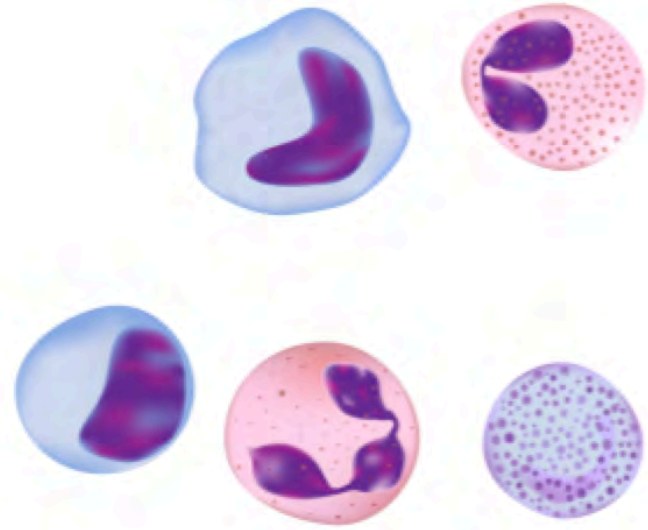


Hepatocellular Carcinoma (HCC)

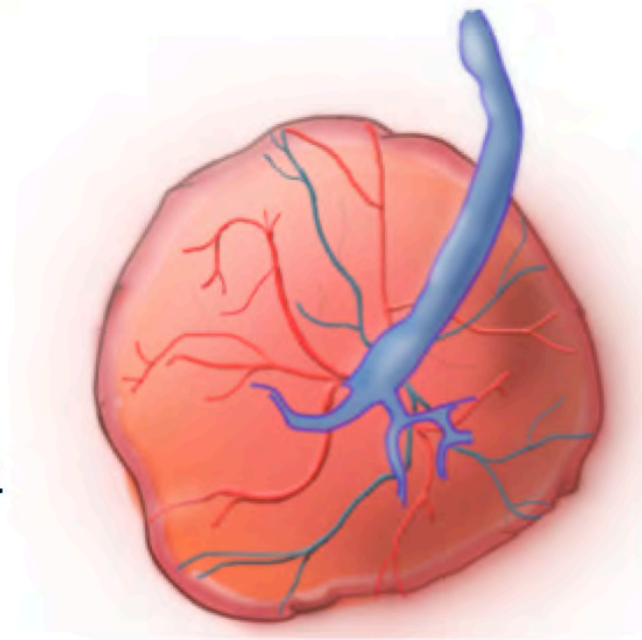
Chan et al PNAS 2013
Jiang et al PNAS 2015



Liver



Blood cells



Placenta



DNA Mixture Experiments



Non-neoplastic
liver at resection
of HCC

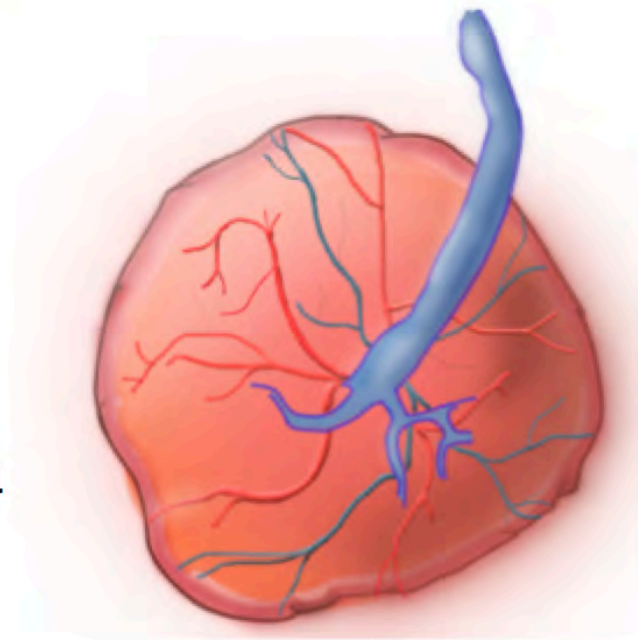
Liver

Buffy coat: 40 yr healthy
non-pregnant woman



Blood cells

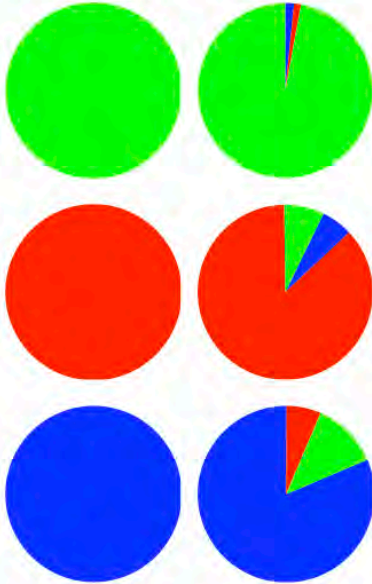
Placenta



Healthy baby
girl at birth

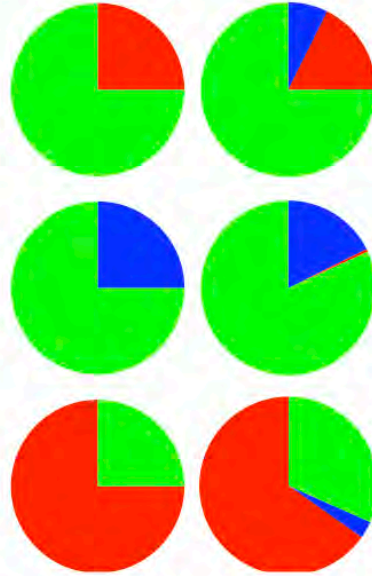
100%
Input

Measured



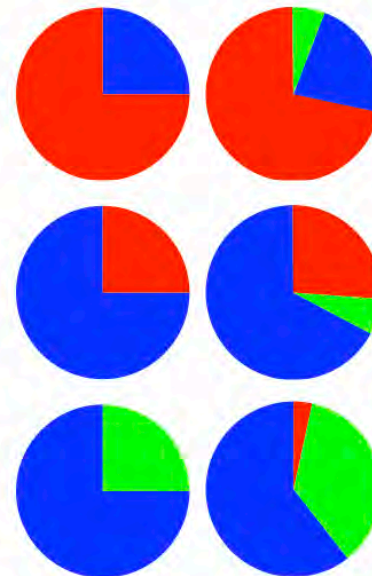
75% + 1
Input

Measured






75% + 1
Input

Measured

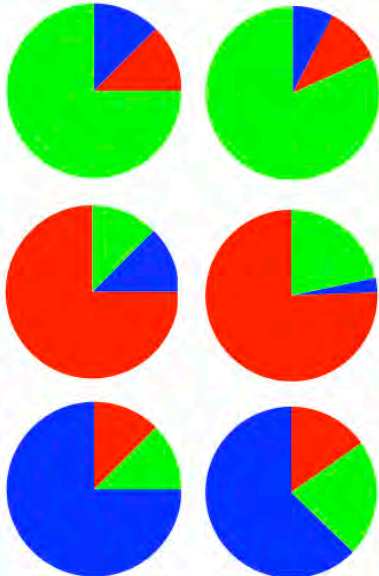


Color scheme

-  Blood cells
-  Placenta
-  Liver

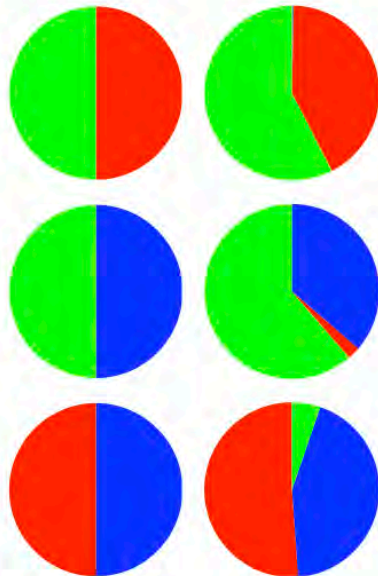
75% + 2
Input

Measured



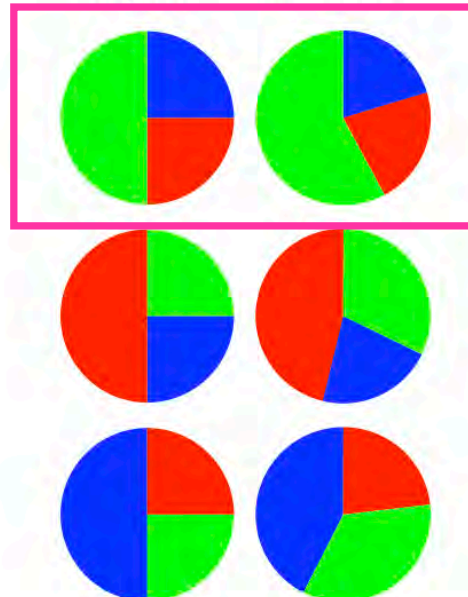
50% + 1
Input

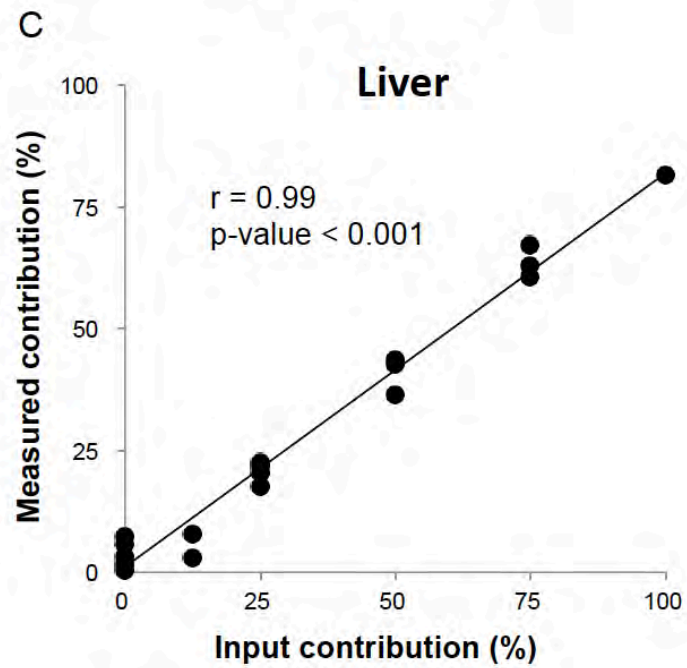
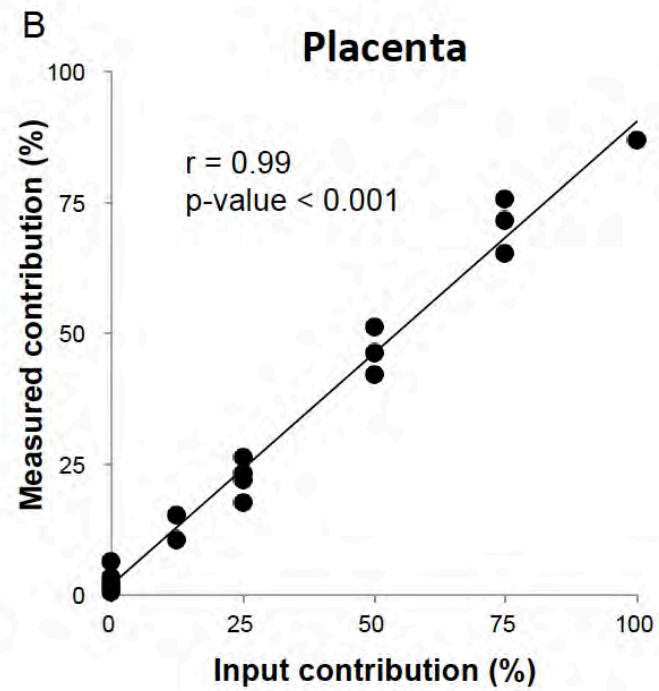
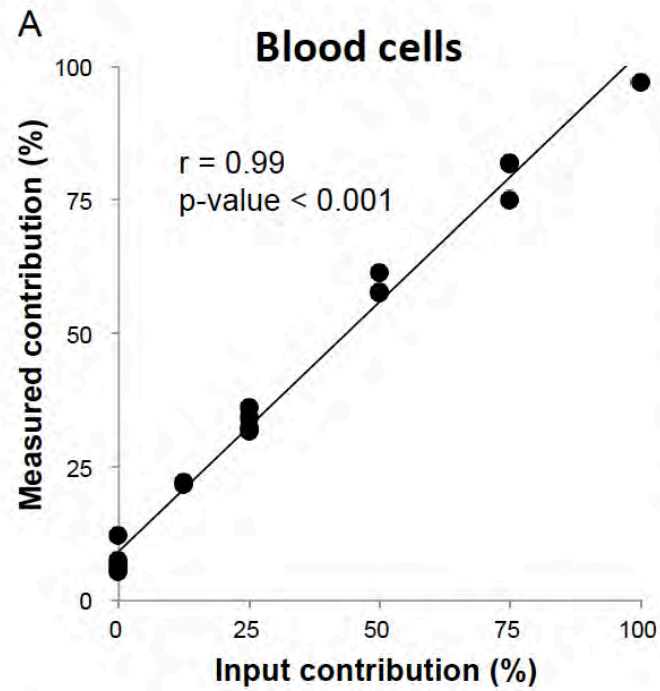
Measured



50% + 2
Input

Measured



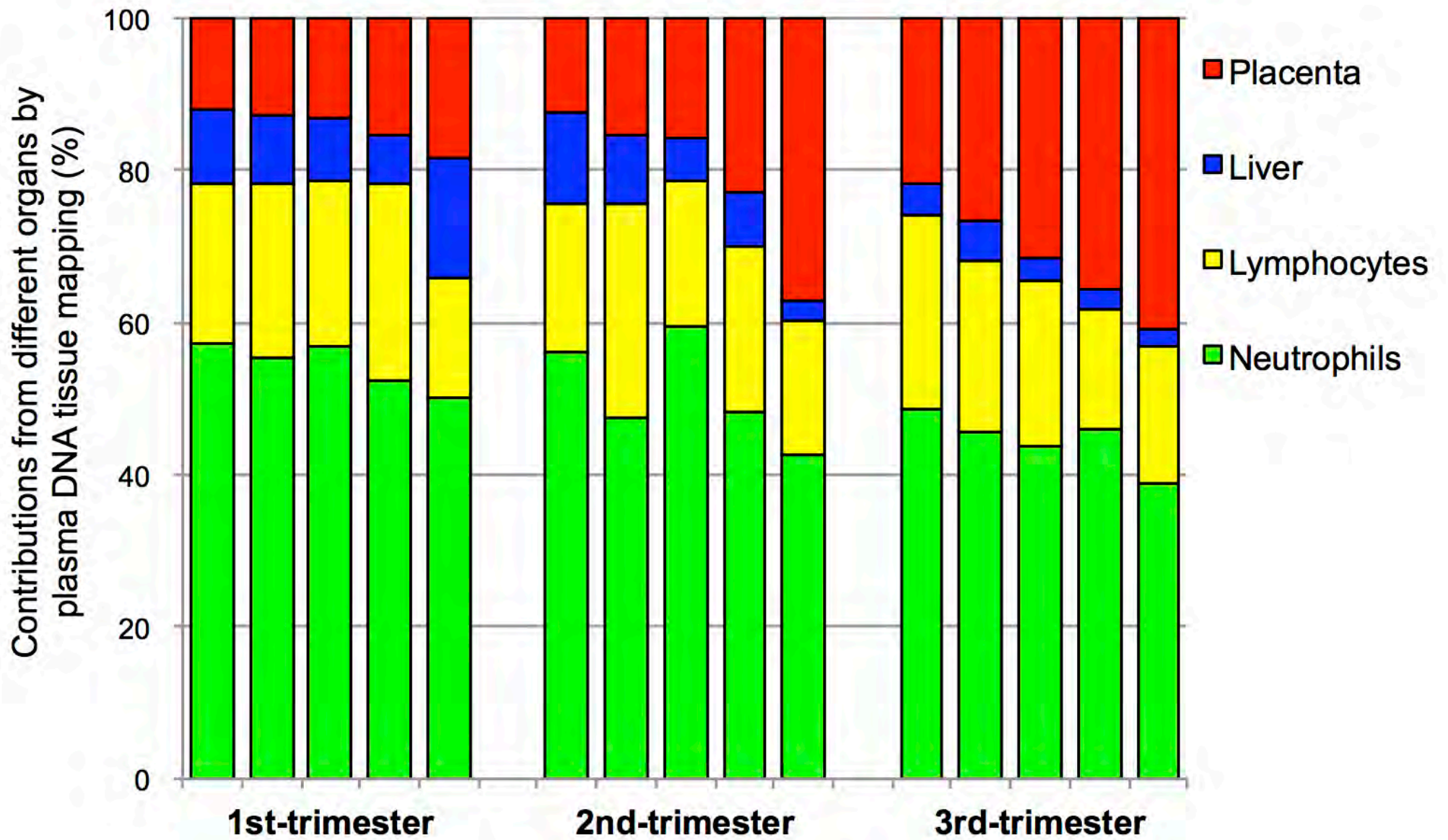


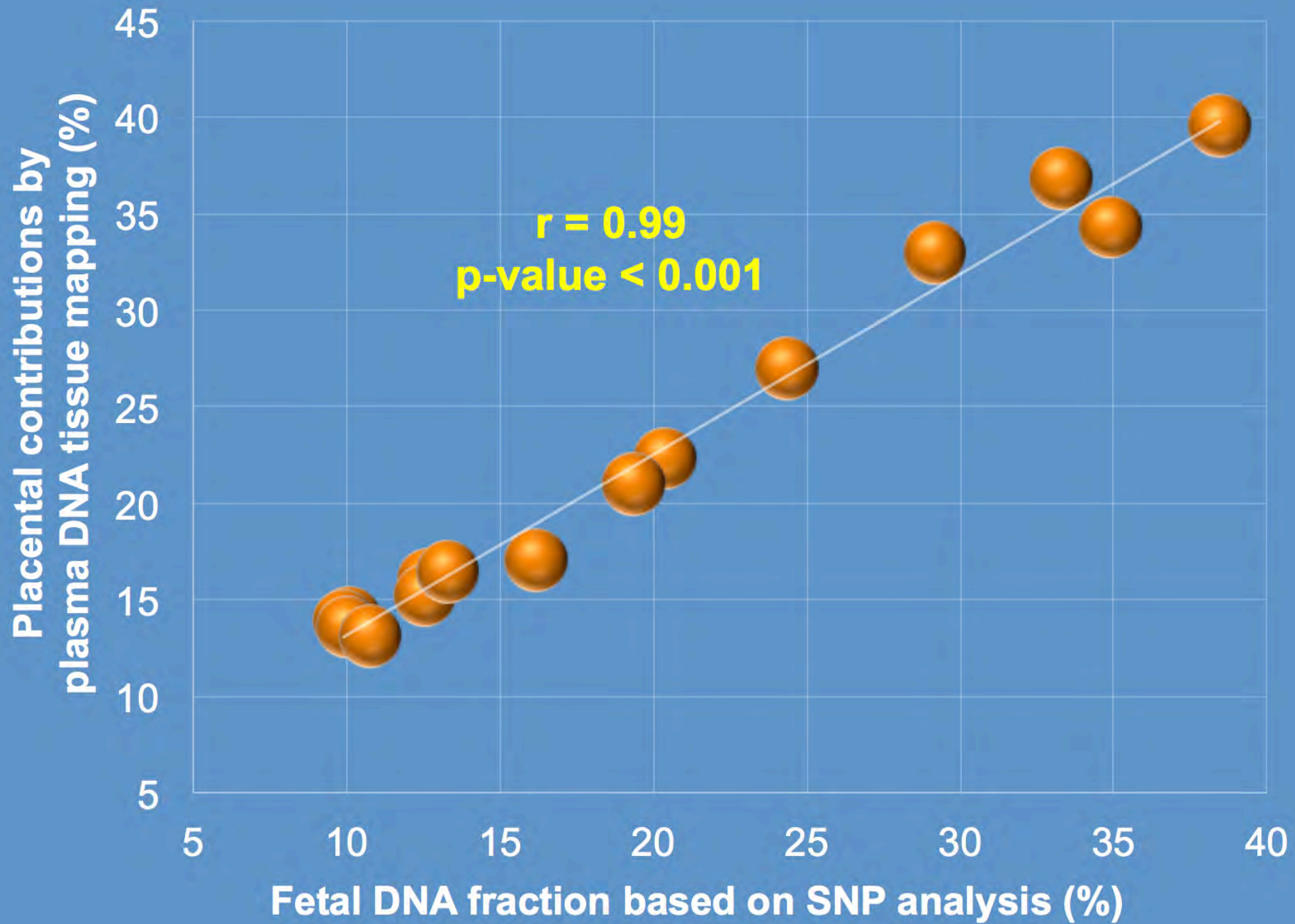
Clinical Scenarios



Pregnancy





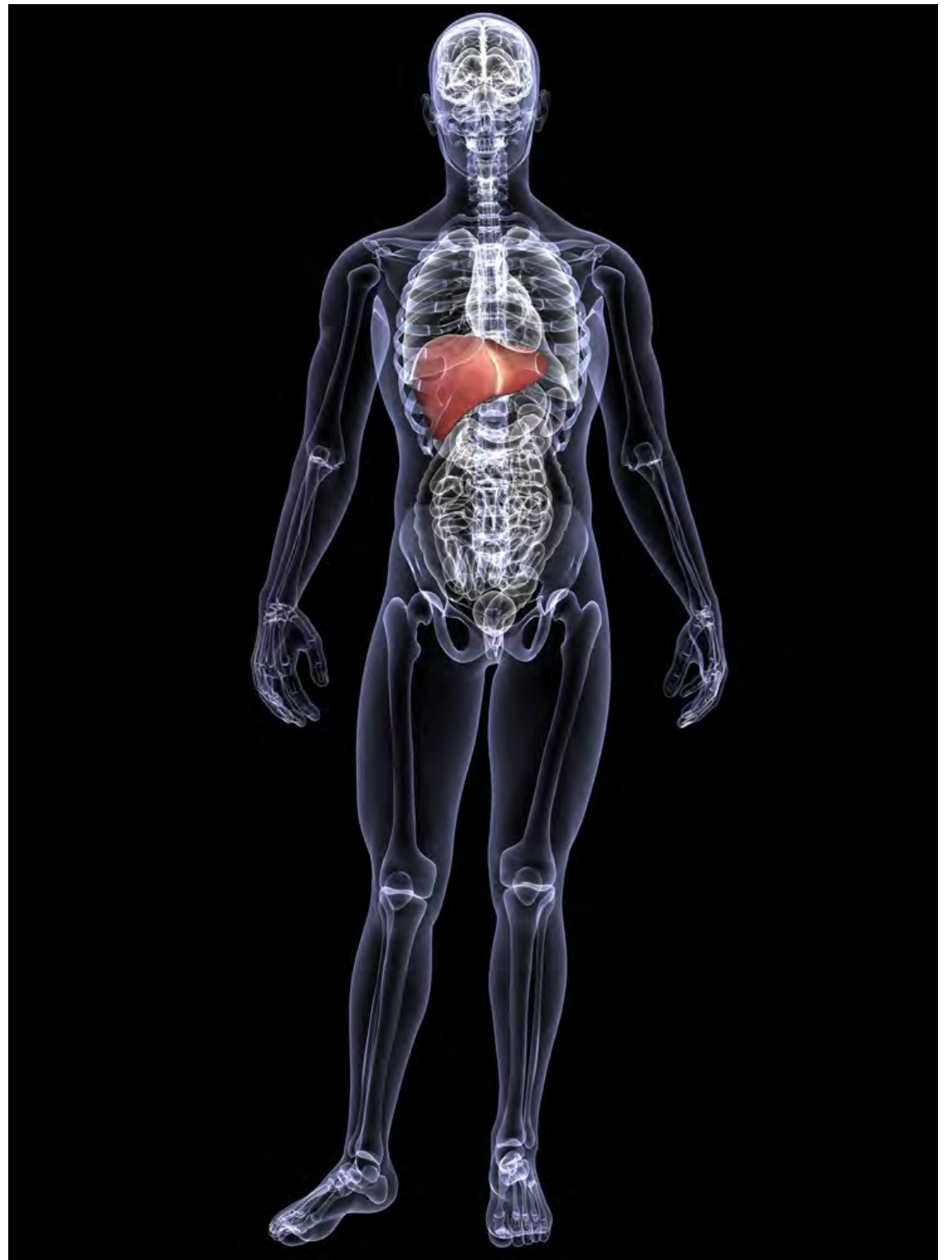


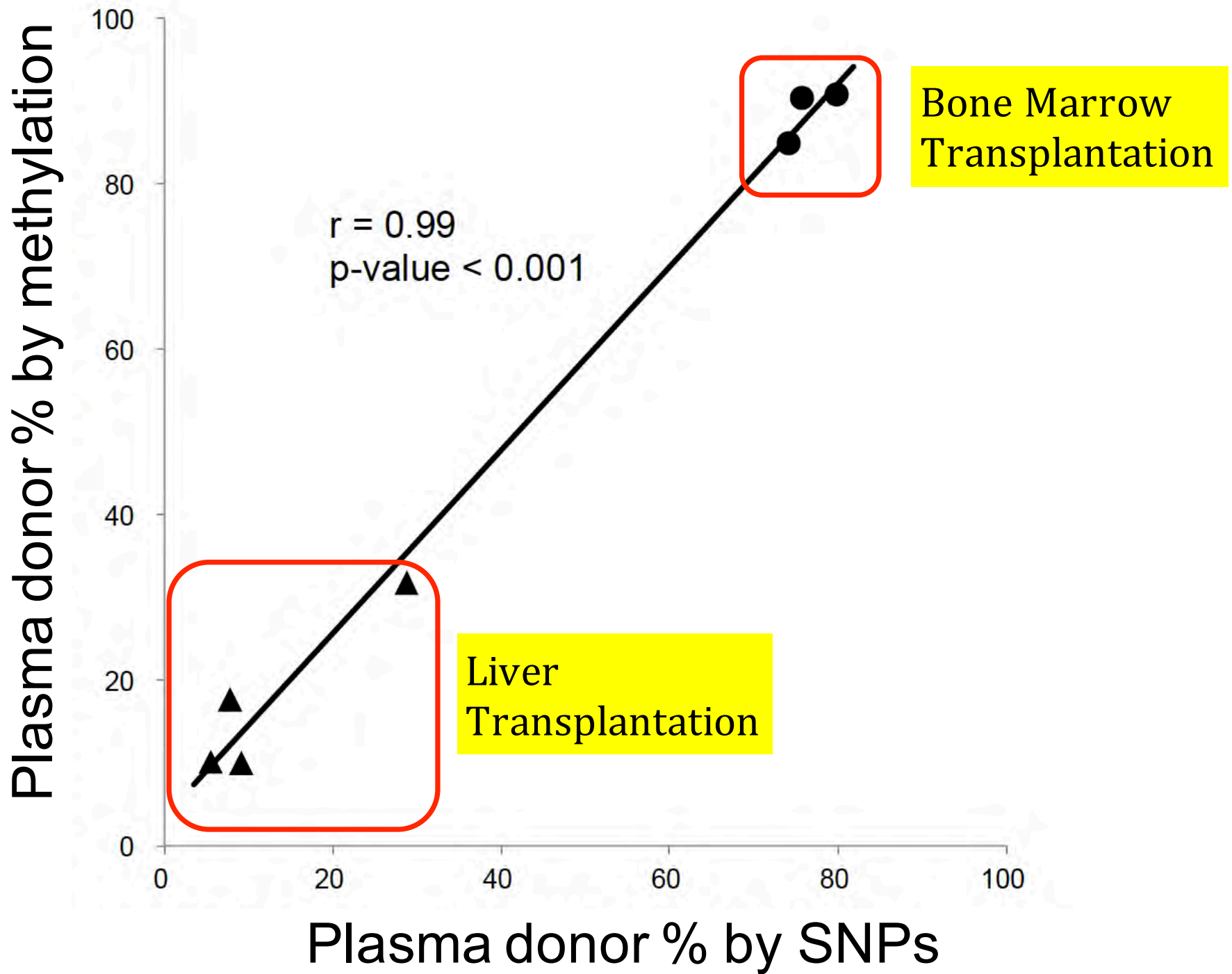
Transplantation

Bone Marrow Transplantation

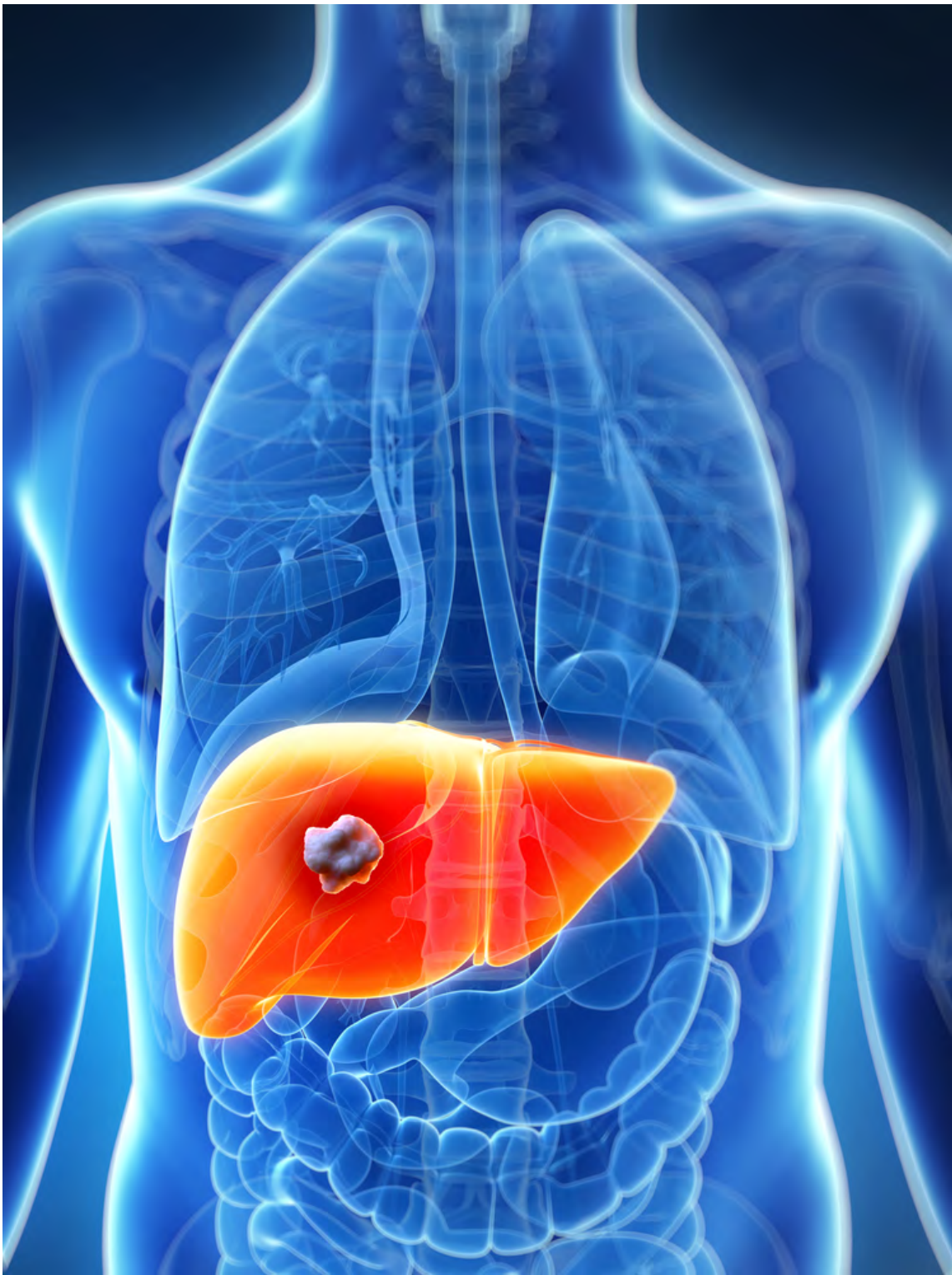


Liver Transplantation





Cancer



Hepatocellular Carcinoma (HCC)

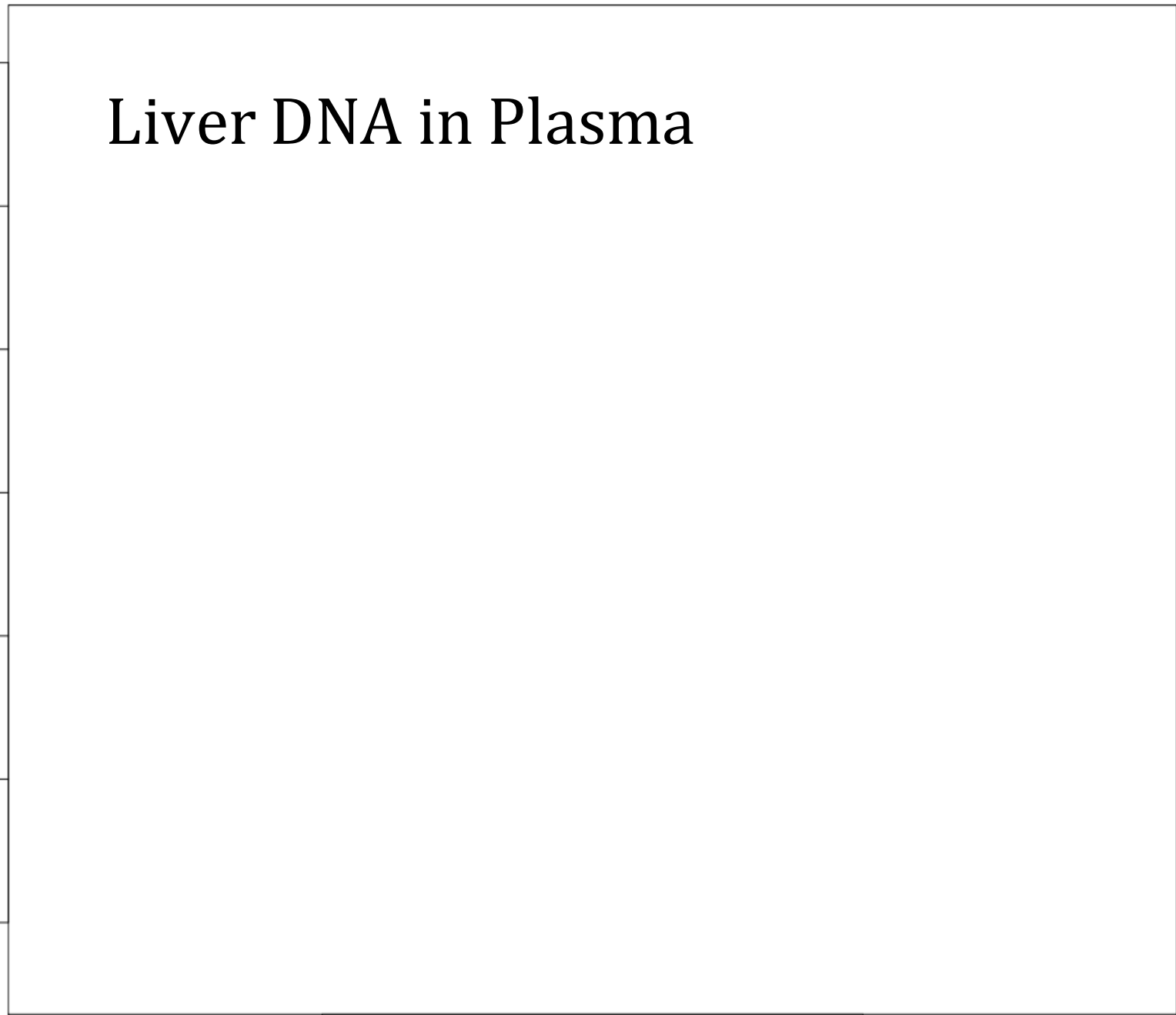
Plasma liver DNA % by methylation

Liver DNA in Plasma

70
60
50
40
30
20
10

Controls

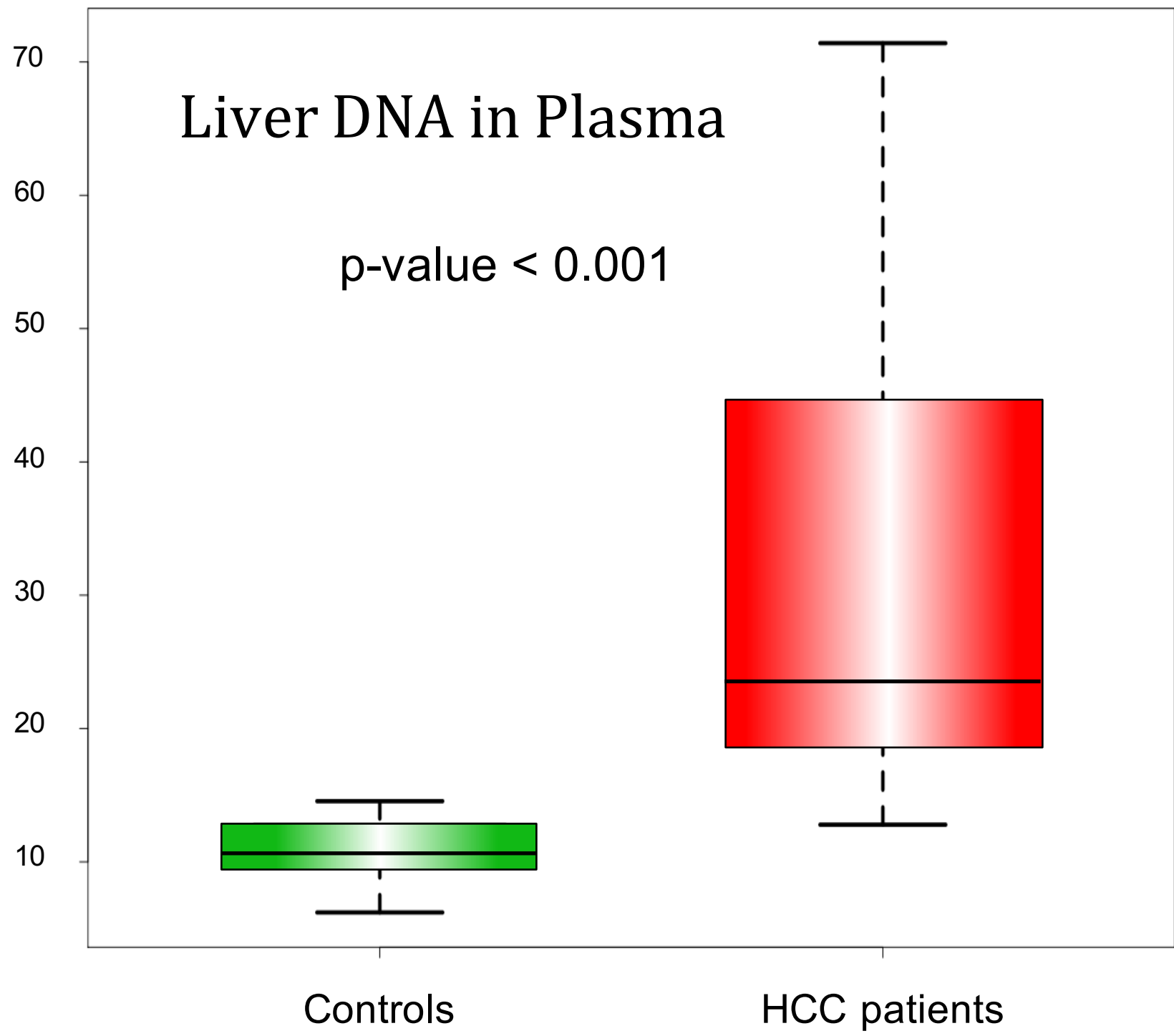
HCC patients

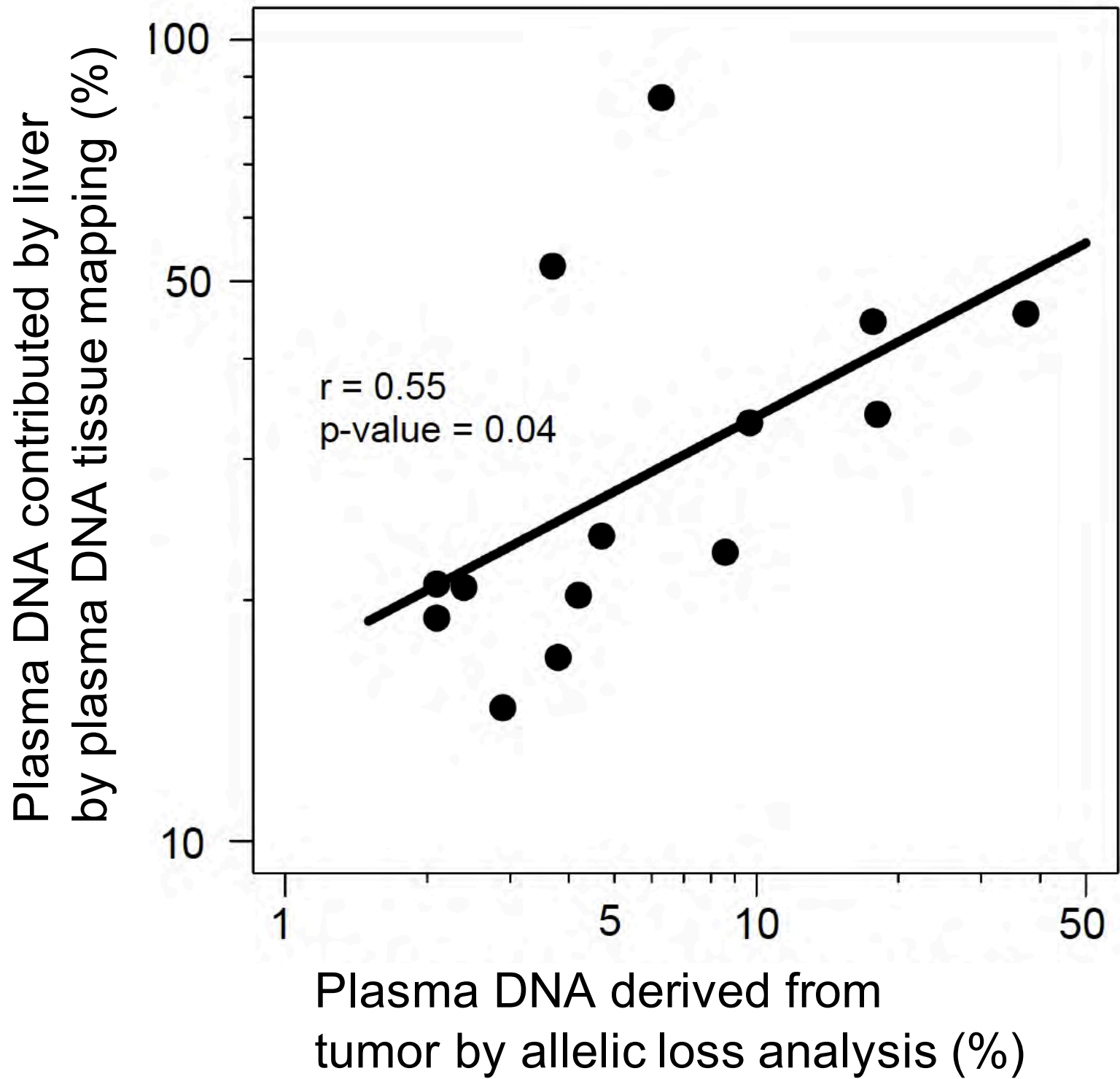


Plasma liver DNA % by methylation

Liver DNA in Plasma

p-value < 0.001





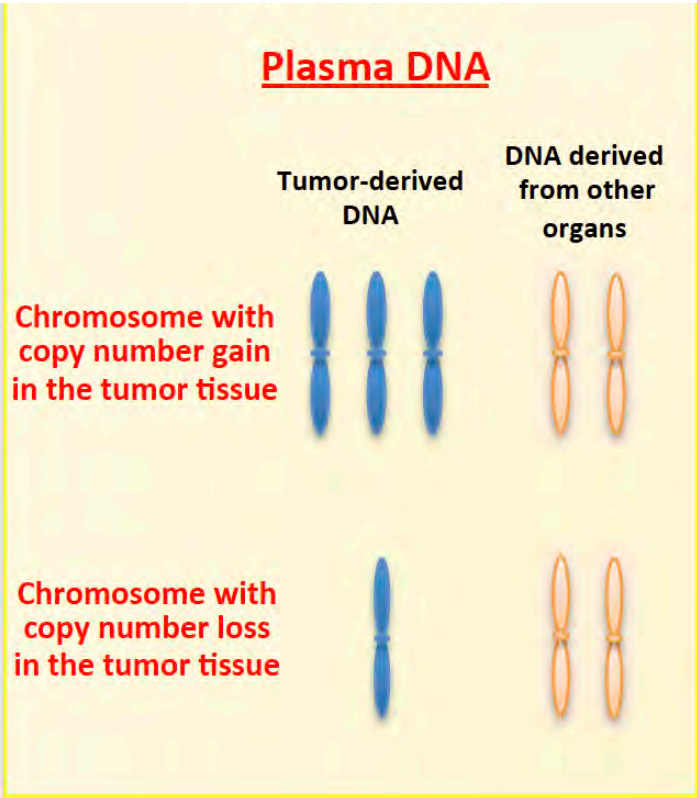
Plasma DNA tissue mapping by genome-wide methylation sequencing for noninvasive prenatal, cancer, and transplantation assessments

Kun Sun^{a,b,1}, Peiyong Jiang^{a,b,1}, K. C. Allen Chan^{a,b,c,1}, John Wong^d, Yvonne K. Y. Cheng^e, Raymond H. S. Liang^f, Wai-kong Chan^g, Edmond S. K. Ma^g, Stephen L. Chan^h, Suk Hang Cheng^{a,b}, Rebecca W. Y. Chan^{a,b}, Yu K. Tong^{a,b}, Simon S. M. Ng^d, Raymond S. M. Wong^{i,j}, David S. C. Huiⁱ, Tse Ngong Leung^k, Tak Y. Leung^e, Paul B. S. Lai^{c,d}, Rossa W. K. Chiu^{a,b}, and Yuk Ming Dennis Lo^{a,b,c,2}

PNAS 2015; 112: E5503-12

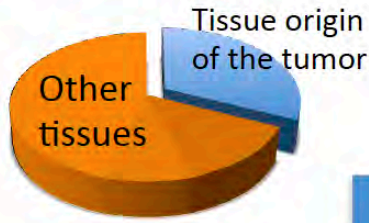
Mapping the Source of Plasma Genomic Aberrations

Cancer Patient

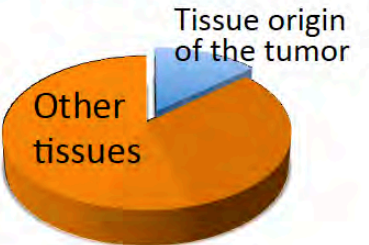


Chromosome-specific plasma DNA tissue mapping

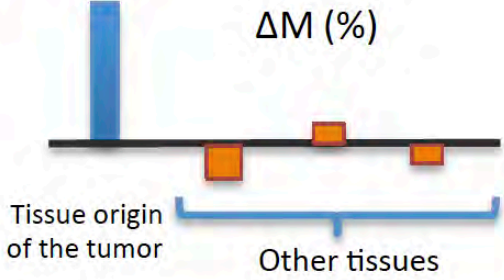
Chromosome with copy number gain in the tumor

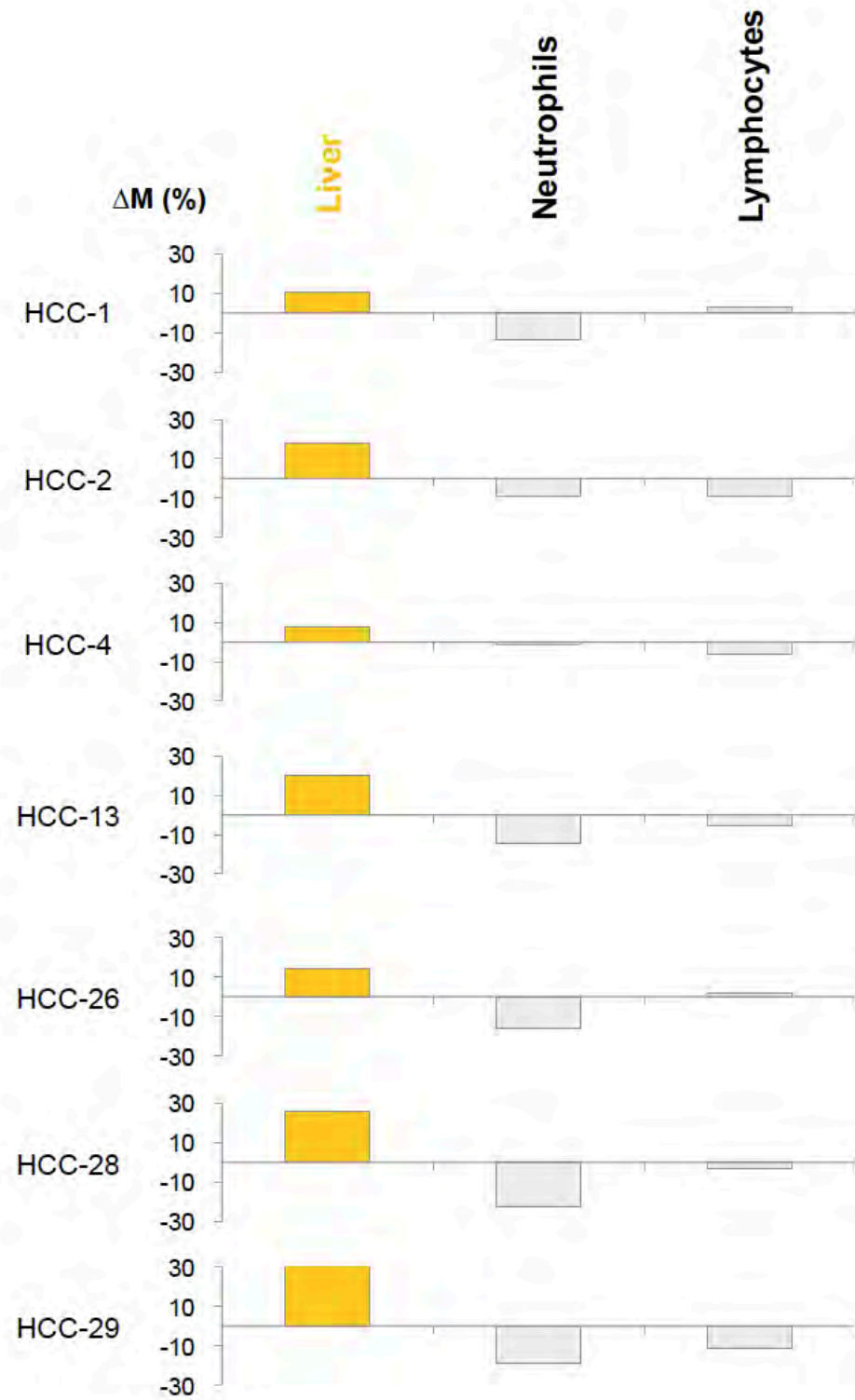


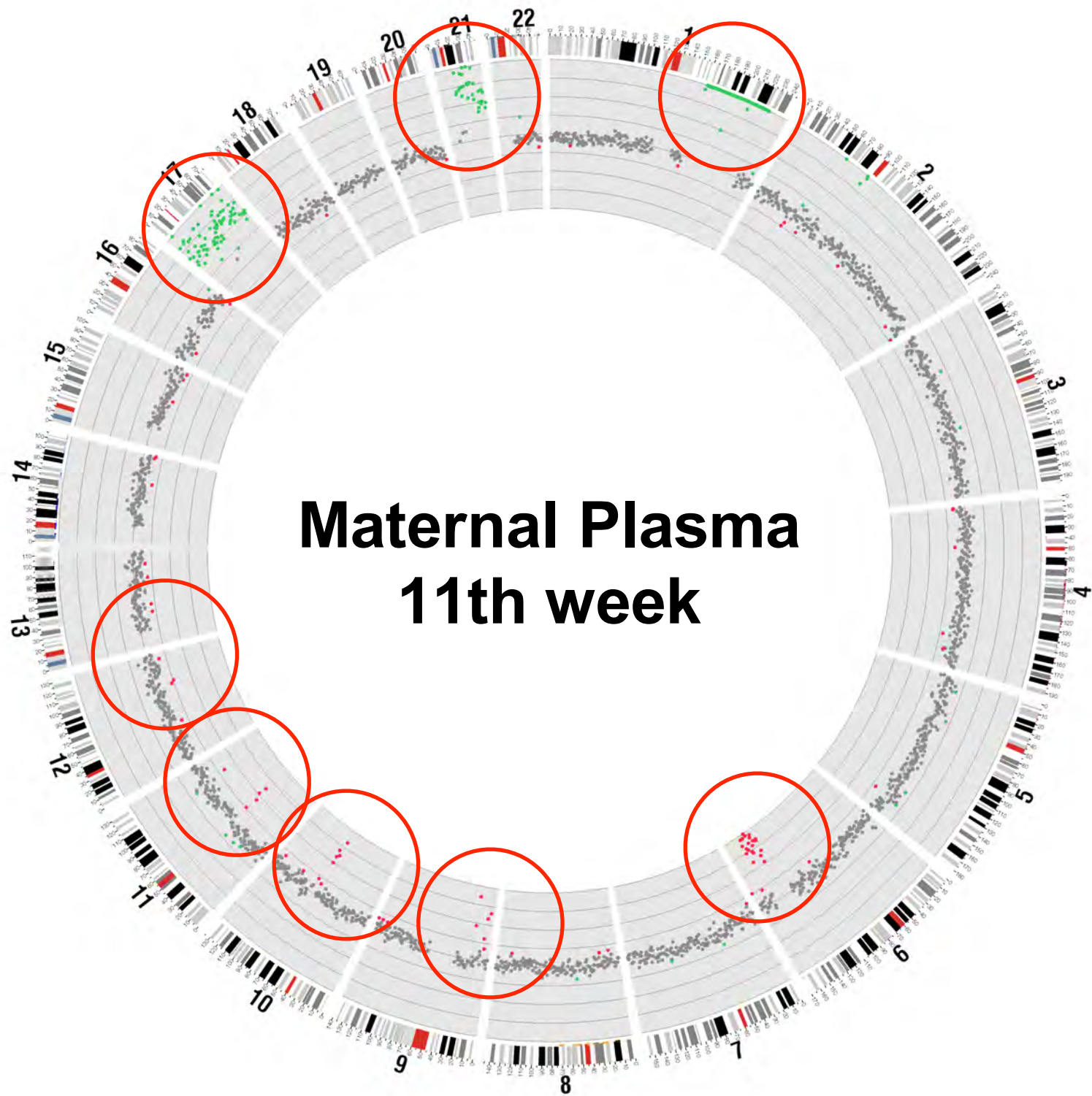
Chromosome with copy number loss in the tumor



Difference in the tissue contributions between the two sets of chromosomes

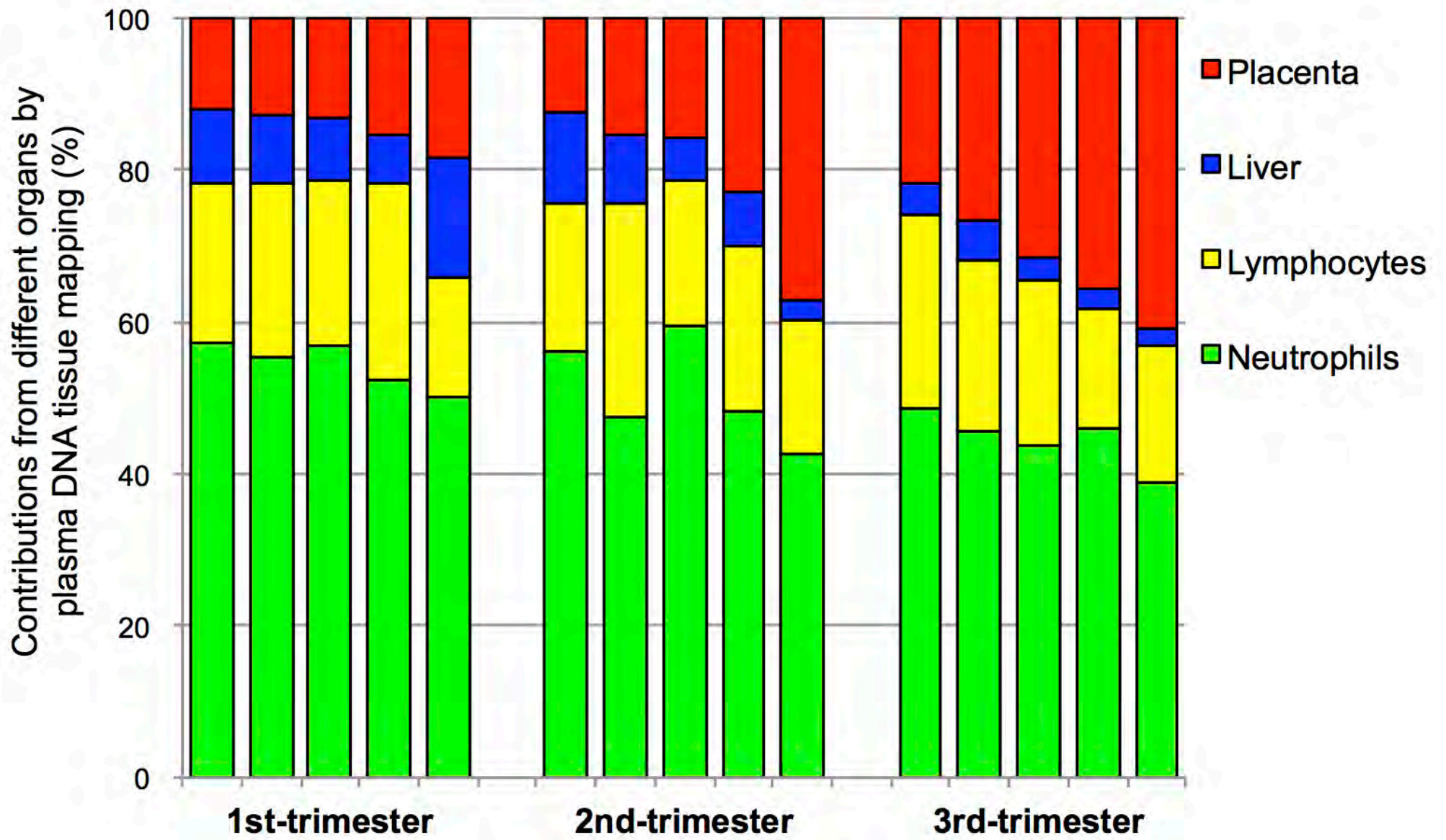


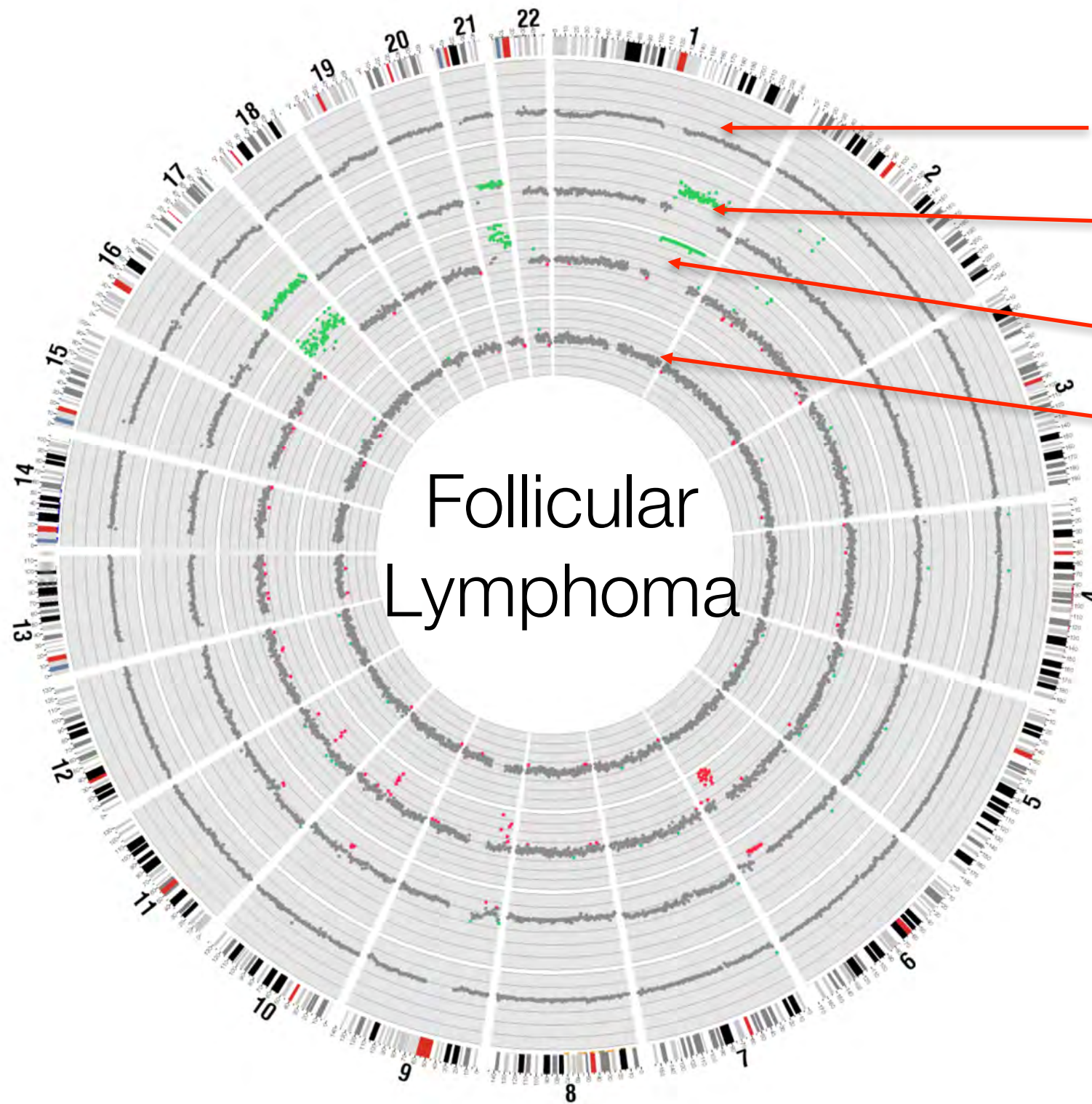




Maternal Plasma 11th week

Organ	Percentage contributions of plasma DNA (%)
Liver	0.6
T cells	6.8
B cells	62.6
Neutrophils	4.4
Placenta	25.5





Buffy coat

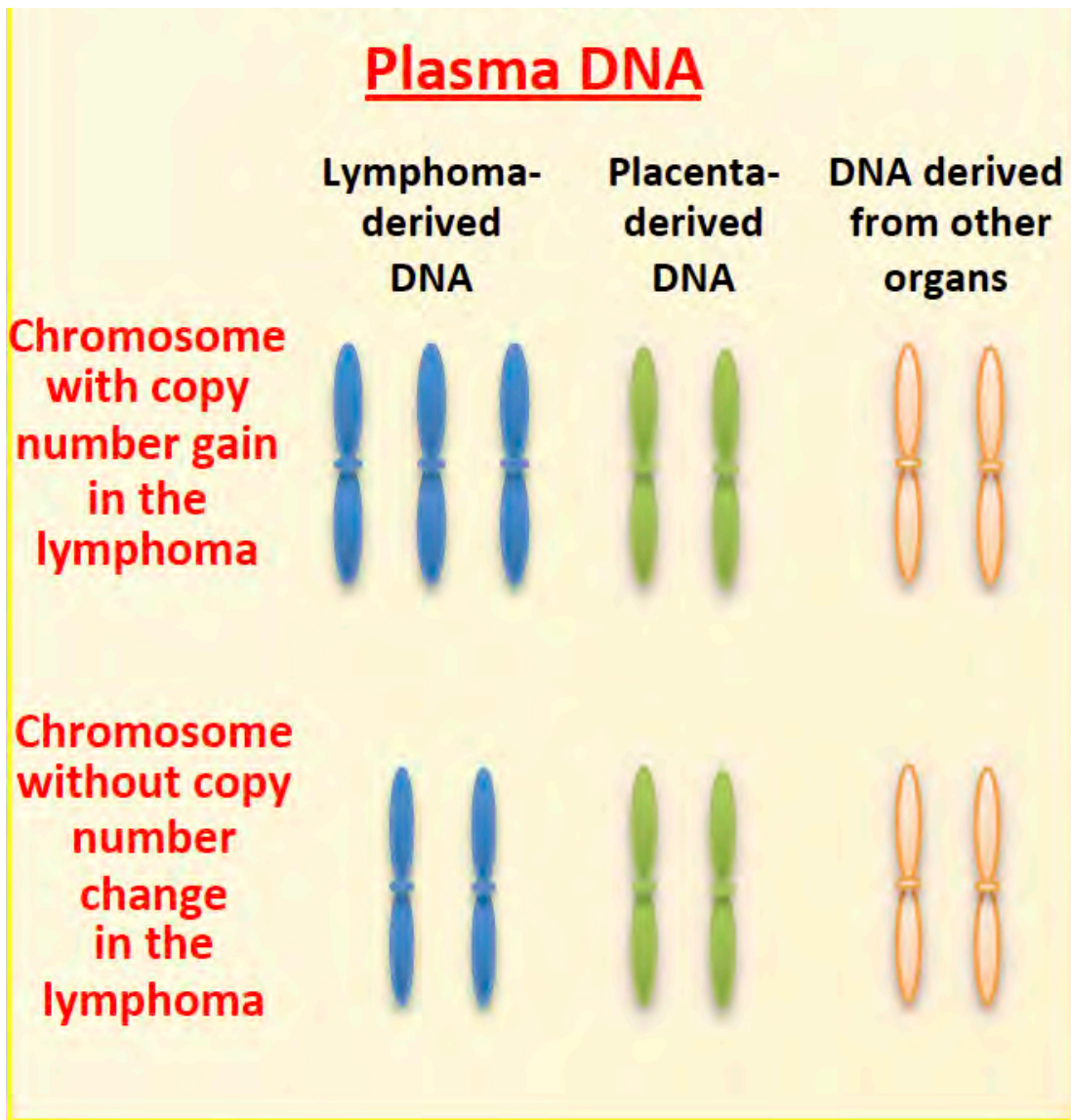
Lymph node

NIPT

Post-treatment

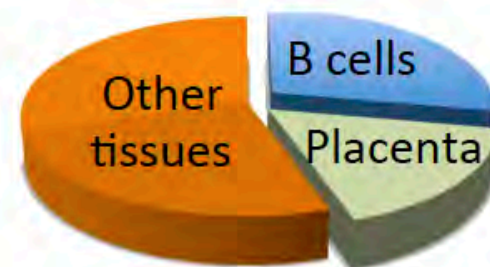
Follicular
Lymphoma

Pregnant woman with lymphoma

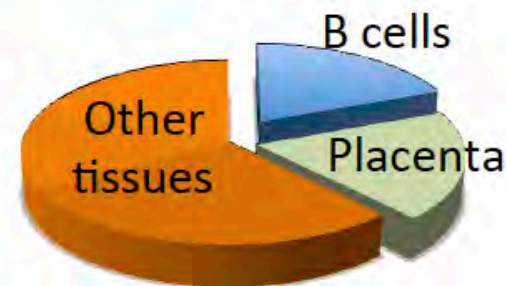


Chromosome-specific plasma DNA tissue mapping

Chromosome with copy number gain in the lymphoma



Chromosome without copy number change in the lymphoma



ΔM (%)

15
5
-5
-15

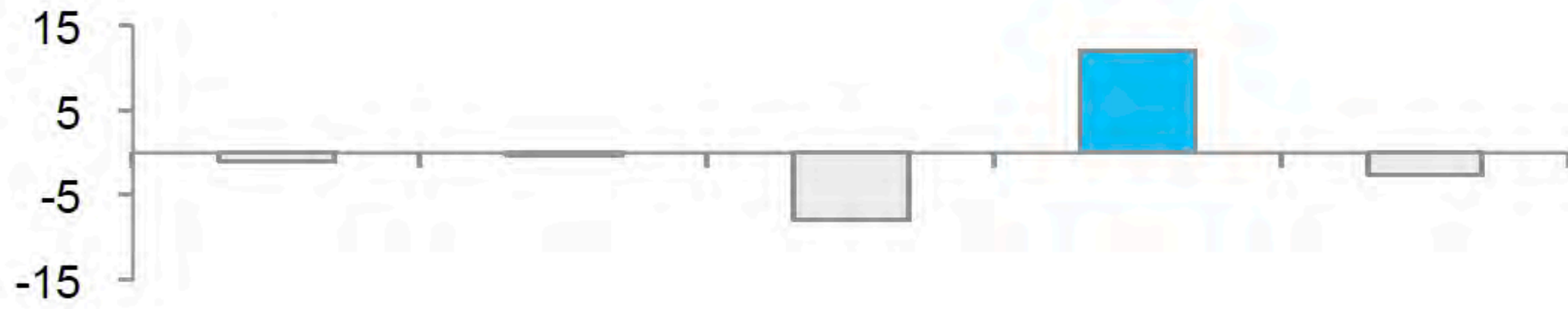
Liver

Neutrophils

T-cells

B-cells

Placenta



From liquid biopsy to anatomy



Conclusion



- ◆ Plasma DNA is a treasure trove for molecular diagnostics
- ◆ Era of non-invasive prenatal testing
- ◆ Universal approach for cancer detection
- ◆ Plasma DNA tissue mapping
- ◆ Many other future applications





Acknowledgements

- ◆ Department of Obstetrics and Gynaecology, CUHK
Tak Yeung Leung, Yvonne Cheng
- ◆ King's College Hospital, UK
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Mary Tang, Elizabeth Lau
- ◆ Hong Kong Polytechnic University
Cesar Wong
- ◆ United Christian Hospital, Hong Kong
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Anthony Chan, Stephen Chan
- ◆ Department of Surgery, CUHK
Paul Lai, John Wong
- ◆ Kwong Wah Hospital, Hong Kong
Wing Cheong Leung
- ◆ Pamela Youde Nethersole Hospital, Hong Kong
Rebecca Tang
- ◆ Tuen Mun Hospital, Hong Kong
Sidney Au Yeung
- ◆ Princess Margaret Hospital, Hong Kong
Helena Lam
- ◆ Yu Yat Kung
- ◆ Boston University
Charles Cantor



HEALTH

Li Ka Shing Institute of Health Sciences 李嘉誠健康科學研究所

Thank you!