Pathologie bij gemetastaseerde ziekte

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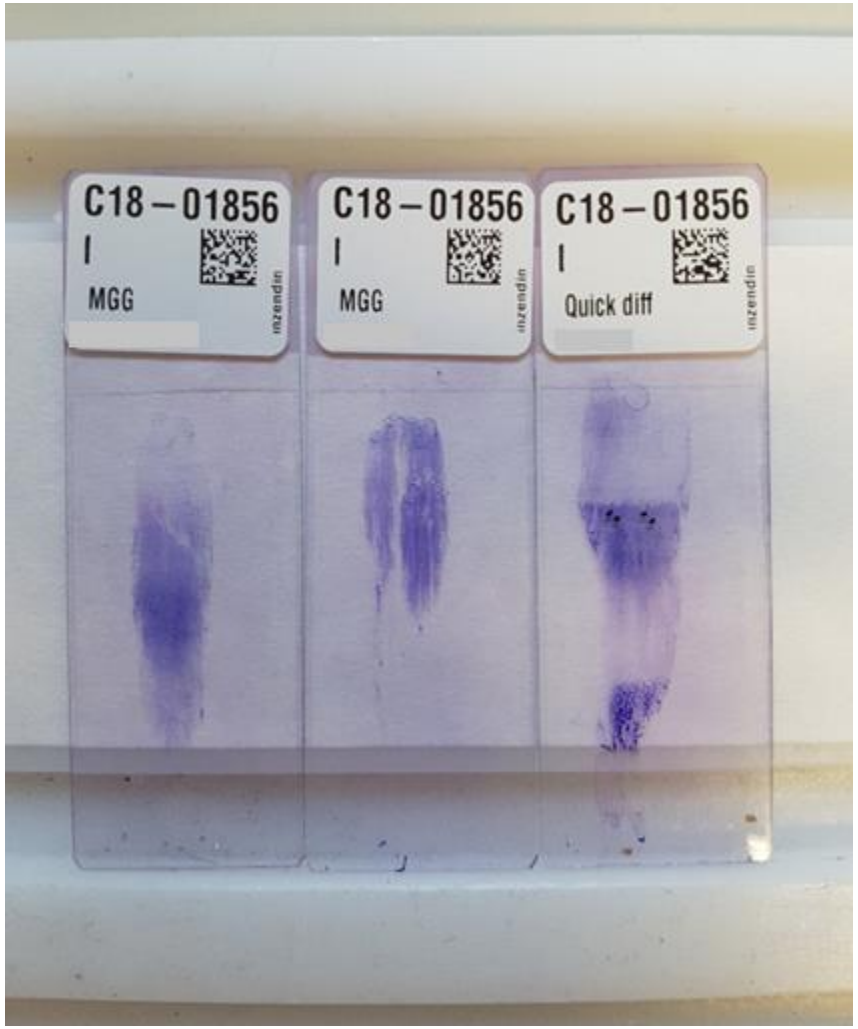
Patholoog
Pathologie DNA, locatie Antonius ziekenhuis

PATHOLOGIE DNA
Partner in pathologie

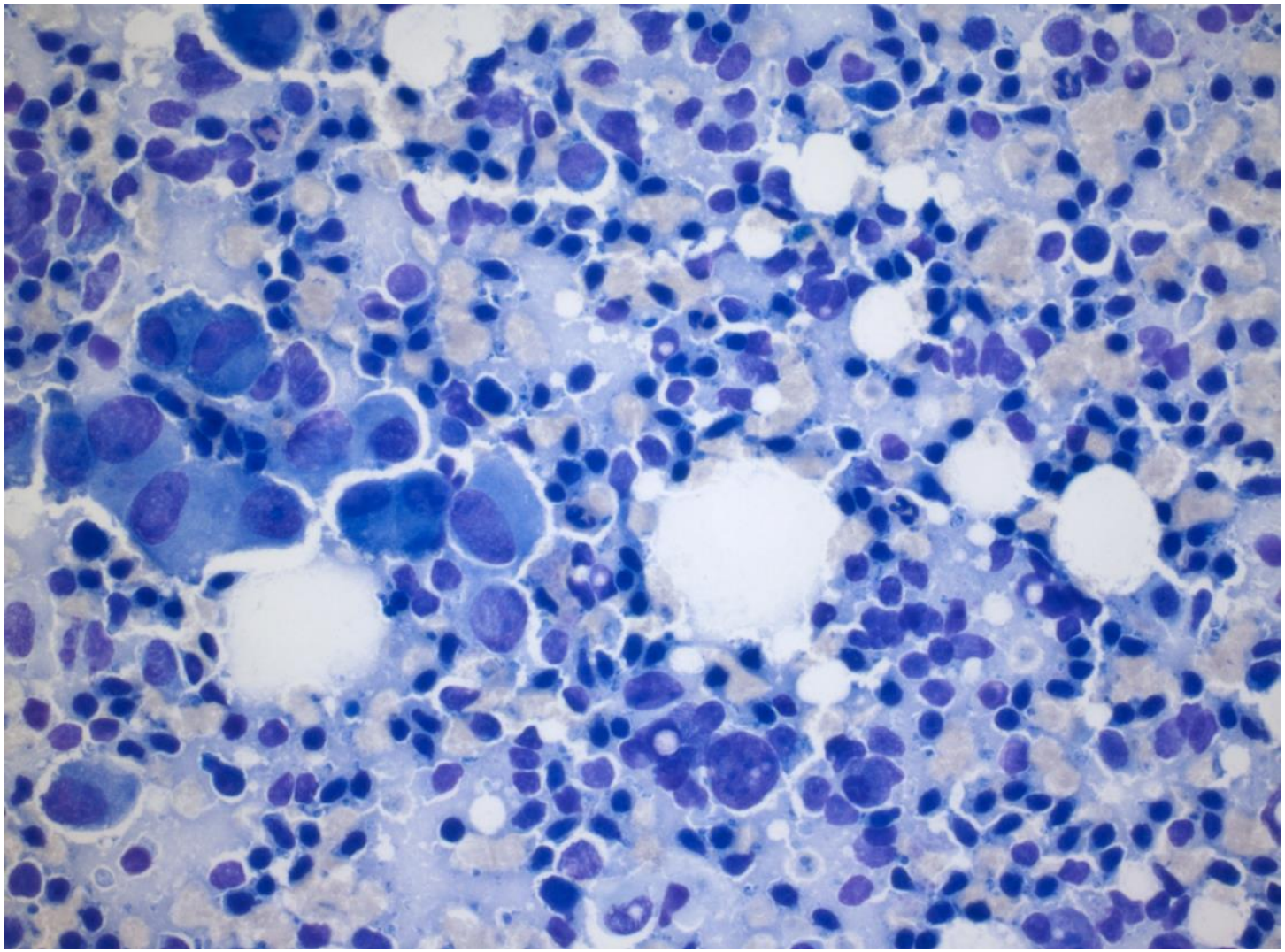
- Introductie pathologie en verschillende technieken
  - Cytologie
  - Histologie
  - Immuunhistochemie
  - Moleculair onderzoek
- Gemetastaseerd mammacarcinoom
- Gemetastaseerd longcarcinoom

Cytologie - celkunde

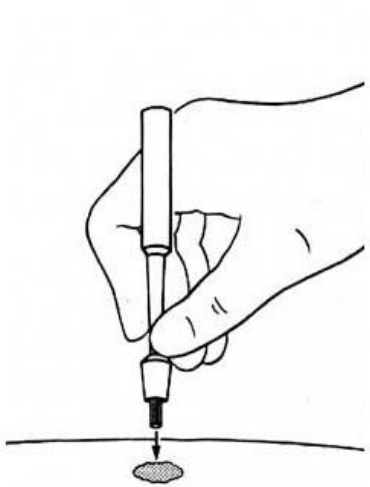
Cytologie glaasjes

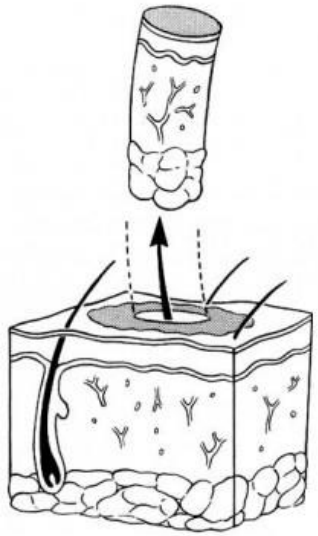


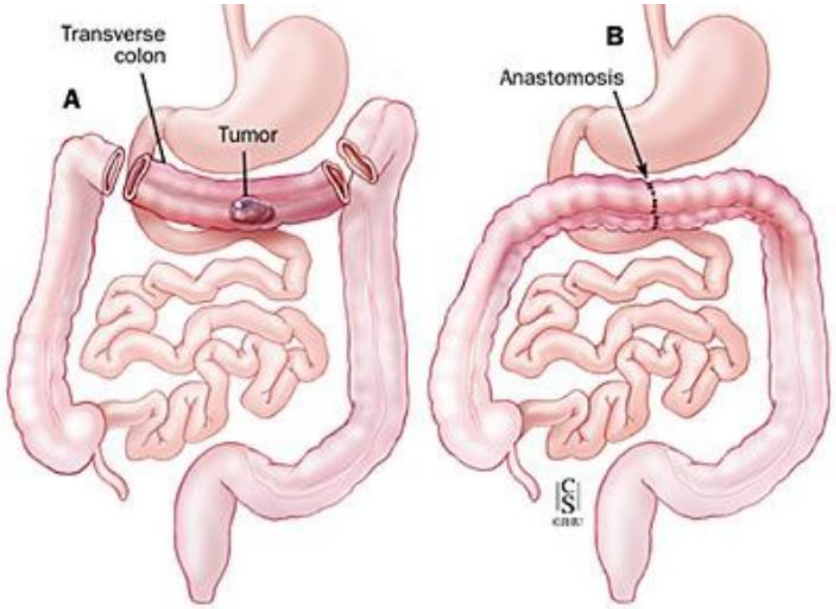
Cytologie



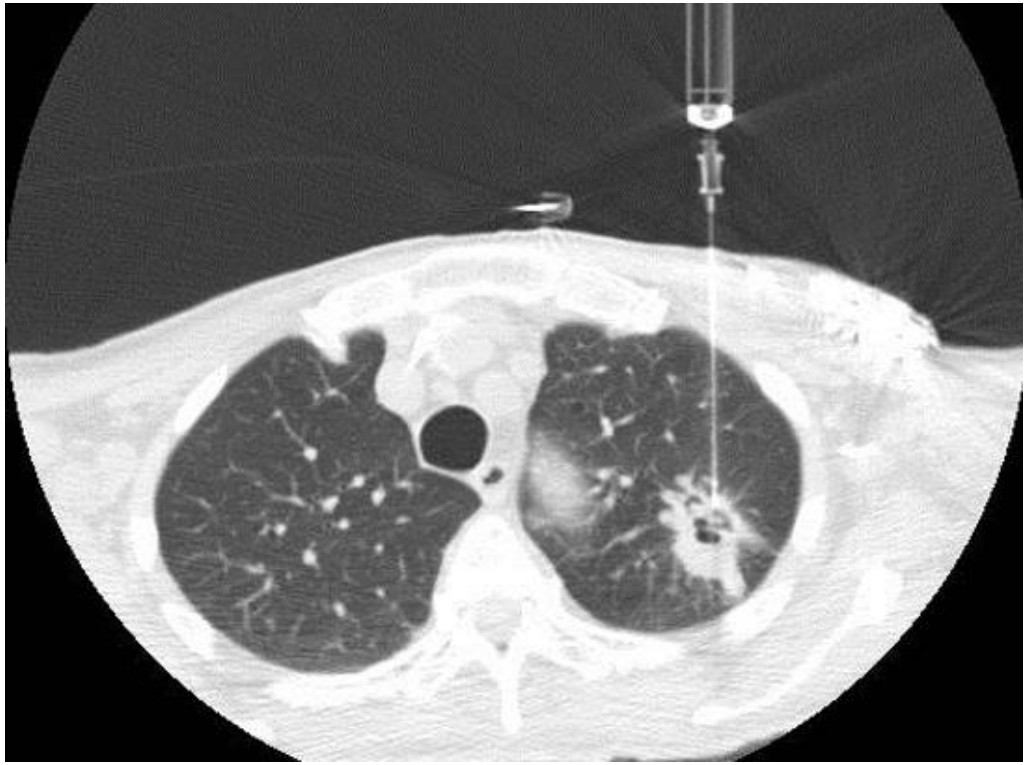
Histologie - weefselonderzoek

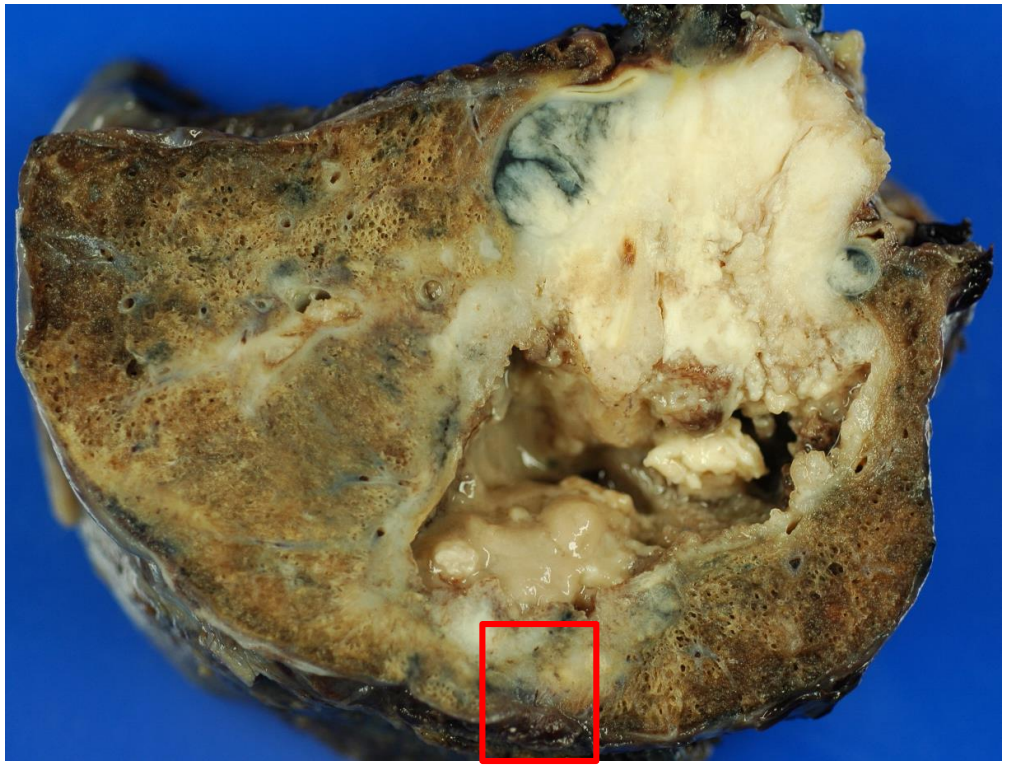






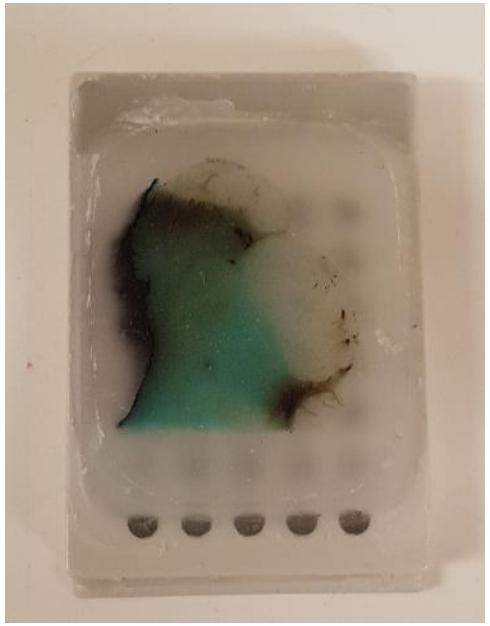
Biopten en resecties





Indozen en doorvoeren

Inbedden in paraffine

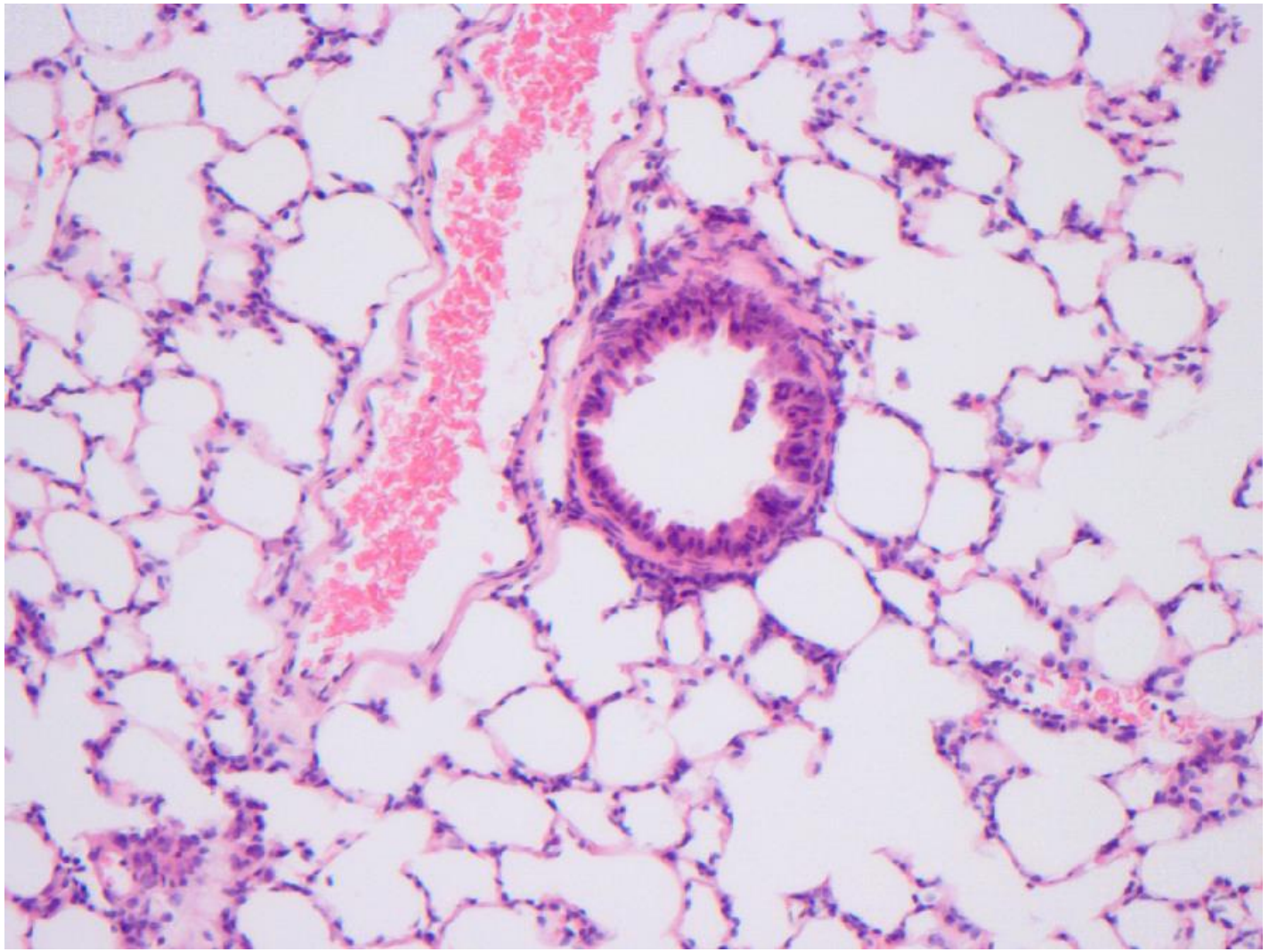


Coupes snijden

Coupes kleuren

Coupes scannen

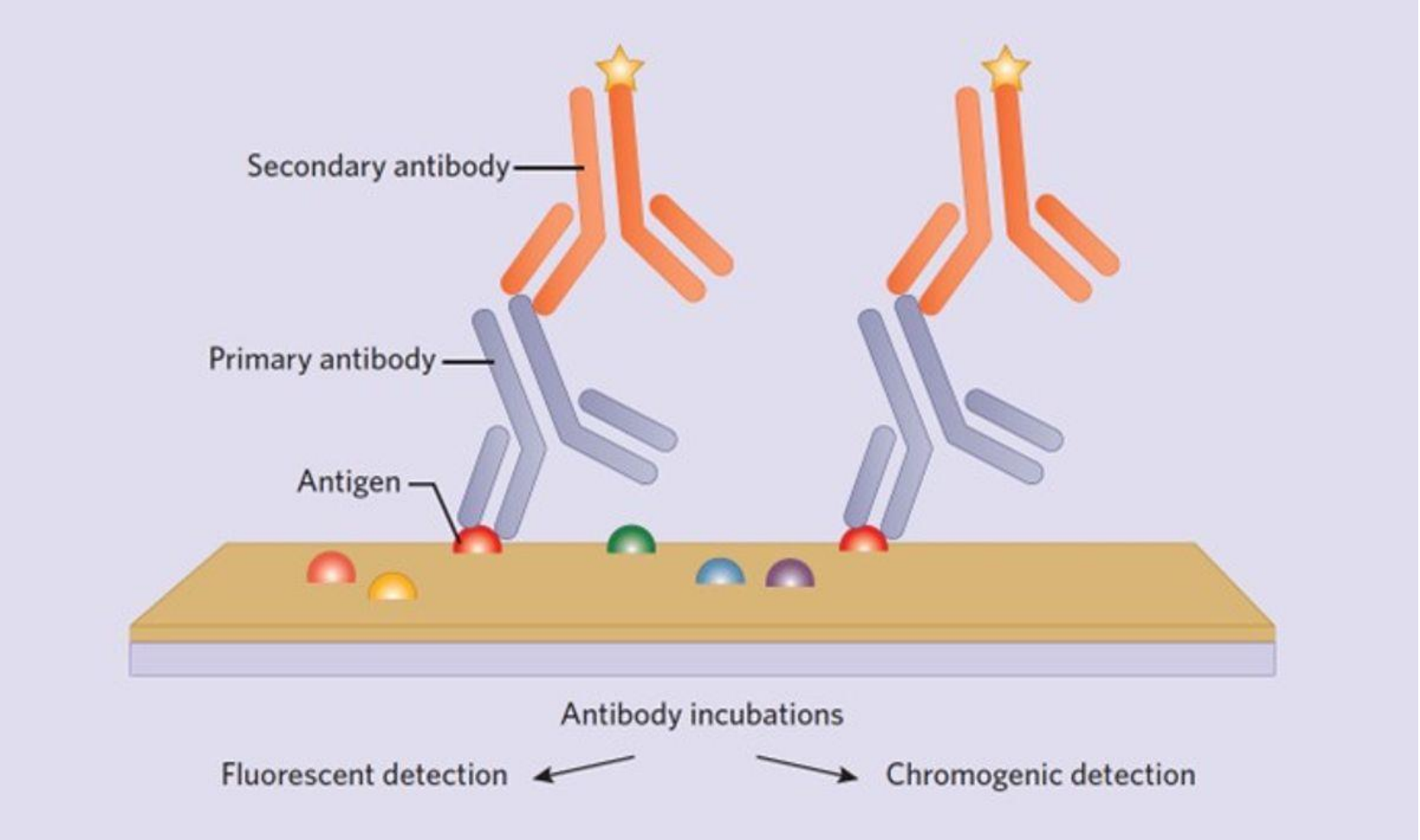
Histologie Long



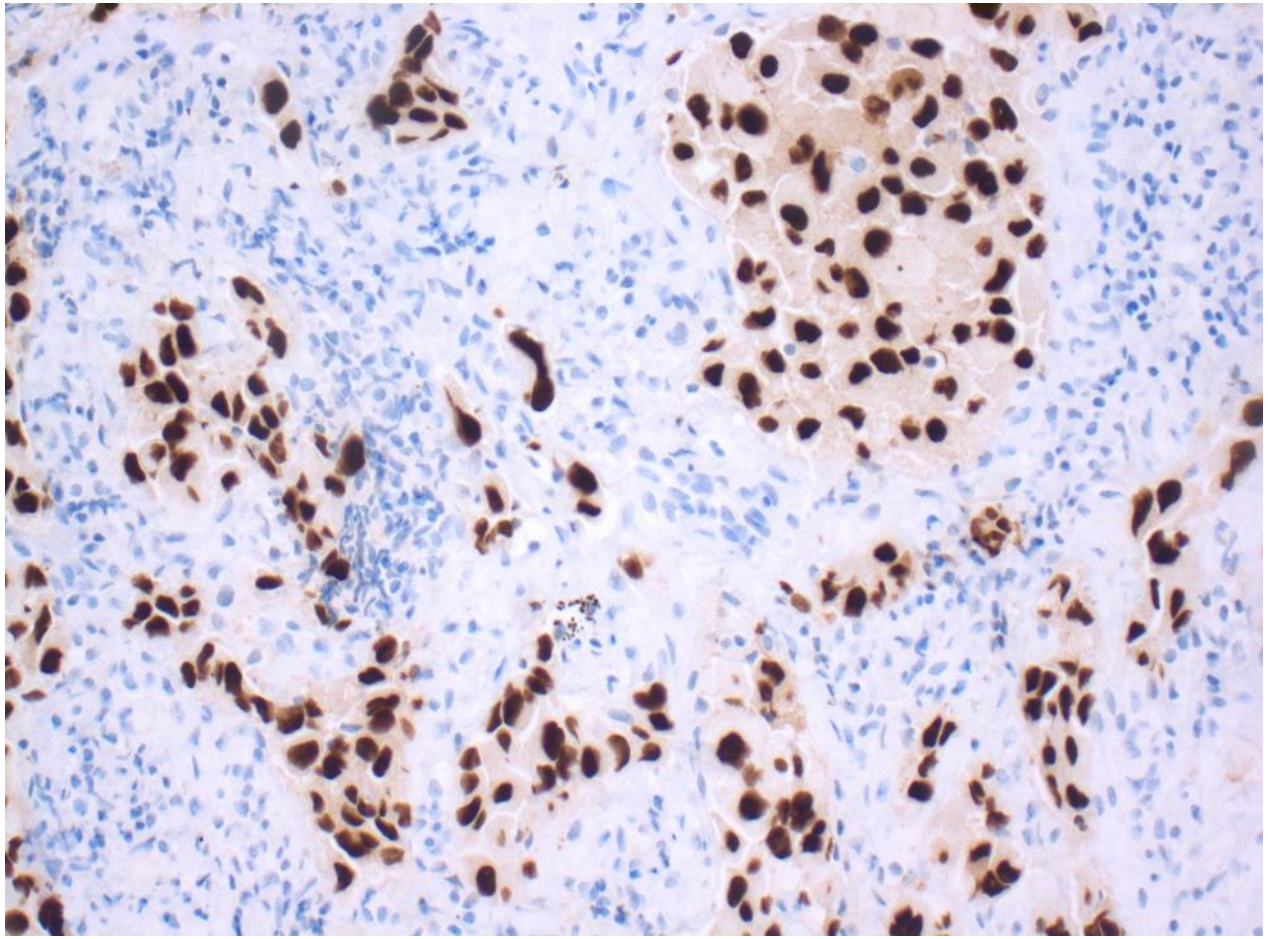
Aanvullende technieken

- Histologische kleuringen
- Immuunhistochemie
- Electronenmicroscopie
- Moleculaire diagnostiek

Immuunhistochemie



Immuunhistochemie

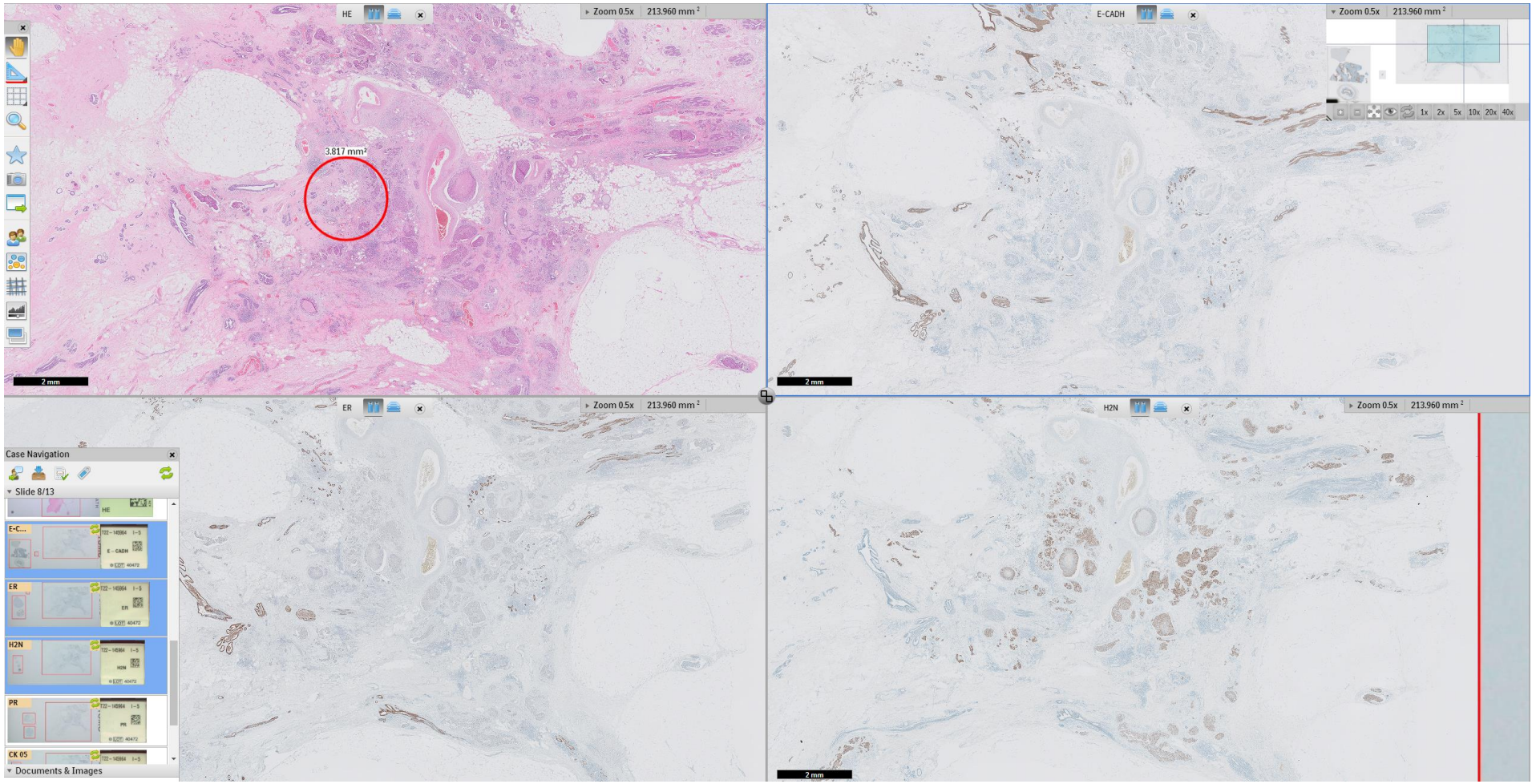


Immuunhistochemie

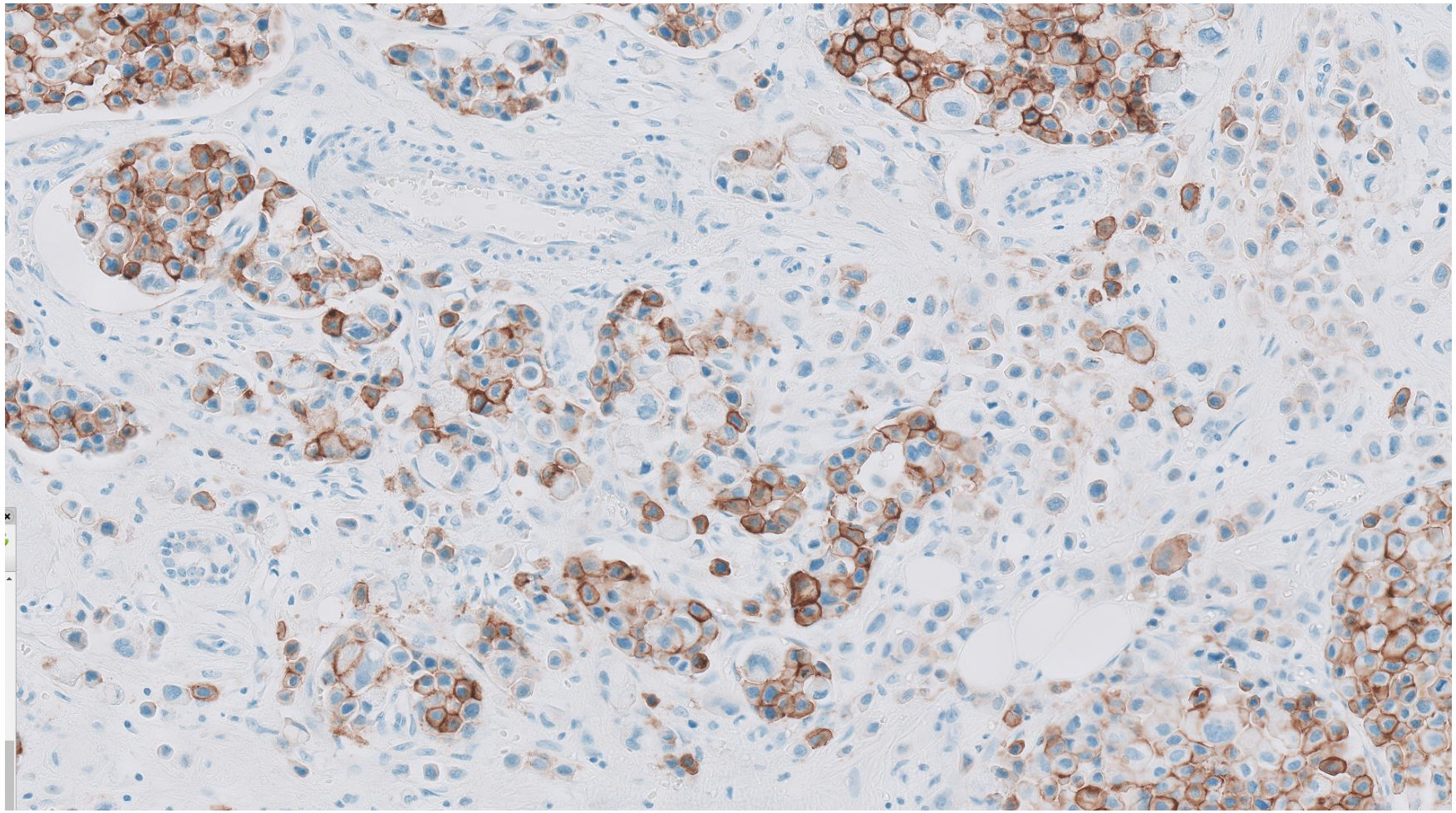
Immunologische kleuringen

ACTINE	CD 01a	CD 99	DOG-1	IgA	MPO	PIN
AFP	CD 01a (R)	CD 117	EBER ISH	IgA FITC	MSH-2	PLAP
ALK	CD 02	CD 123	EBV-LMP1	IgD *	MSH-6	PMS-2
ALK-1 *	CD 03	CD 138	E-CADH	IgE *	MTAP	Podopl
AMACR	CD 03 (KP)*	CD 163	EMA	IgG	MUC-1	PR
Amyloid AA *	CD 04	CDK4	ER	IgG 4	MUC-2	PRAME
AR *	CD 05	CDX2	ERG	IgG 4 (+ IgG)	MUC-4	PRAME (R)
A-synucleine *	CD 07	CEA (M)	F VIII *	IgG FITC	MUC-5AC	PSA
B-APP *	CD 08	CEA (P) *	F 13a	IgM	MUC-6	RB-1
BAP-1	CD 08 (KP)*	CHROMO	GATA-3	IgM FITC	MUM-1	RCC
BCL-2	CD 10	CK 05	GFAP	INHIBI	Myogenine	ROS1
BCL-2 C2	CD 14	CK 5+6*	GLUT-1	KAPPA	Myosin SM	S100
BCL-6	CD 15	CK 5+6-P63*	GLYC A	KAPPA BM *	Napsin A	S100 (R)
BCL-10	CD 20	CK 5+P63	GLYPICAN 3	KAPPA FITC	NF	SATB-2
BerEp4	CD 21	CK 7	GRANZB	KAPPA ISH *	NUT	SMA
Blanco (i)	CD 23	CK 8+18	H2Neu	Ki-67	NKX-3	SMAD-4
BOB-1	CD 25	CK13+CK17 *	H2Neu SISH	Ki-67 (R)	OCT-2	SMARCA4
BRAF	CD 30	CK 14	HBc-Ag *	Ki-67 (KP) *	OCT-4	SMOOTH
BRAF (R)	CD 31	CK 19	HBs-Ag *	LAMBDA	P16	SOX-10
C1q FITC	CD 31+CK BS5	CK 20	HCG	LAMBDA BM *	P16 (R)	SOX-11
C3c FITC	CD 34	CK 34BE12	HHV-8	LAMBDA FITC	P40	Spirocheet
C-Myc	CD 43	CK AE1-3	HMB-45	LAMBDA ISH *	P53	STAT-6
CA IX	CD 44	CK AE1-3 (R)	HMB-45 (R)	LEF-1	P57	SYNAP
CA125	CD 45	CK BS5	HP	LMO2	P63	TdT
CA19.9	CD 56	CK MNF	HSA	MAI *	P120	THYREO
CALCIT	CD 57	CMV	HSV (R)	MDM-2	PAN-TRK *	TIA-1
CALDES	CD 61	CXCL13 *	HSV-I	Mel A	PARVO B19 *	TRPS1
CALPONINE	CD 68	CyclD1	HSV-II *	Mel A (R)	PAX5	Tryptase *
CALRET	CD 68 (R)	DESM (M)	ICOS *	MEL A+Ki-67	PAX8	TTF-1
CATbet	CD 79a	DESM (P) *		MLH-1	PD-1	VIM
				MOC 31	PERIPH *	WT-1

Digitale pathologie

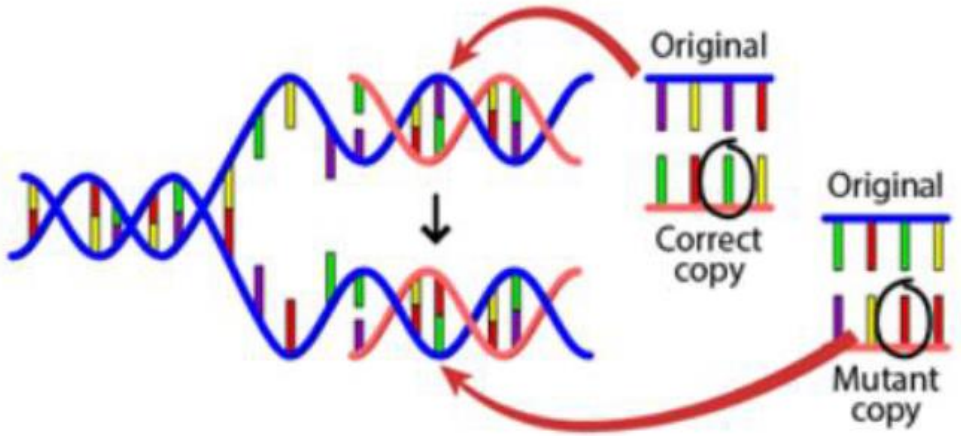


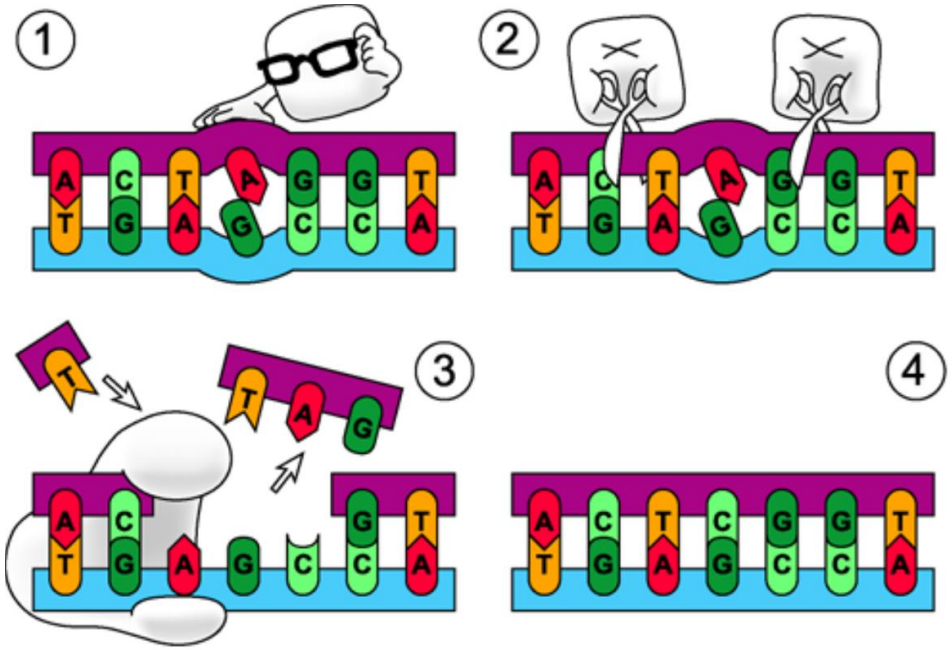
Artificical intelligence: algoritmes



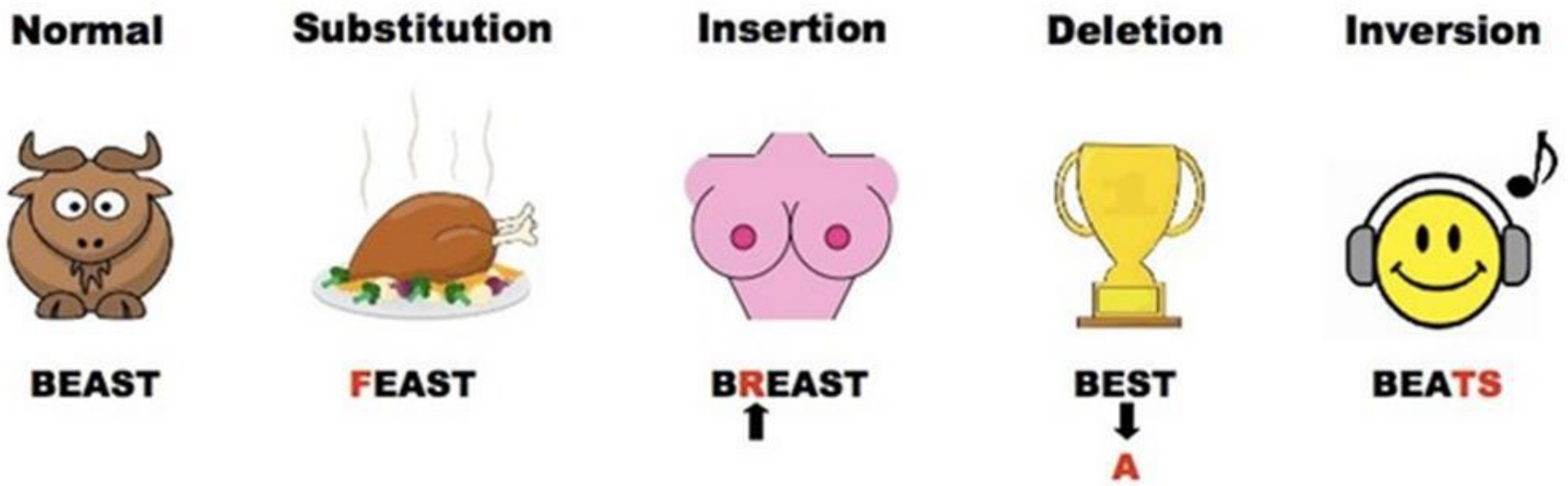
Moleculaire diagnostiek

- Op zoek naar fouten ontstaan tijdens celdeling



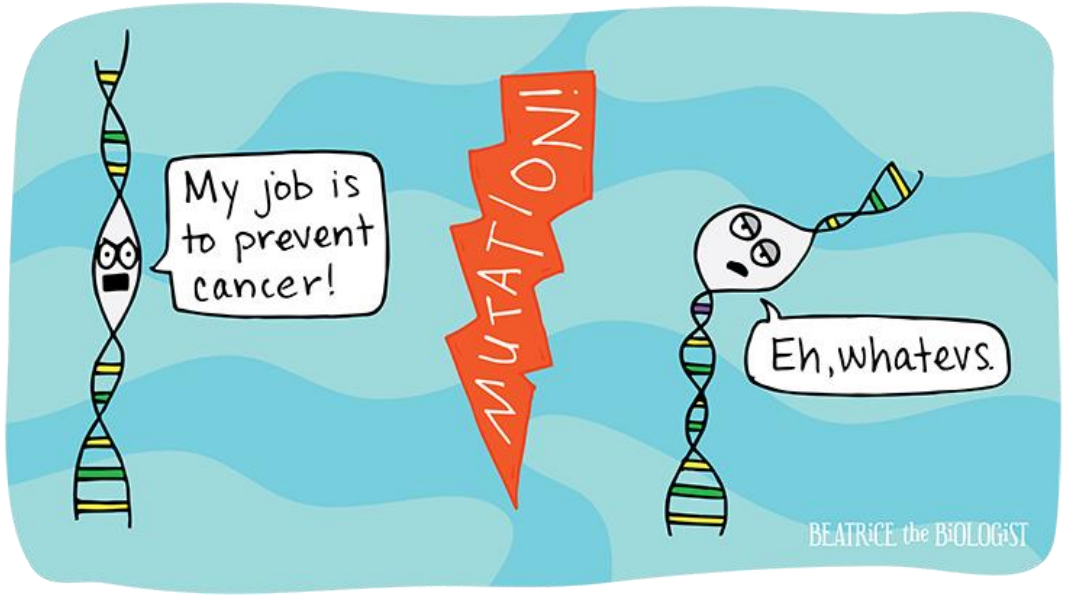


Moleculaire diagnostiek

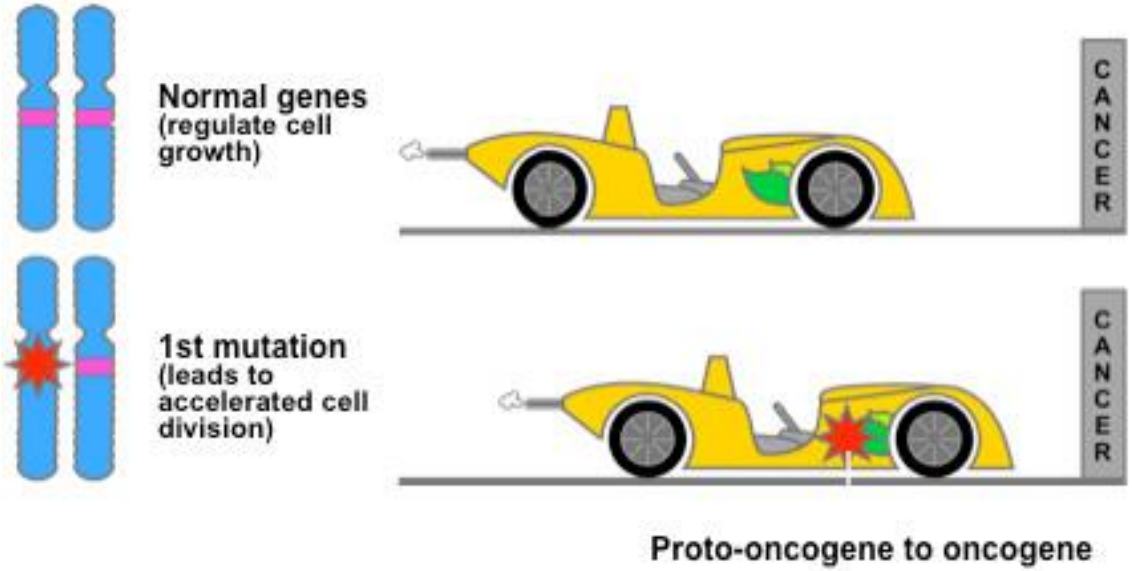


Moleculaire diagnostiek

Tumorsupressorgenen



Oncogenen



Tumorsupressorgenen vs Oncogenen

Table 14.3

Representative Tumor-Suppressor Genes Implicated in Human Tumors

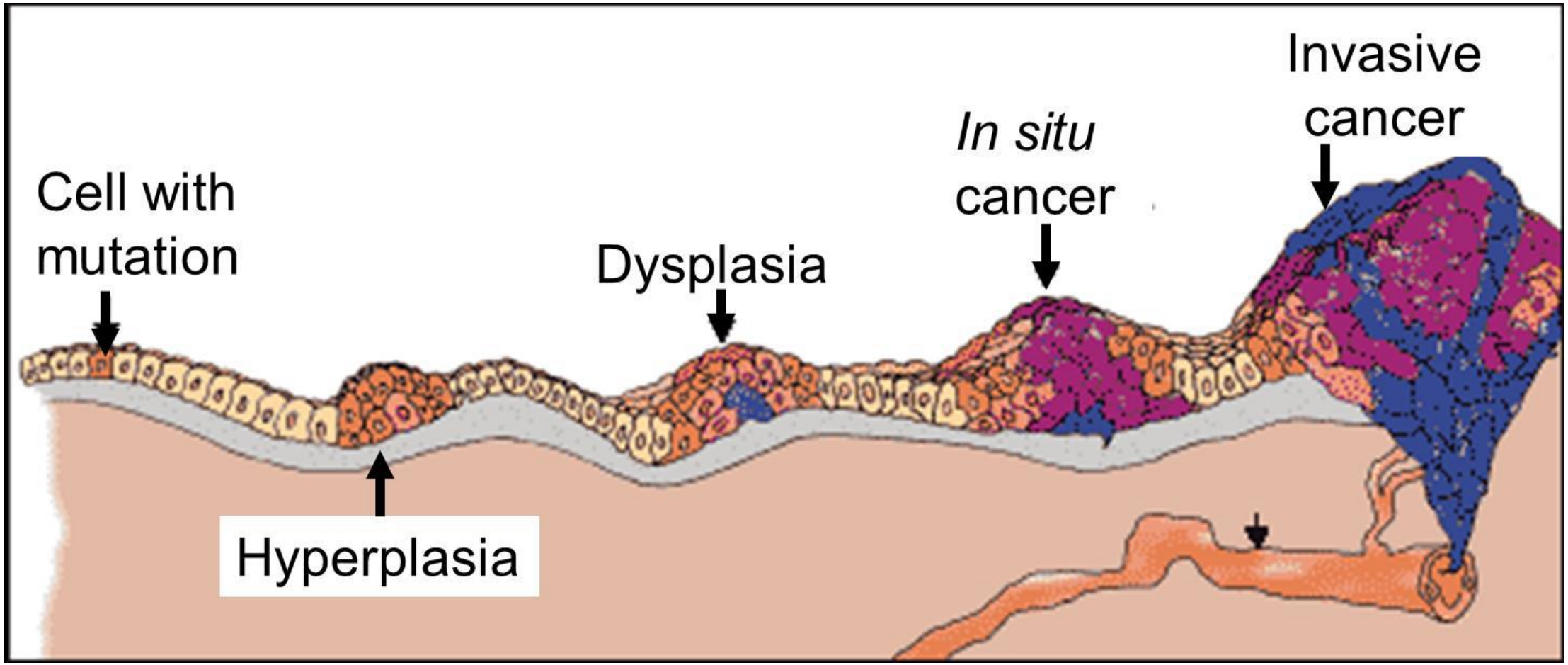
Tumor-Suppressor Gene	Neoplasm(s)
RBI	Retinoblastoma; osteosarcoma; carcinoma of breast, bladder, and lung
p53	Astrocytoma; carcinoma of breast, colon, and lung; osteosarcoma
WT1	Wilms' tumor
DCC	Carcinoma of colon
NF1	Neurofibromatosis type 1
FAP	Carcinoma of colon
MEN-1	Tumors of parathyroid, pancreas, pituitary, and adrenal cortex

Adapted from Bishop, J. M. 1991. Molecular themes in oncogenesis. *Cell* **64**, 235–248.

Target	Disease
HER-2	Breast[a]
BCR/ABL	Chronic myeloid leukemia[a]
C-KIT	Gastrointestinal stromal tumor[a]
EGFR	NSCLC[a]
EGFR	Head and neck, colorectum[a]
EGFR	Pancreas[a]
VEGF	Breast, colorectum[a], kidney
VEGFR, B-Raf	Kidney[a]

© 2008 **Nature Publishing Group** Weinstein, I. B. *et. al.*, Mechanisms of Disease: oncogene addiction a rationale for molecular targeting in cancer therapy, *Nature Clinical Practice Oncology* >b>3, 448-457 (2006).

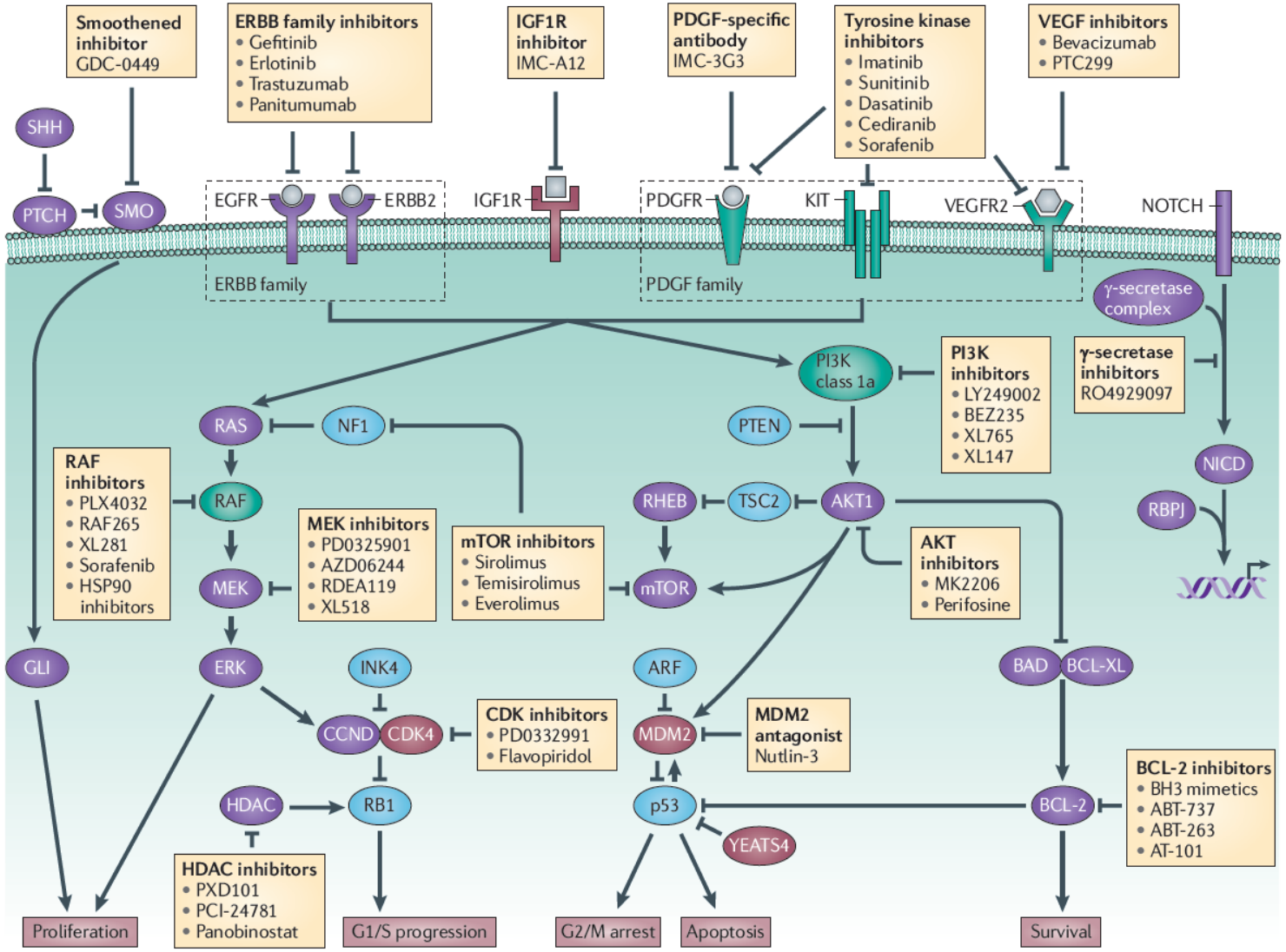
Carcinogenese



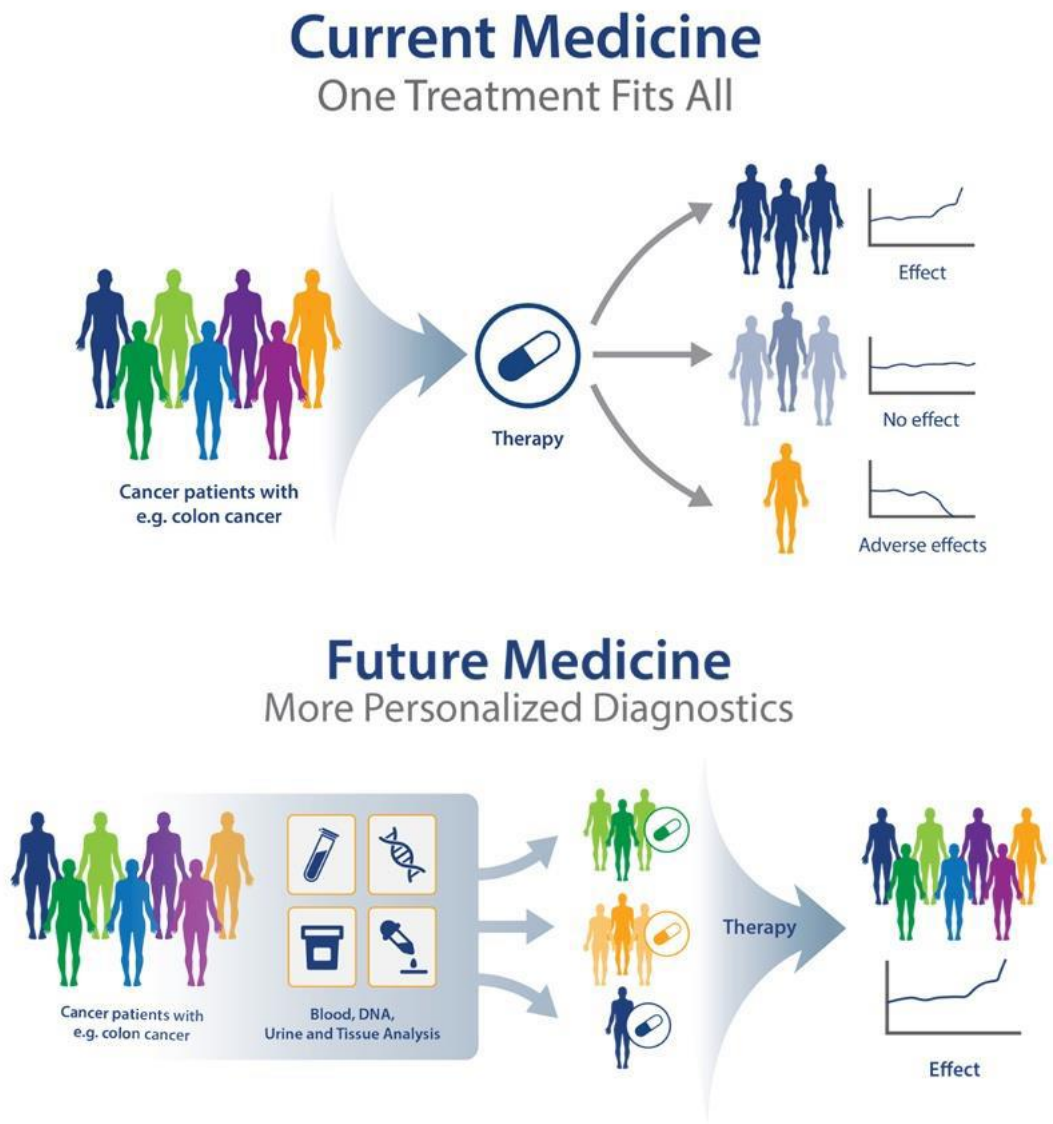
Moleculaire testen

- DNA: Next generation sequencing, whole genome sequencing, whole exome sequencing
- RNA: Next generation sequencing, whole transcriptome sequencing
- Eiwit: Immuunhistochemie en ISH

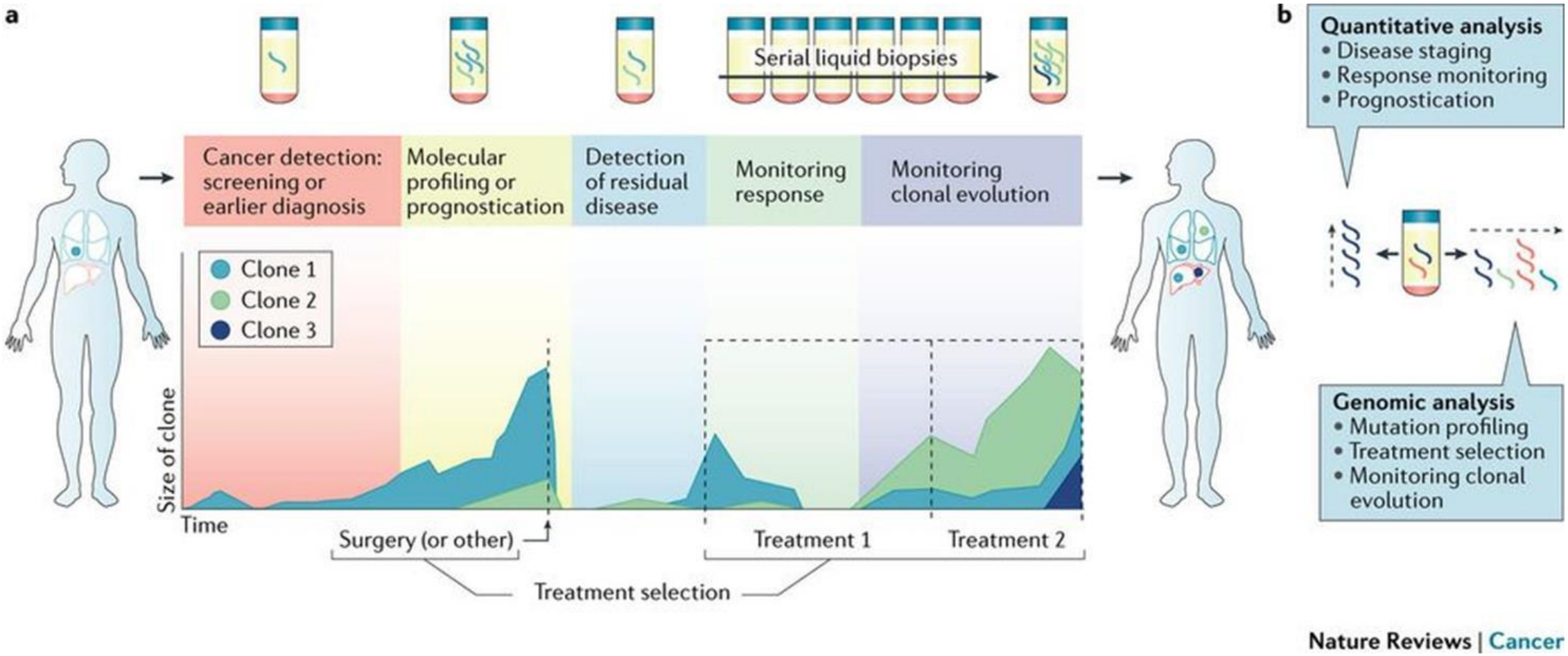
Moleculaire diagnostiek: doel



Personalized medicine

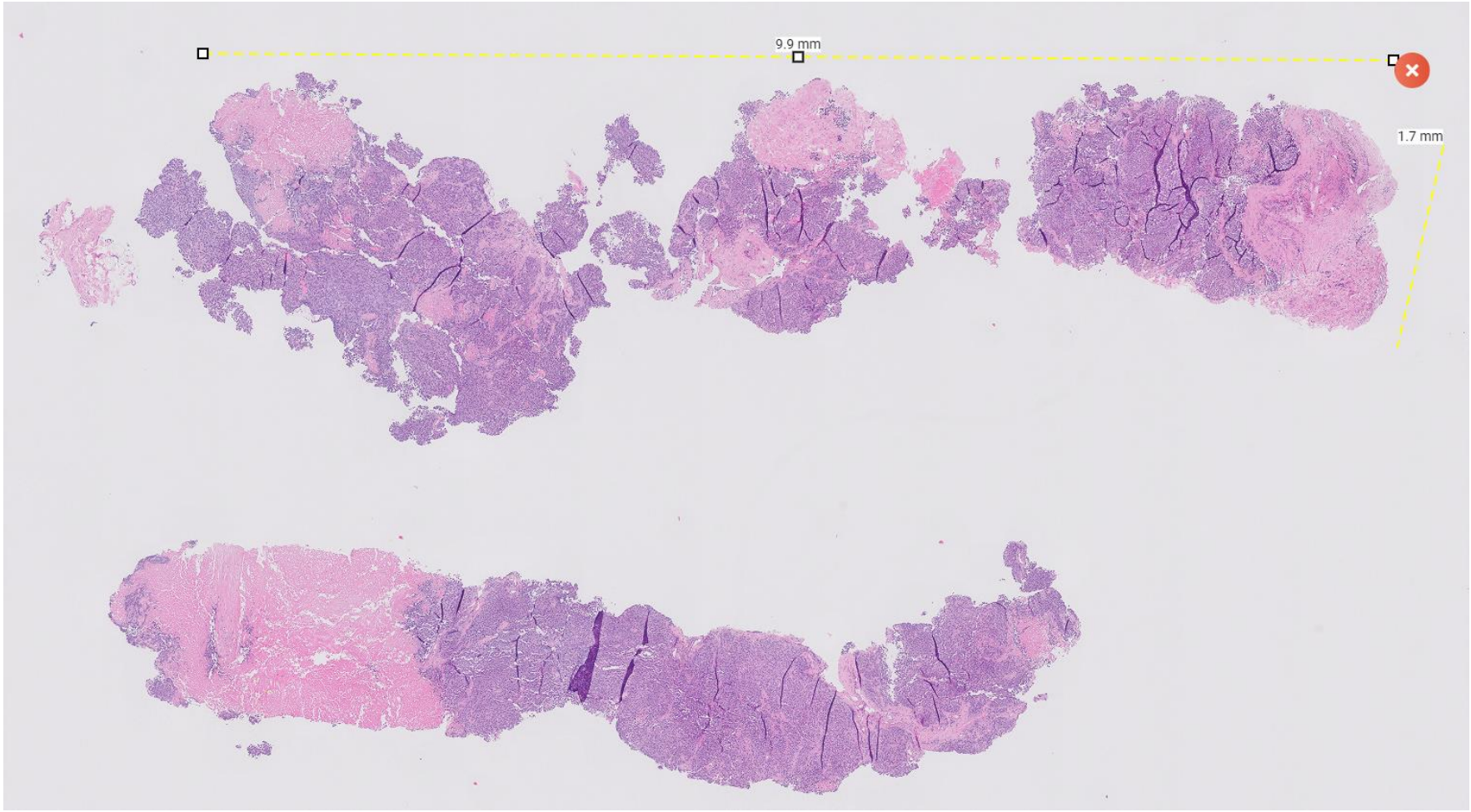


Liquid biopsy

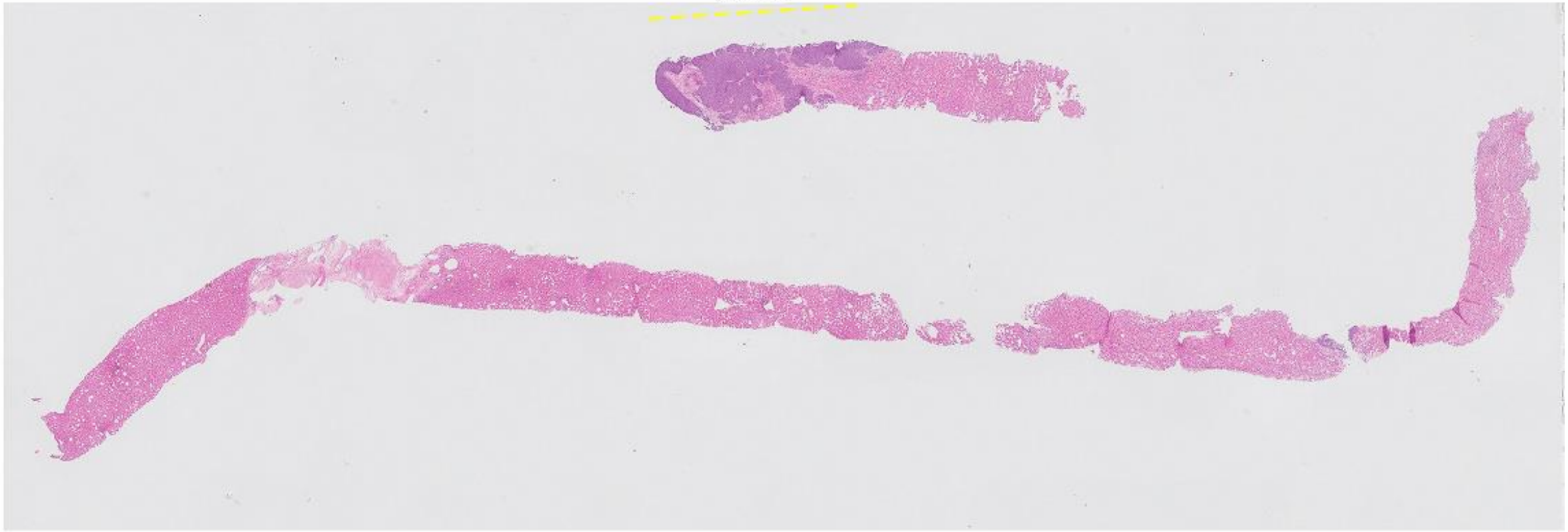


Diagnostiek bij gematastaseerde ziekte

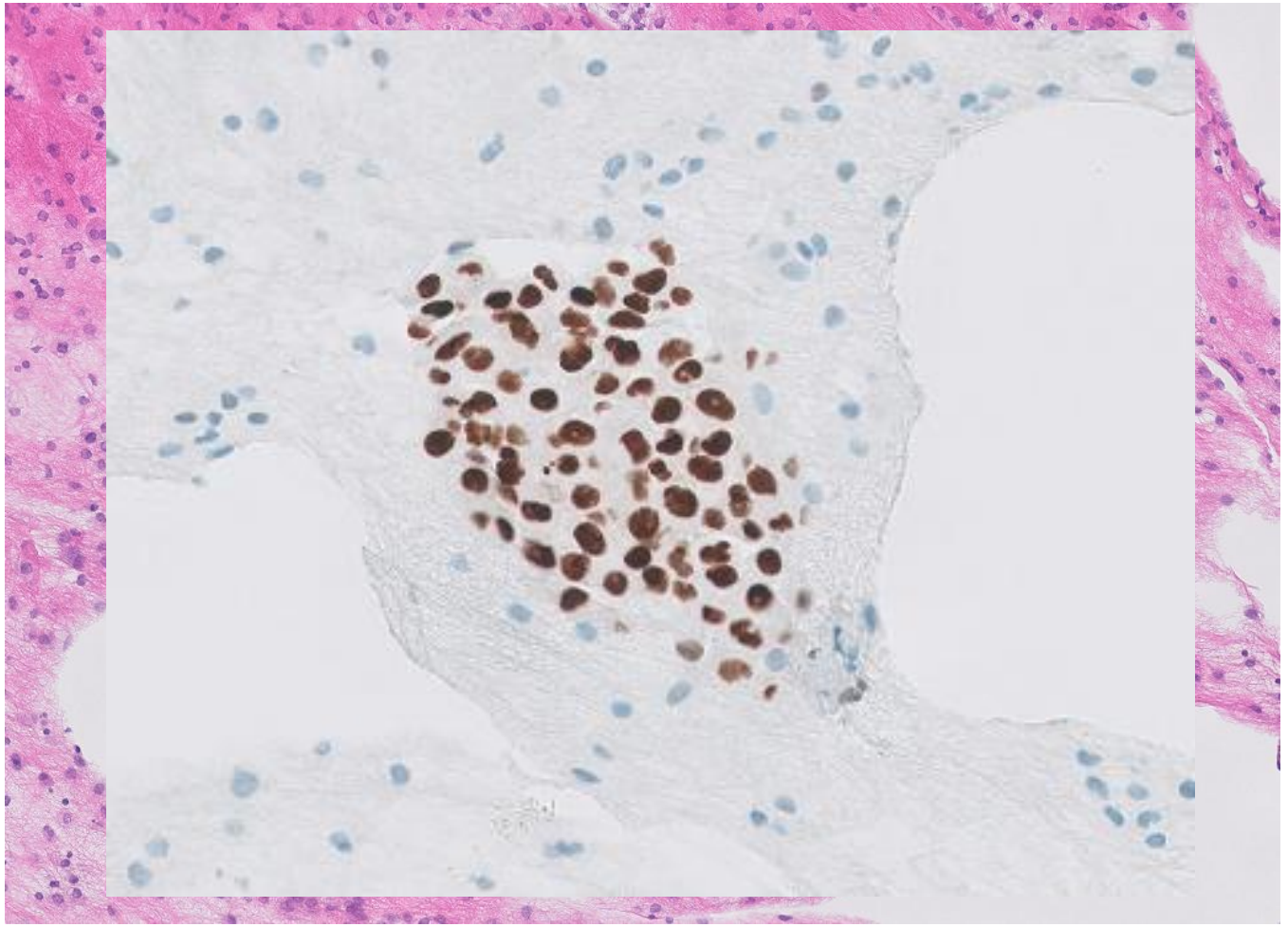
Biopt metastase



Biopt metastase



Punctie metastase



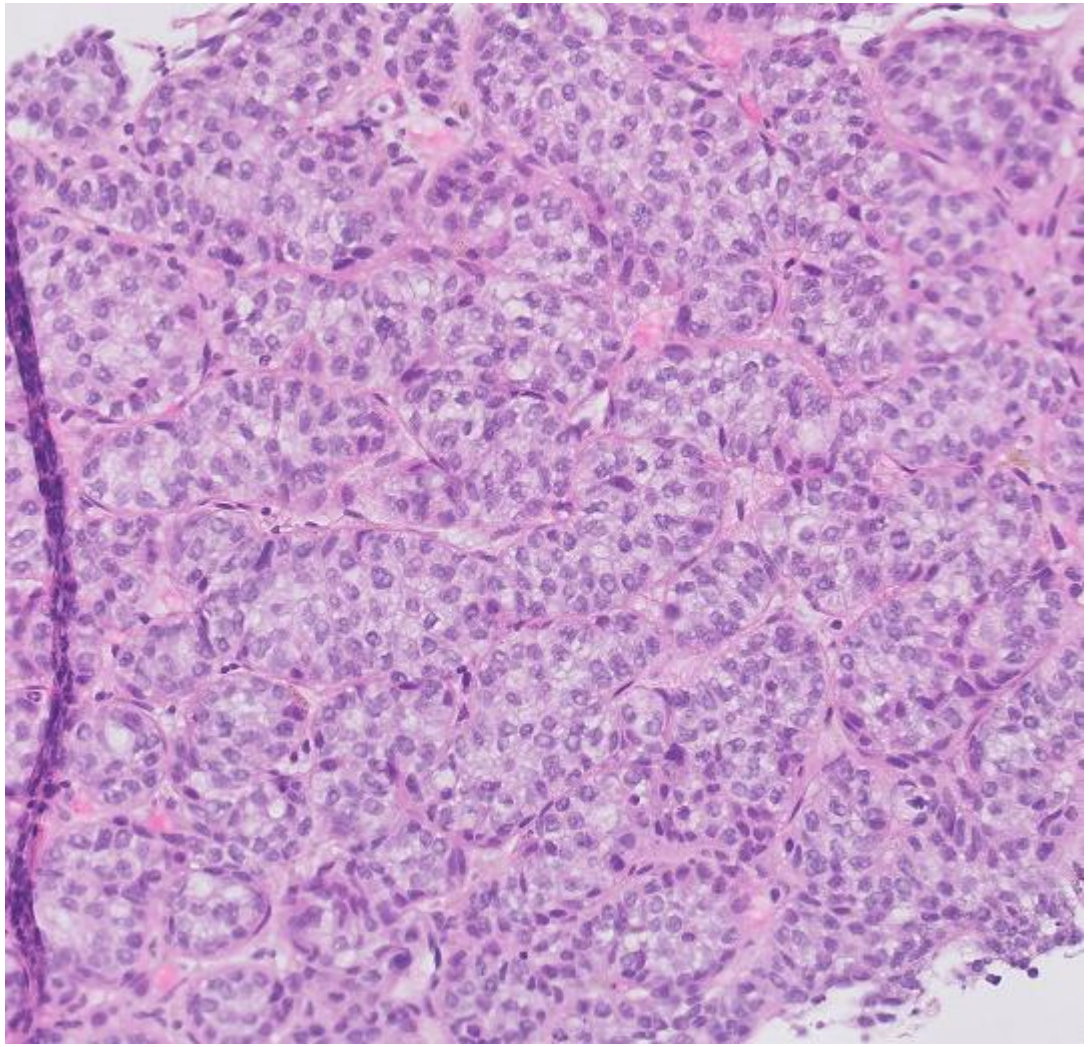
Gemetastaseerd mammacarcinoom

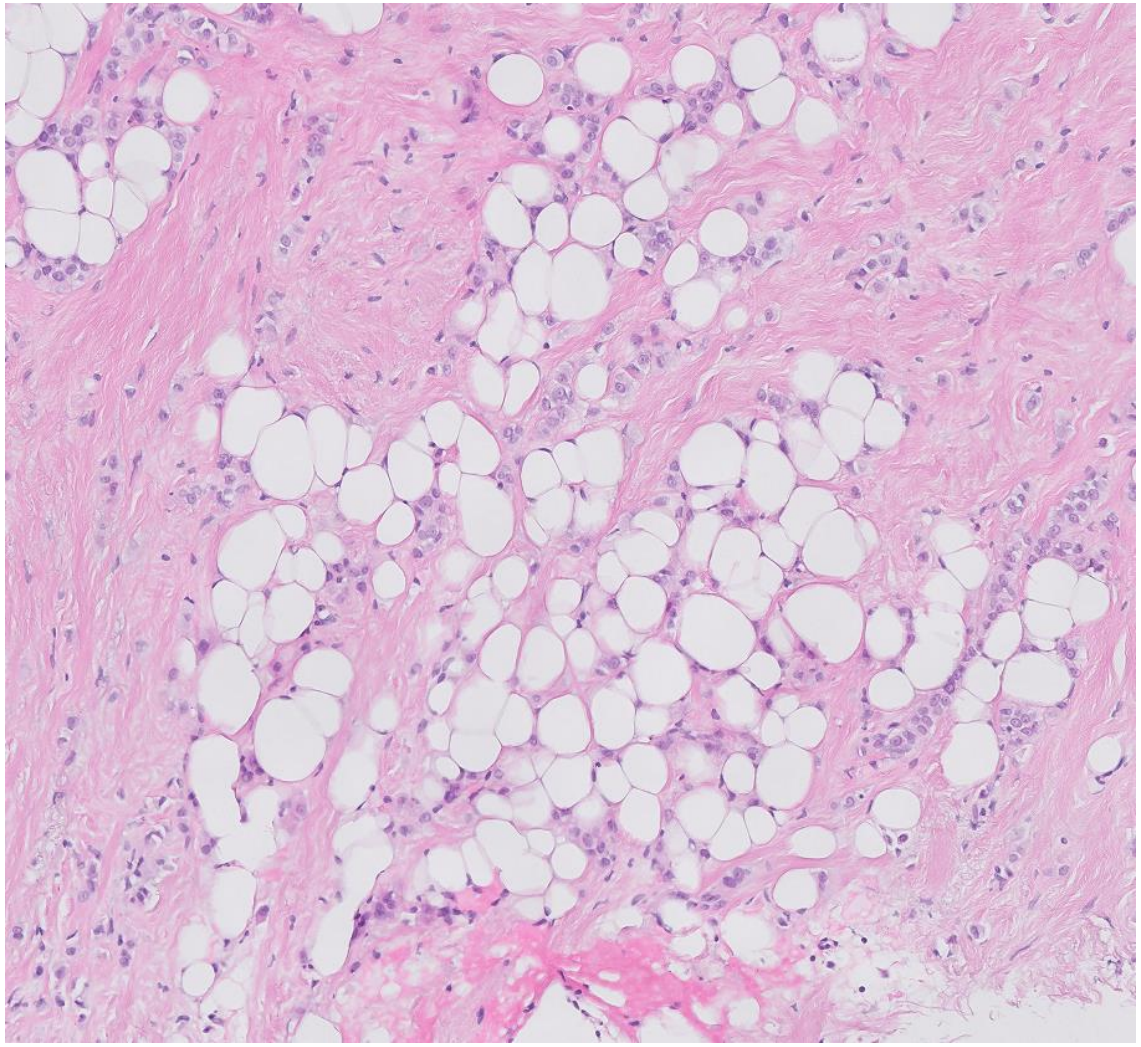
- Type tumor
- ER PR
- HER2
- Mammaprint
- Oncotype
- PD-L1
- PIK3CA
- TILs
- Ki67

Type tumor

- Invasive breast carcinoma of no special type
- Microinvasive carcinoma
- Invasive lobular carcinoma
- Tubular carcinoma
- Cribriform carcinoma
- Mucinous carcinoma
- Mucinous cystadenocarcinoma
- Invasive micropapillary carcinoma
- Carcinoma with apocrine differentiation
- Metaplastic carcinoma
- Acinic cell carcinoma
- Adenoid cystic carcinoma
- Secretory carcinoma
- Mucoepidermoid carcinoma
- Polymorphous adenocarcinoma
- Tall cell carcinoma with reversed polarity
- Neuroendocrine tumour
- Neuroendocrine carcinoma
- Postradiation angiosarcoma of the breast
- Primary angiosarcoma of the breast
- Leiomyosarcoma
- Liposarcoma
- Extranodal marginal zone lymphoma of mucosa-associated lymphoid tissue (MALT lymphoma)
- Follicular lymphoma
- Diffuse large B-cell lymphoma
- Burkitt lymphoma
- Breast implant-associated anaplastic large cell lymphoma
- Malignant adenomyoepithelioma
- Phyllodes tumour

Mammacarcinoom type tumor

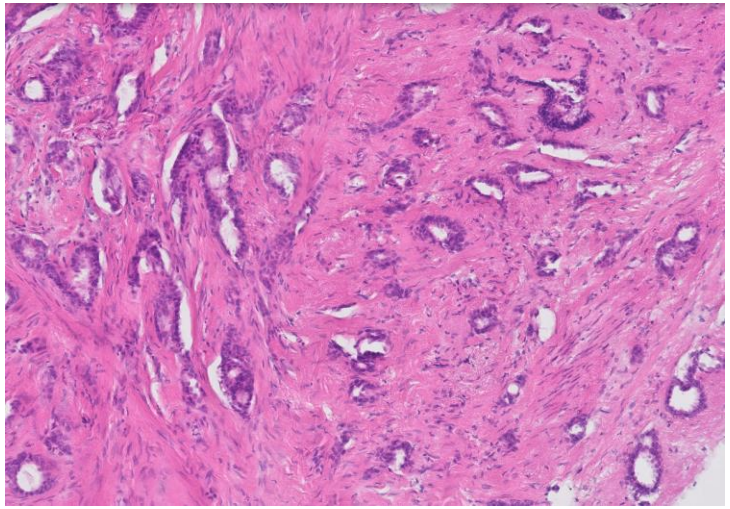


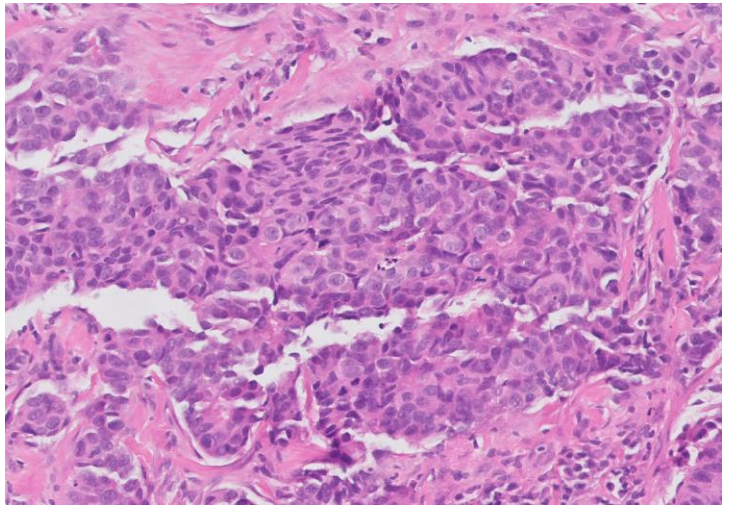


Gradering van de tumor: Bloom and Richardson

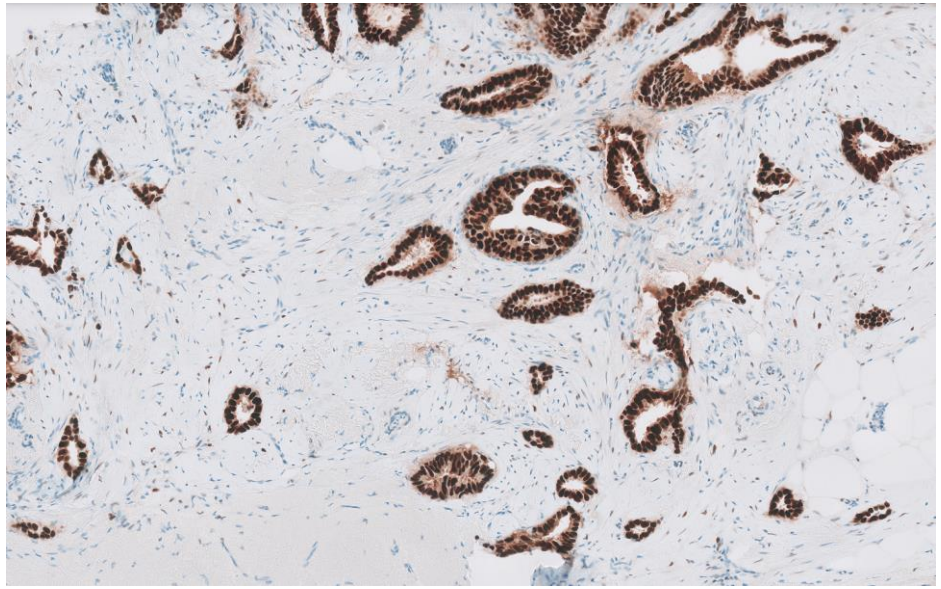
Buisvorming: >75%, 10-75%, <10%
Atypie: score 1, 2 of 3
Mitosen: <7, 8-12, >12

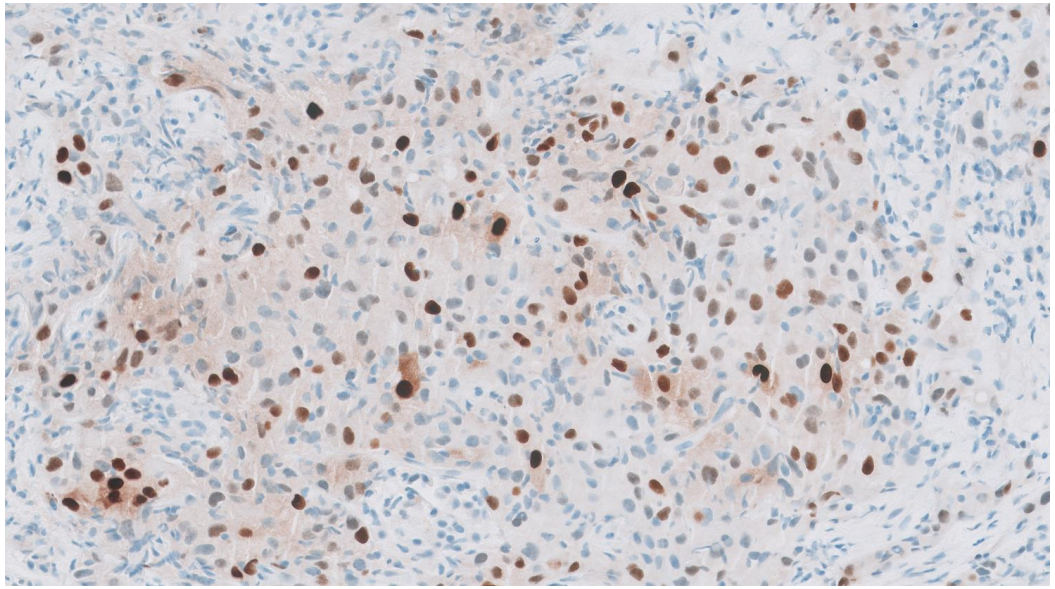
Graad 1: 3-5, Graad 2: 6-7, Graad 3: 8-9





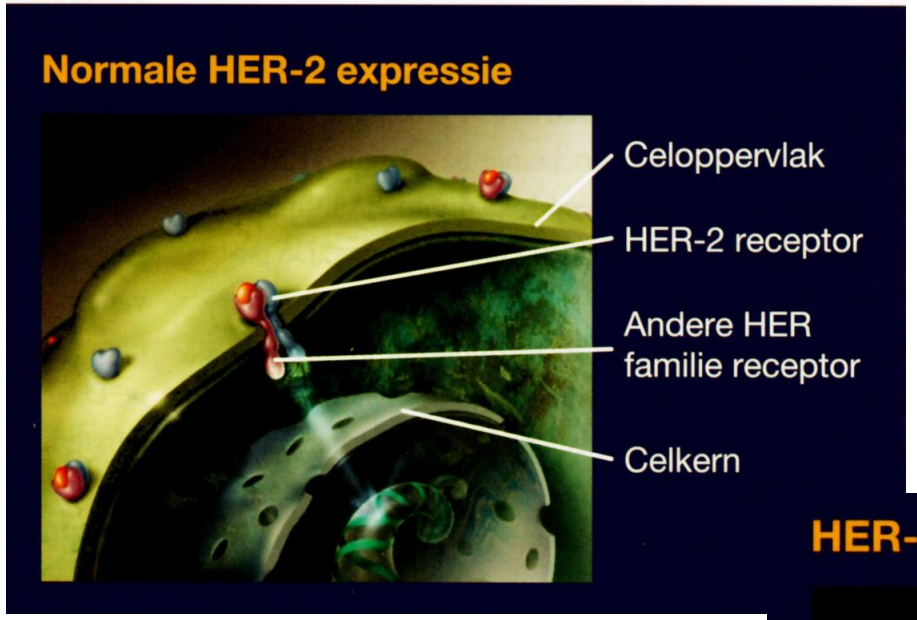
Oestrogeenreceptor en progesteronreceptor

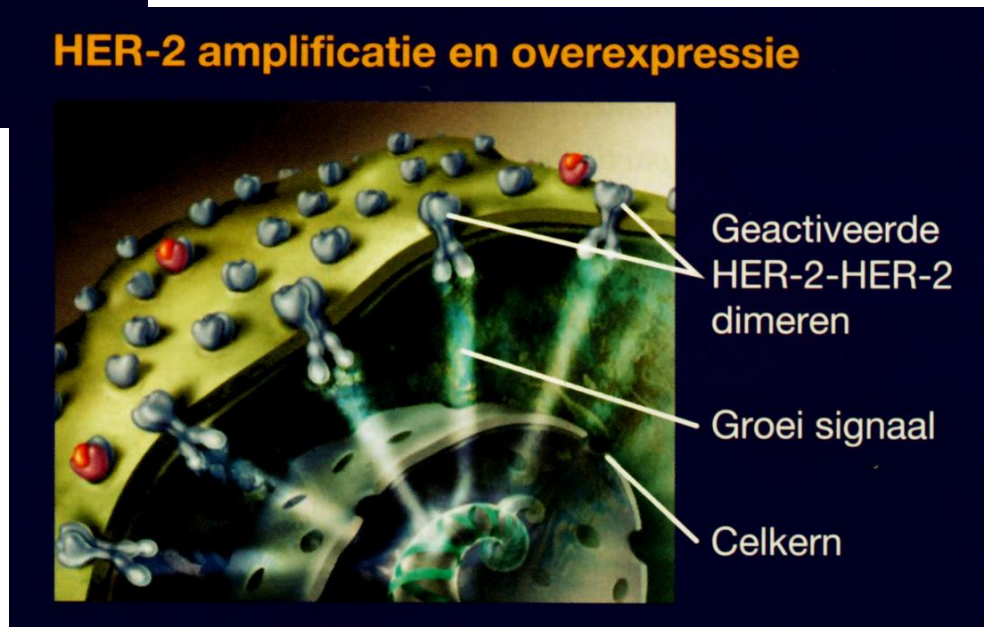




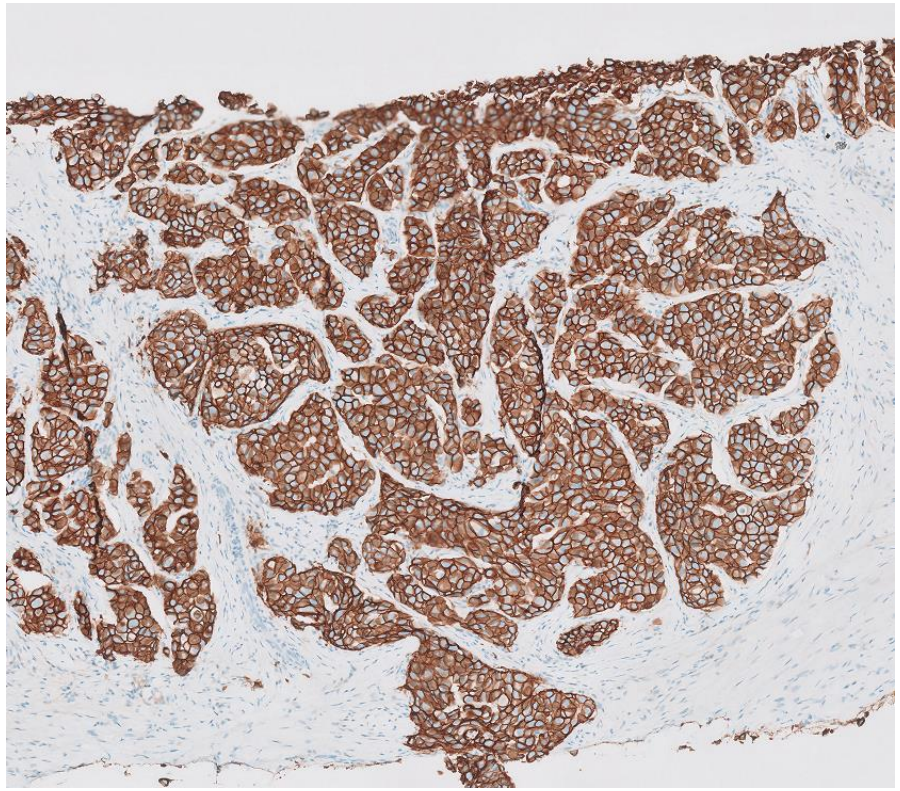
>10% positieve cellen is positief

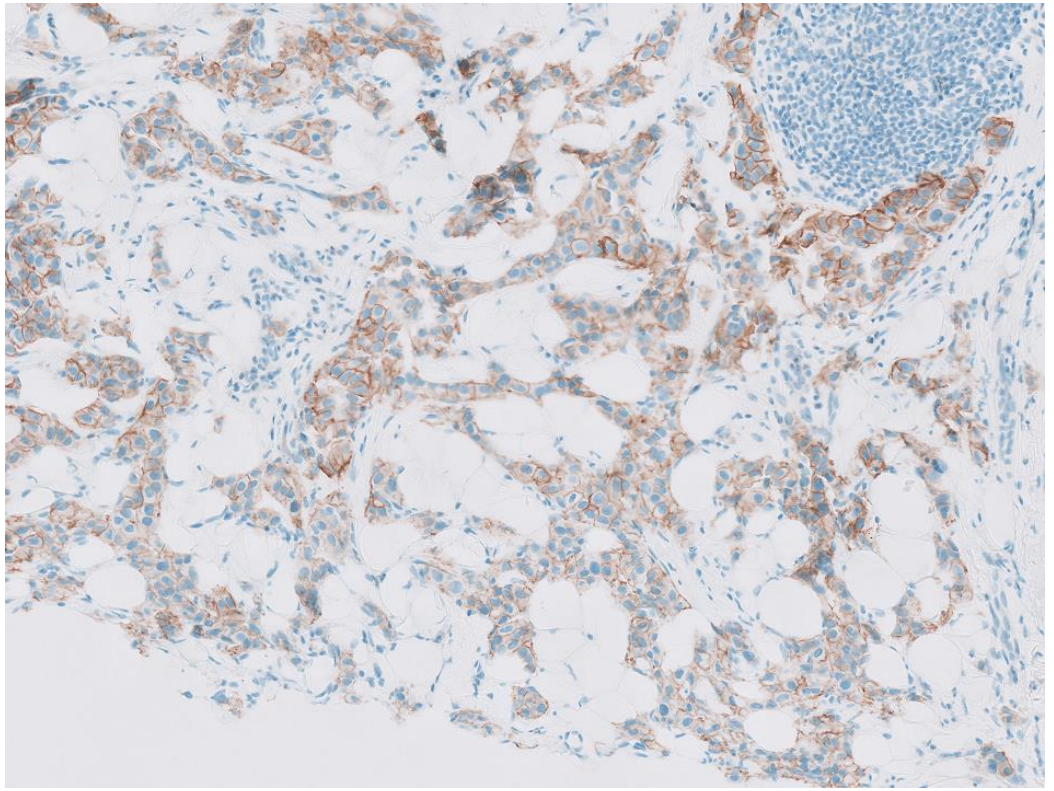
HER2


Normale HER-2 expressie
Celoppervlak
HER-2 receptor
Andere HER familie receptor
Celkern


HER-2 amplificatie en overexpressie
Geactiveerde HER-2-HER-2 dimeren
Groei signaal
Celkern

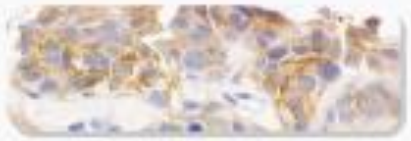
HER2



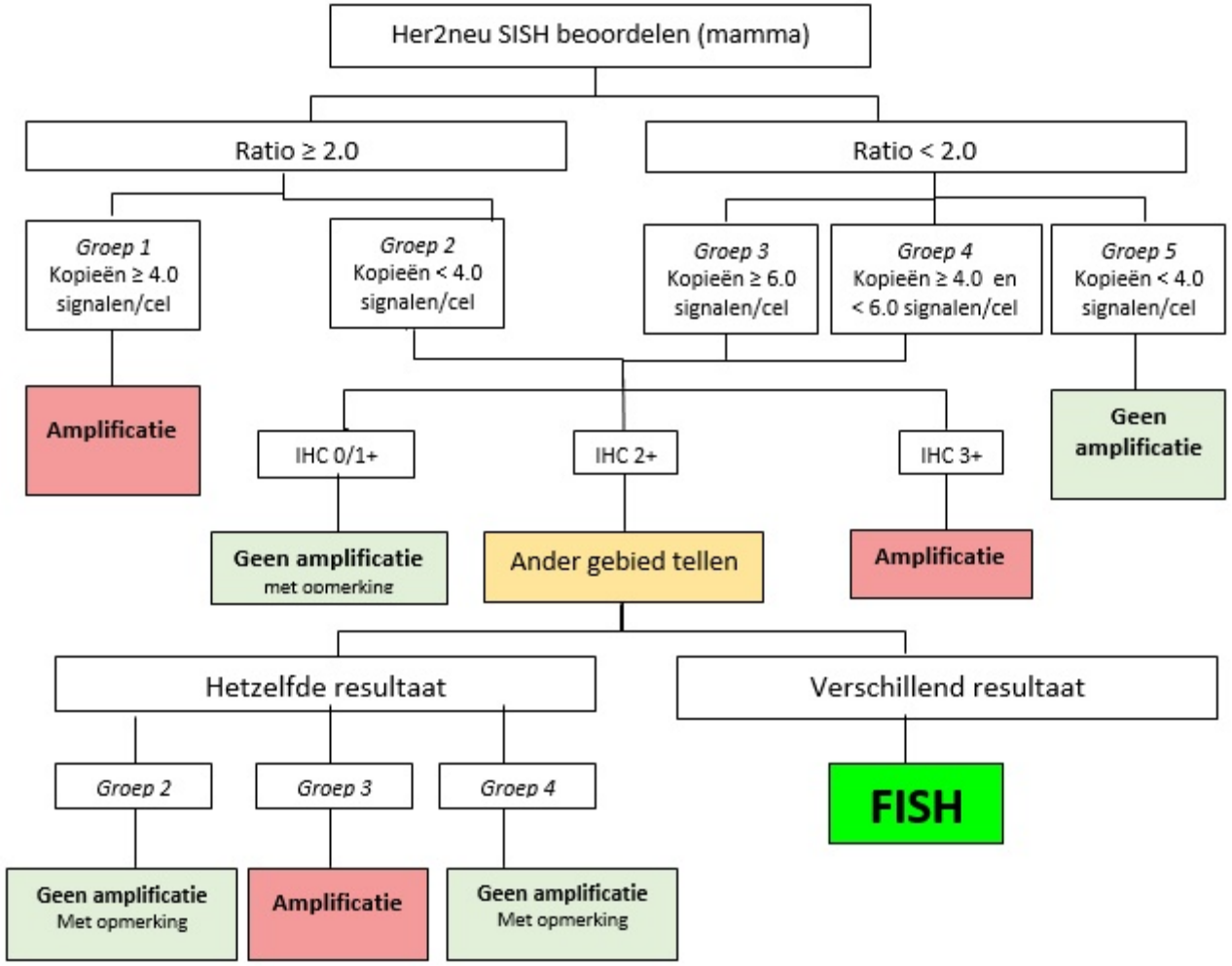


HER2 scoring

Spectrum of HER2 positivity according to ASCO/CAP guidelines

	IHC score	HER2 test intepretation	HER2 status	
	0	No staining or incomplete and faint/barely perceptible membrane staning n ≤10% of tumor cells	Negative	
	1+	Incomplete and faint/barely perceptible membrane staining in >10% of tumor cells	Low	Spectrum of HER2 positivity
	2+	Weak-moderate complete membrane staining in >10% of tumor cells OR intense membrane stainingi n ≤10% of tumor cells	NO ISH amplification? YES	
	3+	Complete and intense membrane staining in >10% of tumor cells	Positive	

SISH


Her2neu SISH beoordelen (mamma)
Ratio ≥ 2.0
Ratio < 2.0
Groep 1
Kopieën ≥ 4.0
signalen/cel
Groep 2
Kopieën < 4.0
signalen/cel
Groep 3
Kopieën ≥ 6.0
signalen/cel
Groep 4
Kopieën ≥ 4.0 en
< 6.0 signalen/cel
Groep 5
Kopieën < 4.0
signalen/cel
Amplificatie
IHC 0/1+
IHC 2+
IHC 3+
Geen amplificatie
Geen amplificatie
met opmerking
Ander gebied tellen
Amplificatie
Hetzelfde resultaat
Verschillend resultaat
Groep 2
Groep 3
Groep 4
FISH
Geen amplificatie
Met opmerking
Amplificatie
Geen amplificatie
Met opmerking

Mammaprint

- >50 jaar
- Klinisch hoog risico

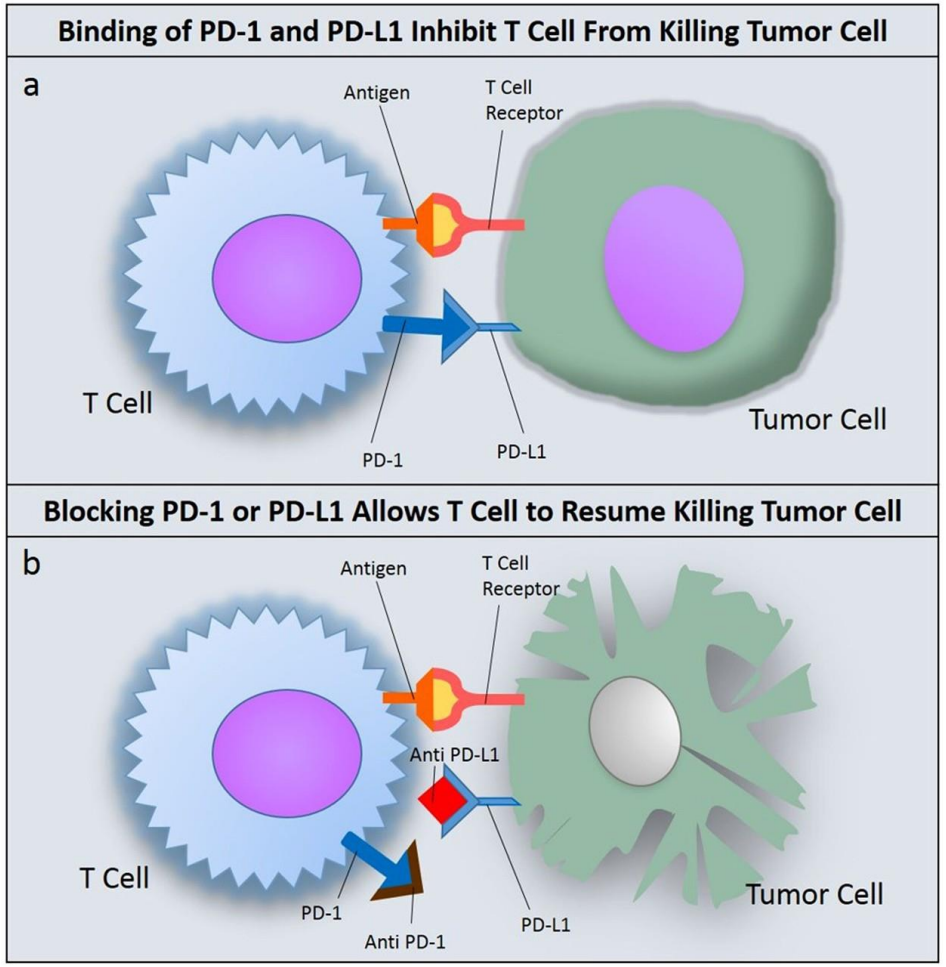
- Moleculaire test
- Mammaprint: high risk of low risk

Oncotype

- Vroeg stadium borstkanker
- Max 3 positieve lymfklieren
- Hormoongevoelige tumor
- HER2 negatief

- Moleculaire test
- Score tussen de 0 en 100

PD-L1



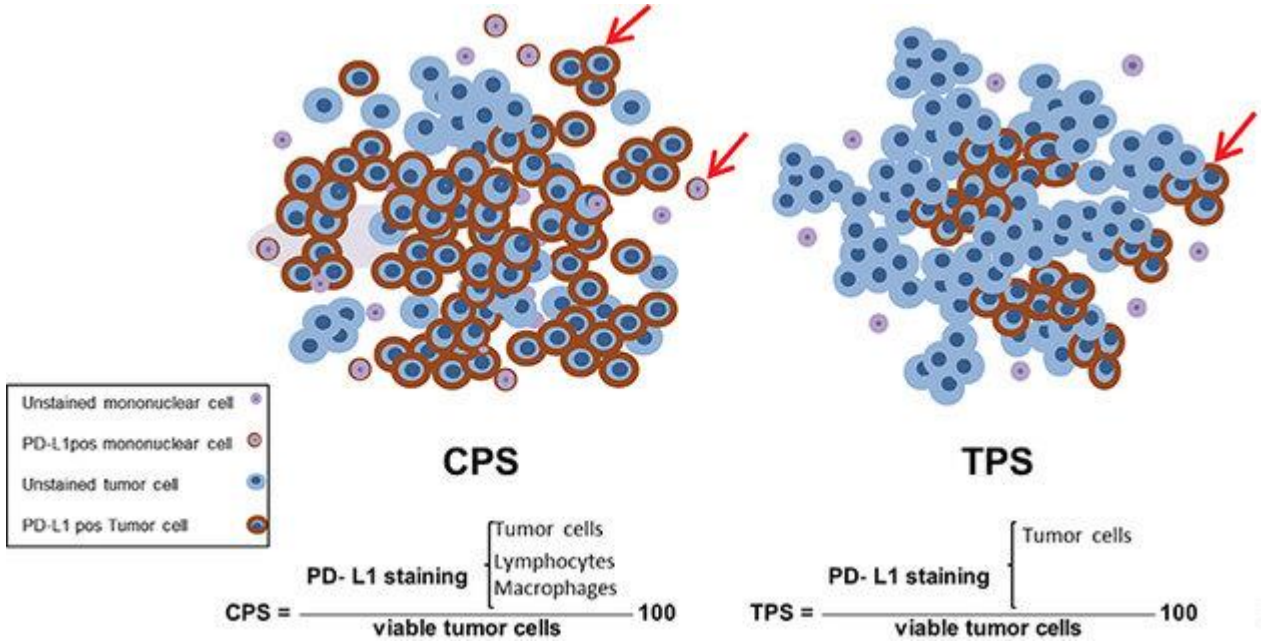
Caldwell, C., Johnson, C.E., Balaji, V.N. *et al.* Identification and Validation of a PD-L1 Binding Peptide for Determination of PDL1 Expression in Tumors. *Sci Rep* **7**, 13682 (2017).

PD-L1 scoring

- Tumor positive score (TPS): totaal percentage positieve tumorcellen

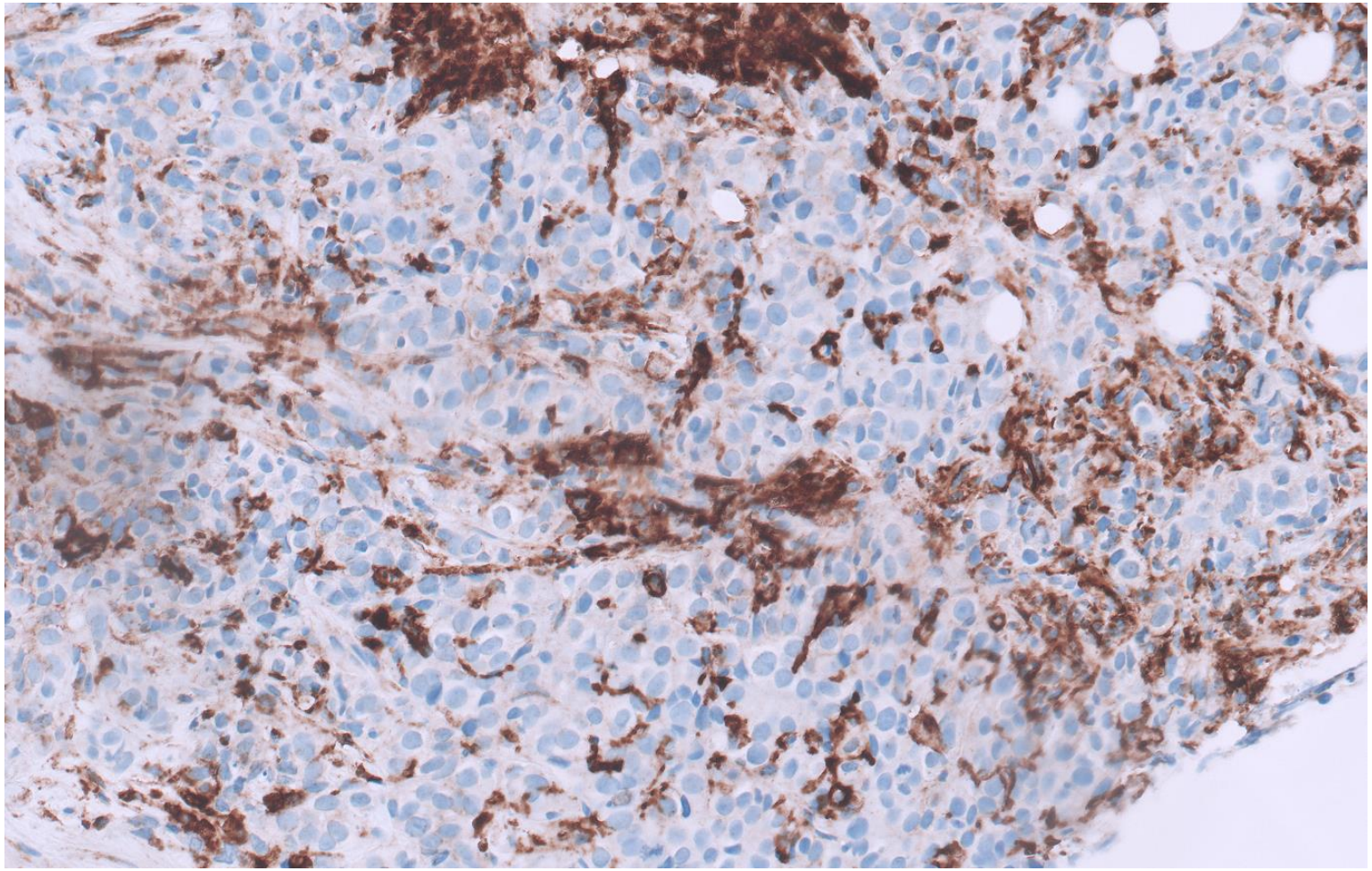
- Combined positive score (CPS):

$$CPS = \frac{\text{PD-L1 positively stained cells (tumor cells, lymphocytes, macrophages)}}{\text{all vital tumor cells}} \times 100$$



https://www.researchgate.net/figure/Combined-positive-score-CPS-of-PD-L1-expression-defined-by-total-number-of-stained_fig1_369336413 [accessed 14 Sept 2024]

PD-L1

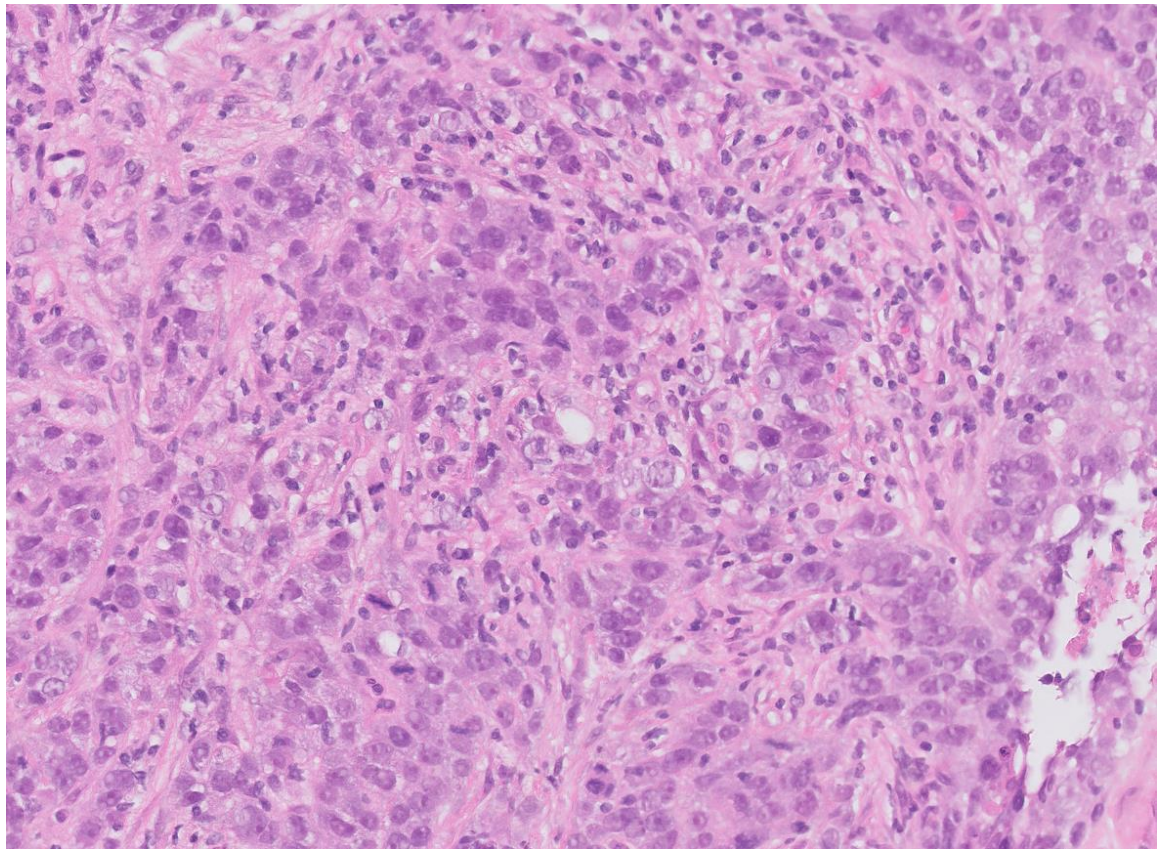


PIK3CA

- NGS onderzoek voor mutatie
- In geval van mutatie kan behandeling gegeven worden met Alpelisib

Tumor infiltrating lymphocytes

- Op zoek naar prognostische markers in triple negatief mamma carcinoom
- OPTImal studie



Home - International TILS Working Group (tilsinbreastcancer.org)

Ki67

- Hogere proliferatie is agressievere tumor
- Wat is een cut-off waarde voor hoge proliferatie?
- Studies lopen nog om te bepalen wat de waarde is

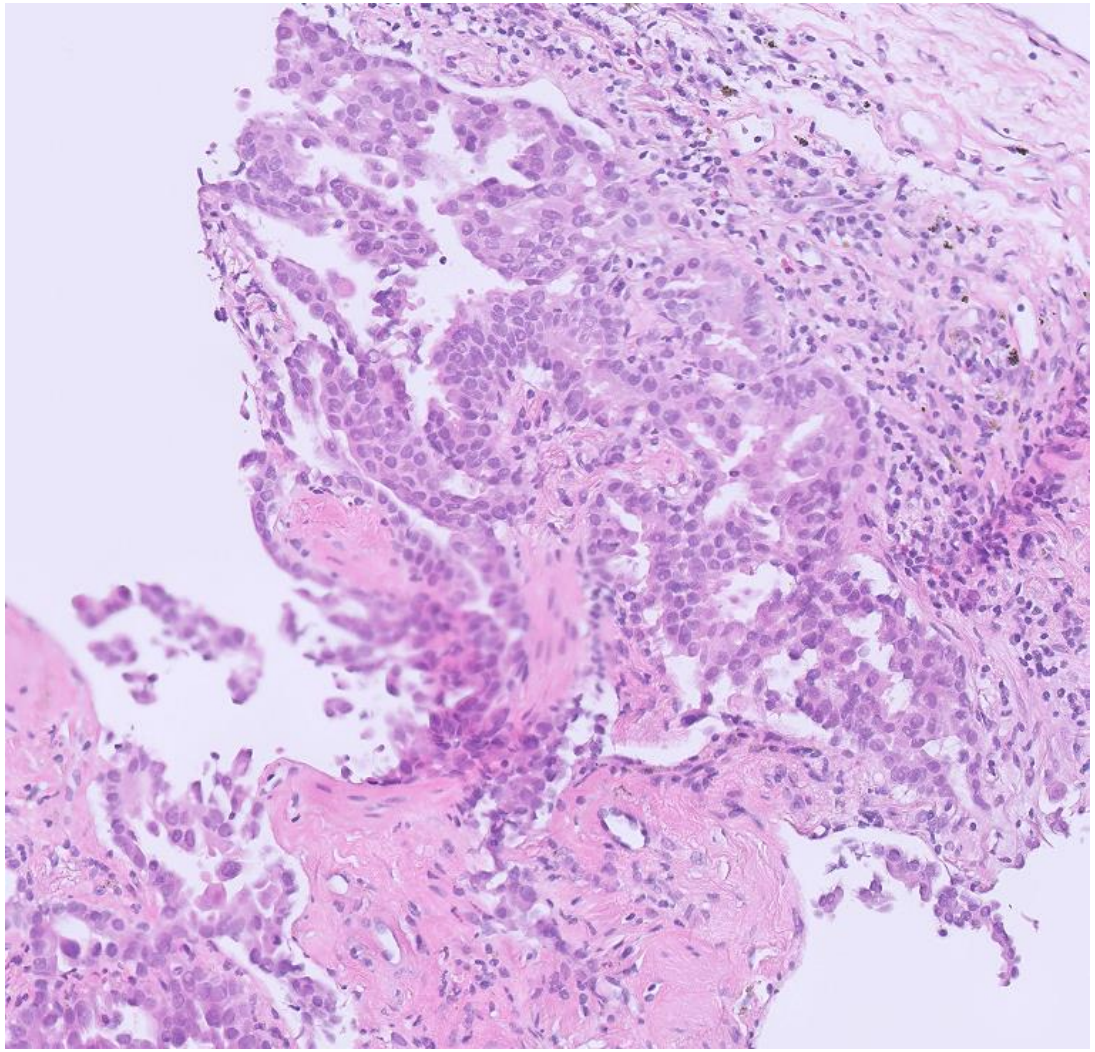
Gemetastaseerd longcarcinoom

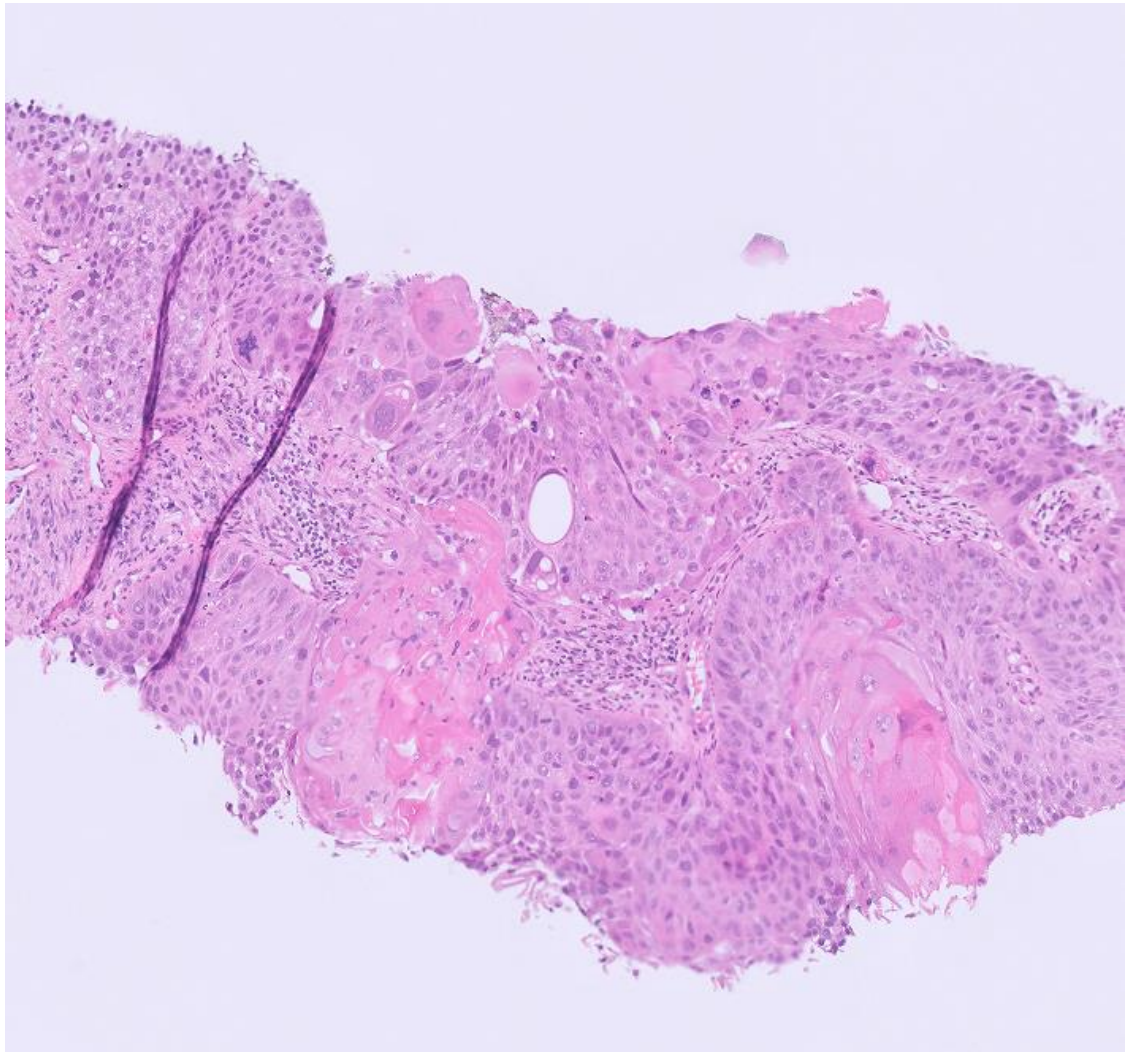
- Type tumor
- PD-L1
- NGS

Type tumor

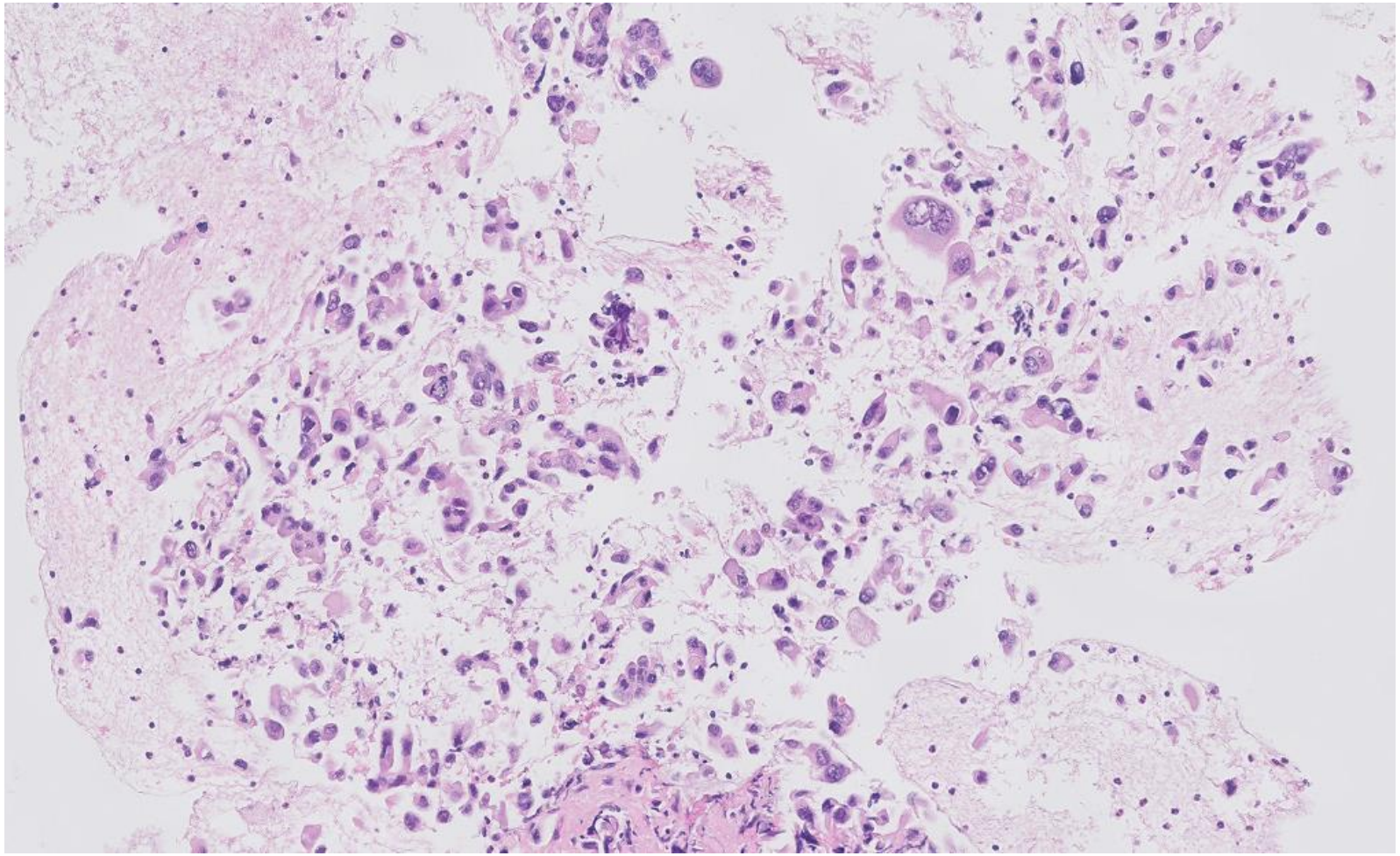
Minimally invasive adenocarcinoma of the lung
Invasive non-mucinous adenocarcinoma of the lung
Invasive mucinous adenocarcinoma of the lung
Colloid adenocarcinoma of the lung
Fetal adenocarcinoma of the lung
Enteric-type adenocarcinoma of the lung
Squamous cell carcinoma of the lung
Lymphoepithelial carcinoma of the lung
Large cell carcinoma of the lung
Adenosquamous carcinoma of the lung
Pleomorphic carcinoma of the lung
Pulmonary blastoma
Carcinosarcoma of the lung
NUT carcinoma of the lung (see NUT carcinoma of the thorax)
Thoracic SMARCA4-deficient undifferentiated tumour
Adenoid cystic carcinoma of the lung
Epithelial-myoepithelial carcinoma of the lung
Mucoepidermoid carcinoma of the lung
Hyalinizing clear cell carcinoma of the lung
Myoepithelioma and myoepithelial carcinoma of the lung
Small cell lung carcinoma
Large cell neuroendocrine carcinoma of the lung
Melanoma of the lung
Meningioma of the lung
Pulmonary artery intimal sarcoma
Congenital peribronchial myofibroblastic tumour
Primary pulmonary myxoid sarcoma with EWSR1-CREB1 fusion
MALT lymphoma of the lung
Pulmonary diffuse large B-cell lymphoma
Lymphomatoid granulomatosis of the lung
Intravascular large B-cell lymphoma of the lung
Localized pleural mesothelioma
Diffuse pleural mesothelioma
Primary effusion lymphoma
Diffuse large B-cell lymphoma associated with chronic inflammation of the pleura

Adenocarcinoom vs plaveiselcelcarcinoom

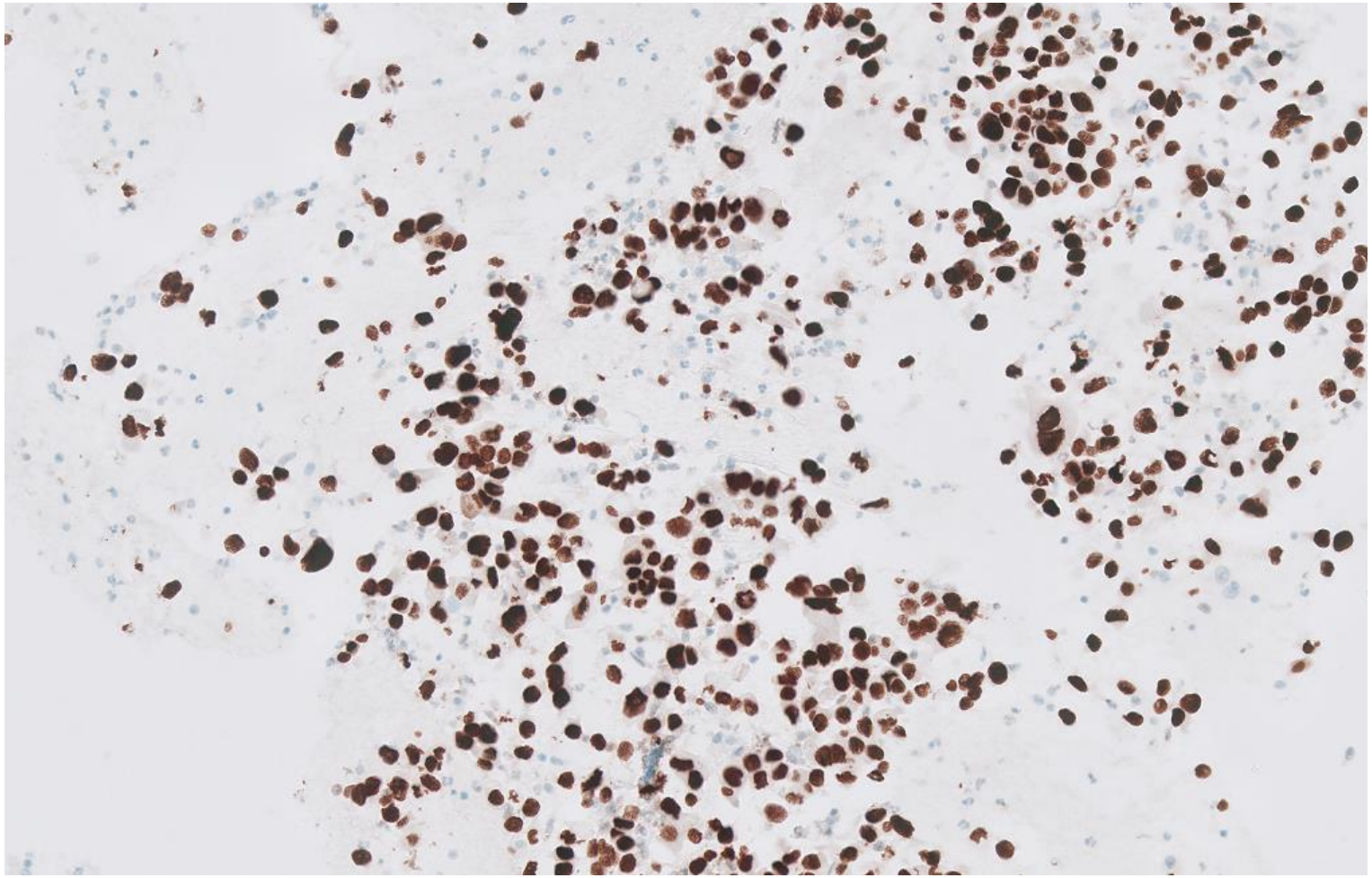




Cytologie pleuravocht



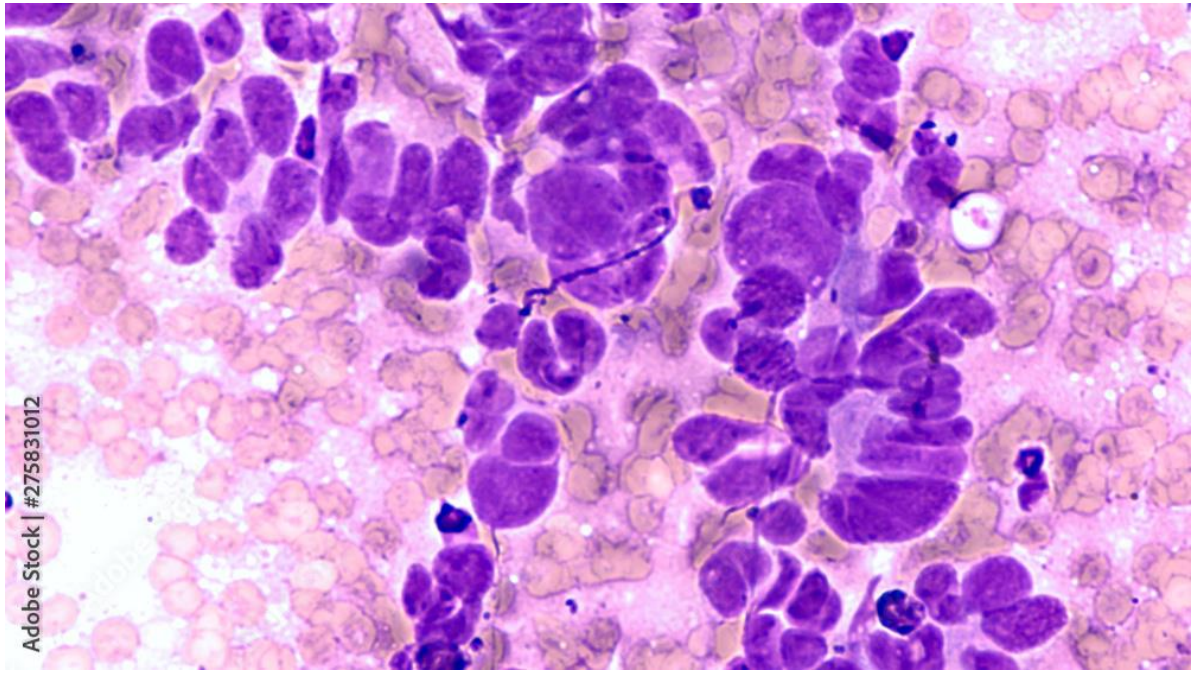
TTF1/p40

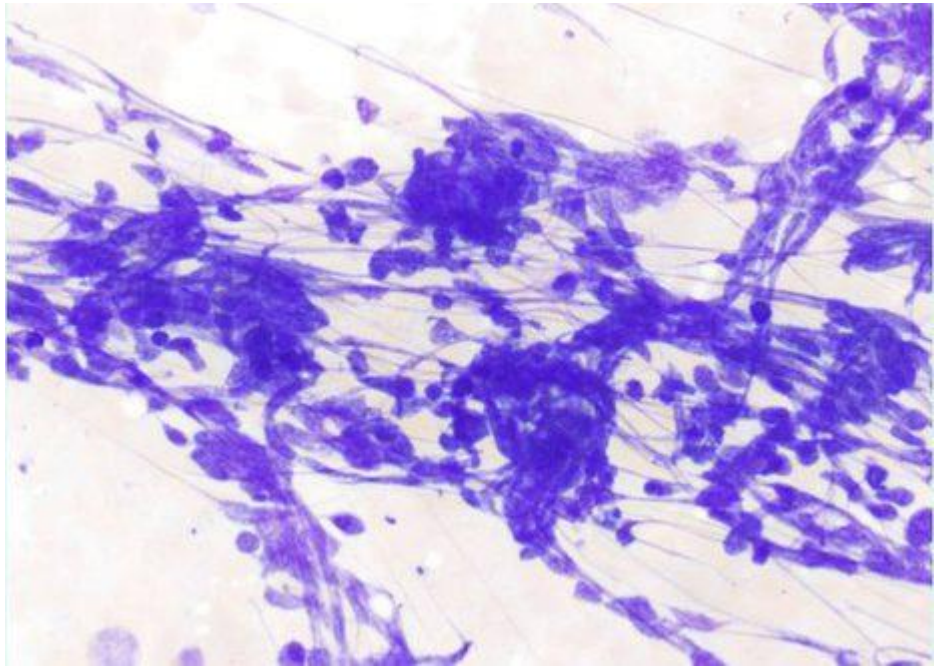


Neuro-endocriene laesies

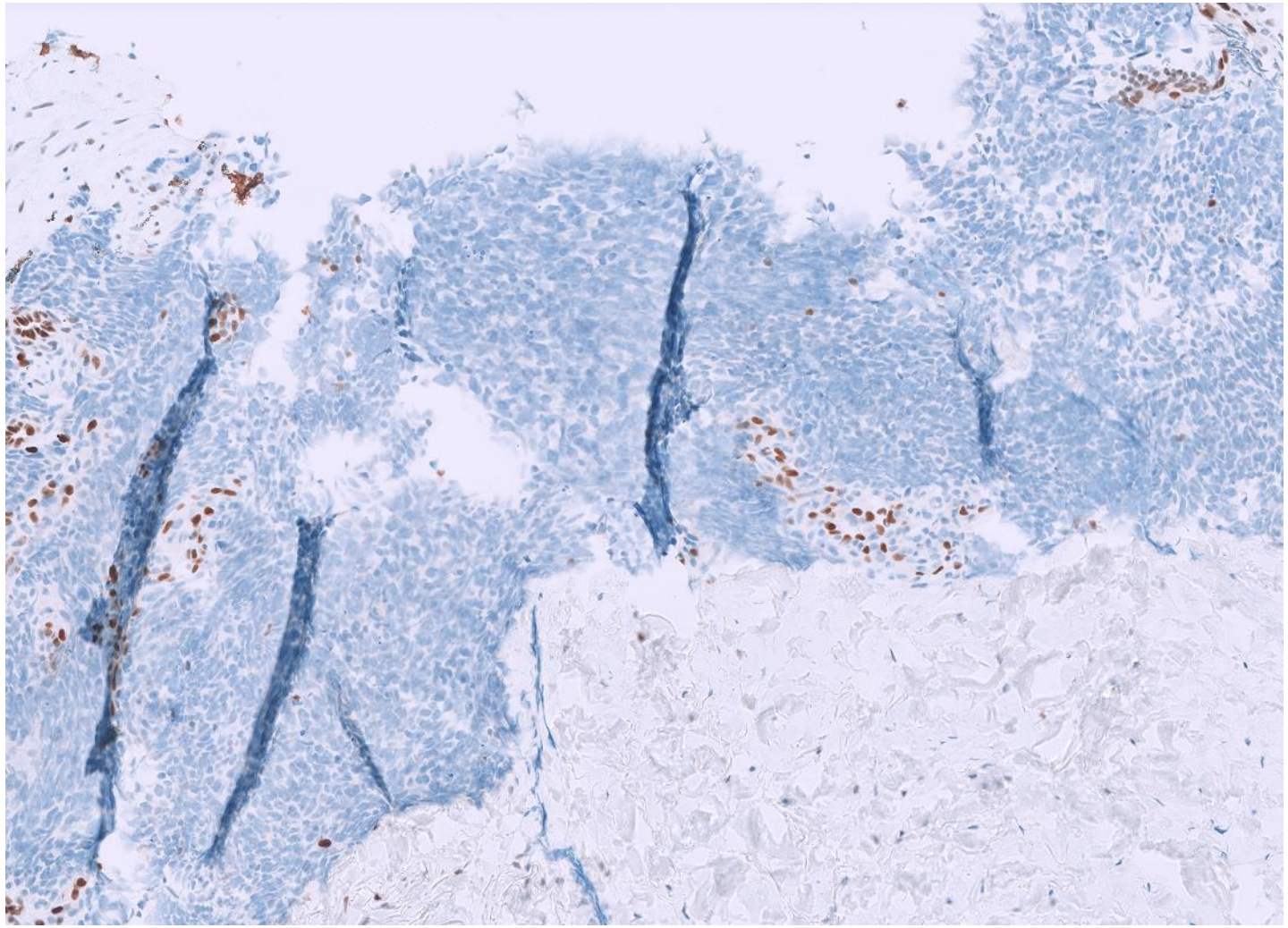
- Carcinoïd
- Atypisch carcinoïd
- Kleincellig carcinoom
- Grootcellig neur-endocrien carcinoom
  - Met of zonder verlies van RB1

Cytologie kleincellig carcinoom





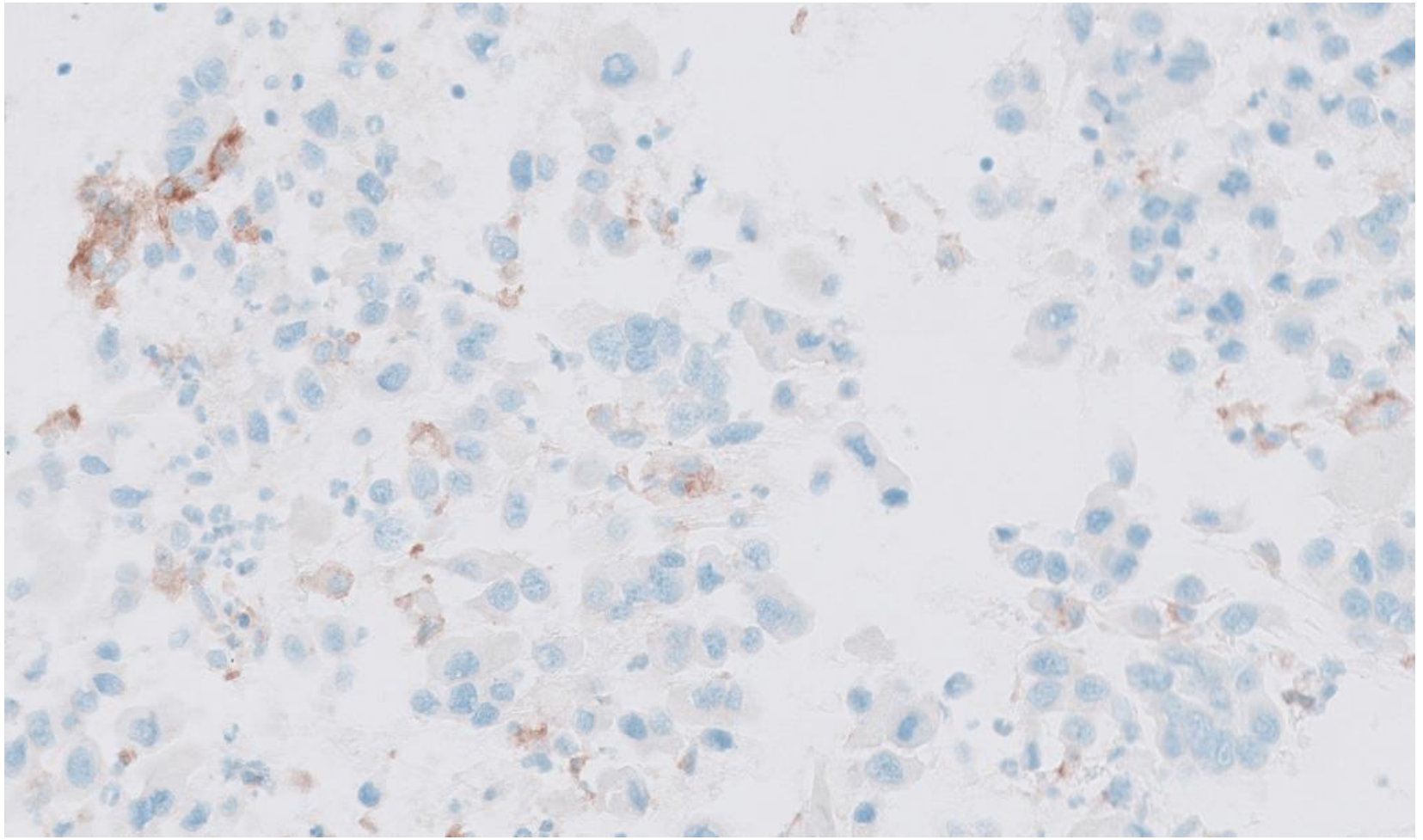
histologie



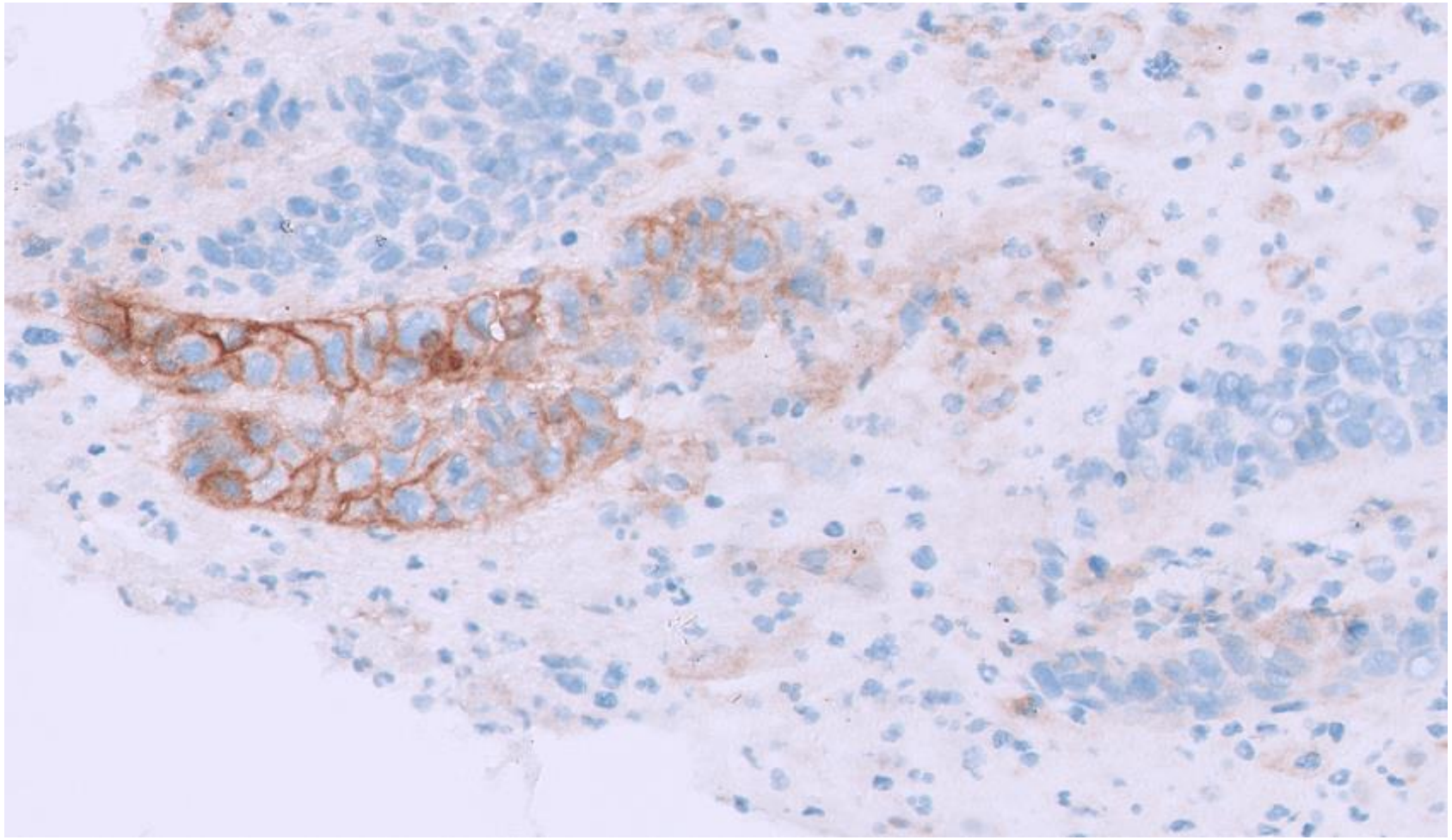
Moleculair onderzoek

- Afhankelijk van subtype tumor en stadiering

PD-L1



PD-L1



NGS

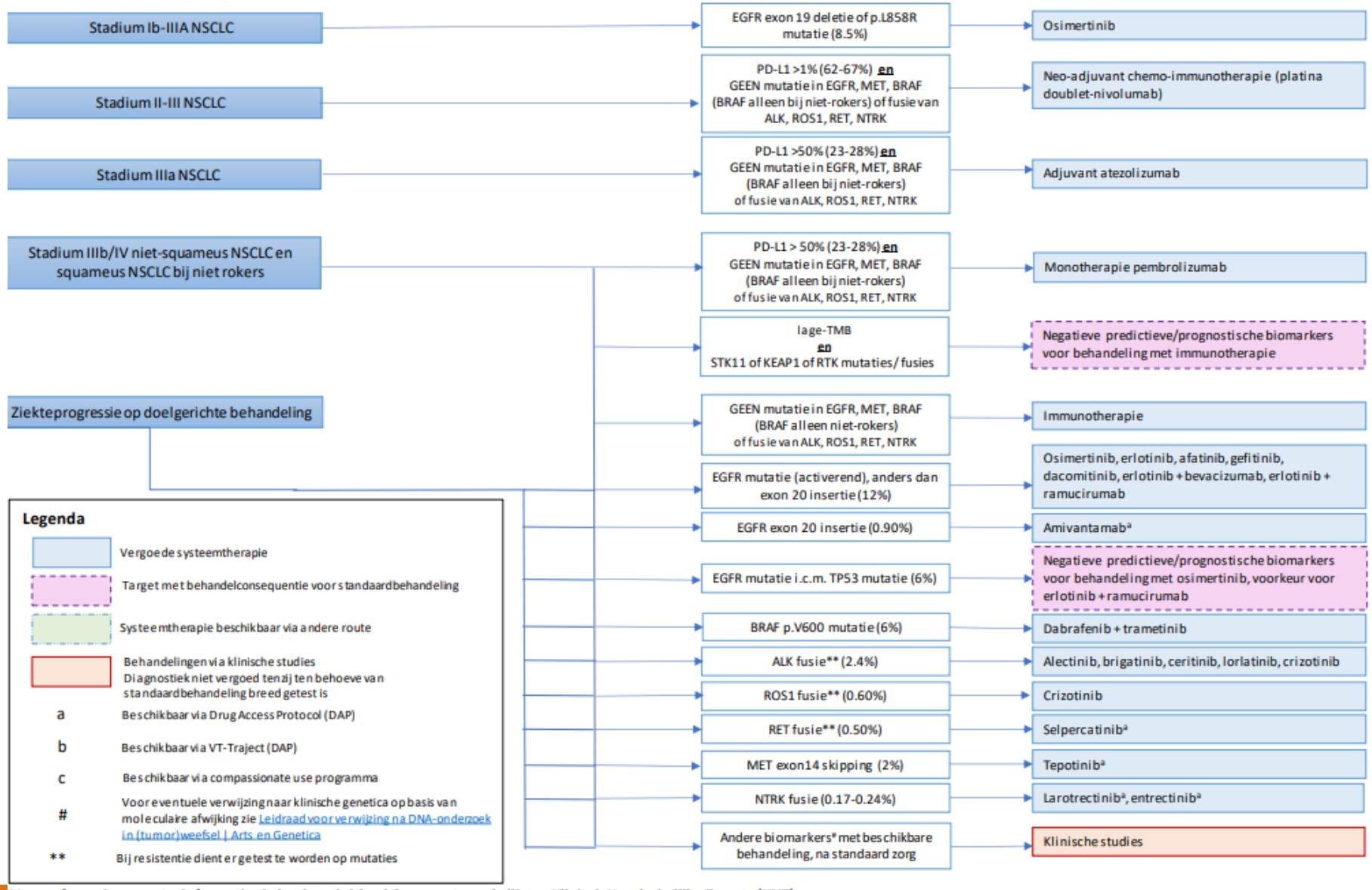
- Voor aanvullende therapie
- Bij niet kleincellig longcarcinoom
- Pre-operatief/postoperatief

- Roker/niet-roker → belangrijk voor inzetten diagnostiek
  - Roker 40% KRAS mutatie
  - Niet roker, direct inzetten RNA onderzoek: >80% vinden we een mutatie

Lijst klinisch noodzakelijke targets

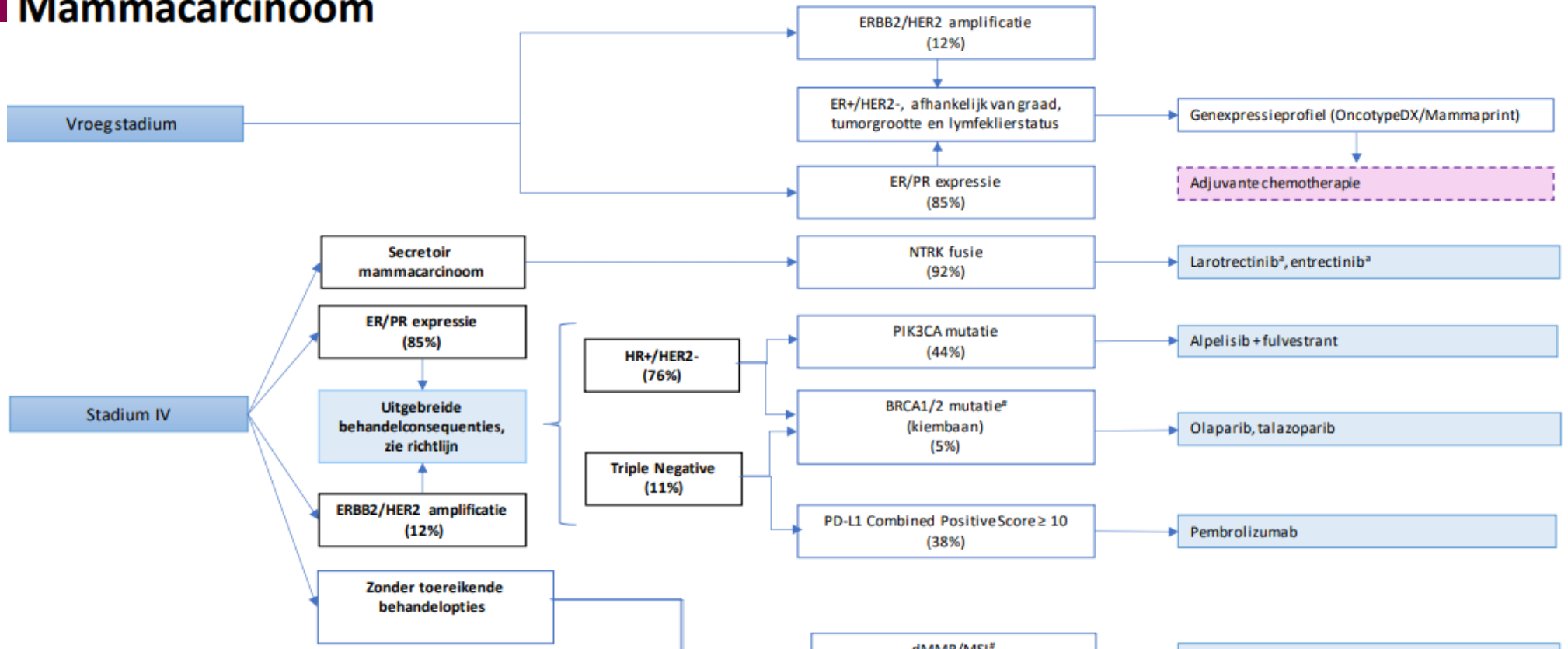
- KNT-lijsten – NVMO
- Landelijke uniformiteit in moleculaire diagnostiek
- Begeleidende uitleg over behandelmogelijkheden

Niet-kleincellig longcarcinoom (NSCLC)



Voor referenties en extra informatie zie het begeleidend document van de lijsten Klinisch Noodzakelijke Targets (KNT).
De meest recente versie is te vinden op https://www.nvalt.nl/vereniging/belangrijke-documenten

Mammacarcinoom



Legenda

- Vergoede systeemtherapie
- Target met behandelconsequentie voor standaardbehandeling
- Systeemtherapie beschikbaar via andere route
- Behandelingen via klinische studies
  Diagnostiek niet vergoed tenzij ten behoeve van standaardbehandeling breed getest is
- a: Beschikbaar via Drug Access Protocol (DAP)
- b: Beschikbaar via VT-Traject (DAP)
- c: Beschikbaar via compassionate use programma
- #: Voor eventuele verwijzing naar klinische genetica op basis van moleculaire afwijking zie Leidraad voor verwijzing na DNA-onderzoek in (tumor)weefsel | Arts en Genetica

Voor referenties en extra informatie zie het begeleidend document van de lijsten Klinisch Noodzakelijke Targets (KNT).