Neue Entwicklungen in histologischer und molekularer Pathologie des Mammakarzinoms

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CHARITÉ

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Neue Entwicklungen in der Pathologie des Mammakarzinoms

Nach Dignitätsdiagnose besteht das zentrales Ziel in der möglichst präzisen Vorhersage des **biologischen Verhaltens** eines individuellen Tumors, d.h.

- Prognose und
- Therapieresponse → TAM, CtX, zielgerichtete Drugs (Herceptin, PARP-Inhibitoren, Check-Point Inhibitoren)

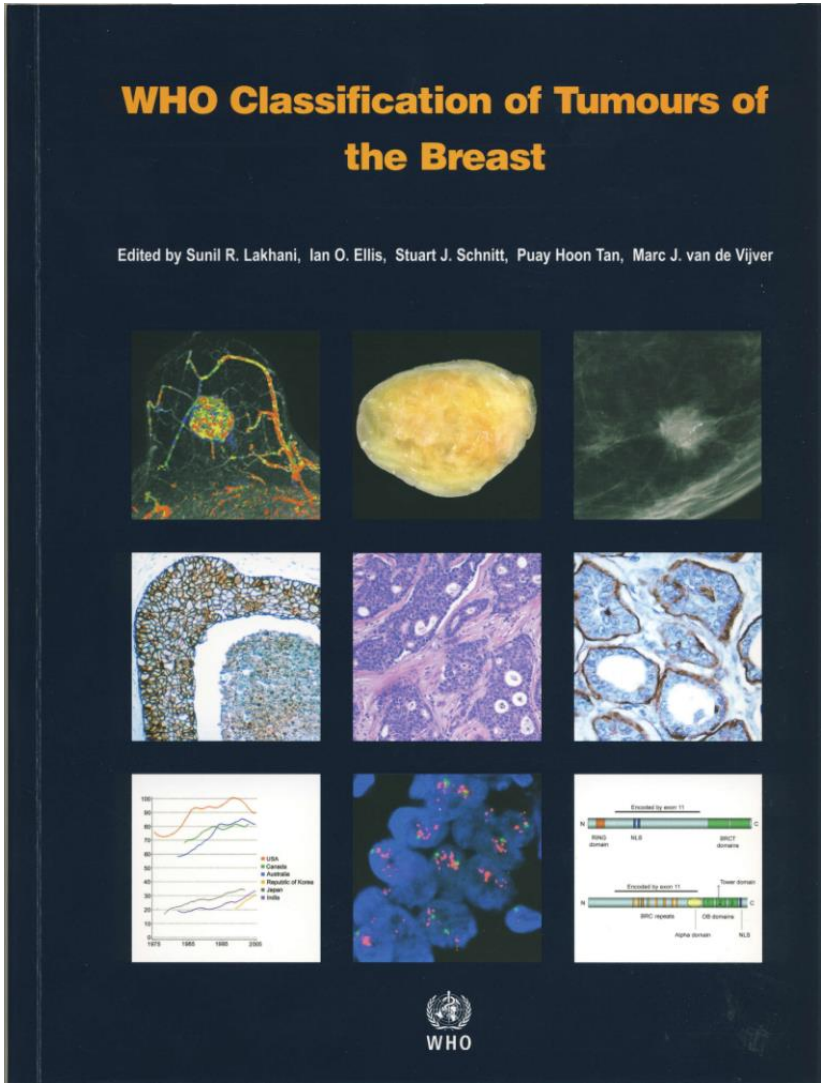
Voraussetzung dafür sind die Bestimmung

- des Tumortyps (konventionell, IHC, molekular)
- von Zielmolekülen (ER, PR, HER2, PD1/PD-L1, etc.)
- von prognostisch/prädiktiven Charakteristika (Genprofile, BRCA1/2, TILs)

Technische Hilfsmittel sind

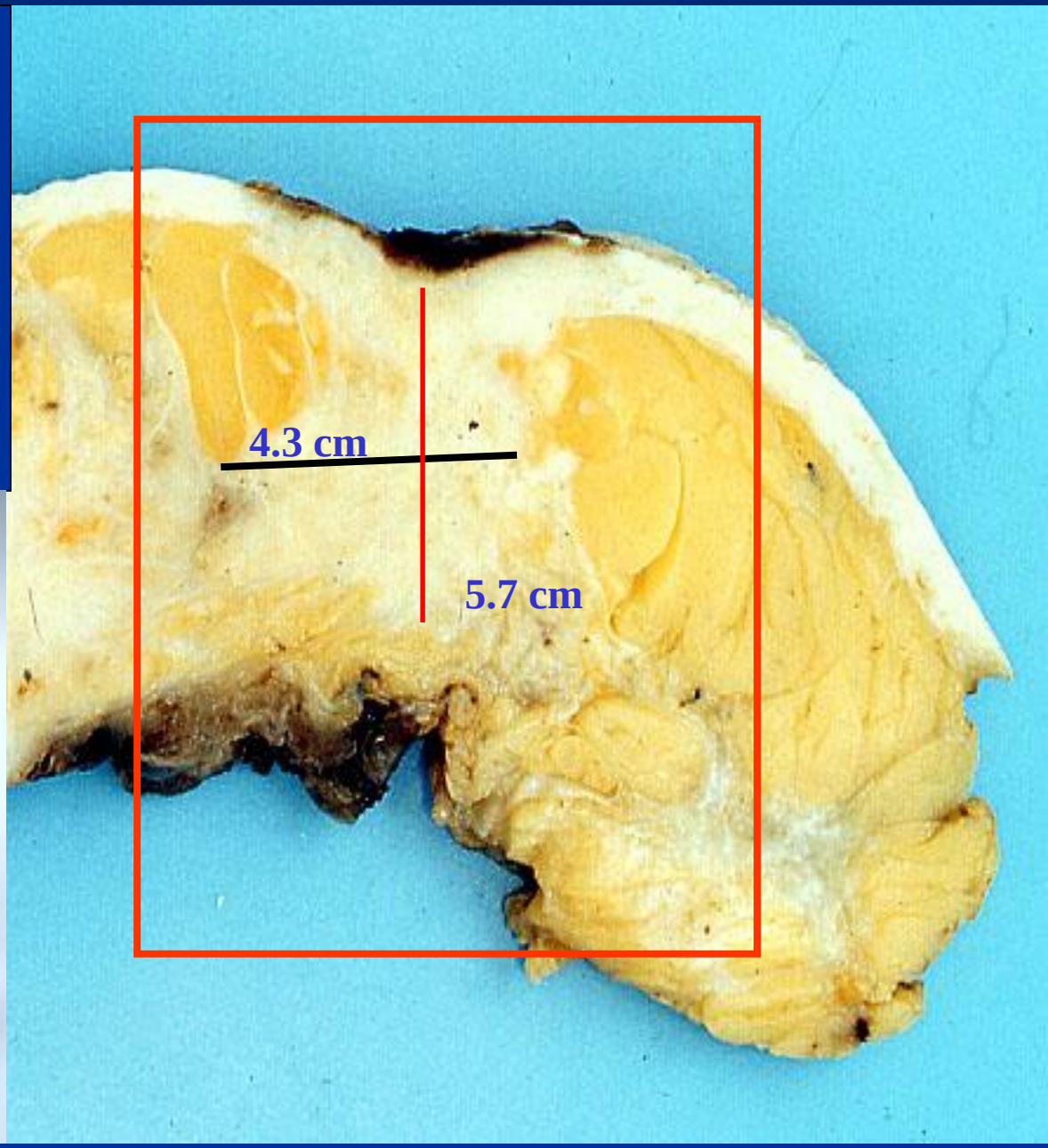
- Morphologie/Immunhistologie/in situ Hybridisierung
- digitale Bildanalyse/virtuelle Mikroskopie
- Multigen-Assays
- Next Generation Sequencing

Die “Bibel” des Pathologen und des Klinikers

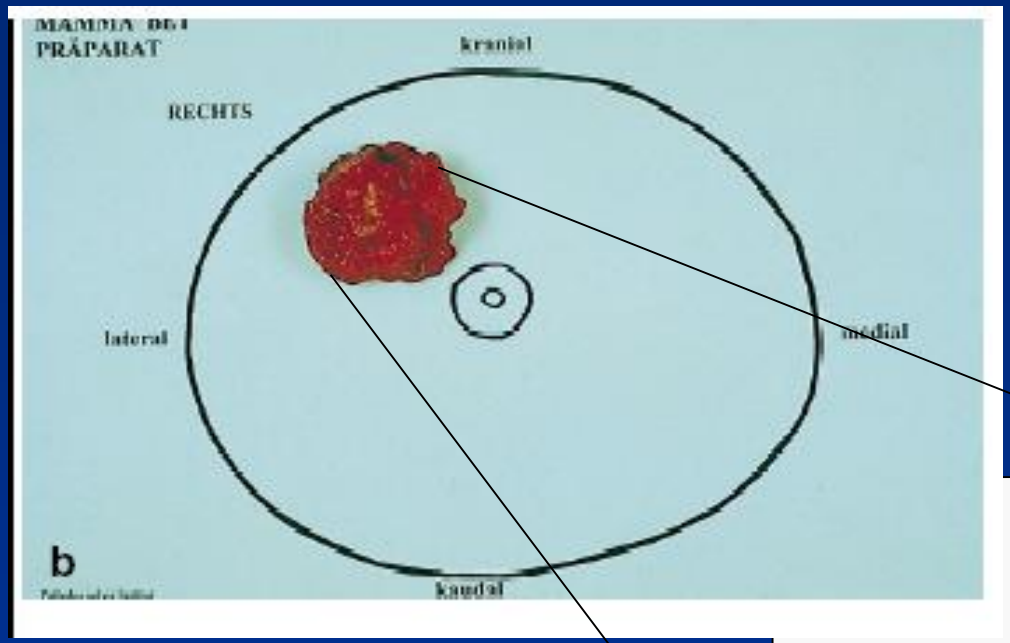


Lyon, 2012

Makro-Dokumentation
des Mamma-Präparat
mit metrischer
Dokumentation
direkt am Bildschirm

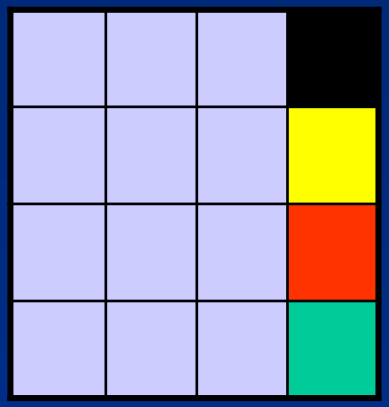


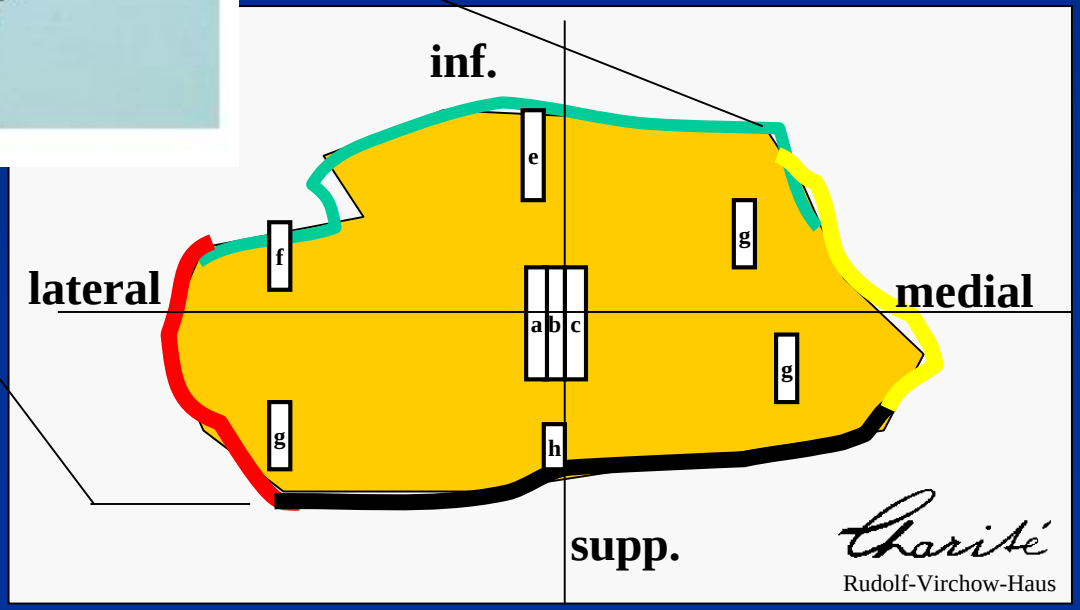
Standardisierte Dokumentation von Mammaexzisionen – Brustzentrum Charité

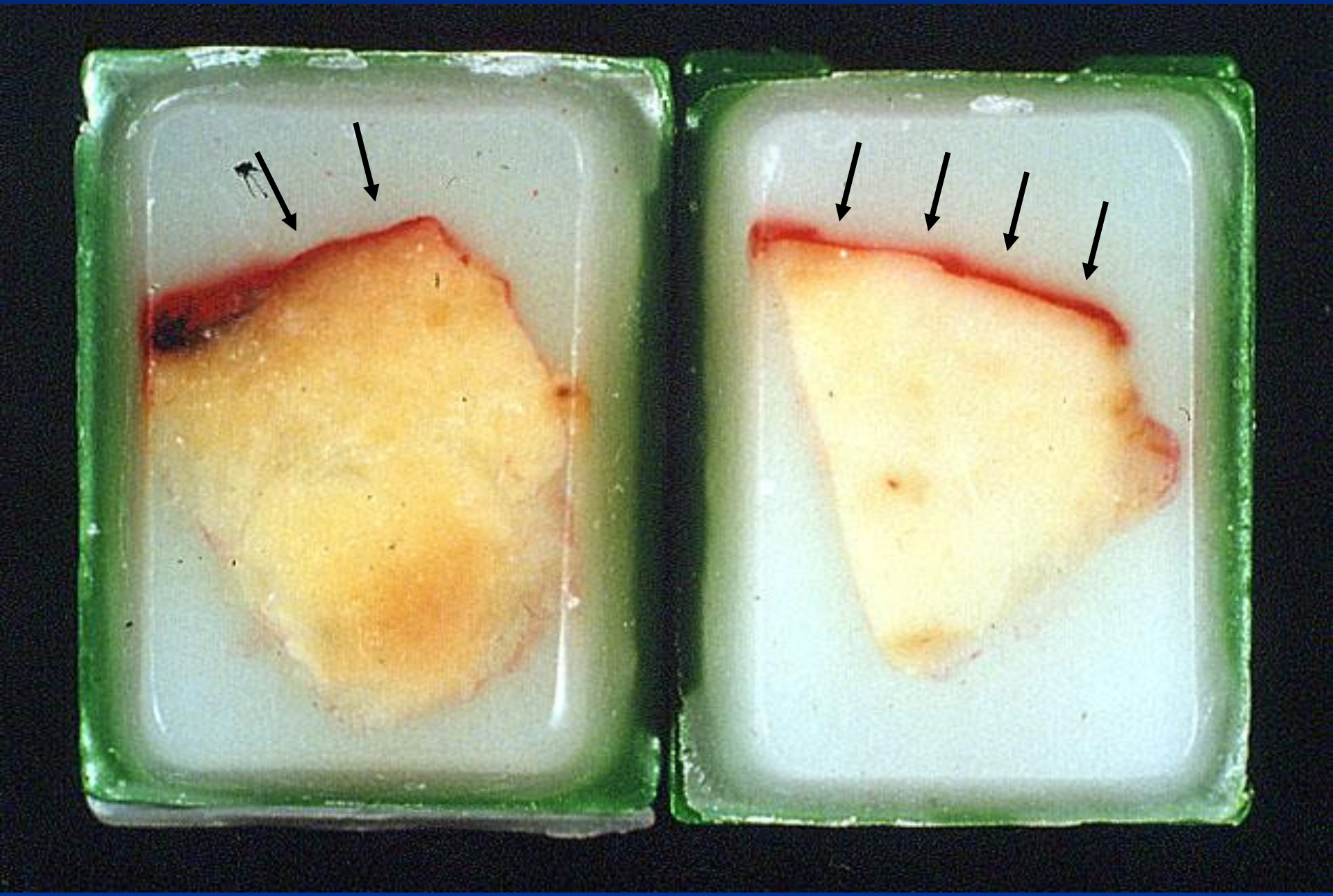


- Mammo-graphie
- Präparate-mammogr.
- rechts
- links

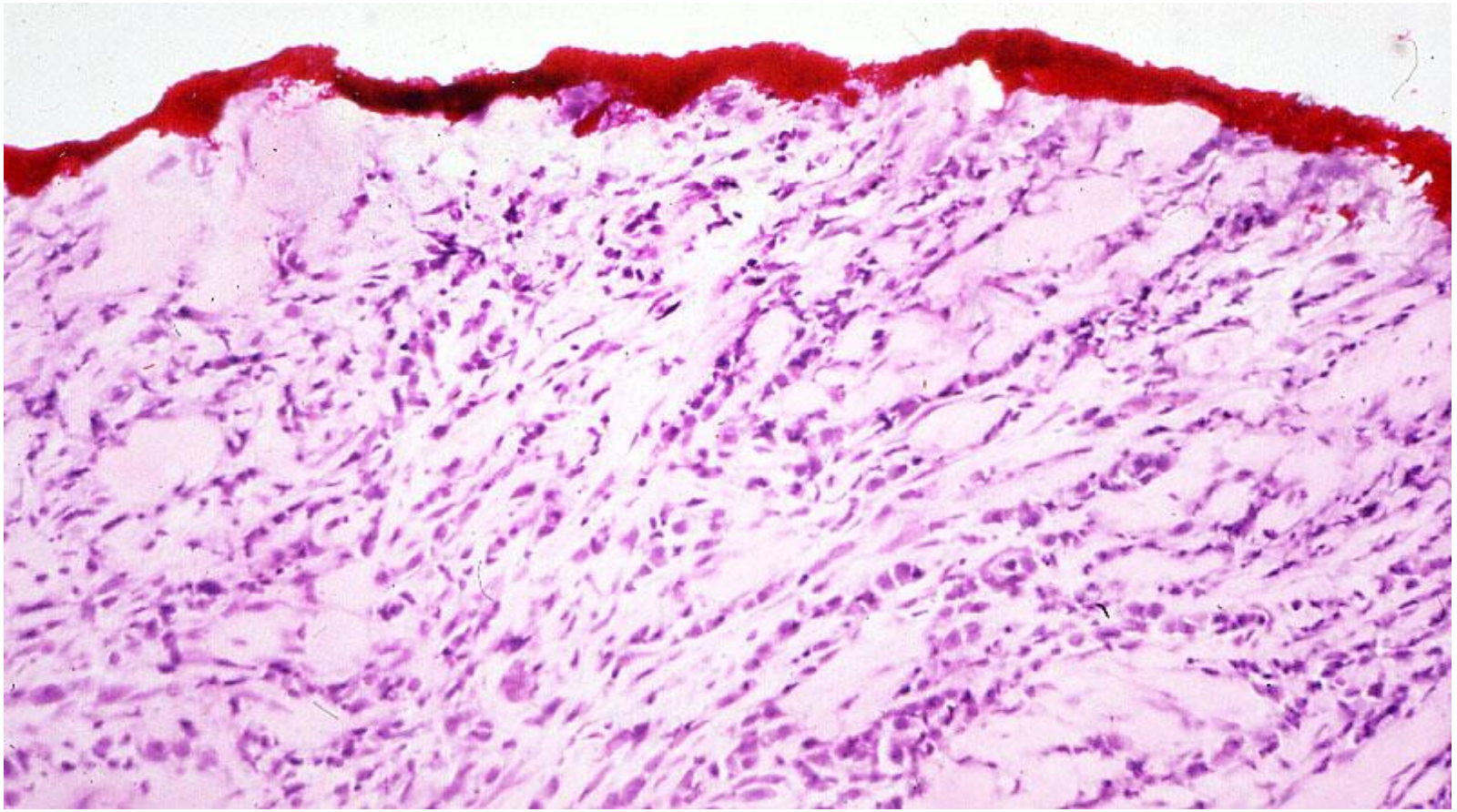
Abstand zum RR - Farbe







Schnellschnittmarkierung beim Mammakarzinom



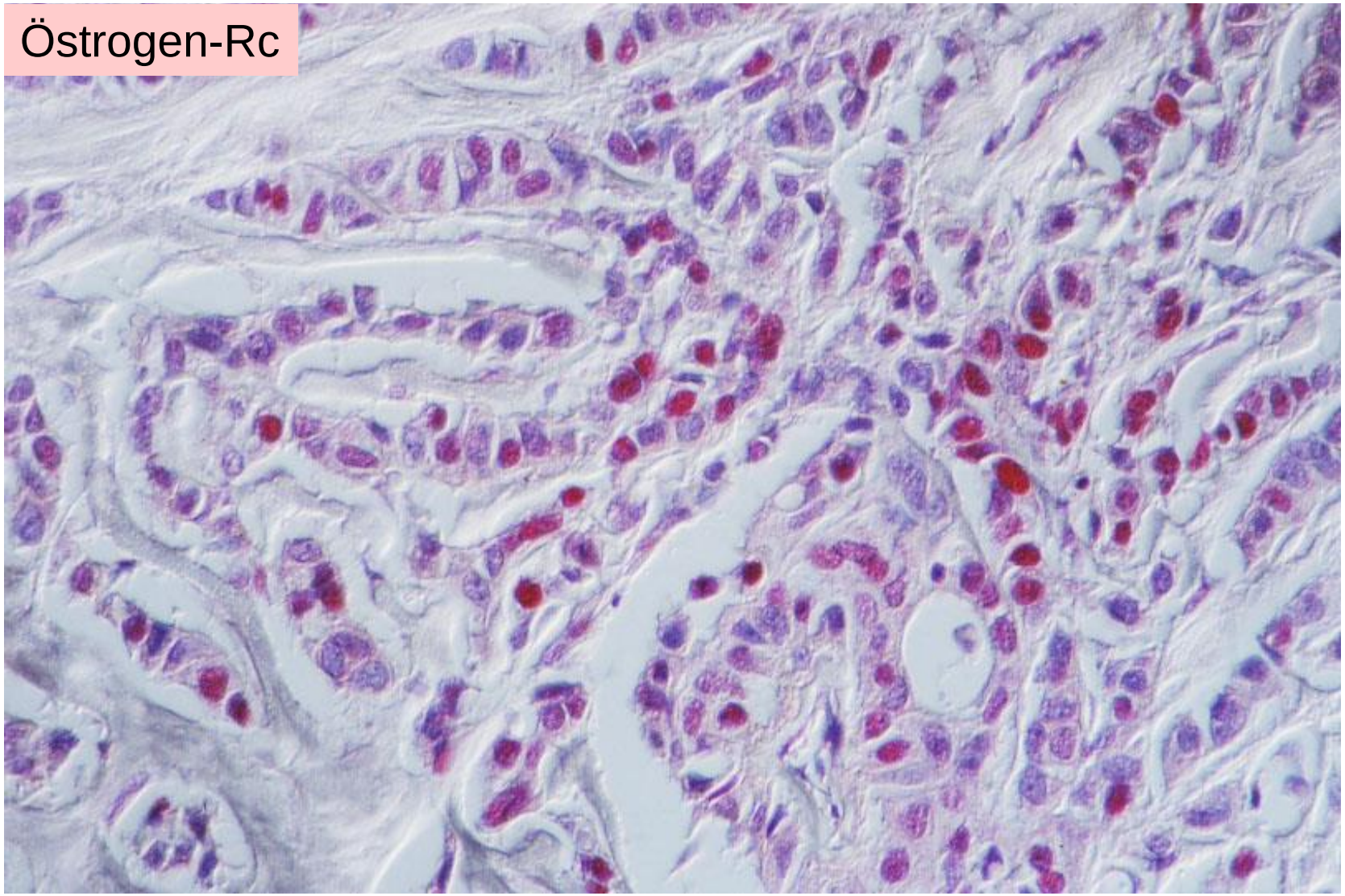
Mikroskopische Untersuchung

- Tumorentität
- Grading
  - Mitosen
  - Kernpleomorphie
  - Tubulusbildung
- Größe invasiver und indraduktaler Tumoranteil
- Peritumorale Angioinvasion (L,V)
- Perineurale Tumorausbreitung
- Beziehung zum Resektionsrand, Residualklassifikation
- Rezeptorstatus (ER, PR)
- Therapierelevante Marker (c-erbB2)
- Lymphknotenstatus
- Sonstige Läsionen
- Ggf. Regressionsgrad nach Sinn

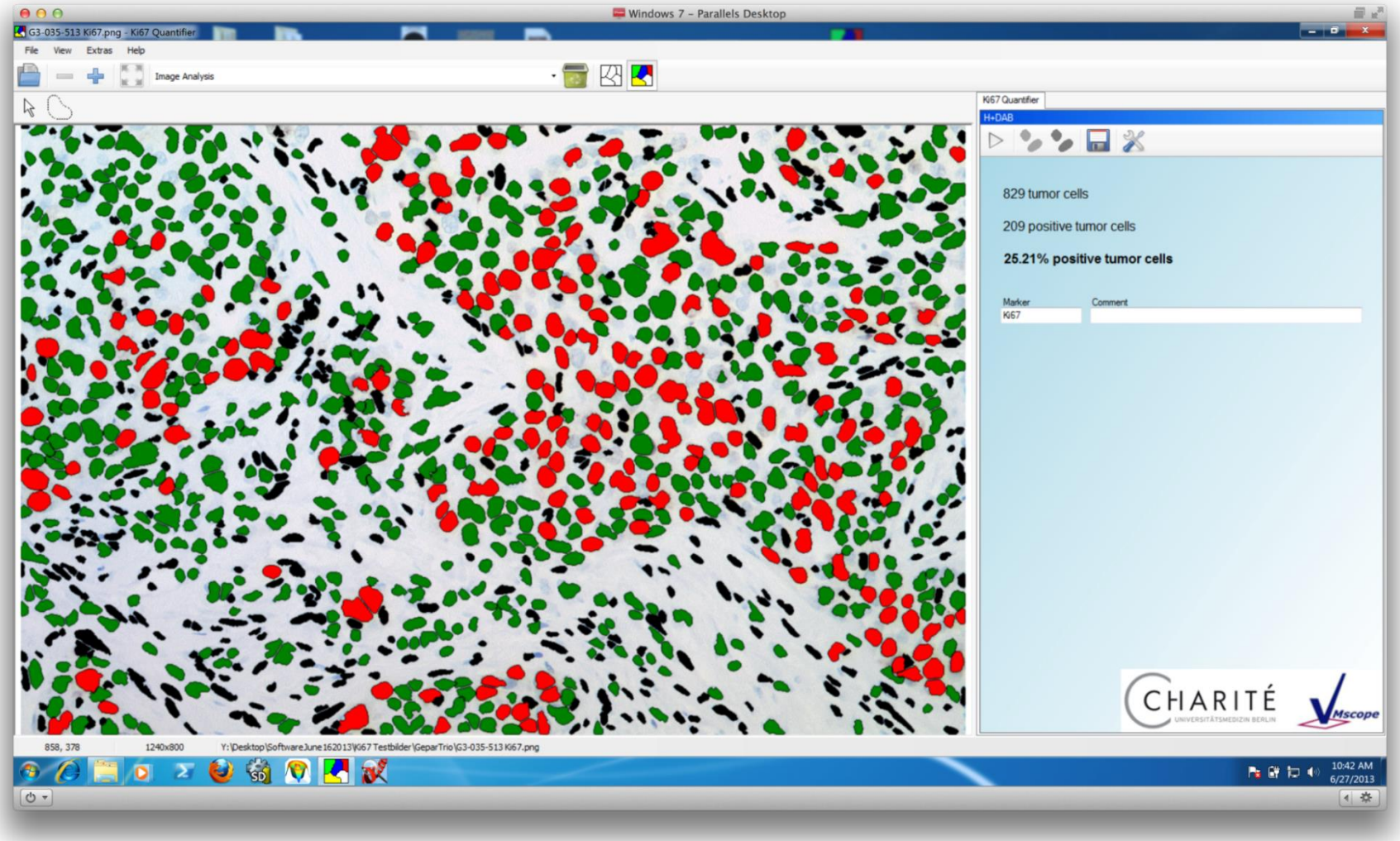
Invasive breast carcinomas (WHO 2012) (w/o microinvasive ca).

Invasive carcinoma of no special type (NST) (prior ductal ca.)	8500/3	
Pleomorphic carcinoma	8522/3	70%
Carcinoma with osteoclast-like stromal giant cells	8035/3	
Carcinoma with choriocarcinomatous features		
Carcinoma with melanotic features		
Invasive lobular carcinoma	8520/3	
Classic lobular carcinoma		
Solid lobular carcinoma		
Alveolar lobular carcinoma		10%
Pleomorphic lobular carcinoma		
Tubulolobular carcinoma		
Mixed lobular carcinoma		
Tubular carcinoma	8211/3	
Cribriform carcinoma	8201/3	5%
Mucinous carcinoma	8480/3	
Carcinoma with medullary features		
Medullary carcinoma	8510/3	
Atypical medullary carcinoma	8513/3	15%
Invasive carcinoma NST with medullary features	8500/3	
Carcinoma with apocrine differentiation		↓
Carcinoma with signet-ring-cell differentiation		
Invasive micropapillary carcinoma	8507/3	rare tumors

Östrogen-Rc



Virtual Microscopy - Pixel based Ki67 Quantifier



green: tumor cell Ki67- red: tumor cell Ki67+ black: non-tumor cell

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Eingangsnummer:	Eingangsdatum	Unser Zeichen	Befundausgang
E29247/16	19.05.2016	bmu/bmu	--.--.----

Patient: **##.##**

PATHOHISTOLOGISCHE BEGUTACHTUNG

(Verwendung zu wissenschaftlichen oder gutachterlichen Zwecken nur mit ausdrücklicher Zustimmung des Institutsdirektors)

Makroskopie
Mamma links: Eine faden- und drahtmarkierte Mamma-DE links, 12,7 g. Größe: 50 mm (medial-lateral) x 35 mm (superior-inferior) x 25 mm (anterior-posterior). Das Präparat lamelliert von lateral nach medial in 7 Scheiben. Auf der Schnittfläche ein tumorsuspektes Areal: 12 mm (medial-lateral) x 8 mm (superior-inferior) x 8 mm (anterior-posterior). Makroskopische Tumorabstände zum Resektionsrand (mm): medial 20 mm, lateral 12 mm, superior 5 mm, inferior 7 mm, anterior 7 mm, posterior 7 mm.

Mikroskopie
Zur Darstellung kommen im Bereich des makroskopisch beschriebenen Tumors atypische Zellverbände. Diese zeigen teils ein solides, teils ein glanduläres Wachstumsmuster (2 Punkte). Sie zeigen eine mäßiggradige Zellkernpleomorphie (2 Punkte). Einzelne Mitosefiguren
(3 Mitosen/10 HPF; SFZ 25; 1 Punkt). Peritumoral kommen unterschiedlich alte Einblutungen und Fettgewebsnekrosen zur Darstellung, passend zu einem vorangegangenen Stanzkanal. Weiterhin kommen atypische intraduktale Epithelproliferate zur Darstellung. Diese weisen ein kribriformes Wachstumsmsuter auf. Fokal Nachweis von Komedonekrosen. Die Zellkerne sind mäßiggradig vergrößert.

Diagnose
Zusammenfassende Diagnose
(unter Einbezug von E42089/16 und E41052/16)

Diagnose: Gut differenziertes (G1) invasives Karzinom of no special type (NST) mit assoziiertem intartumoralen duktalen Carcinoma in situ (DCIS), intermediärer Grad (G2), kribriformer Typ mit Komedonekrosen.

Lokalisation: Mamma links
Art der OP: DE, SLN
Punkte (Elston & Ellis): 5
Tumorgröße (mm): 14

Abstand zum Resektionsrand invasiver Tumoranteil (mm):
med.: >10 lat.: >10 sup.: >4 inf.: >5 ant.: 5 post.: 2,5

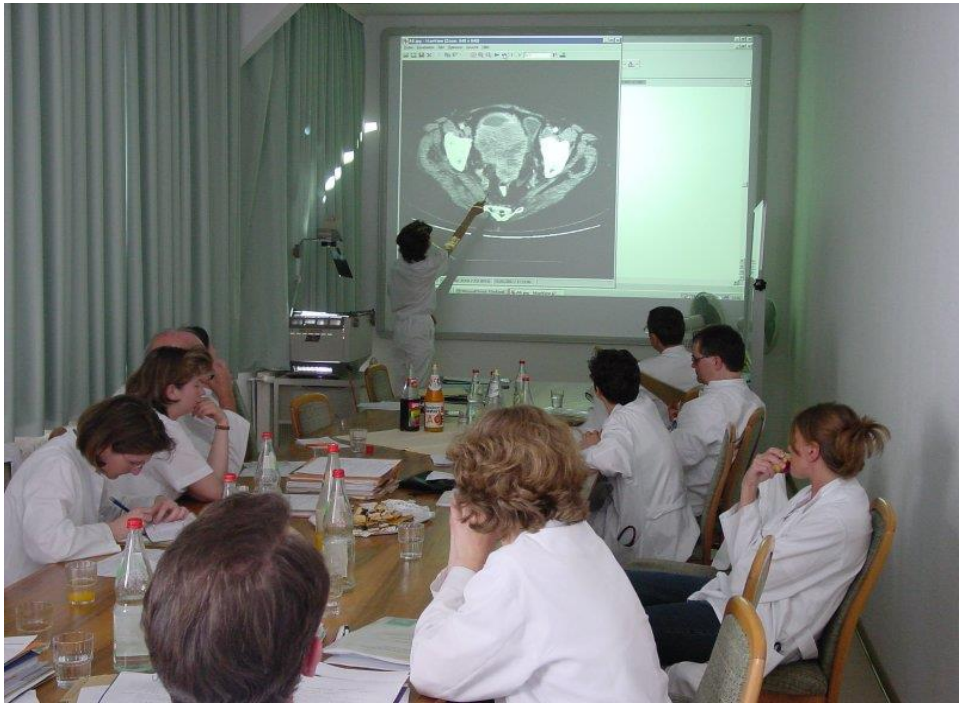
Rezeptorstatus (Stanze): ER: 80 % PR: 5 % HER2(neu): 1+ MIB-1: 5 %

LK-Status LKQ: 0/4 (vorläufig, IH folgt)
LK-Lokalisation: Sentinel

Kommentar
Es folgt die immunhistochemische Aufarbeitung der Sentinel-Lymphknoten, Sie erhalten einen Zusatzbericht. Die bisherige TNM-Verschlüsselung würde wie folgt lauten: pT1c G2 R0 L0 V0.

Prof. Dr. M. Dietel PD Dr. B. Pfitzner

Um die heute zu fordernden Qualitätsansprüche erfüllen zu können, ist eine interdisziplinäre, im Detail standardisierte Zusammenarbeit mit adäquater Dokumentation zwingend notwendig



BRUSTZENTRUM CHARITÉ - MAMMAKONFERENZ

Vorstellung am: 17.09.2003

Name: Becker	**Vorname: Brigitte**	**geb: 06.11.1949**
Bisherige Diagnose:	53 Jahre Mammakarzinom links pT1c pN0 (SN) Stanzbiopsie: Invasives Karzinom ER neg. /PR neg./ c-erbB2 neg. Starke psychische Belastung	
Bisherige Therapie:	Tumorektomie linke Mamma und Sentinel am 11.09.03 Z.n. us-gestützter Stanzbiopsie linke Mamma am 28.06.2003	
Fragestellung:	1) Wiederholung der Immunhisto am OP-Präparat notwendig 2) Radiatio linke Mamma 3) Chemotherapie 4x EC oder 6 x FEC, wenn rezeptornegativ	
Beschluß:		
Verantwortl. Arzt:		
Datum:		
Unterschrift:		

Kernteam: Zustimmung zum Beschluß

Intern. Onkologie:	Hr. Prof. Possinger	☐	Pathologie:	Hr. Prof. Guski	☐
	Hr. OA Dr. Flath	☐		Hr. Dr. Nadjari	☐
	Hr. OA Dr. Reichardt	☐	Radiologie:	Hr. OA Dr. Ehrenstein	☐
	Hr. Prof. Dr. Riess	☐		Fr. Dr. Diekmann	☐
Strahlentherapie:	Hr. Prof. Wust	☐		Hr. Dr. Diekmann	☐
	Fr. OÄ Dr. Ullrich	☐	Andere:		☐
	Fr. OÄ Dr. Hinkelbein	☐			☐
Gyn. / Chirurg.:	Hr. Prof. Blohmer	☐			☐
	Hr. OA Dr. Winzer	☐			☐
	Fr. OÄ PD Dr. Reles	☐			☐
	Fr. OÄ Dr. Kleine-Tebbe	☐			☐

Qualität in der Pathologie

BRCA1/2 Ringversuch

Teilnehmer: n = 34

(aus D, Schweiz, Österreich)

Zertifiziert: n = 26

QuIP

Qualitätssicherungs-Initiative Pathologie
Ringversuche
Immunhistochemie und Molekularpathologie

Geschäftsstelle QuIP | Robert-Koch-Platz 9 | 10115 Berlin

Ringversuch
Teilnahmezertifikat
Molekularpathologie – BRCA1/2

2015

Prof. Dr. med. Manfred Dietel
Universitätsmedizin Berlin, Institut für Pathologie
Charitéplatz 1
10117 Berlin

hat am Ringversuch „BRCA 1/2 Mutationstestung“ erfolgreich teilgenommen

Leitung des Ringversuches:
Prof. Dr. med. Peter Schirmacher, Dr. Volker Endris

Heidelberg, Berlin, Gelsenkirchen 18.06.2015

Prof. Dr. med. P. Schirmacher
Deutsche Gesellschaft für
für Pathologie e.V.

Prof. Dr. med. M. Dietel
Vorsitzender QuIP-Board

Prof. Dr. med. W. Schlake
Bundesverband Deutscher
Pathologen e.V.

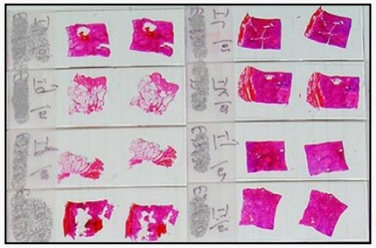
Bestandteil dieser Teilnahmebescheinigung ist die getrennt gefasste, inhaltliche Beurteilung der Untersuchung.

Träger der Ringversuche Immunhistochemie und Molekularpathologie QuIP sind:
Deutsche Gesellschaft für Pathologie e. V. geschaeftsstelle@pathologie-dgp.de, www.pathologie-dgp.de/ und
Bundesverband Deutscher Pathologen e.V, bv@pathologie.de, www.bv-pathologie.de; Robert-Koch-Platz 9, 10115 Berlin

One Day Diagnostics for Breast Cancer

diagnostic biopsy, 10:00 → technician in near-by lab → fixation, rapid tissue processing, 3-5 h

cutting + staining, cover slipping,1h →



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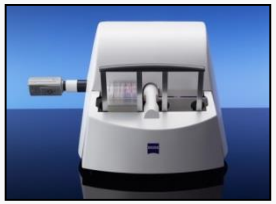
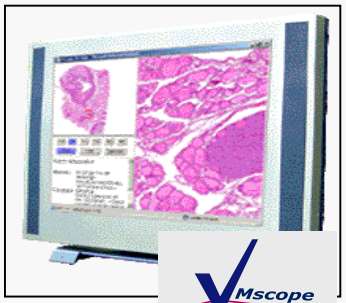


scanning, 5m



Beyond the scope of microscopy

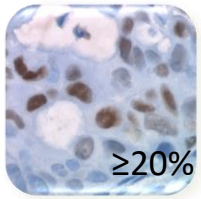
diagnosis

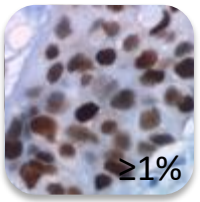
16:30

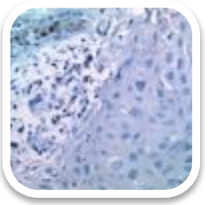
tel, fax, e-mail

happy clinician

Molecular subtypes of breast cancer - St. Gallen 2013

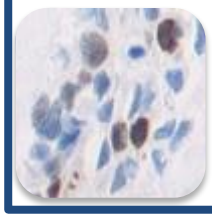


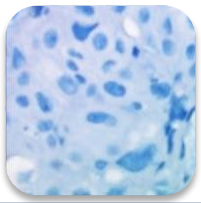


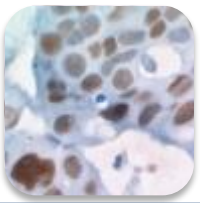


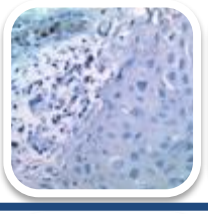
Luminal A
(Ki67 low and PR+)

Endocrine therapy alone



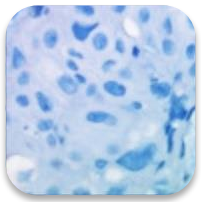


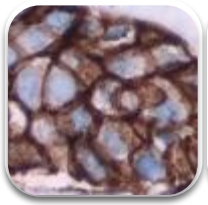




Luminal B (HER2-)
(Ki67 high or PR-/low)

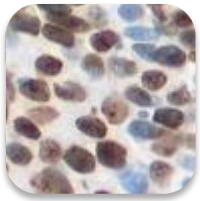
Endocrine ± cytotoxic therapy

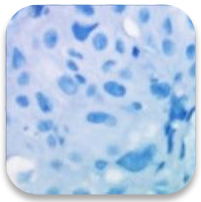


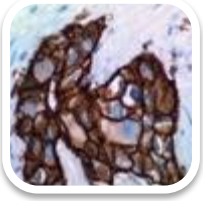


Luminal B/HER2+

Cytotoxics + anti-HER2 + endocrine therapy

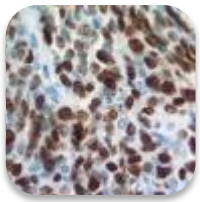


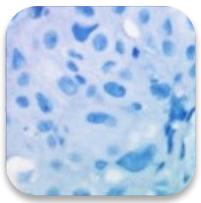


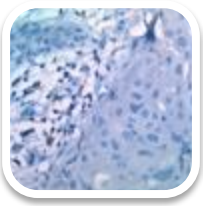


HER2+ (non-luminal)

Cytotoxics + anti-HER2 therapy





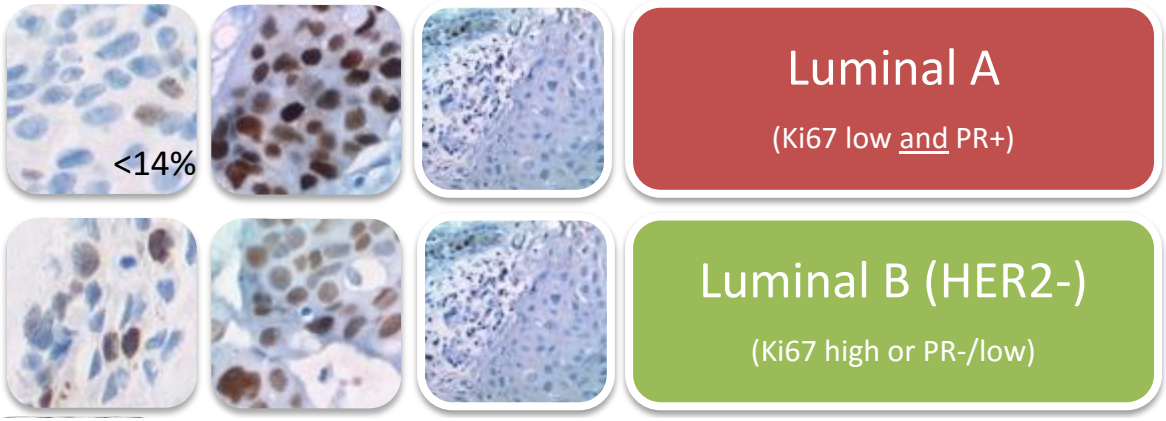


Triple-negative

Cytotoxic therapy

adapted from Goldhirsch Ann Oncol Sept 2013

Therapy options for ER+/HER2- breast cancer



Luminal A

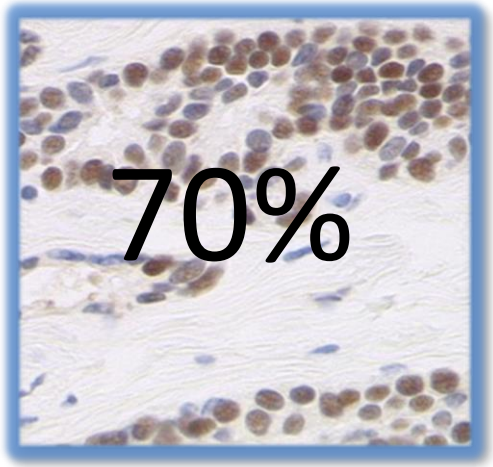
(Ki67 low and PR+)

Luminal B (HER2-)

(Ki67 high or PR-/low)

Endocrine therapy vs. chemoendokrine therapy

Endocrine therapy 5 years vs. 10 years



Opportunities for stratification:

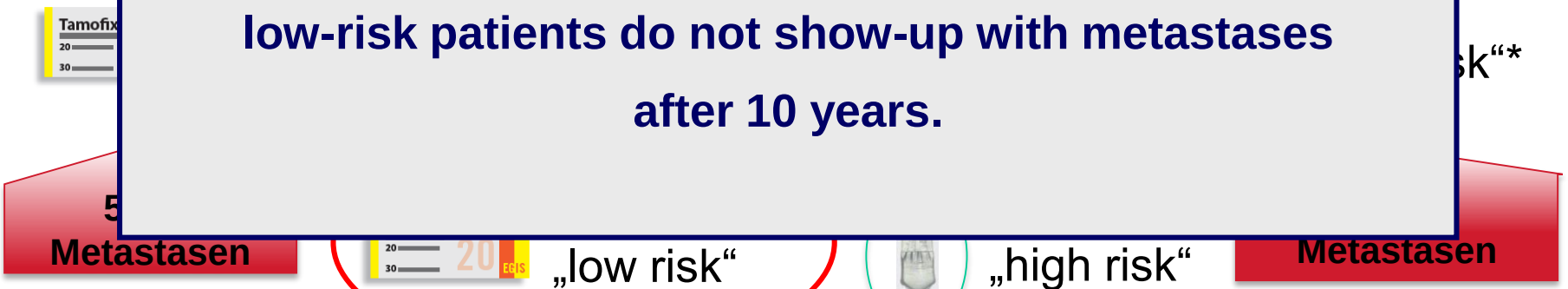
1. clinical pathological parameters
2. Ki67
3. **Oncotype Dx**
4. **EndoPredict**
5. **PAM50**
6. **Mammaprint**
7. uPA-PAI1

Conflict of interest statement:
M. Dietel was Cofounder of Sividon diagnostics.

Stratification by EndoPredict$_{clin}$ ®

1702 Patientinnen in ABCSG 6 & 8

Following the EPclin-based predictive data 96% of the low-risk patients do not show-up with metastases after 10 years.



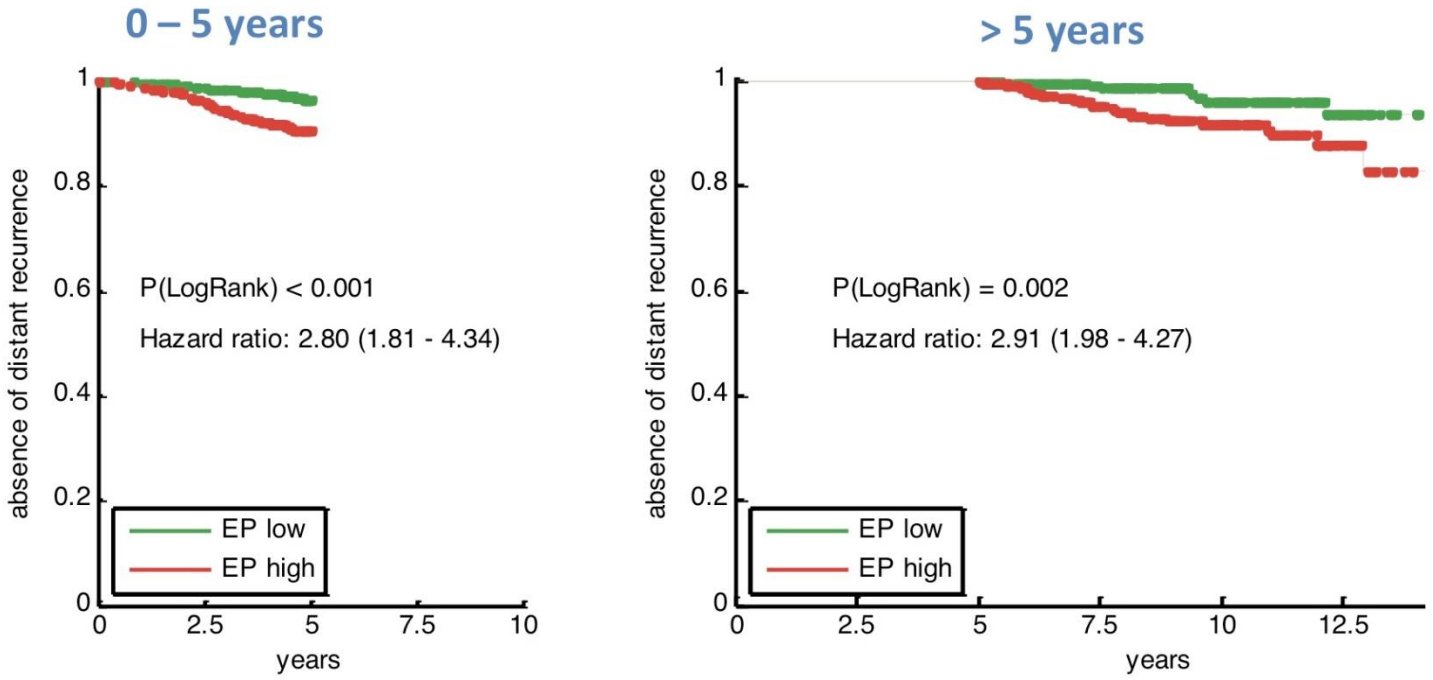
w/o Endopredict these patients may have recieved CTx

4,5 % Metastasen

19,7 % Metastasen

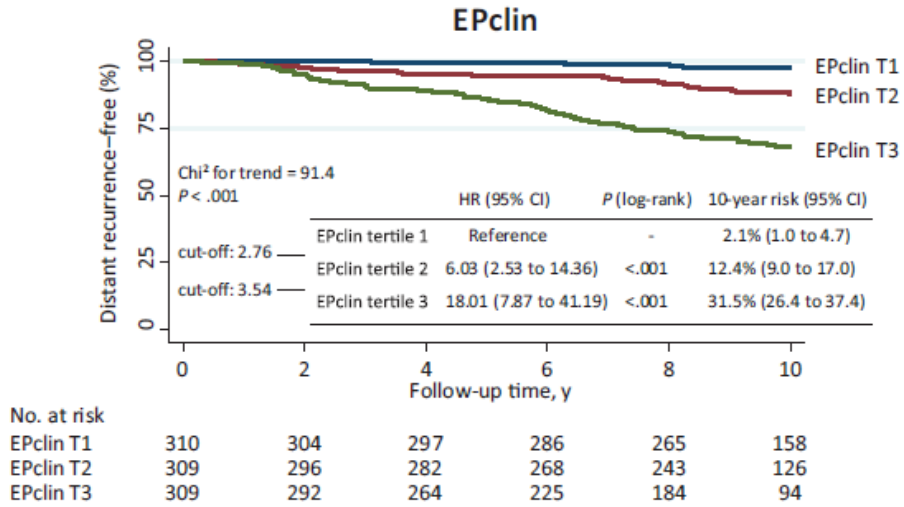
*nach S3-Leitlinien

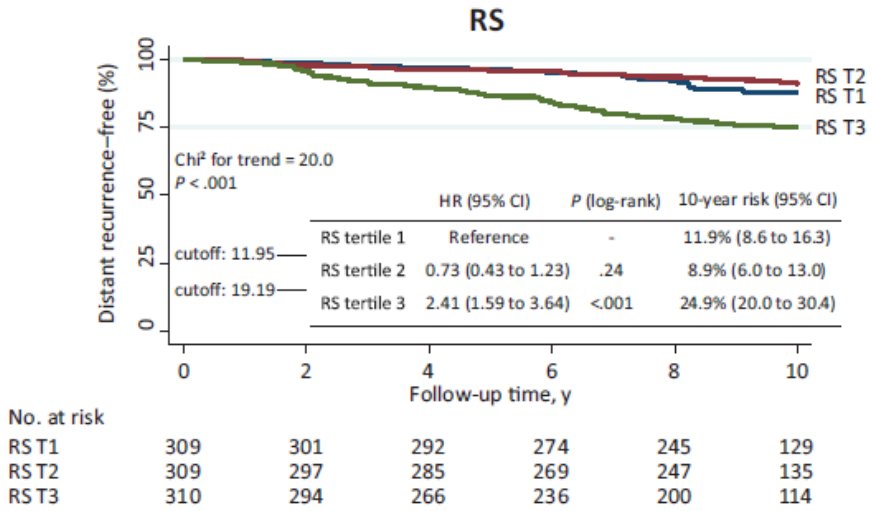
Stratification by EndoPredict$_{clin}$ ®



The EndoPredict low-risk group (49%) had a significant improved clinical outcome **bevor and after 5 years** of follow-up. 96,3% were free of distant metastases between 5 and 10 years in the EP low risk group.

Dubsky, SABCS 2012 / Br J Cancer. 2013;109:2959-64





OXFORD

JNCI J Natl Cancer Inst (2016) 108(11): djw149

doi: 10.1093/jnci/djw149
First published online July 10, 2016
Article

ARTICLE

Comparison of EndoPredict and EPclin With Oncotype DX Recurrence Score for Prediction of Risk of Distant Recurrence After Endocrine Therapy

Richard Buus, Ivana Sestak, Ralf Kronenwett, Carsten Denkert, Peter Dubsky, Kristin Krappmann, Marsel Scheer, Christoph Petry, Jack Cuzick, Mitch Dowsett

Kaplan-Meier estimates for 10-year distant recurrence according to EP, EPclin, and recurrence score, split into tertiles in all patients. Kaplan-Meier curves were calculated and tested for equality using the log-rank test.

The numbers of patients at risk in each group at various time points are given below each graph. All statistical tests were two-sided. CI = confidence interval; EP = EndoPredict; HR = hazard ratio; RS = recurrence score

OXFORD

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Comparison of EndoPredict and EPclin With Oncotype DX Recurrence Score for Prediction of Risk of Distant Recurrence After Endocrine Therapy

Richard Buus, Ivana Sestak, Ralf Kronenwett, Carsten Denkert, Peter Dubsky, Kristin Krappmann, Marsel Scheer, Christoph Petry, Jack Cuzick, Mitch Dowsett

Conclusions: EP and EPclin were highly prognostic for DR in endocrine-treated patients with ER+, HER2-negative disease. EPclin provided more prognostic information than RS. This was partly but not entirely because of EPclin integrating molecular data with nodal status and tumor size.

Conclusions

- EPclin identified a low risk group of patients who may be spared chemotherapy
- EPclin provided more accurate prognostic information than the RS - partly but not entirely due to the EPclin including tumour size and nodal status
- Differences between EPclin and RS were greatest in node positive patients
- The bottom tertile of EPclin in node negative patients identified a group with extremely good prognosis
- The data highlight the importance of the inclusion of clinicopathologic factors (including type of endocrine treatment) for estimates of residual risk of distant recurrence

Prognosefaktoren III – Primäres Mammakarzinom

AGO 2015

Faktor	LoE_{2009}	CTS	AGO
➢Disseminierte Tumorzellen (DTC, im Knochenmark)	I	B	+/-
➢Zirkulierende Tumorzellen (CTC, im Blut, Cell Search®) $	I	B	+/-
➢Therapieentscheidungen basierte auf CTC-Phänotypen	III	C	-
➢ Multigene assay (EndoPredict®, Prosigna® , Oncotype DX®) $ (N-/+, HR+ HER2-)	I	B	+*
➢ 70 gene signature (MammaPrint®), N-/+	II	C	+*
➢ IHC4 (central pathology, published algorithm) #	I	B	+/-

* Sollte nur bei ausgewählten Patientinnen angewandt werden, wenn alle anderen Kriterien keine Therapieentscheidung zulassen

$ Validierte klinische Daten nur verfügbar für diesen Assay

Cuzick et al., J Clin Oncol 29: 4273-4278, 2011

Guidelines Breast
Version 2015.1D

PARP- inhibitors and triple negative BC

Routine **BRCA1/2-testing** as prerequisite for treatment with PARP inhibitors, such as

Olaparib (AZ) Two Phase III clinical trials are currently ongoing:

OlympiAD: Olaparib vs TPC* in MBC with *BRCA* mutations – *fully recruited*

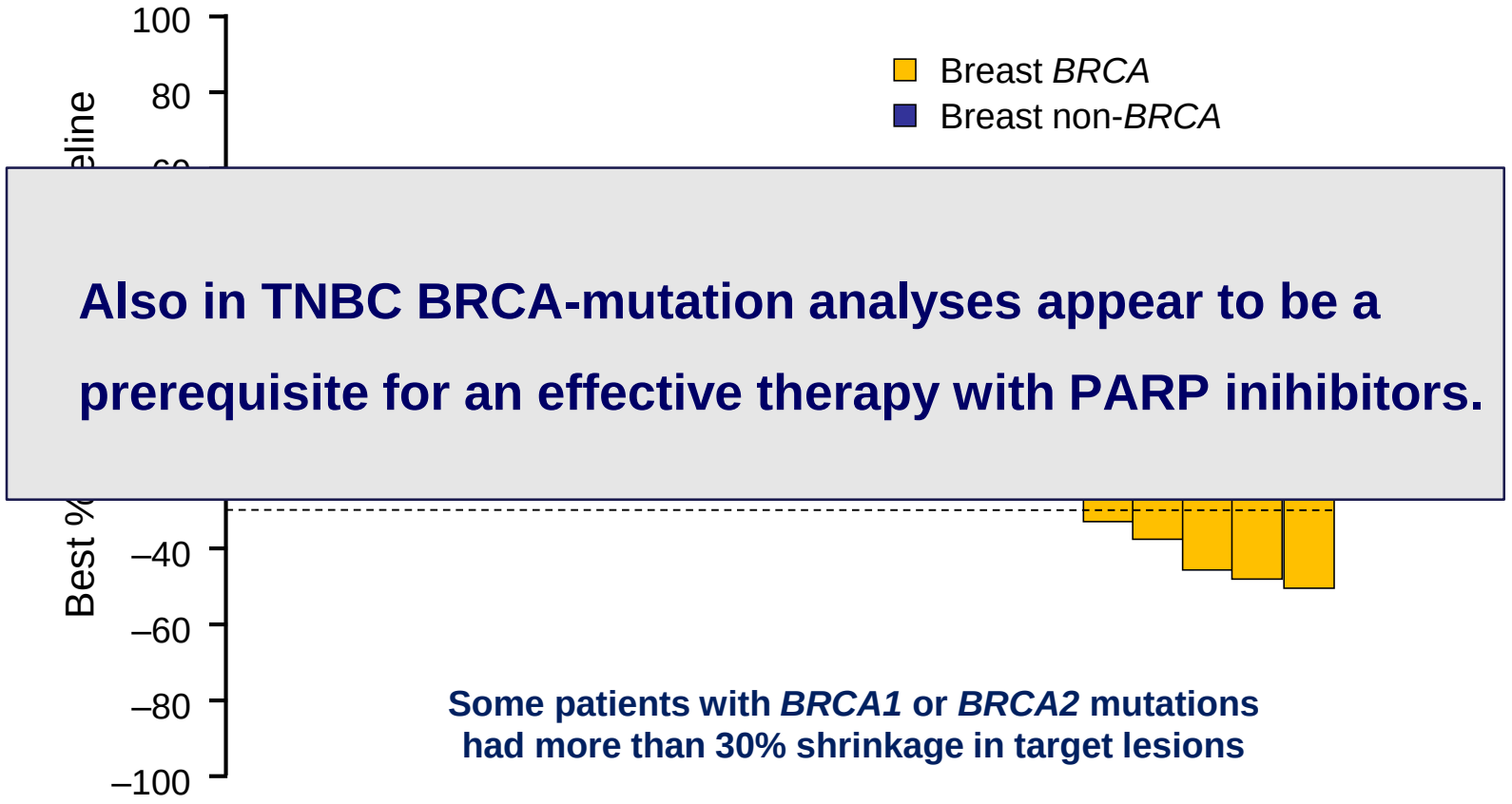
OlympiA: Adjuvant olaparib vs placebo in high-risk TNBC with *BRCA* mutations – *currently recruiting*

Talazoparib (BMN-673, *BioMarin Pharmaceutical Inc.*) is now in phase 3 for metastatic germline BRCA mutated advanced/metastatic breast cancer. Trial estimated to complete in June 2016.

Rucaparib is in phase II clinical trials for metastatic breast and ovarian cancer with known BRCA1 or BRCA2 mutation.

*treatment of physician's choice

Best percentage change in target lesion size by *BRCA* status in breast cancer



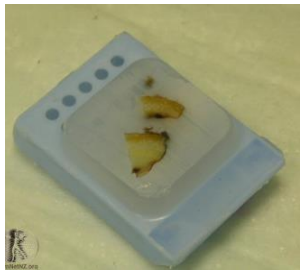
Also in TNBC BRCA-mutation analyses appear to be a prerequisite for an effective therapy with PARP inihibitors.

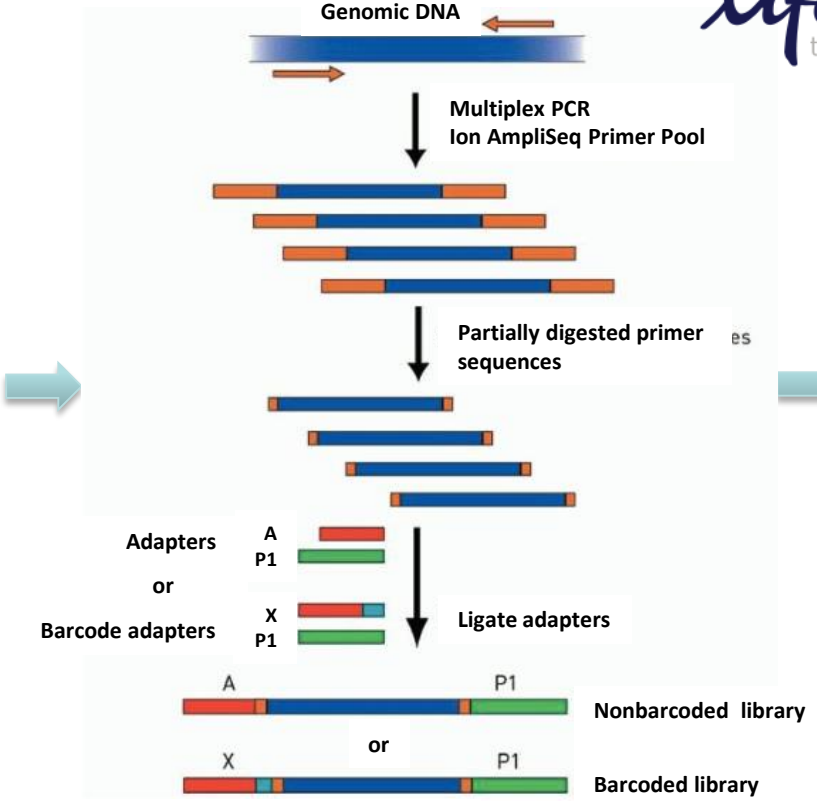
Some patients with *BRCA1* or *BRCA2* mutations had more than 30% shrinkage in target lesions

Median best percentage change from baseline: +10.1% (–50 to 50)
data provided by Astra Zeneca

Gelmon KA et al. *Lancet Oncol*. 2011;12:852–861

Integrating Next Generation Sequencing in Diagnostic Pathology





life technologies™

IonTorrent PGM

Ion AmpliSeq™ Cancer Panel targets 46 critical genes

ABL1	*ERBB4*	*KDR*	*PTPN11*
AKT1	*FBXW7*	*KIT*	*RB1*
ALK	*FGFR1*	*KRAS*	*RET*
APC	*FGFR2*	*MET*	*SMAD4*
ATM	*FGFR3*	*MLH1*	*SMARCB1*
BRAF	*FLT3*	*MPL*	*SMO*
CDH1	*GNAS*	*NOTCH1*	*SRC*
CDKN2A	*HNF1A*	*NPM1*	*STK11*
CSF1R	*HRAS*	*NRAS*	*TP53*
CTNNB1	*IDH1*	*PDGFRA*	*VHL*
EGFR	*JAK2*	*PIK3CA*	
ERBB2	*JAK3*	*PTEN*	

Genome-wide somatic mutations shown in circo plots

Ellis MJ: 2 1 J U N E 2 0 1 2 | VO L 4 8 6 | N AT U R E | 3 5 3

Published Ahead of Print on May 2, 2016 as 10.1200/JCO.2015.64.8931
The latest version is at http://jco.ascopubs.org/cgi/doi/10.1200/JCO.2015.64.8931

JOURNAL OF CLINICAL ONCOLOGY | ORIGINAL REPORT

Pembrolizumab in Patients With Advanced Triple-Negative Breast Cancer: Phase Ib KEYNOTE-012 Study

Rita Nanda, Laura Q.M. Chow, E. Claire Dees, Raanan Berger, Shilpa Gupta, Ravit Geva, Lajos Pusztai, Kumudu Pathiraja, Gursel Aktan, Jonathan D. Cheng, Vassiliki Karantza, and Laurence Buissere

Conclusion

This phase Ib study describes preliminary evidence of clinical activity and a potentially acceptable safety profile of pembrolizumab given every 2 weeks to patients with heavily pretreated, advanced **PD-L1 positive*** TNBC. A single-agent phase II study examining a 200-mg dose given once every 3 weeks

* determined by immunohistochemistry

Summary and Conclusions

- In patients with heavily pretreated, **PD-L1–positive,** advanced **ER+/HER2–** breast cancer
  - Pembrolizumab showed a manageable safety profile
  - Pembrolizumab was associated with an ORR* of 12% and a CBR** of 20%.
    - In the 22 evaluable patients (1 scan after baseline), ORR was 14% and CBR was 23%
  - Responses were durable (range, 8.7+ to 44.3+ weeks)
- Further investigation of immune therapies in ER+/HER2– breast cancer, particularly combination therapies, is warranted

* overall response rate, ** clinical benefit rate

Comprehensive Breast Cancer Diagnostics Depends on Combination of up to date Technologies

Telepathology for 2nd opinion, quality control and objective quantification

In situ hybridization detecting genetic alterations

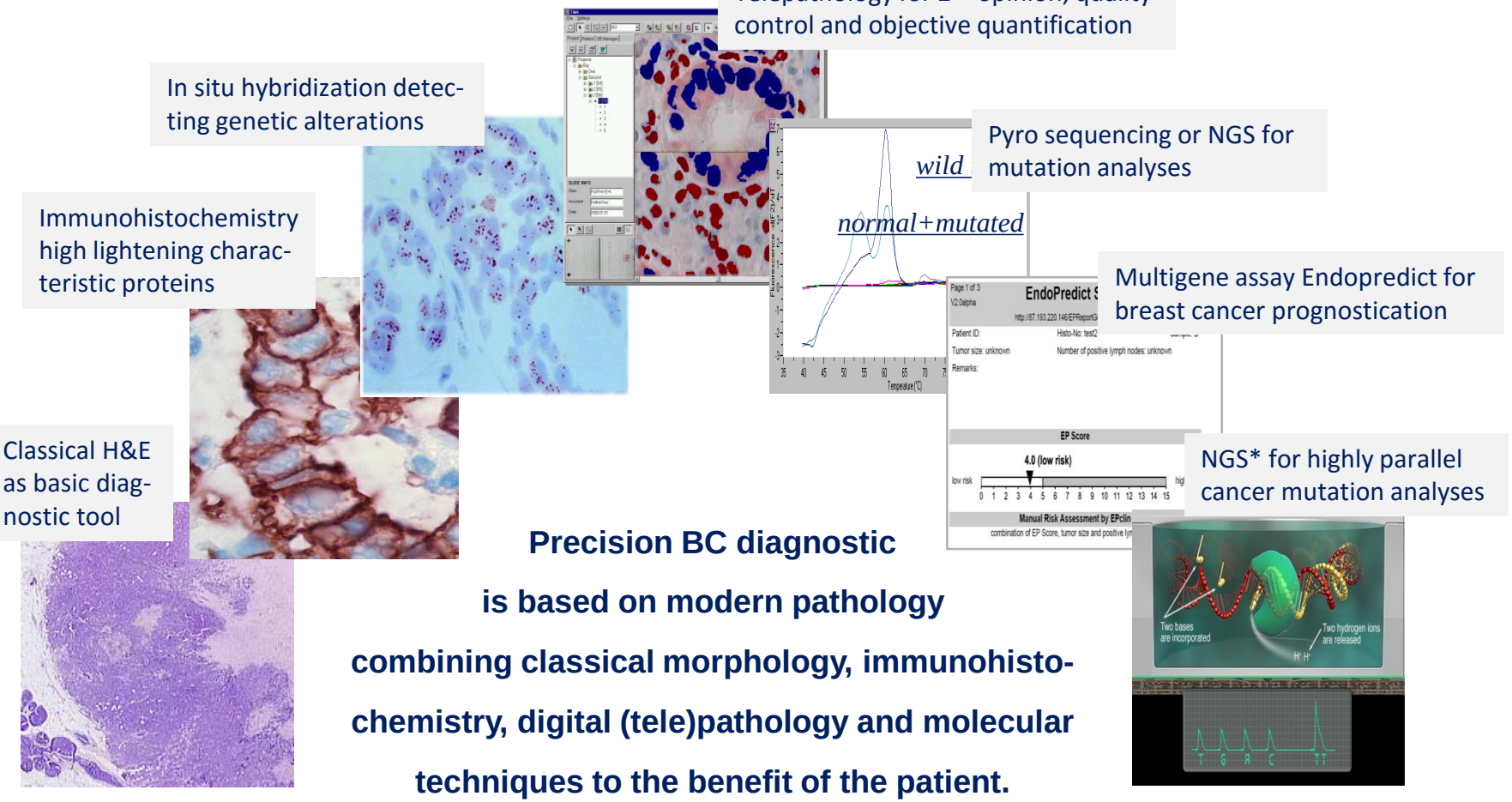
Pyro sequencing or NGS for mutation analyses

Immunohistochemistry high lightening characteristic proteins

Multigene assay Endopredict for breast cancer prognostication

Classical H&E as basic diagnostic tool

NGS* for highly parallel cancer mutation analyses



Precision BC diagnostic is based on modern pathology combining classical morphology, immunohistochemistry, digital (tele)pathology and molecular techniques to the benefit of the patient.

*next generation sequencing


Institut für Pathologie,
Rudolf-Virchow-Haus, Charité
Humboldt-Universität zu Berlin
Berliner Medizin-historisches Museum
Alexander Ufer
INSTITUT FÜR PATHOLOGIE
RUDOLF-VIRCHOW-HAUS
CHARITÉ