The Continuing Dilemma of Ductal Carcinoma in Situ

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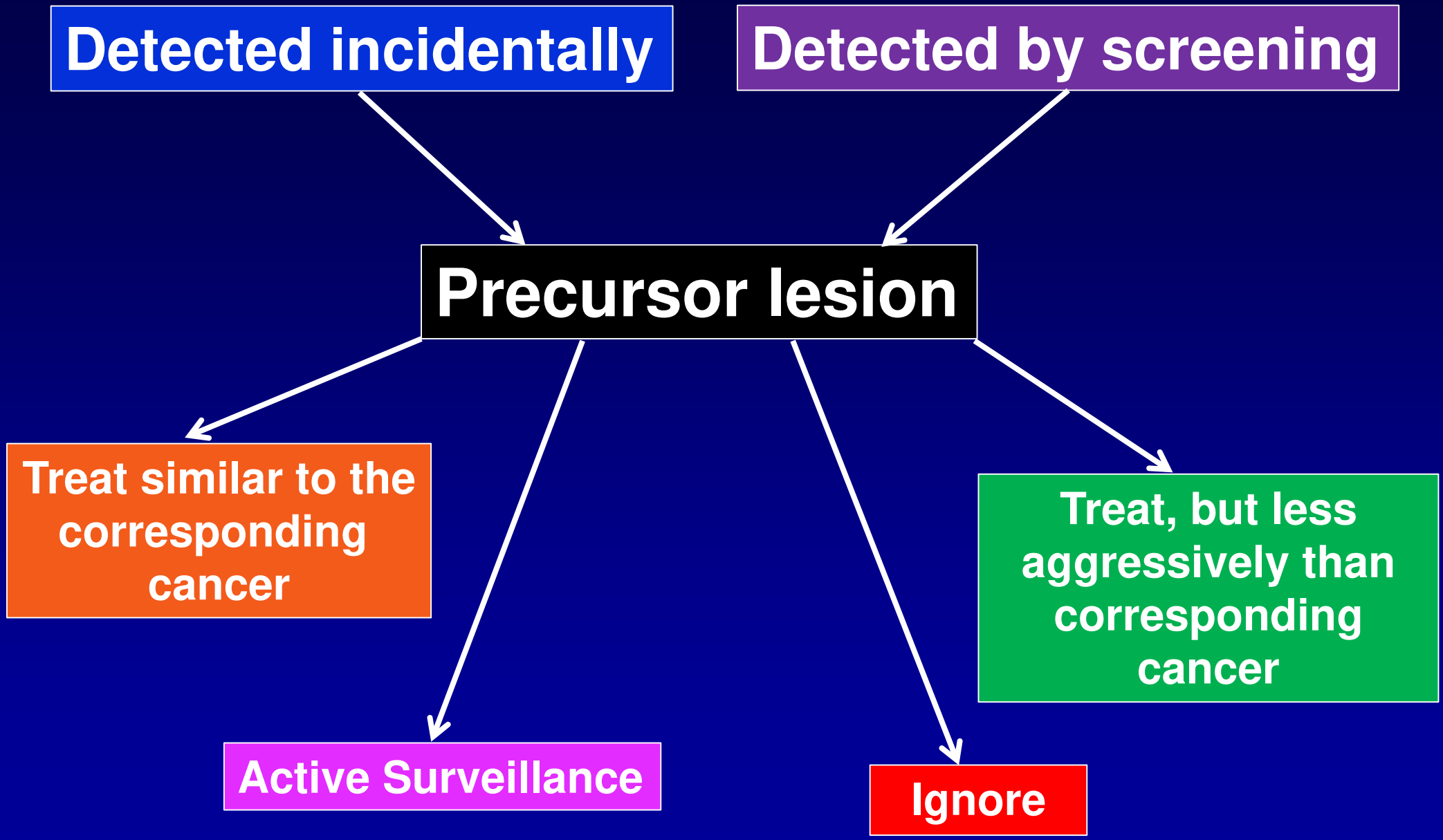
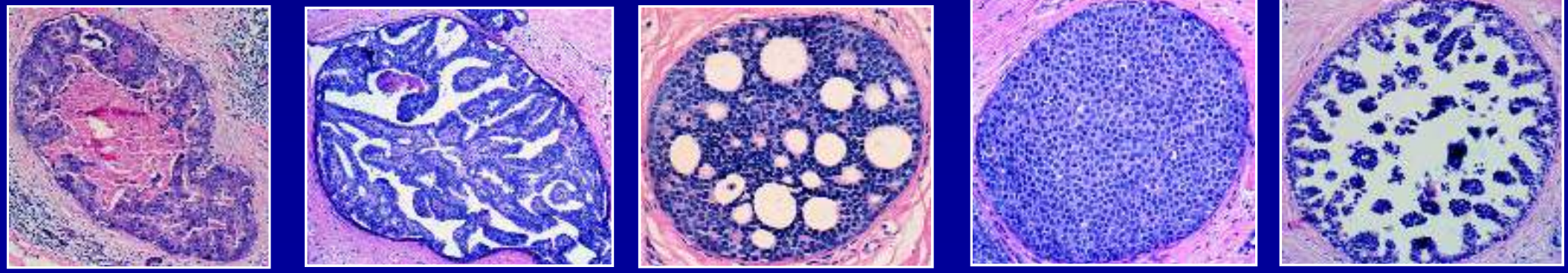
Disclosures

- None


Detected incidentally
Detected by screening
Precursor lesion
Treat similar to the corresponding cancer
Active Surveillance
Ignore
Treat, but less aggressively than corresponding cancer

DCIS

- Heterogeneous group of lesions
- Natural history poorly defined (especially for small, mammographically-detected lesions)
- Optimal treatment controversial



Heterogeneity of DCIS

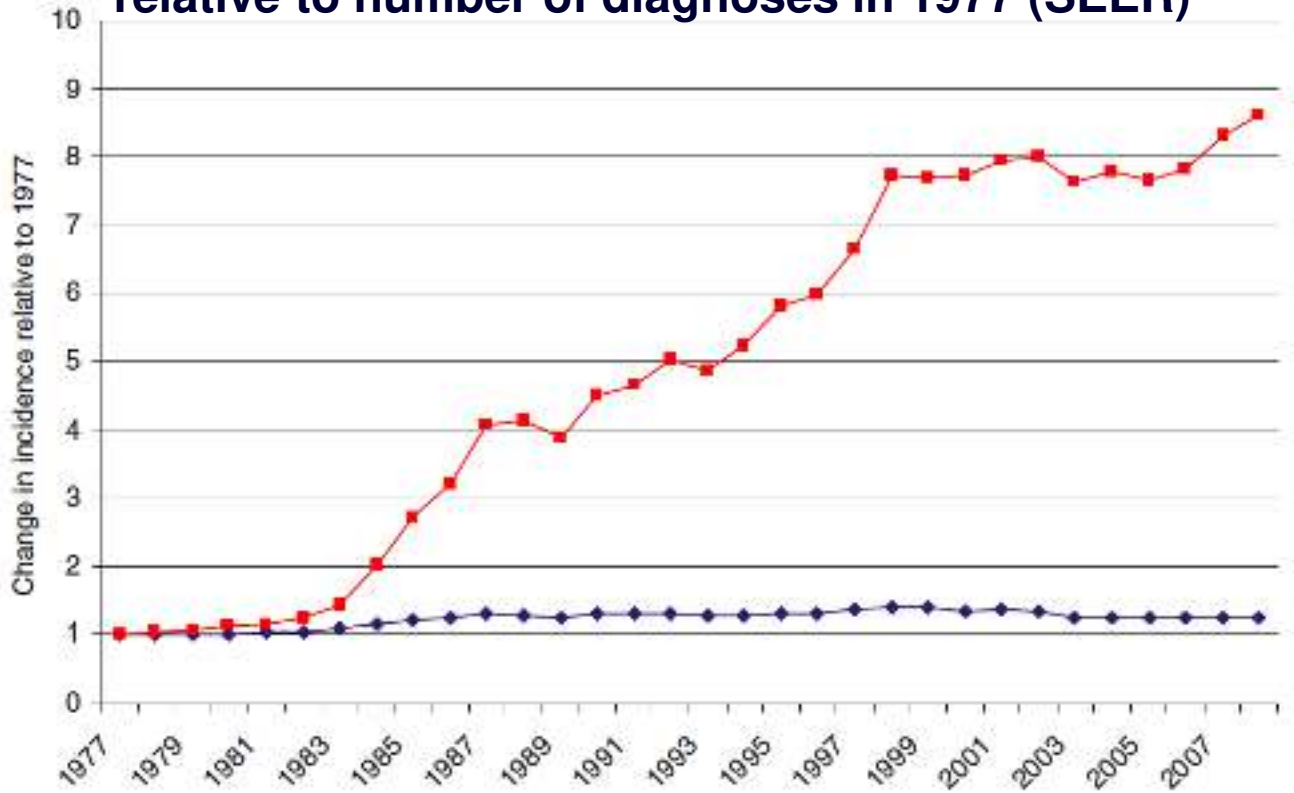
- Presentation
- Distribution in breast
- Pathologic features
- Genetic/molecular alterations
- Clinical behavior

Epidemiology

- Most cases today detected because of microcalcifications on screening mammogram
- Accounts for ~20% of breast “cancers”
- ~61,000 new cases in 2016

Increased Detection of DCIS Due To Mammographic Screening

Age-adjusted incidence of DCIS (red) and invasive breast cancer (blue) relative to number of diagnoses in 1977 (SEER)



Punglia, 2013

How much of this represents “over-diagnosis”?

Reservoir of DCIS in General Population

- Autopsy studies
  - DCIS found in up to 14.7% of women dying of other causes (median 8.9%)
- Reduction mammoplasty studies
  - DCIS found in up to 3% of patients

Reservoir of DCIS in General Population

- These studies undoubtedly underestimate prevalence due to limited sampling
- True prevalence unknown

Natural History of DCIS

	# Benign Bxs Examined	# with DCIS (# with follow-up)	Age (yrs)	Histology	Follow-up	Subsequent Invasive ca
Eusebi, 1994	9,520	55 (55)	27-44	Comedo and non-comedo	1-14 yrs	11 (20%)
Sanders, 2015	11,760	45 (45)	33-74	Low grade	3-42 yrs	16 (36%)
Rosen, 1980	>8,000	30 (15)	Not stated	Non-comedo	1-24 yrs	8 (53%)
Collins, 2005	1,877	13 (13)	41-63	Low, int. and high grades	4-18 yrs	OR 13.5

Caveats:

- Biopsies initially interpreted as benign
- Extent of lesion and adequacy of excision unknown

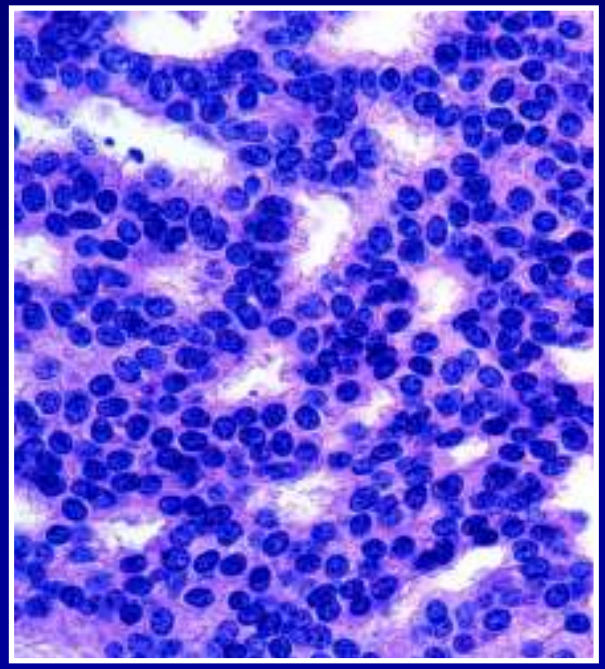
Natural History of DCIS

- Not all DCIS will progress to invasive cancer
- Non-obligate precursor

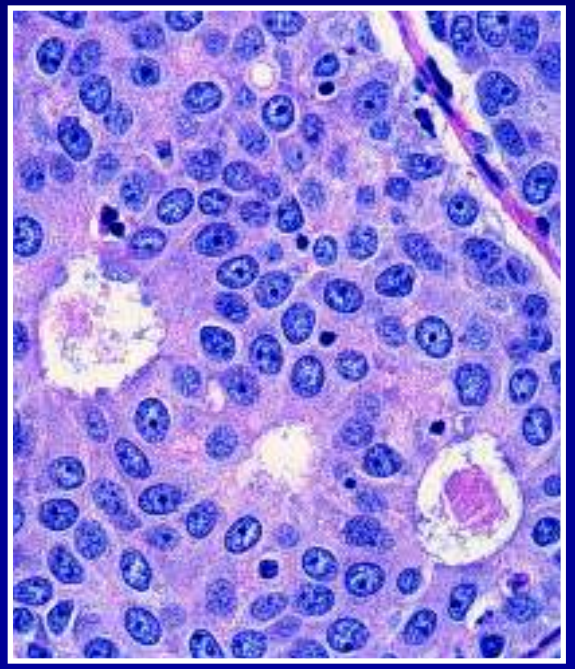
Classification of DCIS

- In current practice, most often classified as low, intermediate or high grade (based on nuclear grade)

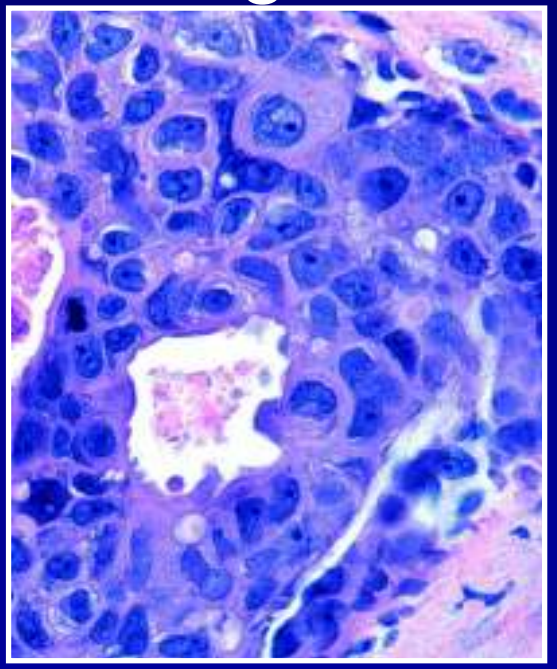
Low



Intermediate



High



Classification of DCIS

- Low grade and high grade DCIS are genetically distinct disorders
  - Low:
    - 16q loss
  - High:
    - 11q, 14q, 8p, 13q losses
    - 17q, 8q, 5p gains
- Low grade DCIS more closely related genetically to LCIS than to high grade DCIS

Classification of DCIS

- Molecular subtypes identified in invasive cancers also observed in DCIS

Molecular Subtypes in DCIS

Using Surrogate IHC Markers*

	NHS (N=263)	UNC (N=229)	CRN (N=371)	Manchester (N=314)
Luminal A	64.4%	65.1%	74.7%	42.6%
Luminal B	13.6%	10.0%	10.8%	28.0%
HER2-E	14.0%	16.6%	10.0%	16.2%
Basal-like/TN	8.0%	8.3%	4.6%	13.1%

**Not all studies used same markers*

Treatment of DCIS

Goals

- Local eradication to prevent the development of invasive breast cancer (prophylactic)
- Ensure sufficient treatment in women at high risk for recurrence/progression
- Avoid over-treatment in women at very low risk for recurrence/progression

Treatment Options

- Mastectomy
- Breast conserving surgery + radiation therapy
- Breast conserving surgery alone
- Endocrine therapy

None offers a survival advantage over the others

Treatment Options

- Confusing to patients
- Treatments offered similar to those for invasive breast cancer, but really not “cancer”
- Clinicians views of DCIS vary
- Patients overestimate their risk
  - >25% think they have at least a moderate chance of DCIS spreading to other parts of the body (Partridge, 2008)

Mastectomy

- Cure rates approach 100%
- Appropriate for patients with extensive disease or those who want to reduce their risk of recurrence to as close to zero as possible
- Does not offer a survival advantage over more conservative treatment
- Over-treatment for most patients encountered in current clinical practice

Breast Conserving Treatment

- Addition of RT to breast conserving surgery reduces risk of local recurrence by ~50% (4 randomized clinical trials)

Breast Conserving Treatment

- Addition of RT to breast conserving surgery reduces risk of local recurrence by ~50% (4 randomized clinical trials)
- ~50% of recurrences are invasive

Breast Conserving Treatment

Role of Endocrine Therapy

- Addition of tamoxifen to breast conserving surgery and RT reduces risk of local recurrence by ~30% (NSABP B-24)
  - Tam benefit limited to women with ER+ DCIS (Allred, 2012)

- Anastrazole
  - Superior to tamoxifen, primarily in women <60 years of age (NSABP B-35)
  - No difference from tamoxifen (IBIS-II DCIS)

Breast Conserving Treatment

- But, the addition of RT and even endocrine therapy is likely over-treatment for some patients

The Continuing DCIS Dilemma

- Only some patients with DCIS will progress to invasive breast cancer

- After decades of research, we still not cannot reproducibly identify which patients are unlikely to progress and, in turn, which patients can be safely managed with excision alone or perhaps even no treatment beyond the diagnostic biopsy

Risk Factors for Local Recurrence

Clinical factors

Young age

Tumor factors

Larger size/extent

High nuclear grade

Comedo necrosis

Volume of DCIS near margin

Molecular subtype

Positive/close margins

Treatment factors

Treatment period

Extent of excision

Use of RT

Use of Endocrine therapy

- **Combinations of factors likely of greater value than individual factors**

Combining Prognostic Factors to Assess Risk

- Informally
- USC/Van Nuys Prognostic Index
- MSKCC Nomogram
- DCIS risk score

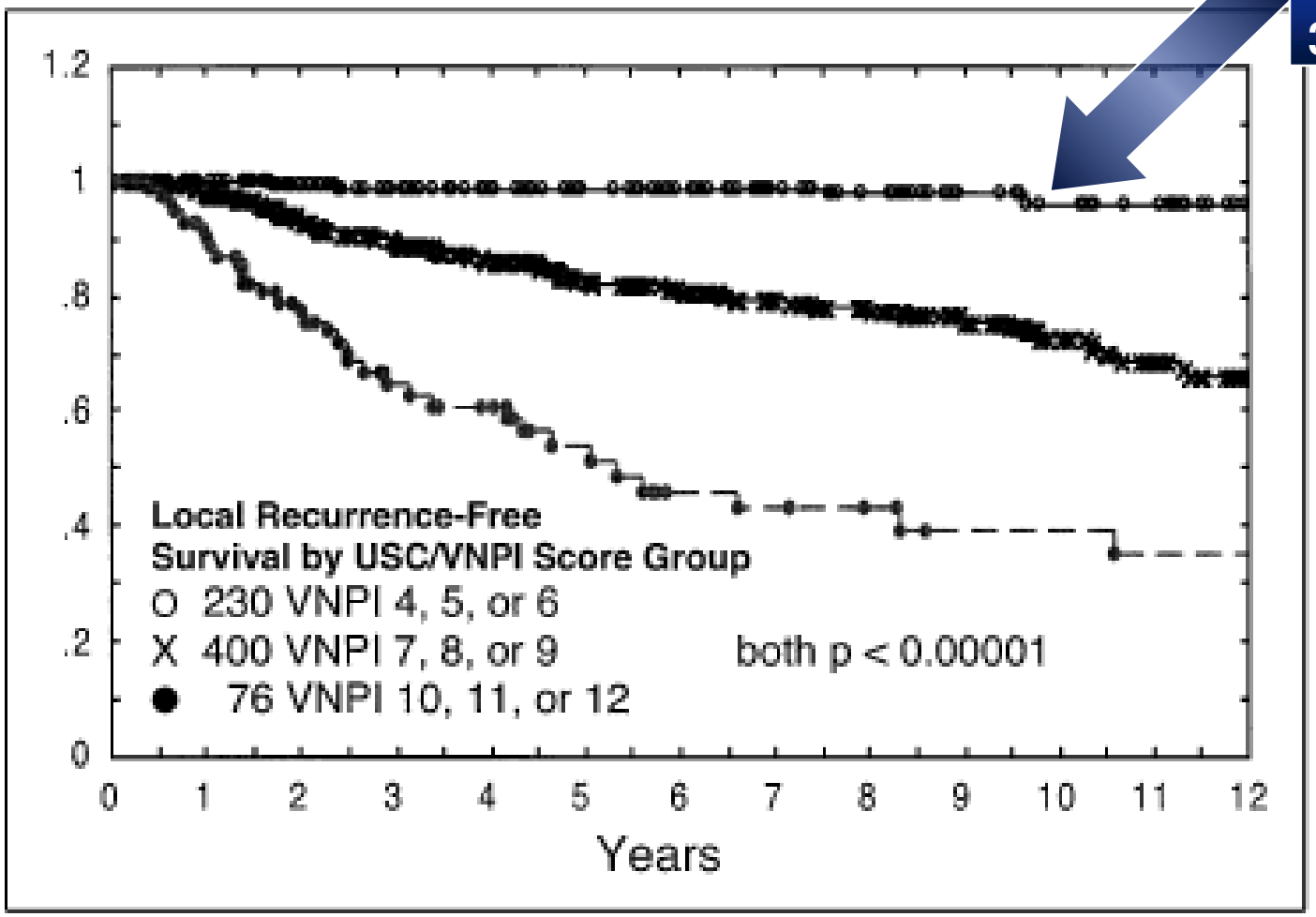
The USC/Van Nuys Prognostic Index

Silverstein, 2003

SCORE	1	2	3
Size (mm)	≤15	16-40	≥41
Margin (mm)	≥10	1-9	<1
Grade	Non-high, no necrosis	Non-high, necrosis	High
Age	>60	40-60	<40

The USC/Van Nuys Prognostic Index

Silverstein, 2003



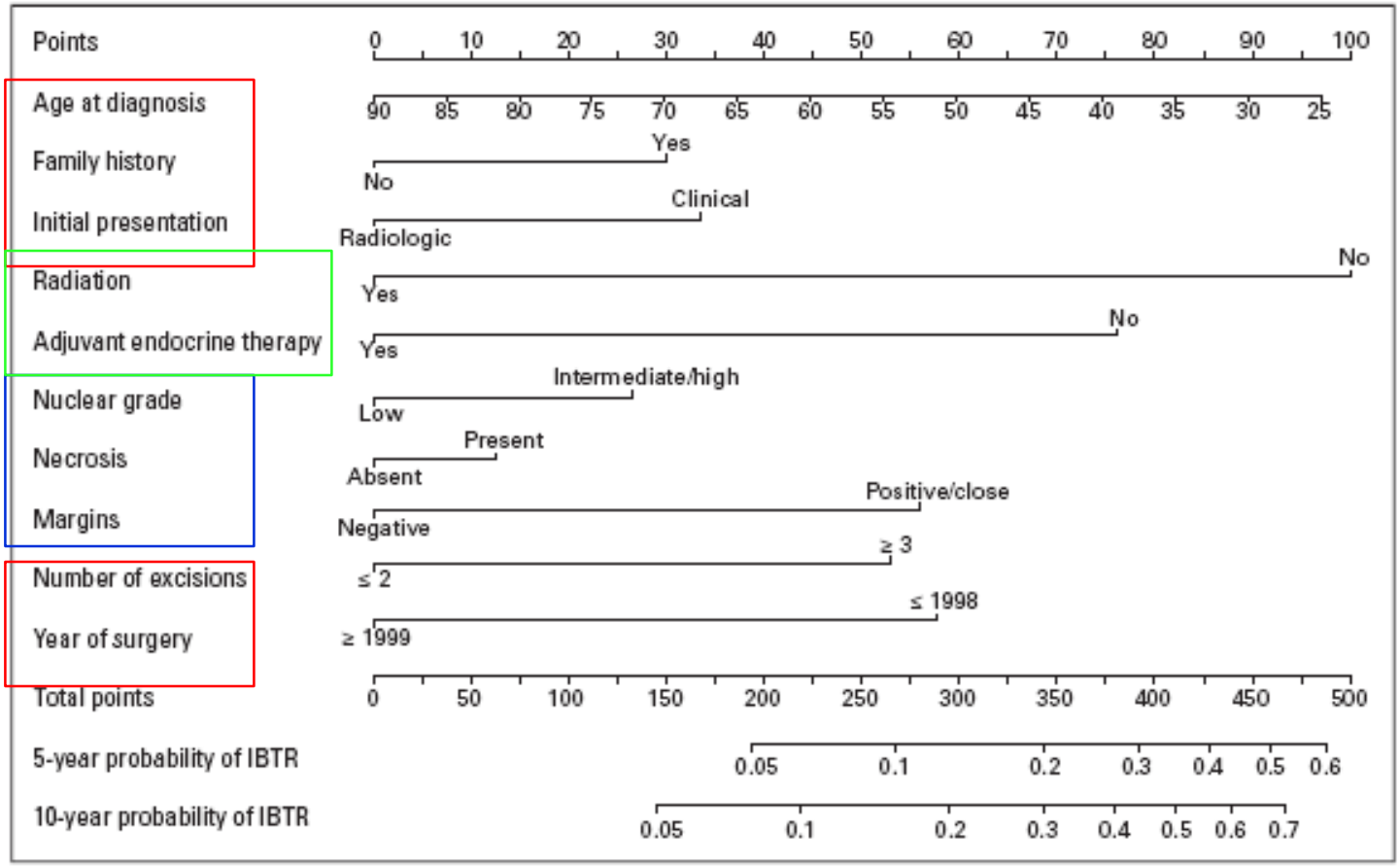
The USC/Van Nuys Prognostic Index
Limitations

- Retrospective study; arbitrary cut points
- Total sequential embedding required to adequately assess size and margins
- Interactions and relative importance of factors unknown
- In practice, can only be applied in minority of cases (largely because of limitations in assessing size)

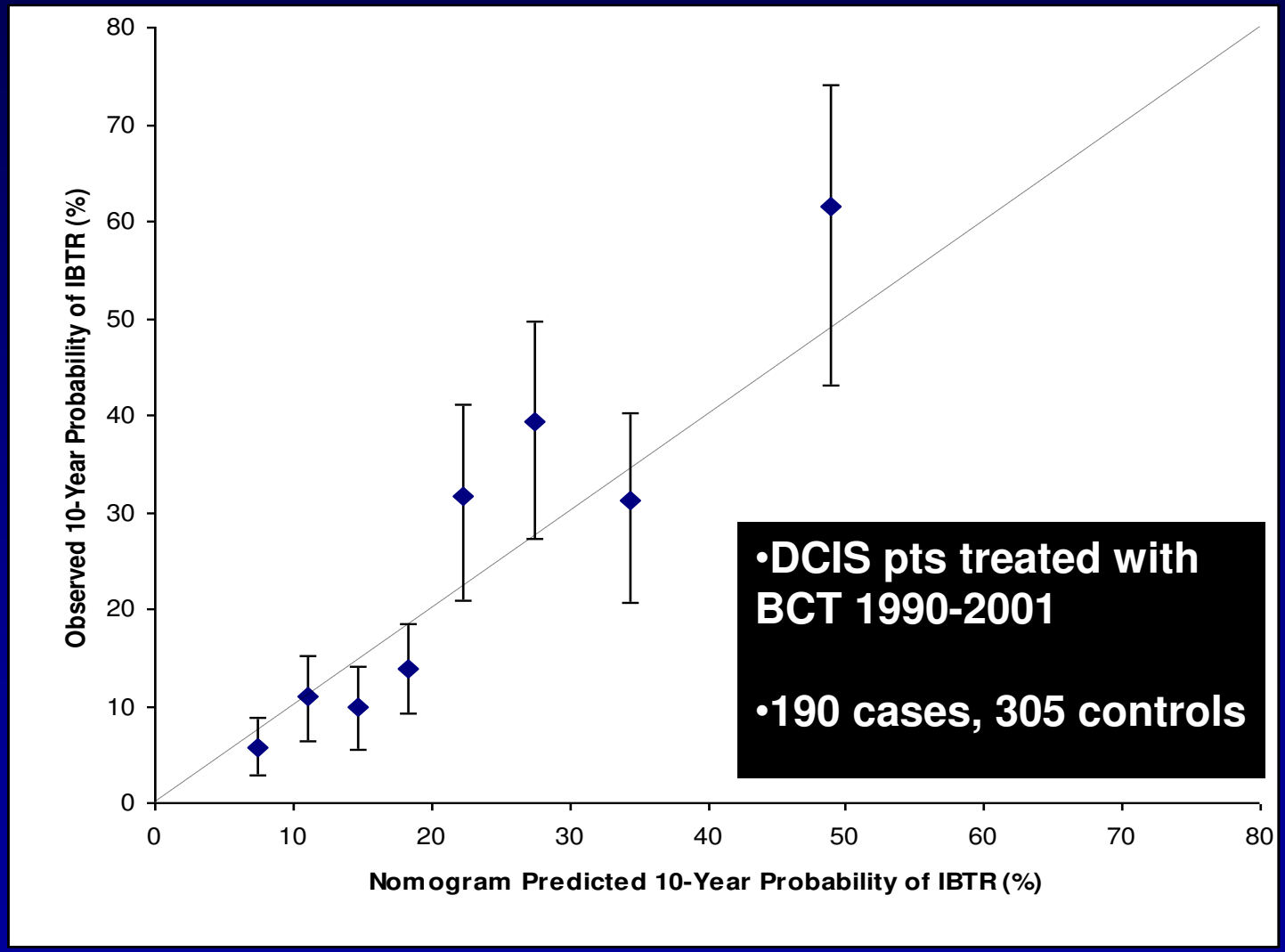
Nomogram for Predicting the Risk of Local Recurrence After Breast-Conserving Surgery for Ductal Carcinoma In Situ

Udo Rudloff, Lindsay M. Jacks, Jessica I. Goldberg, Christine A. Wynveen, Edi Brogi, Sujata Patil, and Kimberly J. Van Zee

JCO, 2010



Observed vs. Nomogram Predicted 10-Year Probability of Local Recurrence in Community-Based Population



Collins, et al, Ann Surg Oncol 2015

DCIS Risk Score

Punglia, BCRT (in press)

- Data from 2762 women with DCIS in NCCN database treated with breast conserving surgery with negative margins used to develop risk score
  - ER status
  - Comedo necrosis
  - Patient age
- Validated in 301 women with DCIS in KPNC database
- C-statistic 0.67 in validation set

Can patients with “low risk” DCIS be safely treated with surgical excision alone?

Prospective Studies

Eight-year update of a prospective study of wide excision alone for small low- or intermediate-grade ductal carcinoma in situ (DCIS)

Julia S. Wong · Yu-Hui Chen · Michele A. Gadd · Rebecca Gelman · Susan C. Lester · Stuart J. Schnitt · Dennis C. Sgroi · Barbara J. Silver · Barbara L. Smith · Susan L. Troyan · Jay R. Harris

Breast Cancer Res Treat 2013

- 158 pts (1995-2002); median F/U 11 yrs
- DCIS ≤2.5 cm, predominant nuclear grade low or intermediate (median size: 8mm)
- Margin width ≥1cm or re-excision without residual DCIS (negative re-excisions in 78%)
- Accrual closed early due to high LR rate
- 10-year estimated cumulative LR rate 15.6%

Local Excision Alone Without Irradiation for Ductal Carcinoma In Situ of the Breast: A Trial of the Eastern Cooperative Oncology Group

Lorie L. Hughes, Molin Wang, David L. Page, Robert Gray, Lawrence J. Solin, Nancy E. Davidson, Mary Ann Lowen, James N. Ingle, Abram Recht, and William C. Wood

JCO 2009

- **Observational study with two arms:**
  - **Low or intermediate grade, ≤2.5 cm (n=565)**
  - **High grade (NG3 + necrosis), ≤1cm (n=105)**
- **Minimum margin width 3mm**
- **Specimen totally, sequentially embedded**
- **Post-excision magnification mammogram negative for microcalcifications**
- **Tamoxifen allowed (~30% in each group)**

ECOG E5194

Local Recurrence at 5, 10 and 12 years

Cohort	5 yrs	10 yrs	12 yrs
Low/Intermediate grade	6%	12.5%	14.4%
High grade	15%	24.6%	24.6%

ECOG E5194

Local Recurrence at 5, 10 and 12 years

Cohort	5 yrs	10 yrs	12 yrs
Low/Intermediate grade	6%	12.5%	14.4%
High grade	15%	24.6%	24.6%

Median size: 6mm

Negative margin width >5mm: 70%

RTOG 9804: A Prospective Randomized Trial for Good-Risk Ductal Carcinoma In Situ Comparing Radiotherapy With Observation

Beryl McCormick, Kathryn Winter, Clifford Hudis, Henry Mark Kuerer, Eileen Rakovitch, Barbara L. Smith, Nour Sneige, Jennifer Moughan, Amit Shah, Isabelle Germain, Alan C. Hartford, Afshin Rashtian, Eleanor M. Walker, Albert Yuen, Eric A. Strom, Jeannette L. Wilcox, Laura A. Vallow, William Small Jr, Anthony T. Pu, Kevin Kerlin, and Julia White

JCO, 2015

- **Same entry criteria as low/intermediate grade arm of ECOG trial**
- **Randomized to conservative surgery alone or with radiation**
- **Whole breast irradiation without boost**
- **Tamoxifen in 62%**

RTOG 9804: A Prospective Randomized Trial for Good-Risk Ductal Carcinoma In Situ Comparing Radiotherapy With Observation

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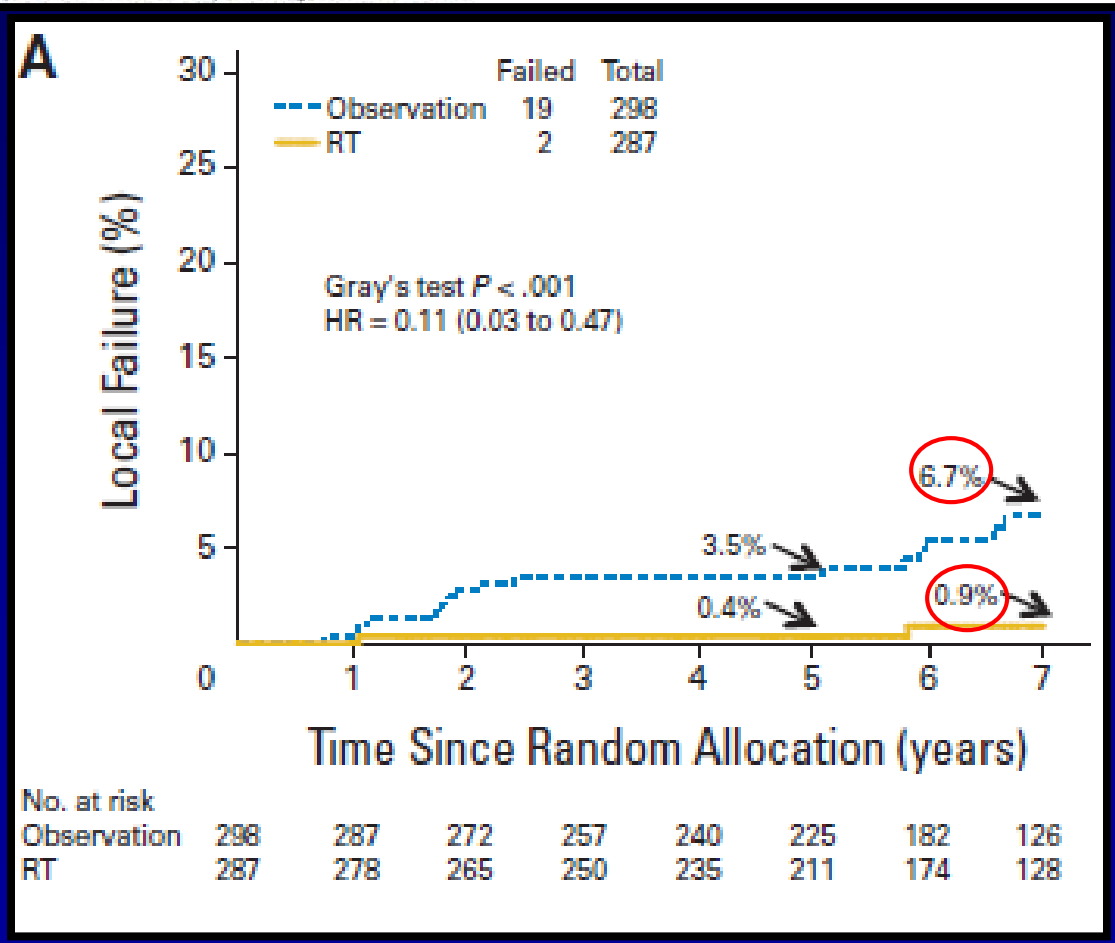
JCO, 2015



RTOG 9804

Two Possible Conclusions

- Even among patients with “low risk” DCIS, breast irradiation significantly reduces the risk of local recurrence
- Among patients with “low risk” DCIS, the 7-yr rate of local recurrence is low, even without radiation (~1%/year)

Local Recurrence Rates in Prospective Studies of "Low Risk" DCIS Treated by Excision Alone

	Local Recurrence Rate
Harvard	15.6% (10 yrs)
ECOG 5194	14.4% (12 yrs)
RTOG 9804	6.7% (7yrs) (~10% at 10 yrs, est)

Can patients with "low risk" DCIS be safely treated with surgical excision alone?

The Bottom Line

- Prospective studies have been unable to identify a subset of patients with "low risk DCIS" treated with surgical excision alone who have local recurrence rates of <10-15% after long-term follow-up based on conventional clinical-pathologic criteria

Can patients with "low risk" DCIS be safely treated with surgical excision alone?

The Bottom Line

- Views of what is an acceptably low local recurrence rate vary
  - RTOG 9804: Local recurrence rate in patients treated with excision alone (~1%/year) similar to that for patients with LCIS

Biomarkers and Risk of Local Recurrence

ER

HER2

COX2

p16

p53

Ki67

Caveolin 1

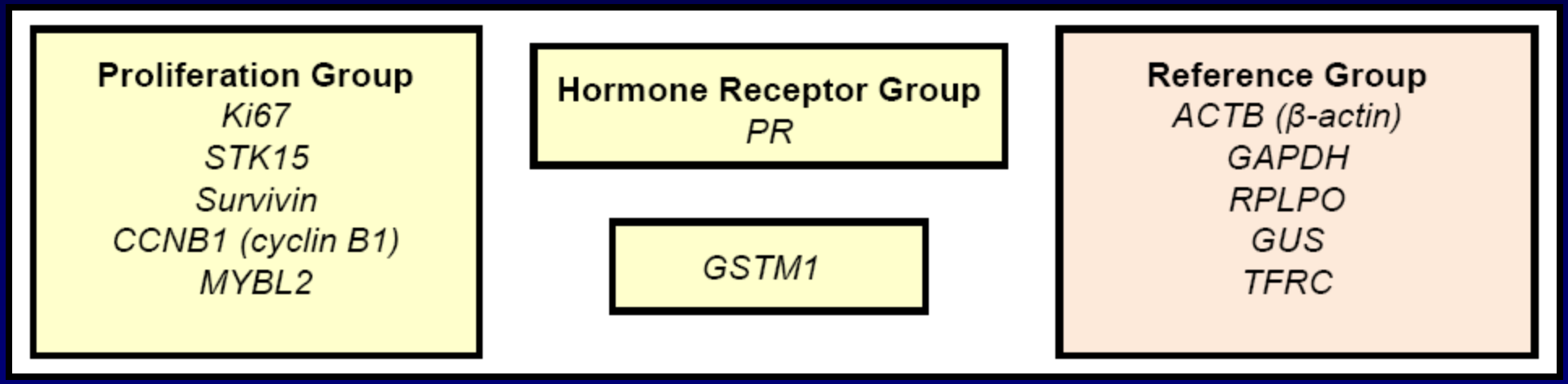
others

Biomarkers and Risk of Local Recurrence

ER

- The only biomarker that should be used in clinical practice (outside the setting of a clinical trial)
- Used as a predictive factor (not as a prognostic factor for local recurrence)

Oncotype DCIS Score



- **ECOG E5194**
  - Selected population (n=327)
  - BCS alone, 1997-2002

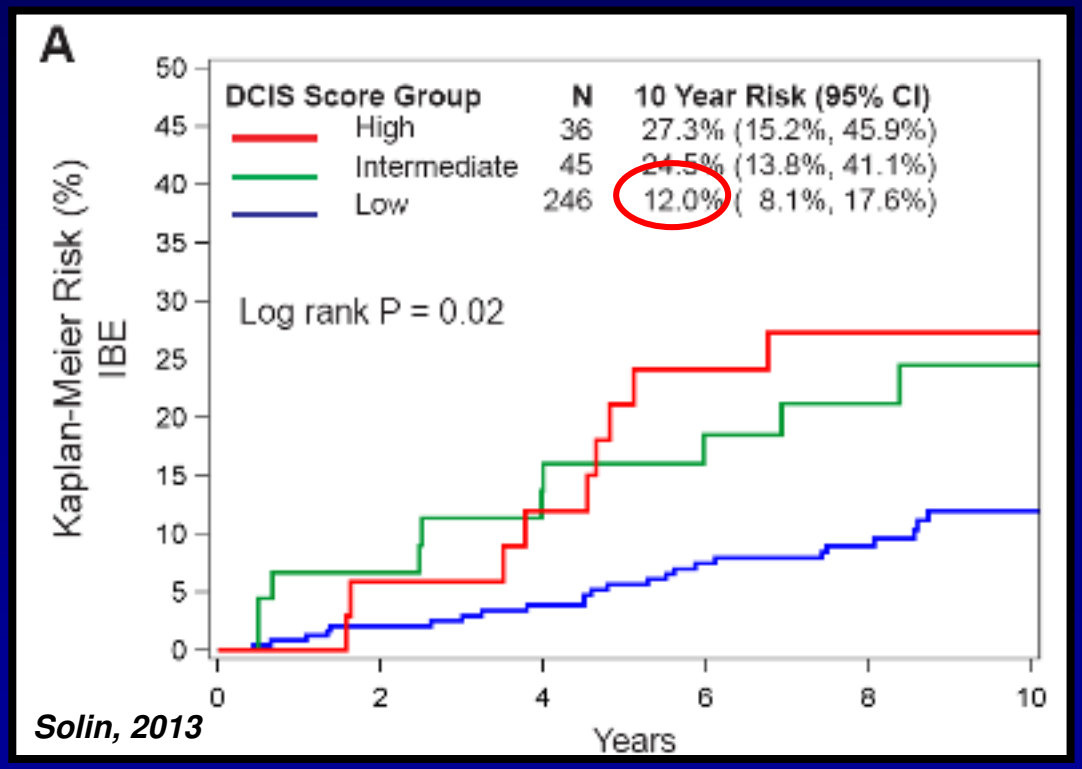
  Solin, 2013
- **Ontario DCIS cohort**
  - General population-based cohort (n=571)
  - BCS alone, 1994-2003

  Rakovitch, 2015

Oncotype DCIS Score

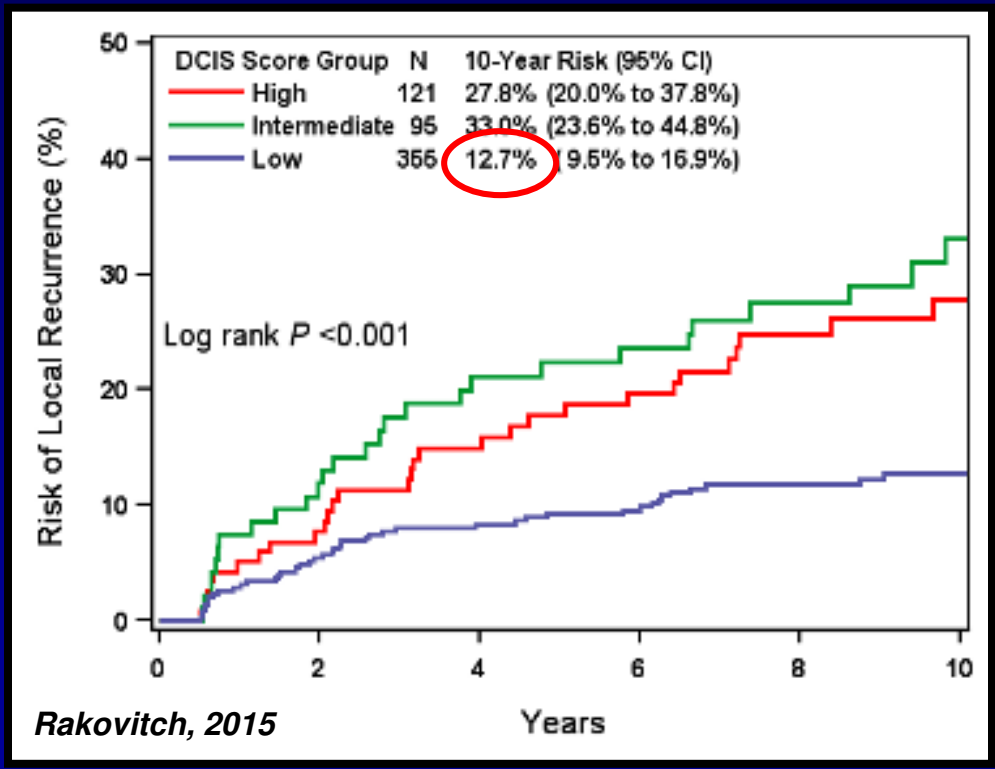
Ipsilateral Breast Events

E5194



Solin, 2013

Ontario DCIS Cohort



Rakovitch, 2015

Integrating DCIS Score (DS) with Clinico-pathologic Factors

Rakovitch, ASCO 2017

Meta-analysis of data from E51994 and Ontario DCIS Cohort (773 pts)

10 yr LR rates (%)

Tumor Size	Age	Low Risk DS	Intermediate Risk DS	High Risk DS
≤1 cm	≥50	7.2	11.3	14.6
	<50	10.2	15.8	19.6
1.1-2.5 cm	≥50	10.1	13.9	19.5
	<50	14.5	18.9	23.2

Other Management Strategies Currently Under Study

Two Ends of the Spectrum

- Trastuzumab for HER2+ DCIS
  - NSABP B-43 (radiation vs radiation plus concurrent trastuzumab [2 doses] after lumpectomy)
- Active surveillance for "low risk" DCIS

Active Surveillance Trials for DCIS

	COMET	LORIS[a]	LORD
Inclusion criteria			
Age (year)	≥40	≥46	≥45
Nuclear grade	Low and intermediate	Low and intermediate	Low
Morphology	Calcifications only	Calcifications only	Calcifications only
Hormone receptor status	ER and/or PR positive, plus HER2 negative if performed	N/A	N/A
Biopsy technique	VACB and/or surgical biopsy	At least 12 gauge VACB and/or surgical biopsy	6 samples with 8–9 gauge or 12 samples with 10–11 gauge VACB
Exclusion criteria			
History of cancer	Exclude if invasive breast cancer	Exclude if invasive breast cancer or ipsilateral DCIS	Exclude if any cancer except in situ of the cervix or basal carcinoma of the skin
Symptomatic	Exclude	Exclude	Exclude
Comedonecrosis	Exclude	Exclude	N/A
Synchronous invasive cancer	Exclude	Exclude	Exclude
Bilateral DCIS at presentation	Include	Include	Exclude
High risk	Include	Exclude if high risk per NICE guidelines[23]	Exclude if family with BRCA 1/2 mutation
History of chemoprevention	Exclude	N/A	N/A

Grimm, 2017

LORIS Trial

- Prospective randomized non-inferiority trial comparing surgical excision with active surveillance (annual mammograms for 10 years) for women with low risk DCIS
- Planned accrual 932 patients over 6 years
- Primary endpoint: Development of ipsilateral invasive cancer

LORIS Trial

- **Eligibility:**
  - **Age ≥46 yrs**
  - **Mammographically-detected or incidental DCIS**
  - **Low risk DCIS on 11g vacuum assisted needle biopsy, confirmed by central pathology review**
    - **Low to intermediate nuclear grade**
    - **No comedo necrosis**

LORIS
The Low Risk DCIS Trial

The COMET Trial

(Comparison of Operative vs Medical Endocrine Therapy)
PI: Shelley Hwang

- **Prospective randomized non-inferiority trial comparing guideline concordant care with active surveillance for women with low risk DCIS on CNB**
  - **Age ≥ 40 years**
  - **Low or intermediate grade DCIS**
  - **No comedo necrosis**
  - **ER+ and/or PR+**
  - **2 pathologists agree on diagnosis**

The COMET Trial

(Comparison of Operative vs Medical Endocrine Therapy)
PI: Shelley Hwang

- Patients in both groups offered option of endocrine therapy
- Planned accrual: 892 pts at 100 sites over 2 years
- Primary endpoint: Development of ipsilateral invasive cancer within 2 years

Active Surveillance Trials for DCIS

	COMET	LORIS[a]	LORD
Inclusion criteria			
Age (year)	≥40	≥46	≥45
Nuclear grade	Low and intermediate	Low and intermediate	Low
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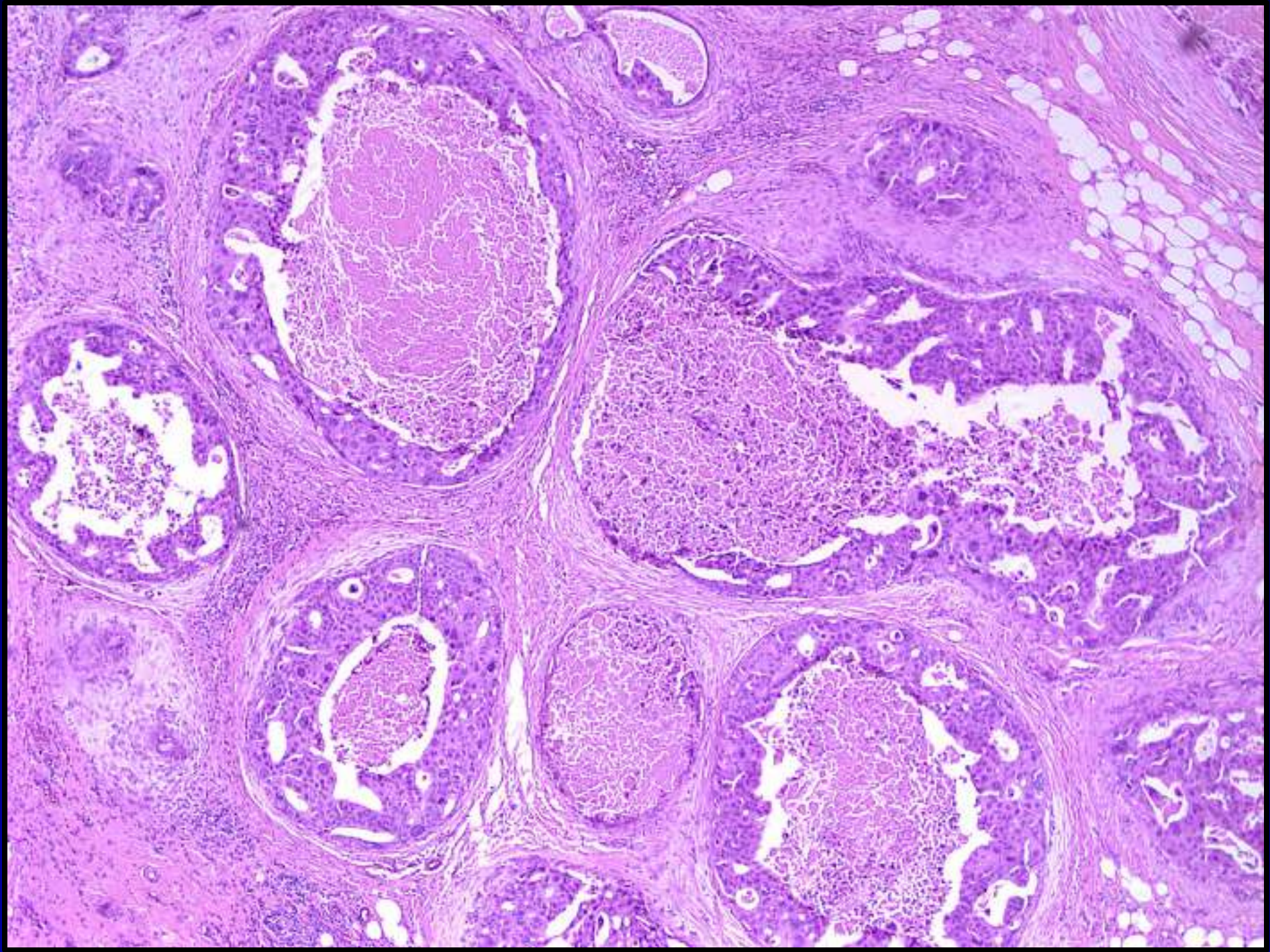
Grimm, 2017

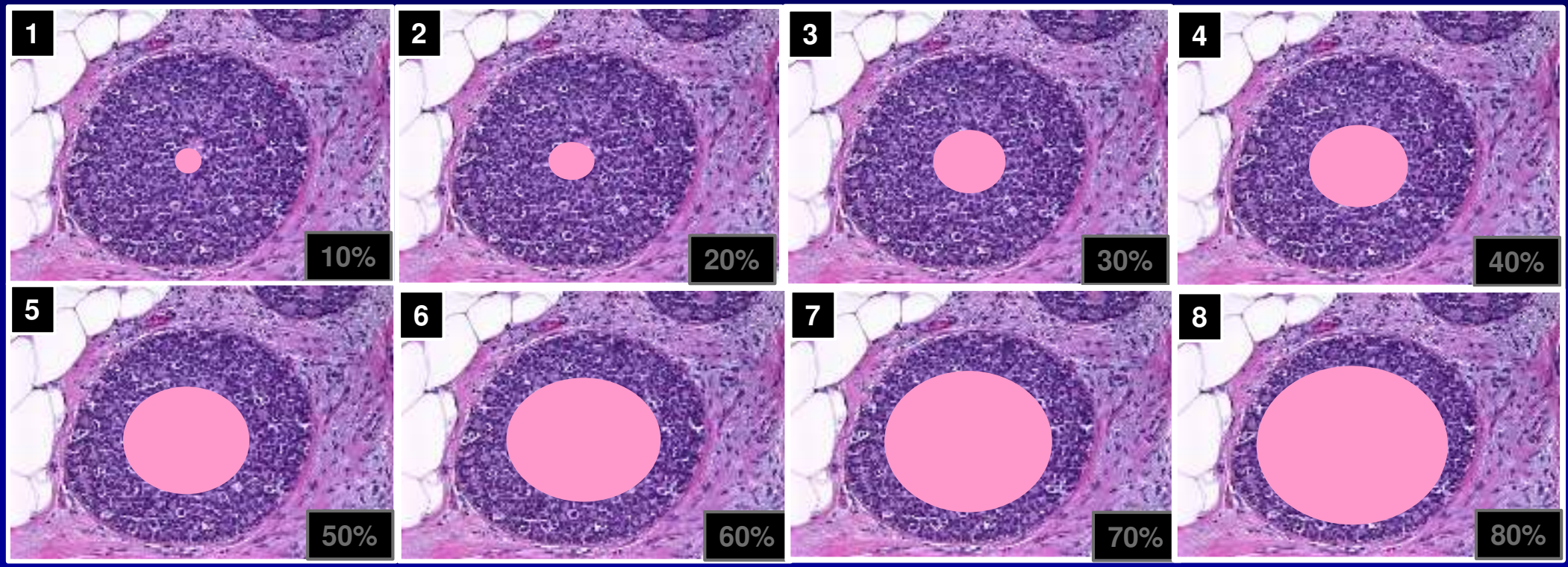


Variability in Diagnostic Threshold for Comedo Necrosis

Harrison, USCAP 2018

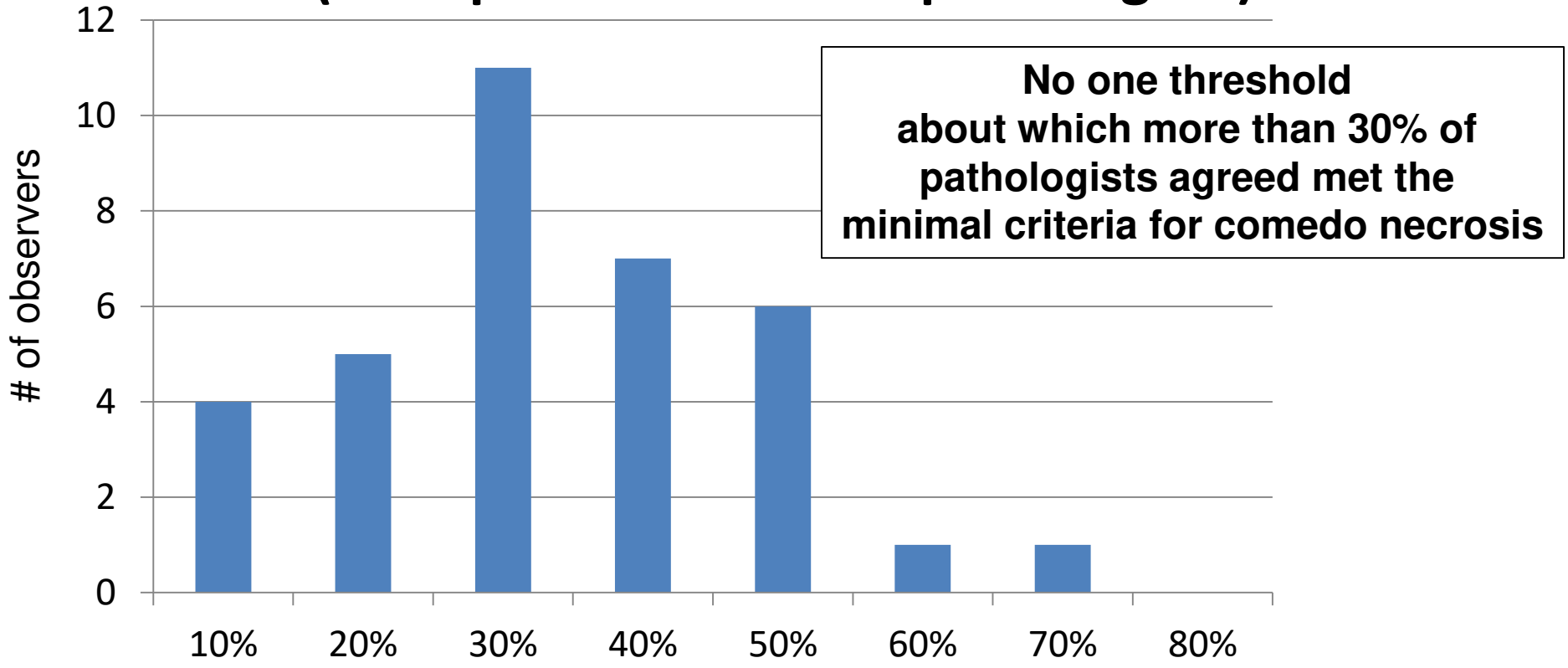
- Eight replicate histologic images of a duct with low nuclear grade, solid pattern DCIS
- To simulate necrosis, superimposed pink circle of various diameters representing 10-80% of duct diameter in 10% increments
- 35 experienced breast pathologists

Which image represents minimum amount of necrosis required for “comedo” necrosis?



Proportion of Duct Diameter With Necrosis Required for a Diagnosis of "Comedo Necrosis"

(35 experienced breast pathologists)



Implications for Active Surveillance Trials

- Low threshold: Exclude potentially suitable candidates
- High threshold: Include patients who may not be appropriate study subjects
- Definition of “comedo necrosis” requires standardization

Upgrade of DCIS to Invasive Cancer

- How often does CNB underestimate invasion in patients with DCIS?

Ductal Carcinoma in Situ at Core-Needle Biopsy: Meta-Analysis of Underestimation and Predictors of Invasive Breast Cancer[1]

Brennan, Radiology, 2011

- **52 studies**
- **7350 cases of DCIS on core biopsy followed by excision**
- **Pooled underestimation (upgrade) rate: 25.9%**

Ductal Carcinoma in Situ at Core-Needle Biopsy: Meta-Analysis of Underestimation and Predictors of Invasive Breast Cancer[1]

Brennan, Radiology, 2011

- **But, what is the underestimation (upgrade) rate for patients who would be eligible for the trials of active surveillance?**

Concordance between vacuum assisted biopsy and postoperative histology: Implications for the proposed Low Risk DCIS Trial (LORIS)

S. Soumian [a,b,e], E.T. Verghese [c,e], M. Booth [c], N. Sharma [c], S. Chaudhri [a], S. Bradley [a], S. Umranikar [a], R.A. Millican-Slater [c], A.M. Hanby [c,d], A. Francis [a,*]

EJSO, 2013

225 DCIS cases diagnosed by VAB, 2001-2010

Table 3
Details of upgrade rates for patients eligible for Low Risk DCIS Trial (LORIS).

DCIS	Number *n* (%)	Upgrade to intermediate grade DCIS *n* (%)	Upgrade to high grade DCIS *n* (%)	Upgrade to invasive cancer *n* (%)
LORIS cases*	19 (100)	3 (16)	1 (5)	0 (0)

***includes only low grade cases**

Do LORIS Trial Eligibility Criteria Identify a Ductal Carcinoma In Situ Patient Population at Low Risk of Upgrade to Invasive Carcinoma?

Melissa Pilewskie, MD[1], Michelle Stempel, MPH[1], Hope Rosenfeld, BS[1], Anne Eaton, MS[2], Kimberly J. Van Zee, MS, MD[1], and Monica Morrow, MD[1]

Ann Surg Oncol 2016

- **296 LORIS-eligible patients identified between 2009-2012** (≥46 yo, non-high grade DCIS on vacuum assisted CNB for screen-detected mammographic microcalcs)
  - Invasive carcinoma at surgery in **58 (20%)**
    - » 31% T1b or larger
    - » 21% high grade
    - » 3% TNBC
    - » 9% HER2+
    - » 5% node+
    - » 90% received RT
    - » 18% recommended for chemo

Surgical Upstaging Rates for Vacuum Assisted Biopsy Proven DCIS: Implications for Active Surveillance Trials

Lars J. Grimm, MD, MHS[1], Marc D. Ryser, PhD[2], Ann H. Partridge, MD, MPH[3], Alastair M. Thompson, MD[4], Jeremy S. Thomas, MBBS[5], Jelle Wesseling, MD, PhD[6], and E. Shelley Hwang, MD, MPH[7]

Ann Surg Oncol, 2017

TABLE 2 Patient demographics and DCIS characteristics for the entire study population and cases eligible for the COMET, LORIS, and LORD trials (eligibility criteria for each trial are listed in Table 1)

	Entire population ($n = 307$) no. (%)	COMET ($n = 81$) no. (%)	LORIS ($n = 74$) no. (%)	LORD ($n = 10$) no. (%)
Age (year, mean [range])	60.3 (33.5–86.7)	61.3 (44.1–86.7)	62.5 (44.1–86.7)	63.1 (51.5–80.6)
Race				
Caucasian	198 (64)	54 (67)	52 (70)	6 (60)
Black	96 (31)	27 (33)	22 (30)	4 (40)
Other	13 (5)	0 (0)	0 (0)	0 (0)
ER-positive	249 (81)	80 (99)	71 (96)	10 (100)
PR-positive	216 (70)	77 (95)	68 (92)	10 (100)
Nuclear grade				
Low	15 (5)	12 (15)	12 (16)	10 (100)
Intermediate	95 (31)	69 (85)	62 (84)	0 (0)
High	197 (64)	0 (0)	0 (0)	0 (0)
Comedonecrosis	157 (51)	0 (0)	0 (0)	0 (0)
Upgrade to high-grade DCIS	10 (3)	6 (7)	5 (7)	1 (10)
Upstage to invasive disease	53 (17)	5 (6)	5 (7)	1 (10)

What drives the progression of DCIS to invasive breast cancer?

Gene Expression and Genomic Alterations in DCIS and Invasive Cancer

- Gene expression
  - Very few genes differentially expressed in DCIS and invasive cancer
  - No clear "DCIS" or "invasive" signature
- Genomic alterations
  - Genomic profiles of DCIS and invasive cancer generally similar for lesions of equivalent grade
  - Low grade DCIS more similar to low grade invasive cancer than to high grade DCIS

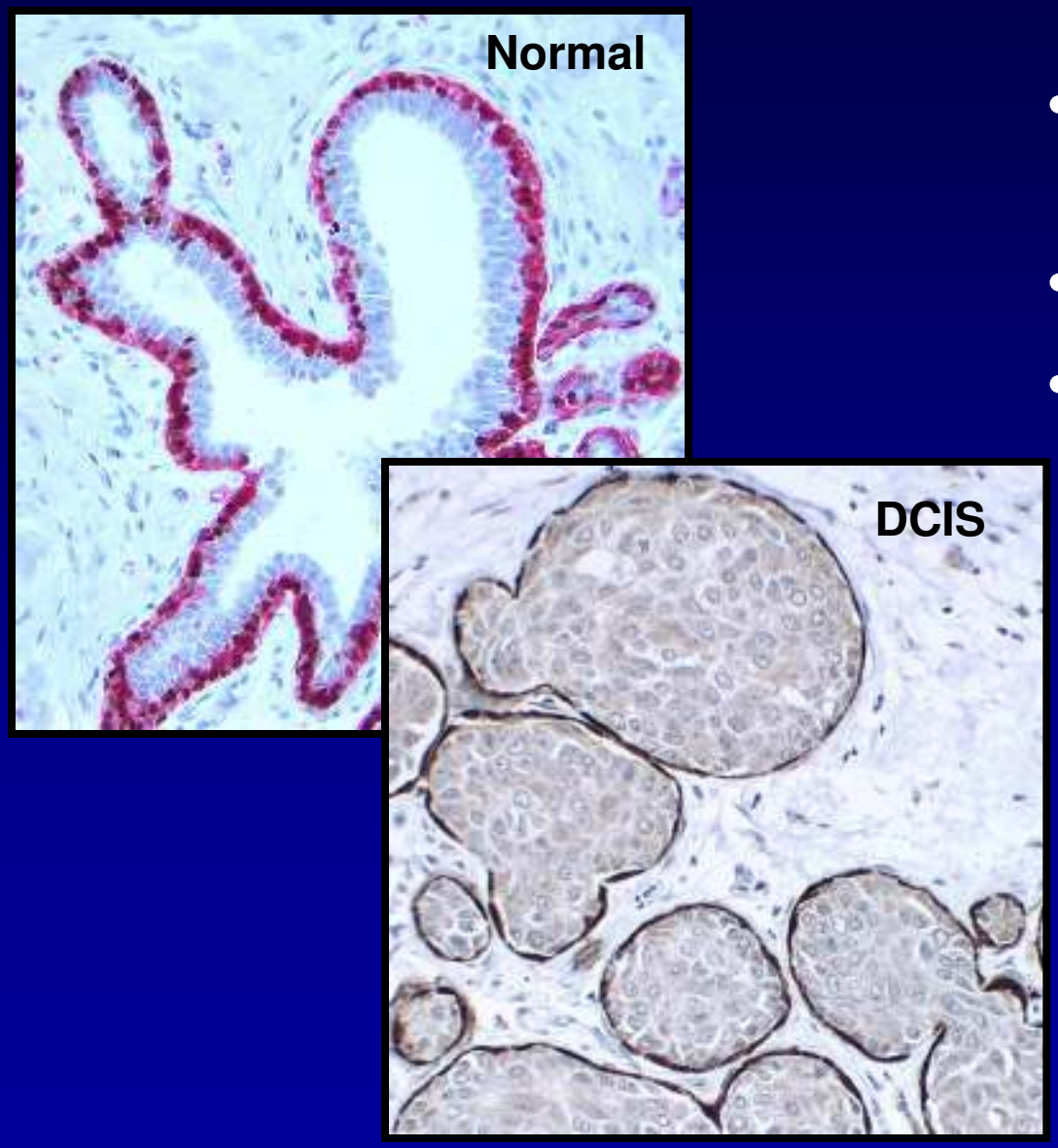
Possible Explanations

- Only a very small number of genes associated with the progression of DCIS to invasive breast cancer
- Progression of DCIS to invasive breast cancer strongly dependent upon epigenetic and/or microenvironmental factors (perhaps even more so than on molecular/genetic changes in DCIS cells themselves)

Potential Microenvironmental Factors in DCIS Progression

- Myoepithelial cells
- Stroma
- Immune cells (TILs)

Myoepithelial Cells

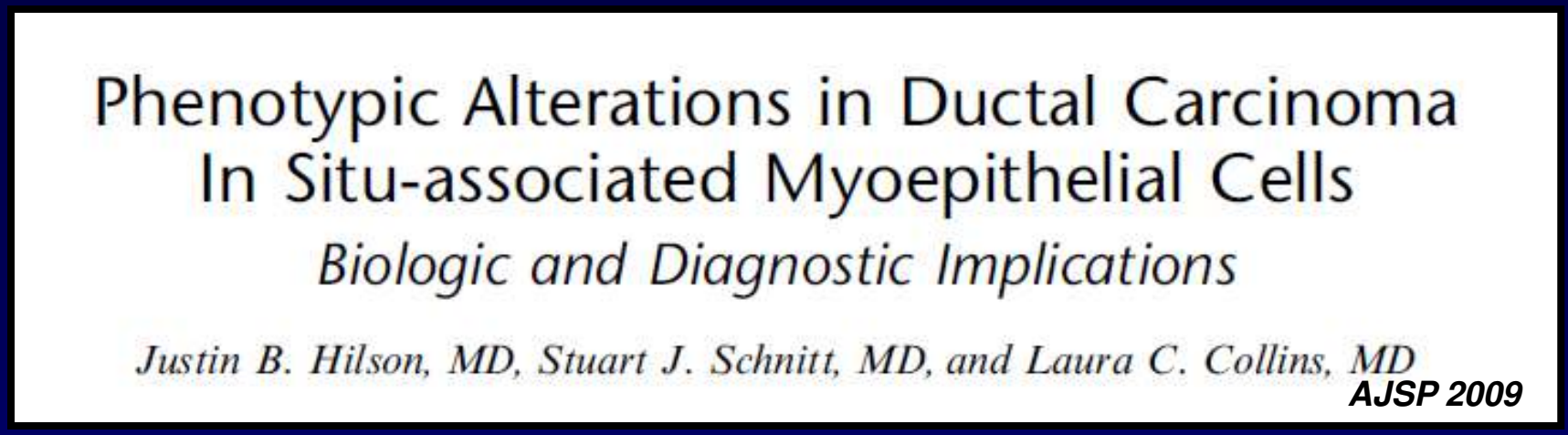


- Surround entire ductal lobular system
- Physiologic functions
- Natural tumor suppressor function
  - Maintenance of basement membrane
  - Physical barrier between benign epithelium/DCIS and stroma
  - Paracrine effects on epithelial cells, stromal cells and endothelial cells

Myoepithelial Cells Associated with DCIS

Polyak Lab, DFCI

- Compared with MEC from reduction mammaplasty specimens, DCIS-associated MEC show:
  - Downregulation of a variety of genes involved in normal functions
    - Oxytocin receptor, laminin, thrombospondin
  - Upregulation of genes for chemokines that enhance epithelial cell proliferation, migration, invasion
    - SDF1/CXCL12 and CXCL14
  - Epigenetic changes



- **DCIS-associated MEC often show immunophenotypic alterations when compare with normal MEC**
- **?Altered state of differentiation**
- **?Altered tumor suppressor capability**

Myoepithelial Cell Differentiation Markers in Ductal Carcinoma *in Situ* Progression

Tanya D. Russell,* Sonali Jindal,† Samiat Agunbiade,* Dexiang Gao,‡ Megan Troxell,§¶ Virginia F. Borges,*‖ and Pepper Schedin*†¶

Am J Pathol 2015

- **MCF10DCIS.com cells injected into mammary ducts of immunodeficient mice**
  - **Alterations in DCIS-associated MEC occur *before* invasion**
  - **Reduced expression of p63, calponin and SMA**
    - **p63 > calponin > SMA**
- **Similar findings in limited number of human DCIS samples**

Tumor-associated myoepithelial cells promote the invasive progression of ductal carcinoma *in situ* through activation of TGFβ signaling

Received for publication, January 3, 2017, and in revised form, May 12, 2017 Published, Papers in Press, May 16, 2017, DOI 10.1074/jbc.M117.775080

Pang-Kuo Lo[‡], Yongshu Zhang[‡], Yuan Yao[‡], Benjamin Wolfson[‡], Justine Yu[‡], Shu-Yan Han[‡§], Nadire Duru[‡], and Qun Zhou[‡1]

J Biol Chem, 2017

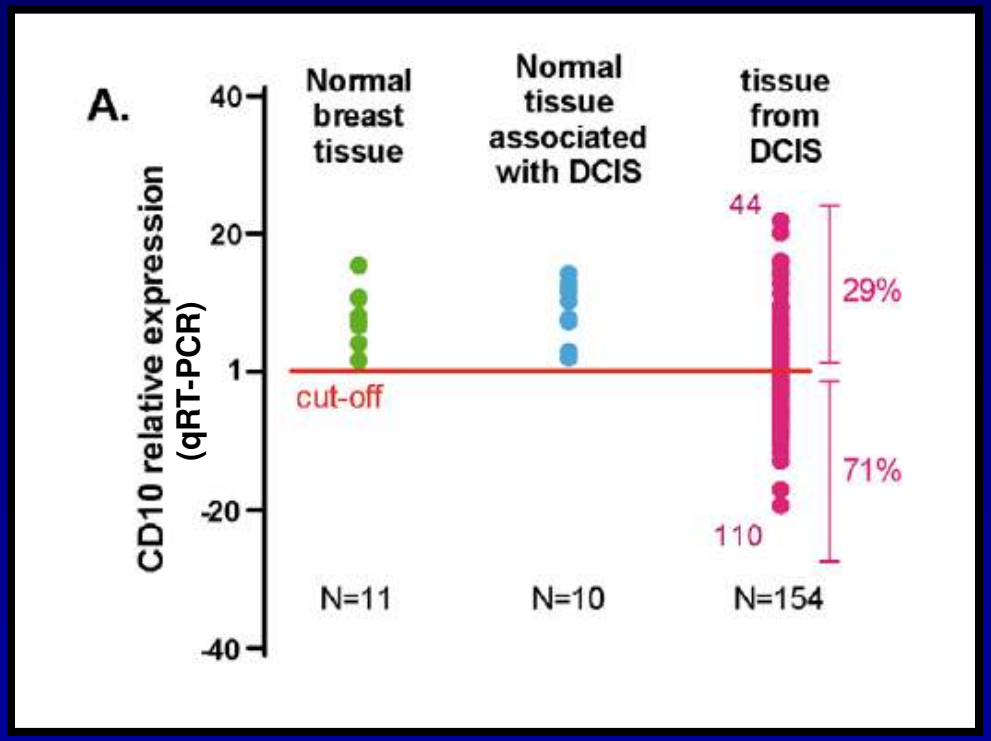
- **Co-culture of tumor-associated MEC and MCF10DCIS.com cells**
  - **Stimulated MEC to secrete TGFβ-1 resulting in activation of TGFβ/Smads pathway in DCIS cells**
    - » **Promoted EMT, basal-like phenotype, stem cell properties, migration and invasiveness of DCIS cells**
  - **miR-10b-5p downstream mediator of TGFβ signaling**
- **Xenografts**
  - **Tumor-associated MEC enhanced DCIS to IDC progression**

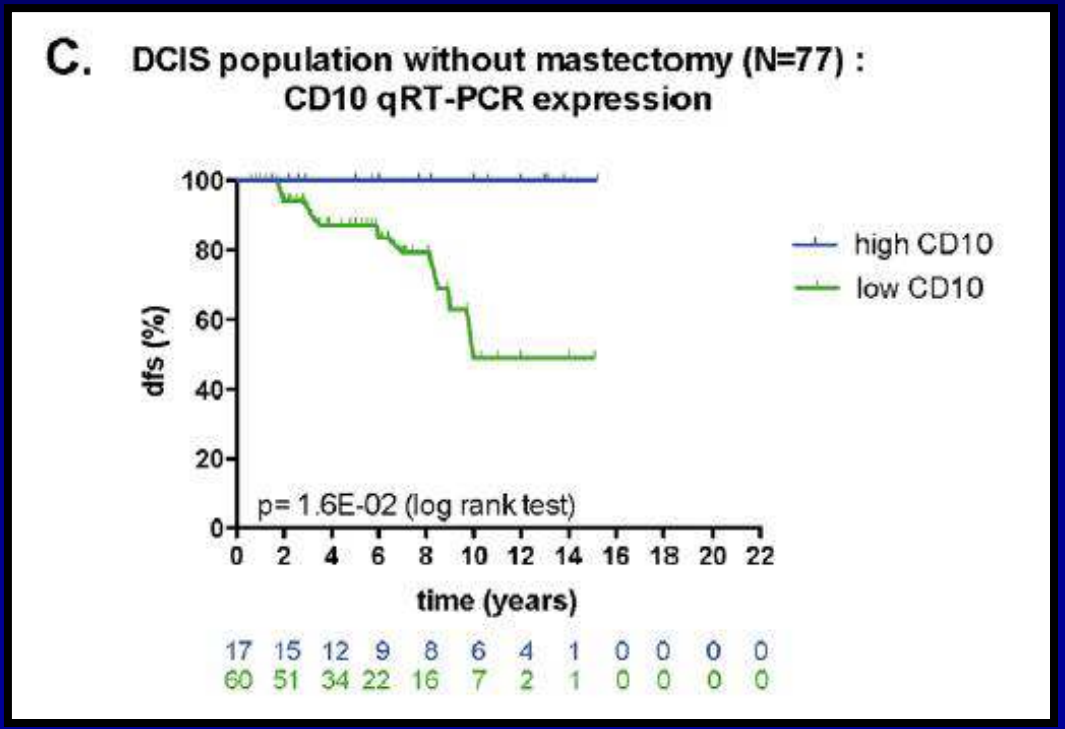
Is Altered Expression of Myoepithelial Cell Markers Clinically Important?

Low CD10 mRNA Expression Identifies High-Risk Ductal Carcinoma In Situ (DCIS)

Jérôme Toussaint[1☯], Virginie Durbecq[1☯], Sevilay Altintas[3], Valérie Doriath[2], Ghizlane Rouas[1], Marianne Paesmans[1], Philippe Bedard[1], Benjamin Haibe-Kains[1], Wiebren A. Tjalma[3], Denis Larsimont[1], Martine Piccart[1,2], Christos Sotiriou[1,2*]

PLoS One, 2010





Independent prognostic factor in MVA: HR 2.39 (95% CI, 1.52-3.76)

Altered Microenvironment Promotes Progression of Preinvasive Breast Cancer: Myoepithelial Expression of αvβ6 Integrin in DCIS Identifies High-risk Patients and Predicts Recurrence

Michael D. Allen[1], Gareth J. Thomas[3], Sarah Clark[1], Marwa M. Dawoud[1], Sabarinath Vallath[1], Sarah J. Payne[1], Jennifer J. Gomm[1], Sally A. Dreger[1], Sarah Dickinson[1], Dylan R. Edwards[4], Caroline J. Pennington[4], Ivana Sestak[2], Jack Cuzick[2], John F. Marshall[1], Ian R. Hart[1], and J. Louise Jones[1]

Clin Cancer Res, 2014

- **Upregulation of αVβ6 integrin in DCIS-associated MEC associated with switch from tumor-suppressor to tumor-promoting activity via TGFβ and MMP9 signaling**
- **Case-control study nested within UK/ANZ cohort (52 case-control pairs)**
  - **High MEC expression of αVβ6 integrin by IHC significantly associated with local recurrence and shorter time to recurrence independent of DCIS size, grade and patient age**

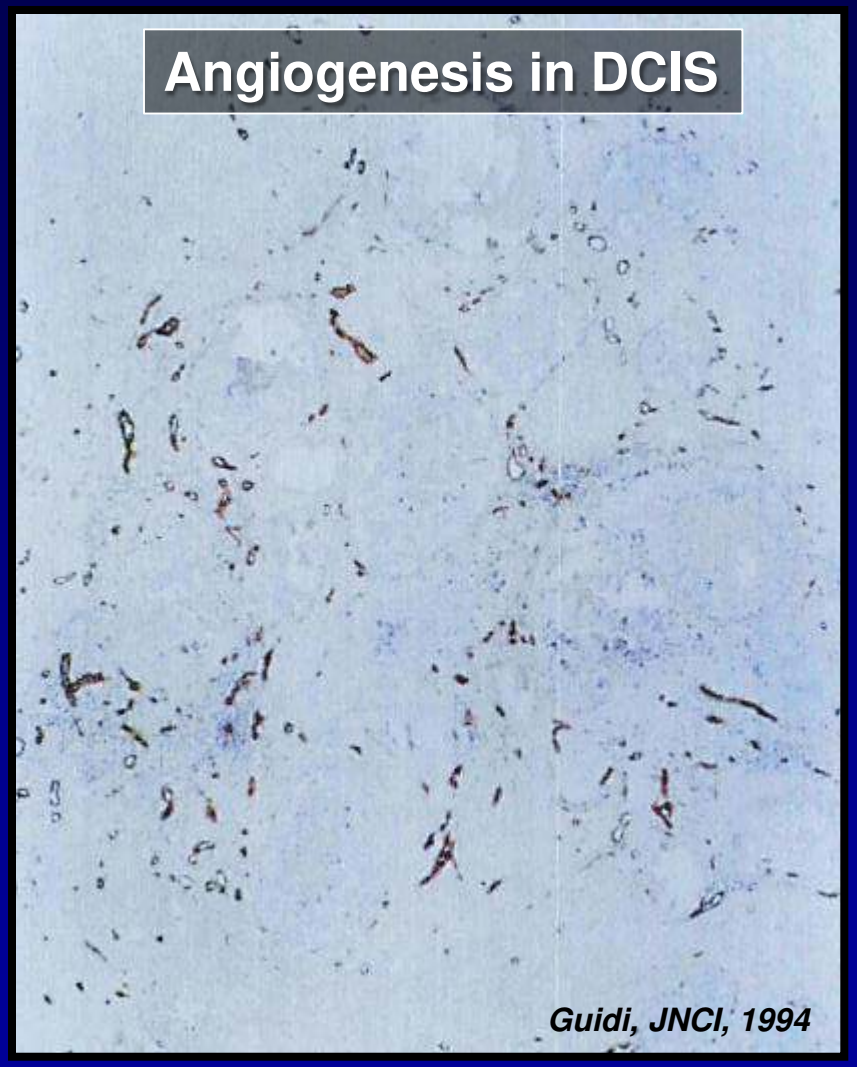
Summary

- Loss of normal myoepithelial cell function may be a key determinant of progression of DCIS to invasive breast cancer
- Identifying the molecular underpinnings of normal myoepithelial cell differentiation and the aberrations that occur in DCIS may identify predictors of invasion and, possibly, targets for prevention

Microenvironmental Factors in DCIS Progression

- Myoepithelial cells
- **Stroma**
- Immune cells (TILs)

Stromal Alterations in DCIS



- Stromal alterations characteristic of invasive cancers are already manifested in some DCIS lesions
  - » Stromal angiogenesis
  - » Increased stromal expression of mRNA for stromal matrix proteins (collagen type I, total fibronectin, ED-A+ fibronectin, versican, decorin, thrombospondin)
- Increased expression of some MMPs

Guidi, JNCI, 1994
Brown, Clin Cancer Res, 1999
Jacobs, Hum Pathol, 2002
Hotary, Genes Dev, 2006

Analysis of stromal signatures in the tumor microenvironment of ductal carcinoma in situ

M. Sharma · A. H. Beck · J. A. Webster · I. Espinosa · K. Montgomery · S. Varma · M. van de Rijn · K. C. Jensen · R. B. West

Breast Cancer Res Treat 2010

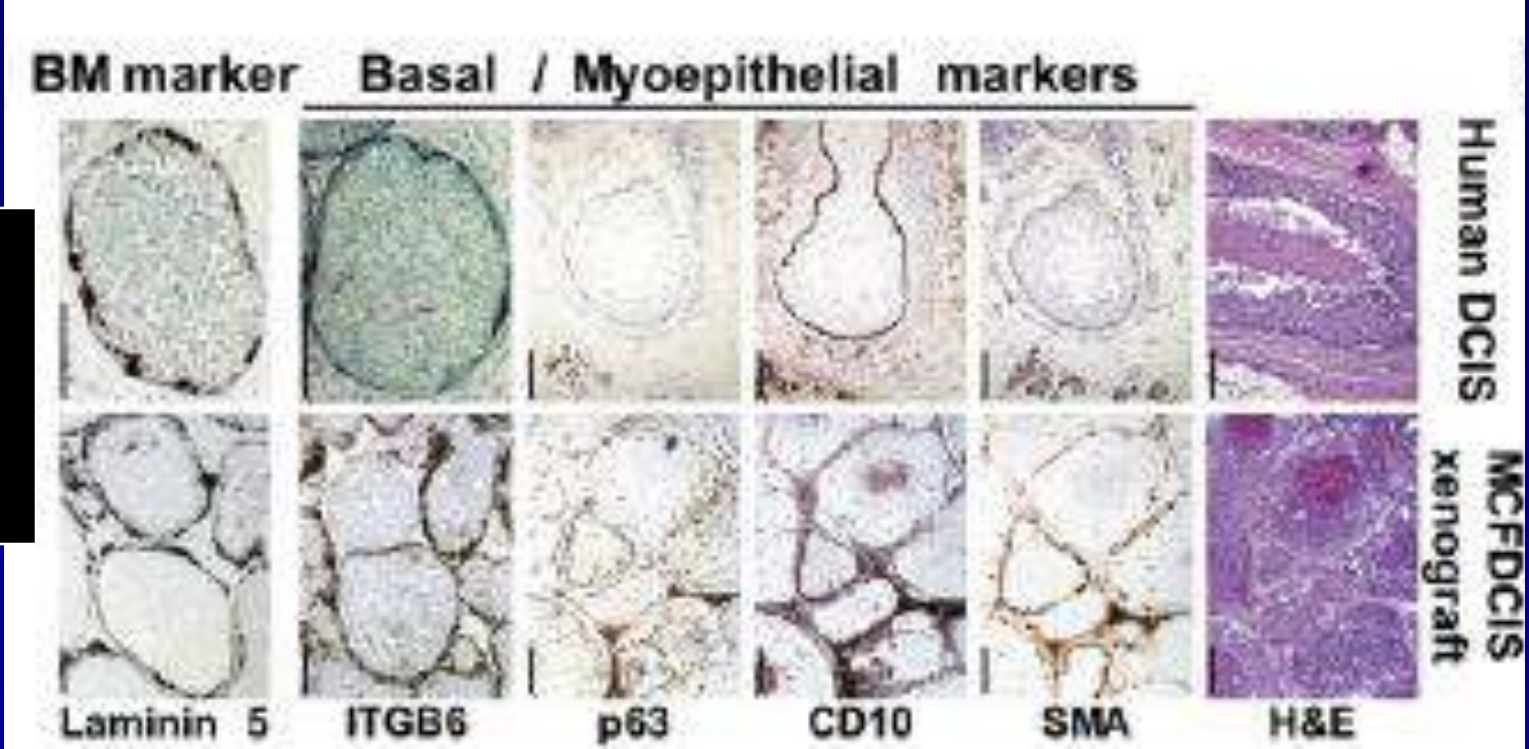
- **Two distinct gene expression signatures identified in stroma of invasive cancers also seen in stroma of DCIS**
  - **Macrophage (CSF1) response**
  - **Fibroblastic (DTF-like) response**
- **Macrophage response signature associated with high grade, ER/PR negative DCIS**
- **Prognostic significance not yet studied**

Regulation of In Situ to Invasive Breast Carcinoma Transition

Cancer Cell, 2008

Min Hu,[1,4] Jun Yao,[1,4] Danielle K. Carroll,[4] Stanislawa Weremowicz,[3,4] Haiyan Chen,[2,5] Daniel Carrasco,[1] Andrea Richardson,[3,4] Shelia Violette,[6] Tatiana Nikolskaya,[7] Yuri Nikolsky,[7] Erica L. Bauerlein,[1,4] William C. Hahn,[1,4] Rebecca S. Gelman,[2,5] Craig Allred,[8] Mina J. Bissell,[10] Stuart Schnitt,[4,9] and Kornelia Polyak[1,4,*]

Injection of MCFDCIS cells into nude mice produces a lesion histologically similar to human DCIS



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Cells Injected into Nude Mice	Histology
MCFDCIS alone	DCIS
MCFDCIS + Normal MEC	DCIS
MCFDCIS + activated fibroblasts	Invasive ca
MCFDCIS + activated fibroblasts + MEC	DCIS

Summary

- In these experimental models, progression to invasive carcinoma and tumor growth
  - promoted by fibroblasts
  - inhibited by MEC
- Results highlight potential importance of microenvironment in breast tumor progression

Microenvironmental Factors in DCIS Progression

- Myoepithelial cells
- Stroma
- **Immune cells (TILs)**

Immune Microenvironment of DCIS

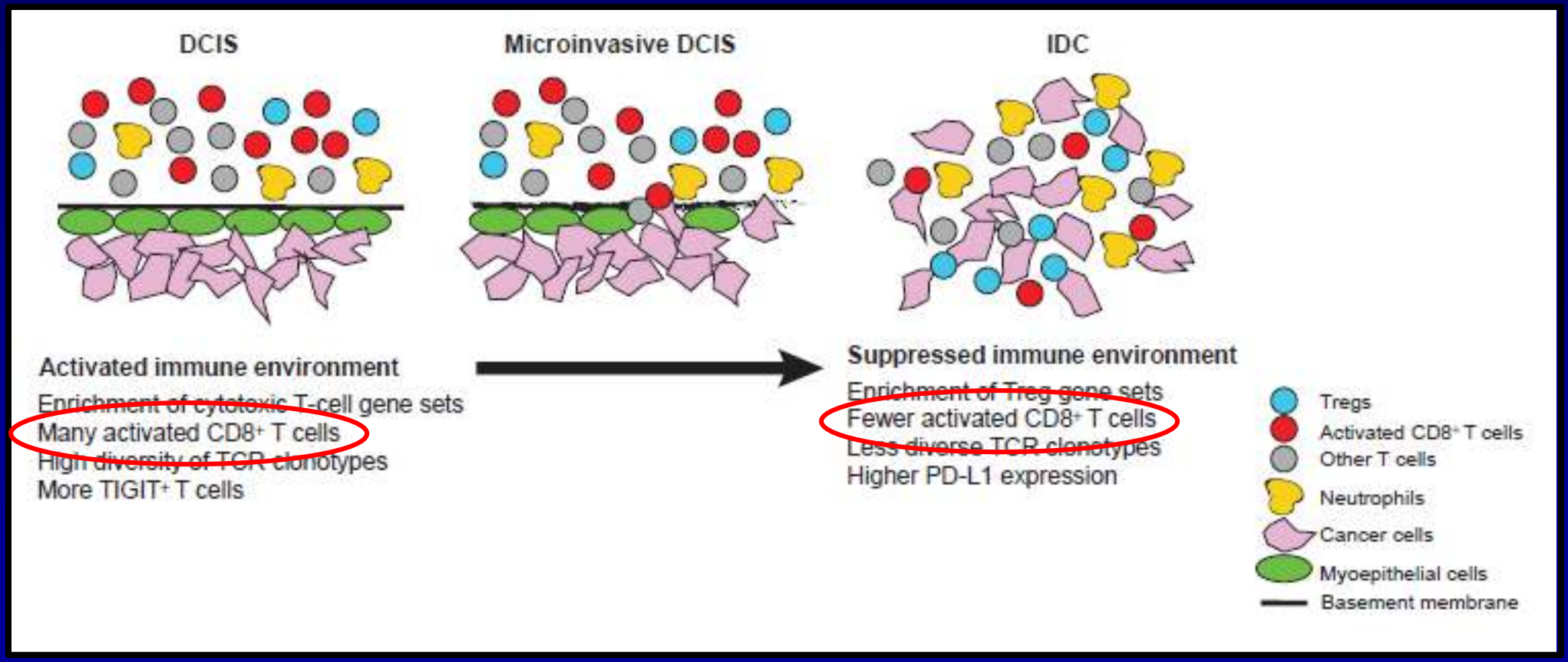
- Some TILs common (86% of cases in one study; mean 5%)
  - ***Large numbers*** of TILs associated with high nuclear grade, ER-, HER2+, TN, *TP53* mutations, fraction genome altered, telomeric imbalances
- Some PD-L1+ TILs common (81% of cases in one study)
  - ***Large numbers*** of PD-L1+ TILs associated with high nuclear grade, ER-, HER2+
- PD-L1 staining of DCIS cells infrequent (0-11% of cases)
  - associated with high nuclear grade, ER-, HER2+
- No association between periductal TILs and ipsilateral breast events (one study of 1488 cases)

Thompson, Mod Pathol 2016
Hendry, Clin Cancer Res, 2017
Pruneri, Ann Oncol 2017
Miligy, Histpathol, 2017

Immune Escape in Breast Cancer During *In Situ* to Invasive Carcinoma Transition

Carlos R. Gil Del Alcazar[1,2,3], Sung Jin Huh[1,2,3], Muhammad B. Ekram[1,2,3], Anne Trinh[1,2,3], Lin L. Liu[4,5], Francisco Beca[1,2,3], Xiaoyuan Zi[6,7], Minsuk Kwak[6], Helga Bergholtz[8], Ying Su[1,2,3], Lina Ding[1,2,3], Hege G. Russnes[8], Andrea L. Richardson[9,10,11], Kirsten Babski[12], Elizabeth Min Hui Kim[12], Charles H. McDonnell III[12], Jon Wagner[12], Ron Rowberry[12], Gordon J. Freeman[1,3], Deborah Dillon[10,11], Therese Sorlie[8], Lisa M. Coussens[13], Judy E. Garber[1,2,3], Rong Fan[6], Kristie Bobolis[12], D. Craig Allred[14], Joon Jeong[15], So Yeon Park[16], Franziska Michor[4,5], and Kornelia Polyak[1,2,3,17,18]

Cancer Discovery, 2017

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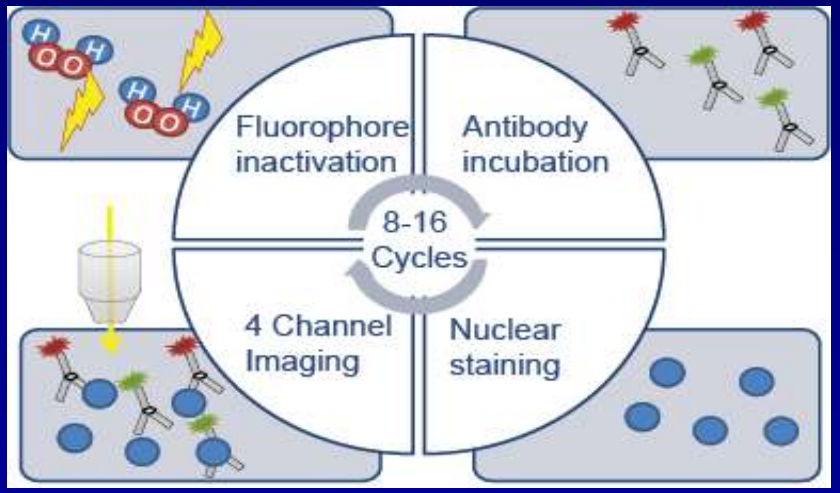
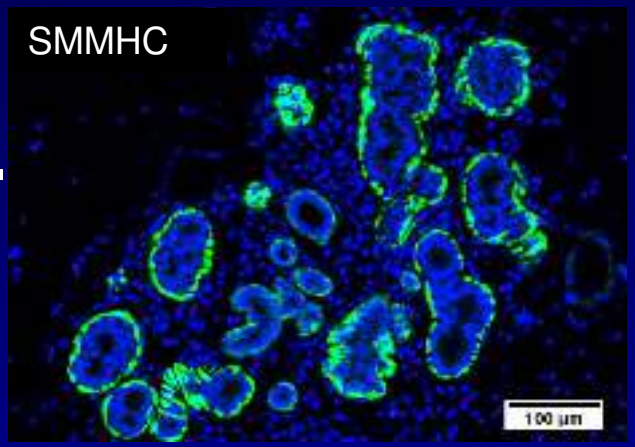
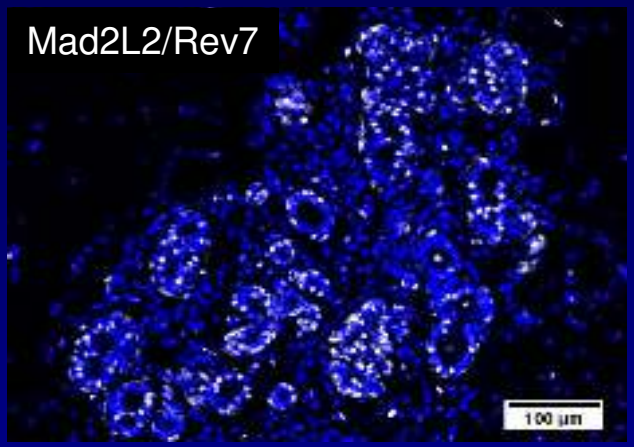
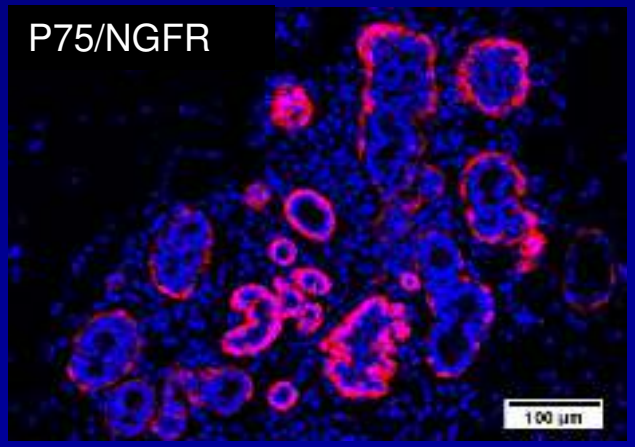
Some New Approaches

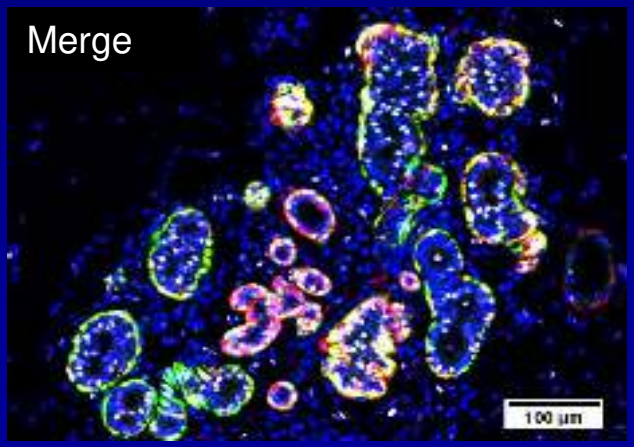
- Cyclic Immunofluorescence (CyCIF)
  - Multiplexed IF
  - MEC phenotypes
  - Immune microenv.







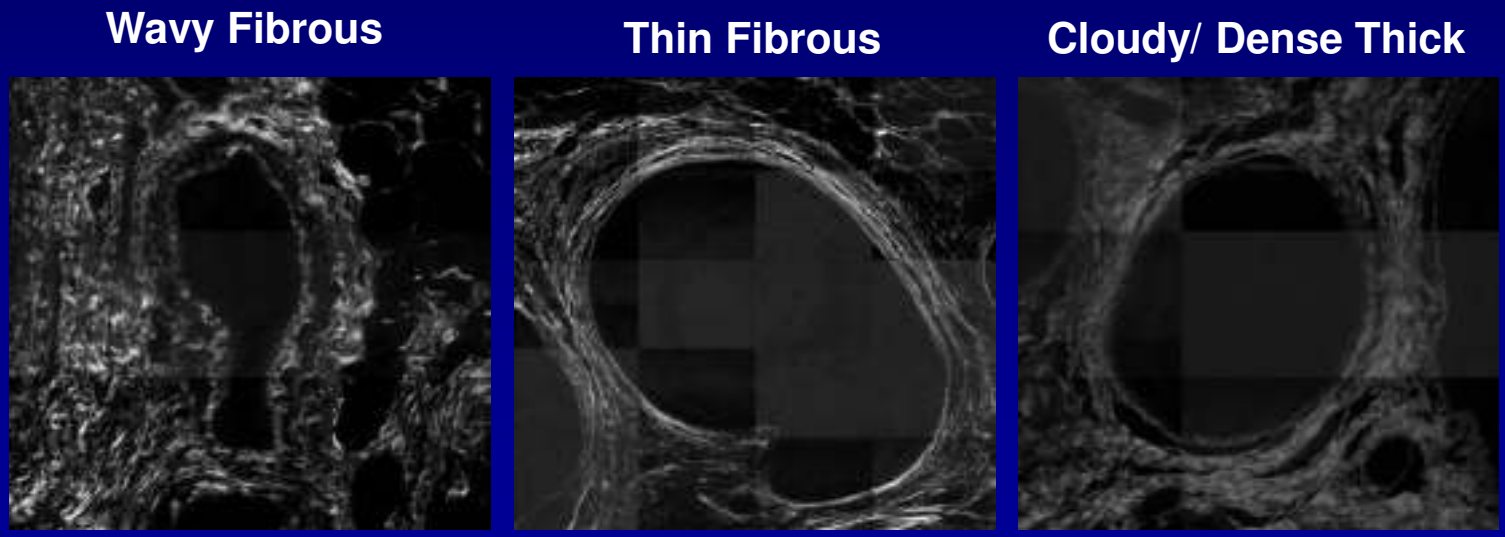




Courtesy of Dr. Sandro Santagata

Some New Approaches

- **Biophysical properties of DCIS stroma using second-harmonic generation (SHG) microscopy**
  - **Assessment of collagen structure**



Catherine Park, M.D., UCSF

Some New Approaches

- Combined genomic and morphometric analysis (NHS, Rob West)
- Assessment of intra-lesional heterogeneity and clonal selection
- Pre-cancer Atlas (PCA)

Management of DCIS

Two Perspectives

- **Arguments for more aggressive treatment**
  - Radiation reduces local recurrence risk even in "low risk" DCIS
  - Any local recurrence is psychologically devastating for patients; viewed as treatment failure
  - Half of recurrences after breast conserving treatment are invasive

- **Arguments for less aggressive treatment**
  - Local recurrence of DCIS is inconsequential
  - Only important clinical endpoint is development of potentially lethal invasive breast cancer
  - Most invasive breast cancers are small, mammographically-detected, N- lesions amenable to treatment

Conclusions

- Although great progress has been made in the treatment of patients with DCIS over the past two decades, much remains to be done

Conclusions

- In particular, accurate risk stratification remains elusive; communication of risk to patients remains problematic

Conclusions

- A better understanding of the molecular alterations associated with the progression of DCIS to invasive breast cancer will hopefully lead to
  - new methods to distinguish those patients with DCIS who are likely to recur or progress from those who are not
  - identification of new targets for treatment and prevention