Recommendations for assessing tumor-infiltrating lymphocytes (TILs) in breast carcinoma in situ

Guidelines for TILs assessment from the «International Immuno-Oncology Biomarker Working Group»

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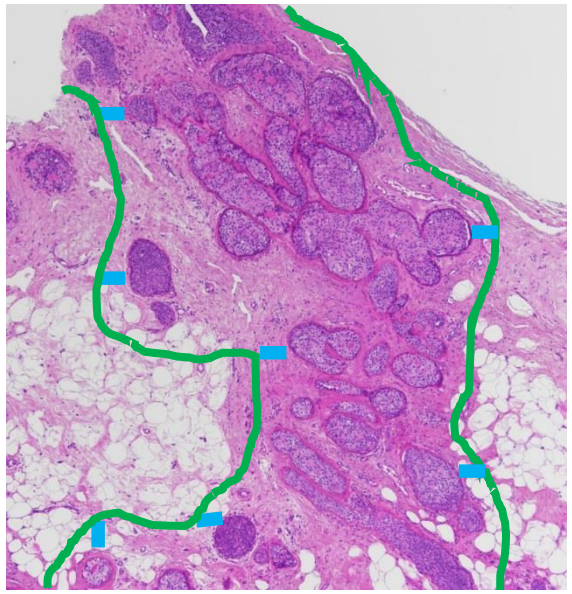
These recommendations are valid for both ductal and lobular carcinoma in situ
Ductal carcinoma in situ (DCIS) is used as an example

- TILs in DCIS should be assessed **on surgical specimens** whenever possible;
  H&E slides representing the entire specimen should be submitted for TIL assessment (especially valid for multicentric studies);
  TILs should be assessed **on the entire surgical specimen**

- These recommendations are related to the assessment of TILs on hematoxyllin-and-eosin **(H&E)-stained sections** of formalin-fixed, paraffin-embedded DCIS tissue; the sections should be 4-5 µm thick

1. Identifying the area for TIL evaluation

- TILs in DCIS should be assessed **within the specialized tumor stroma**;
  as specialized tumor stroma is considered the area
  limited by the external borders of the tumor (DCIS) nests,
  extended for 2 High-Power Fields (HPFs, 40x) towards the tumor-adjacent stroma



extension over 2 HPFs

limits of the specialized stroma area to be considered for TIL assessment

2. Areas to be excluded from TIL evaluation

- **ASSESS ONLY** the TILs **WITHIN THE STROMA** →
  → do not assess the intraepithelial TILs (within the tumor nests)

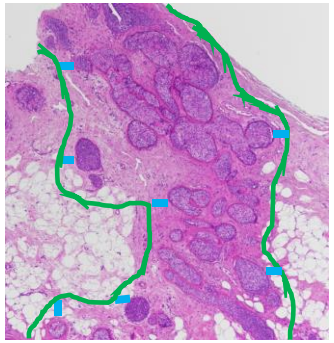
- **DO NOT ASSESS :**
  - TILs in the invasive tumor areas, whatever the size (micro-invasion or bigger)
  - TILs in the necrotic areas or the areas with artefacts
  - TILs in the hyalinisation areas
  - TILs around normal mammary structures

3. Cells to and not to assess

- **TILs are :** small mature lymphocytes
  « activated » lymphocytes (« lymphoblastoid », like in some infections)
  lymphoplasmocytoid cells
  plasmocytes

- **TILs are not :** stromal mesenchymal cells
  macrophages
  neutrophils
  mastocytes
  altered tumor cells (apoptotic, the cells altered by tissue processing etc.)

4. How to assess TILs in DCIS

- **DO THIS** ➡  **ON EACH SLIDE**

- **DETERMINE THE PERCENTAGE (%) OF THE STROMAL SURFACE OCCUPIED BY TILs, ON EACH AREA DELINEATED FOR TIL ASSESSMENT**

DO NOT FOCUS ON THE « HOT SPOTS »
(the areas with the densest TILs)

- **DERIVE THE AVERAGE % FOR ALL THE AREAS EVALUATED**

This is the TIL SCORE of a DCIS

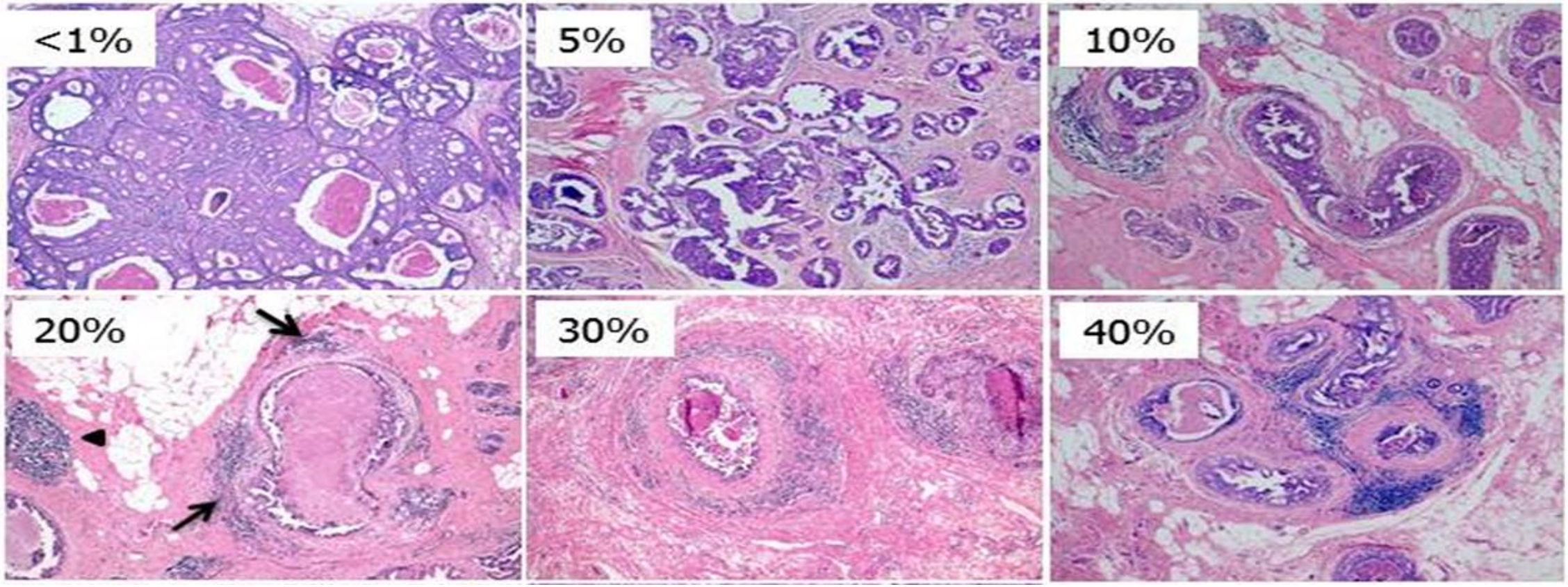
5. Assess TILs % as continuous parameter

- **Report TIL scores as a continuous parameter (0- 100%)**

- In case of DCIS associated to a micro-invasive carcinoma (µCa)
  report the TIL score of the µCa but do not include it when deriving the DCIS TIL score

- **Examples of TILs scores in DCIS are given on the next page**

EXAMPLES of TIL SCORES in DCIS (1)



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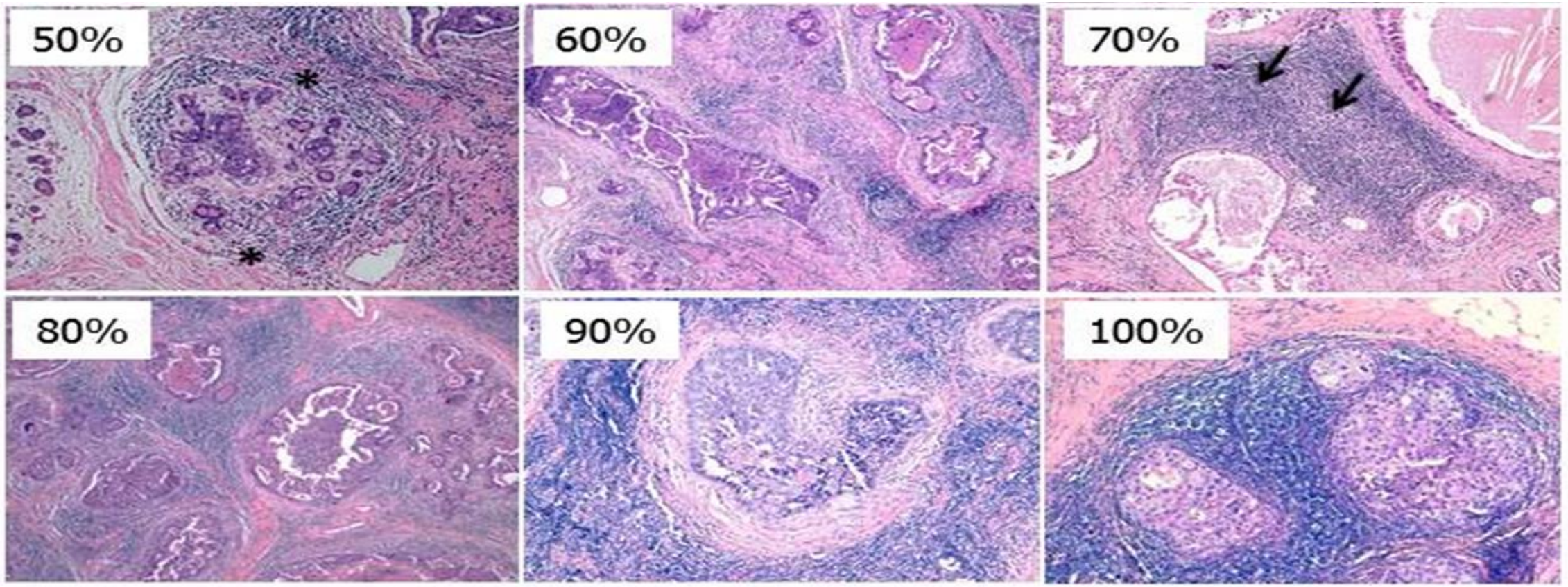
EXAMPLES of TIL SCORES in DCIS (2)



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