Immunohistochemistry approach for challenging melanocytic tumours

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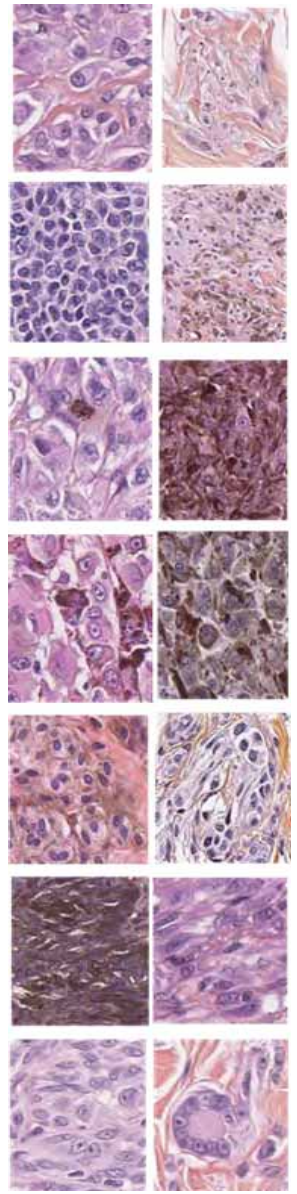
CENTRE DE LUTTE CONTRE LE CANCER LEON BERARD
Chercher et soigner jusqu'à la guérison



Financial disclosures: none

Lecture

- Classical IHC approach with basic antibodies

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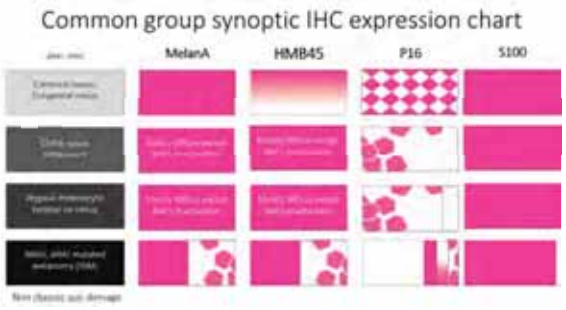
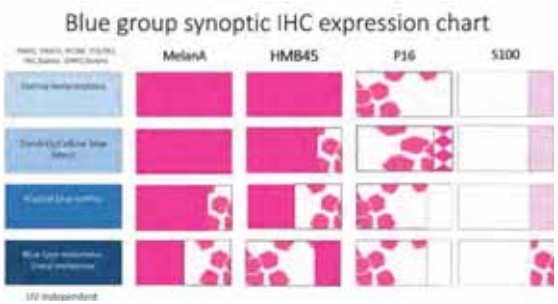
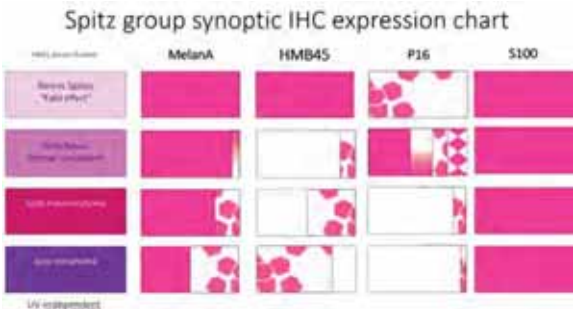
- Classical IHC approach with basic antibodies
- Contextual/combined IHC diagnosis approach







Lecture

- Classical IHC approach with basic antibodies
- Contextual/combined IHC diagnosis approach
- Case analysis approach

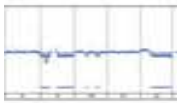
Integrative diagnosis approach of melanocytic proliferations

Embryogenesis

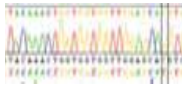
Clinical features

Microscopy/morphology

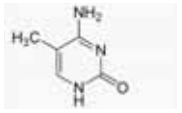
Immunophenotype



Genomic profile



Mutation status



Methylation profiles

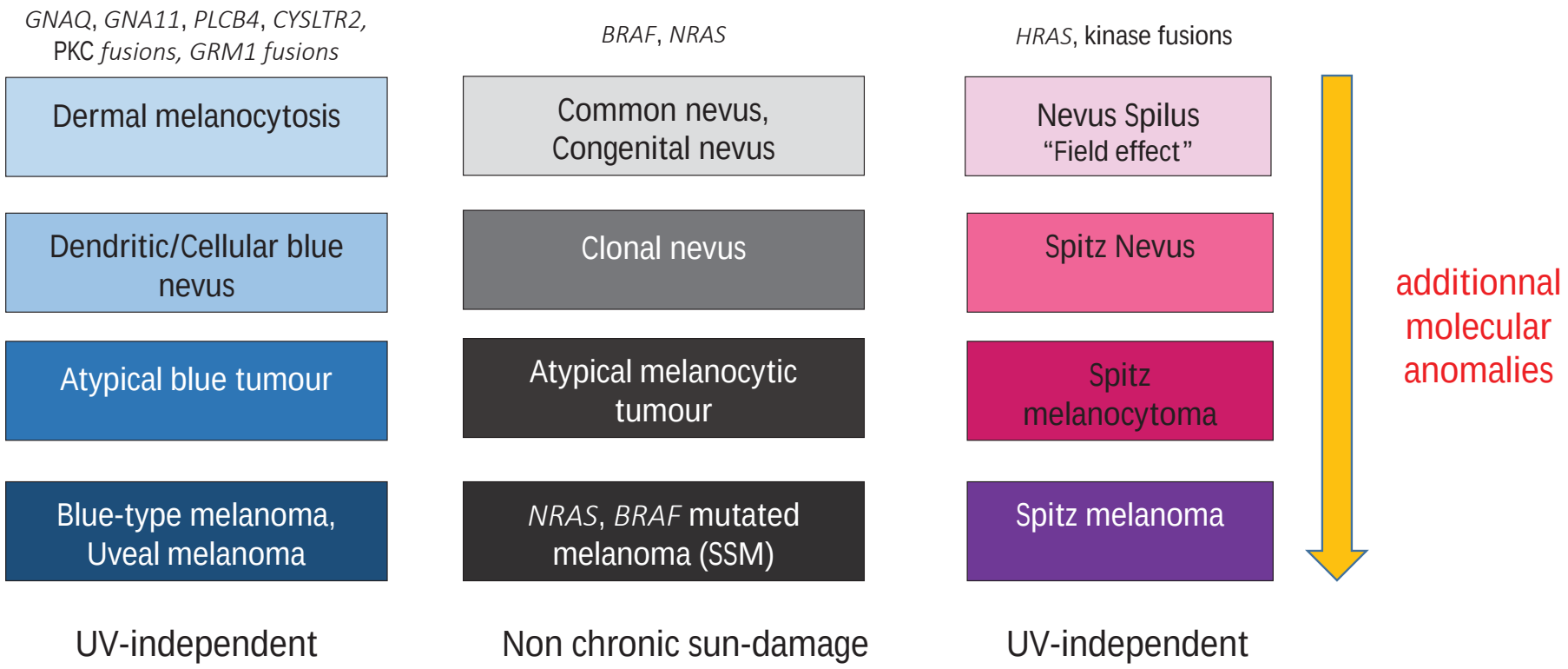
Clinical evolution

Immunohistochemistry

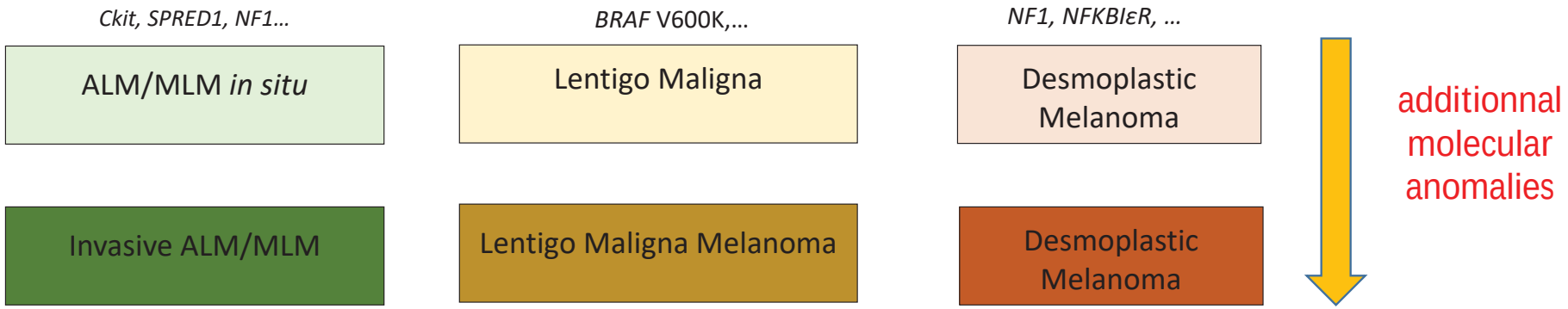
Immunohistochemistry

It’s all about chosing the adapted antibody(ies) for the selected task(s)

Integrative classification of melanocytic tumors
Nevus to melanoma groups



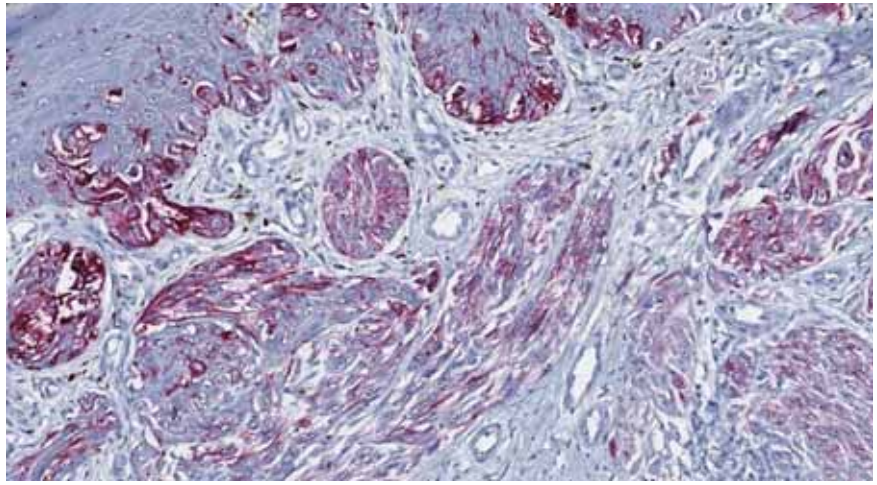
Melanoma arising from melanocyte (without nevus)



Immunohistochemistry
Melanocytic » antibodies

Specificity vs Sensitivity

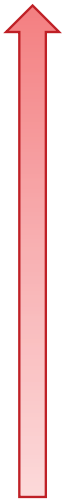
- S100 Protein
- MelanA
- HMB45
- MiTF
- SOX10



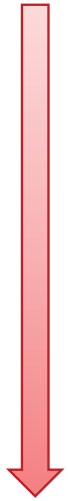
HMB45

Immunohistochemistry
Melanocytic » antibodies

Sensitivity



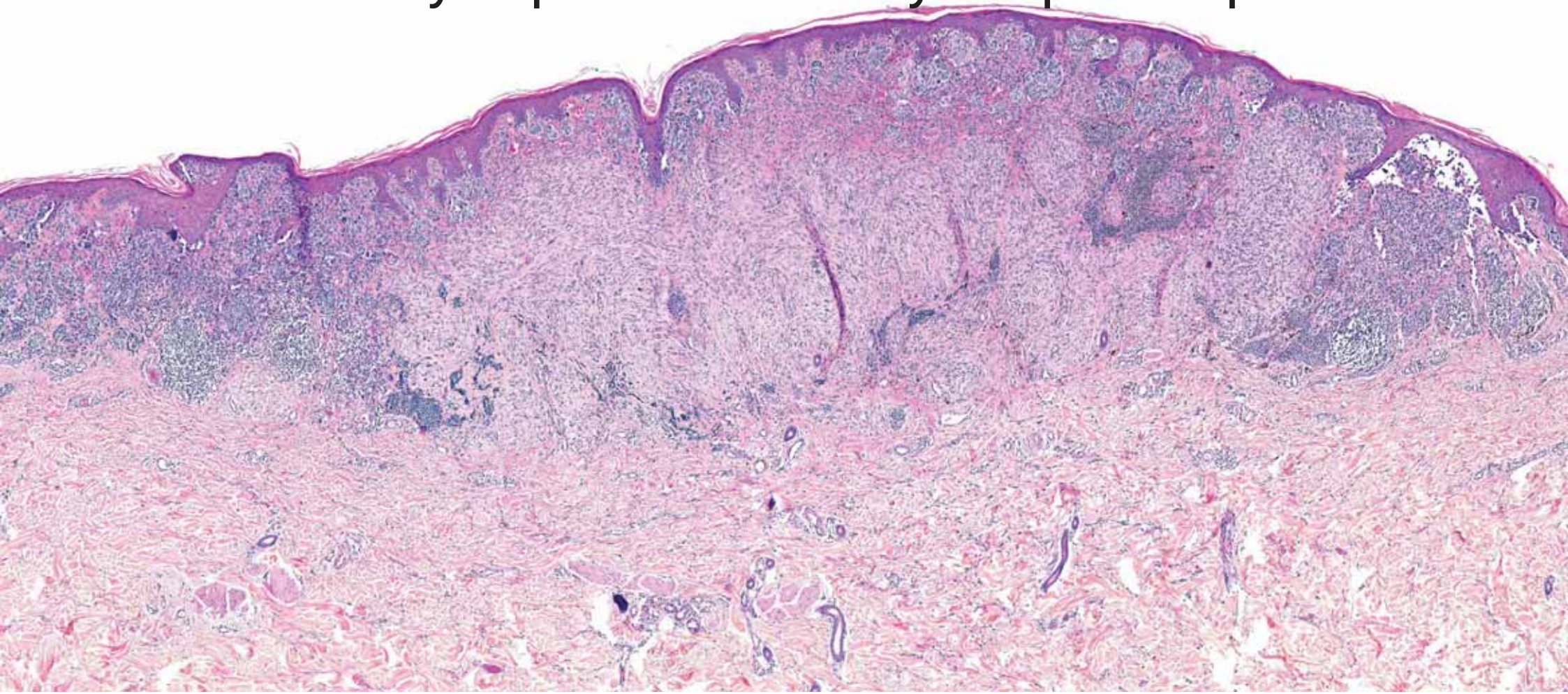
Antibody	Topography of staining
S100 Protein	Cytoplasm + nuclear
Sox10	nuclear
MART1-MelanA	Cytoplasm
MiTF	nuclear
Tyrosinase	Cytoplasm
HMB45	Cytoplasm



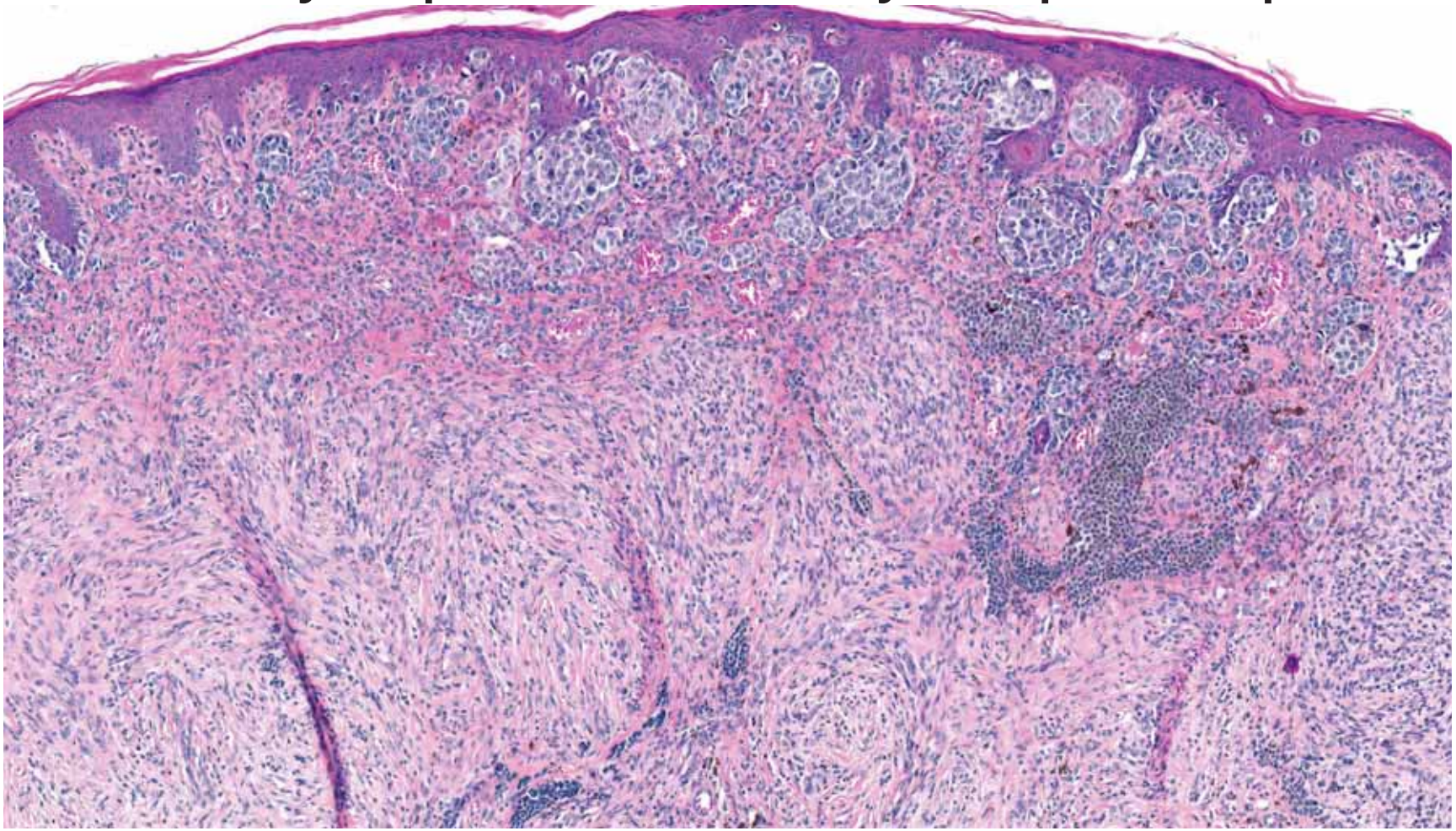
Specificity

Adapted from L Lowe

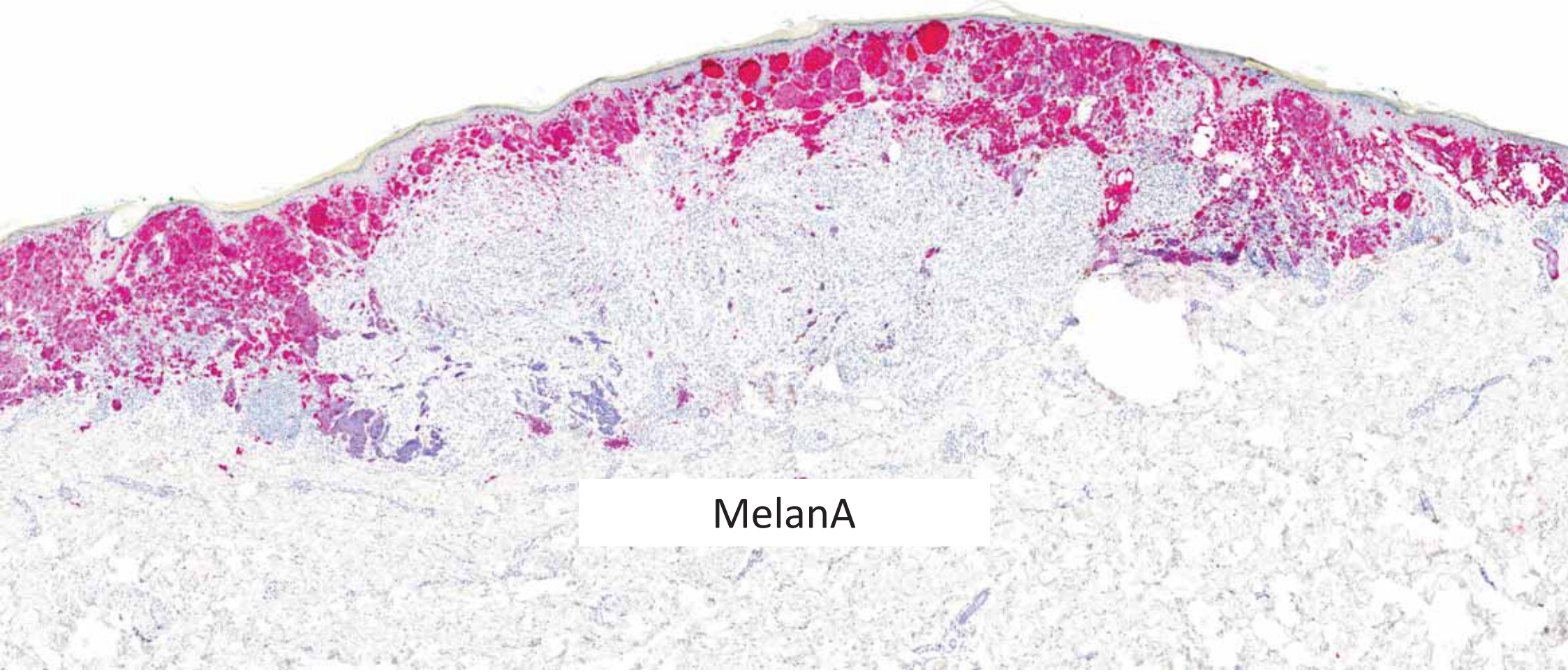
The less a melanocyte is differentiated (spindly)
the less they express melanocytic specific proteins



The less a melanocyte is differentiated (spindly) the less they express melanocytic specific proteins

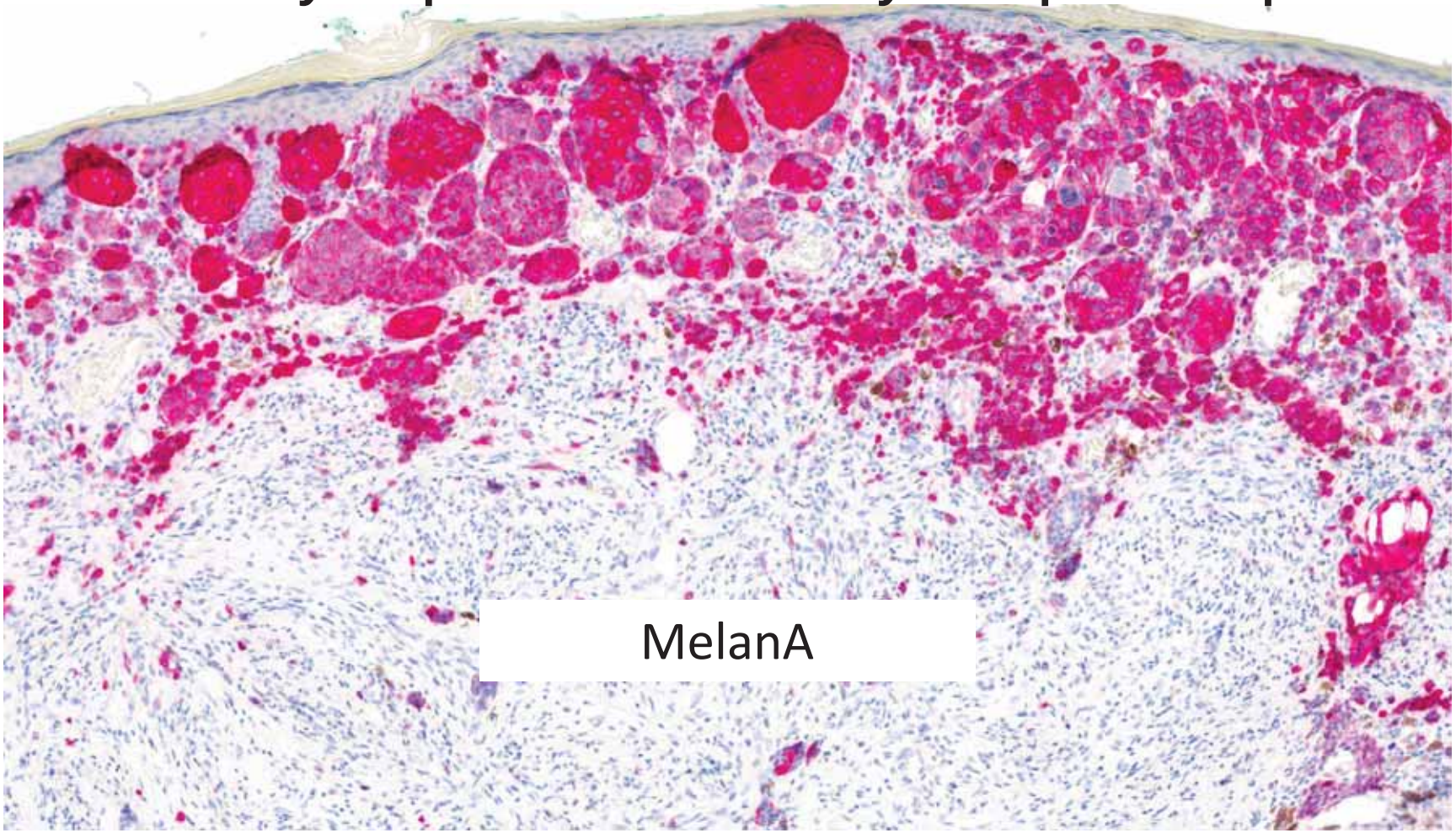


The less a melanocyte is differentiated (spindly) the less they express melanocytic specific proteins



MelanA

The less a melanocyte is differentiated (spindly) the less they express melanocytic specific proteins



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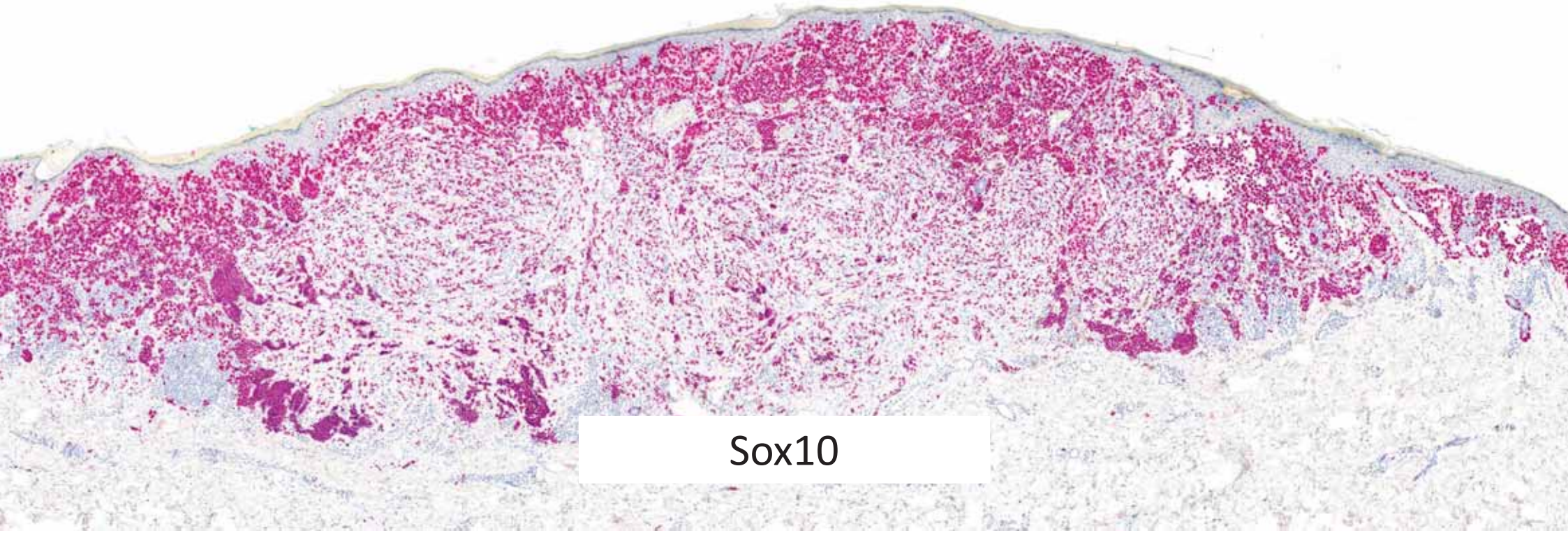
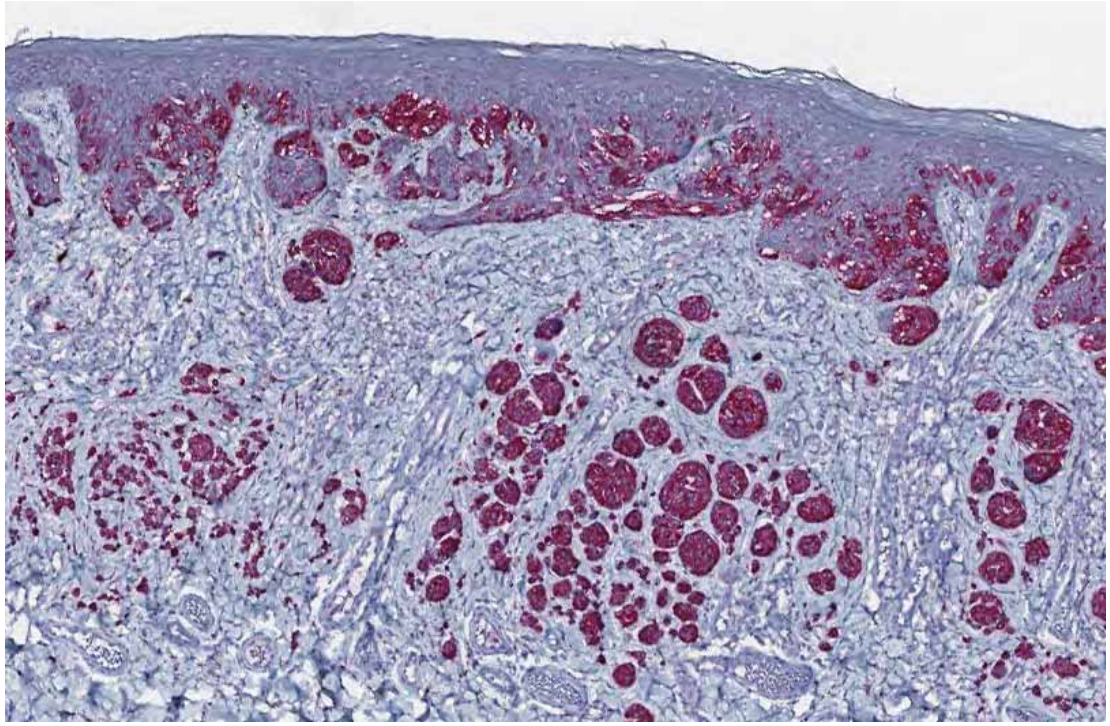


Molecular immunohistochemistry: « Driver-specific » antibodies All tumoral cells are positive

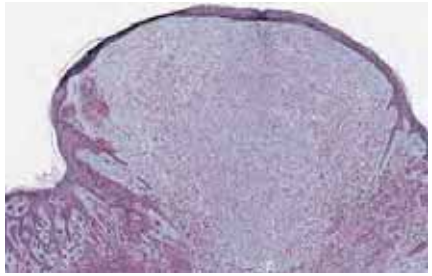
- BRAF V600E
- NRAS Q61R
- ALK
- ROS1
- Pan-TRK
- MET
- RET



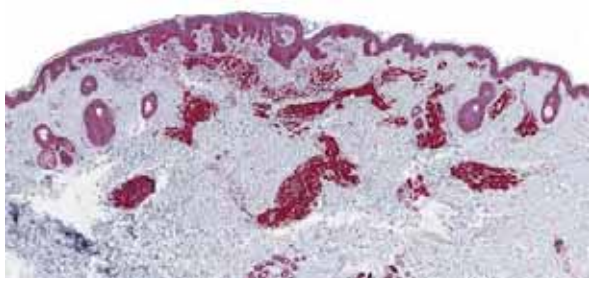
Pan-TRK

Molecular immunohistochemistry: « secondary events» antibodies Frequent clonal loss/positivity

- P16
- BAP1
- PRKAR1A
- Beta catenin
- CyclinD1



BAP1

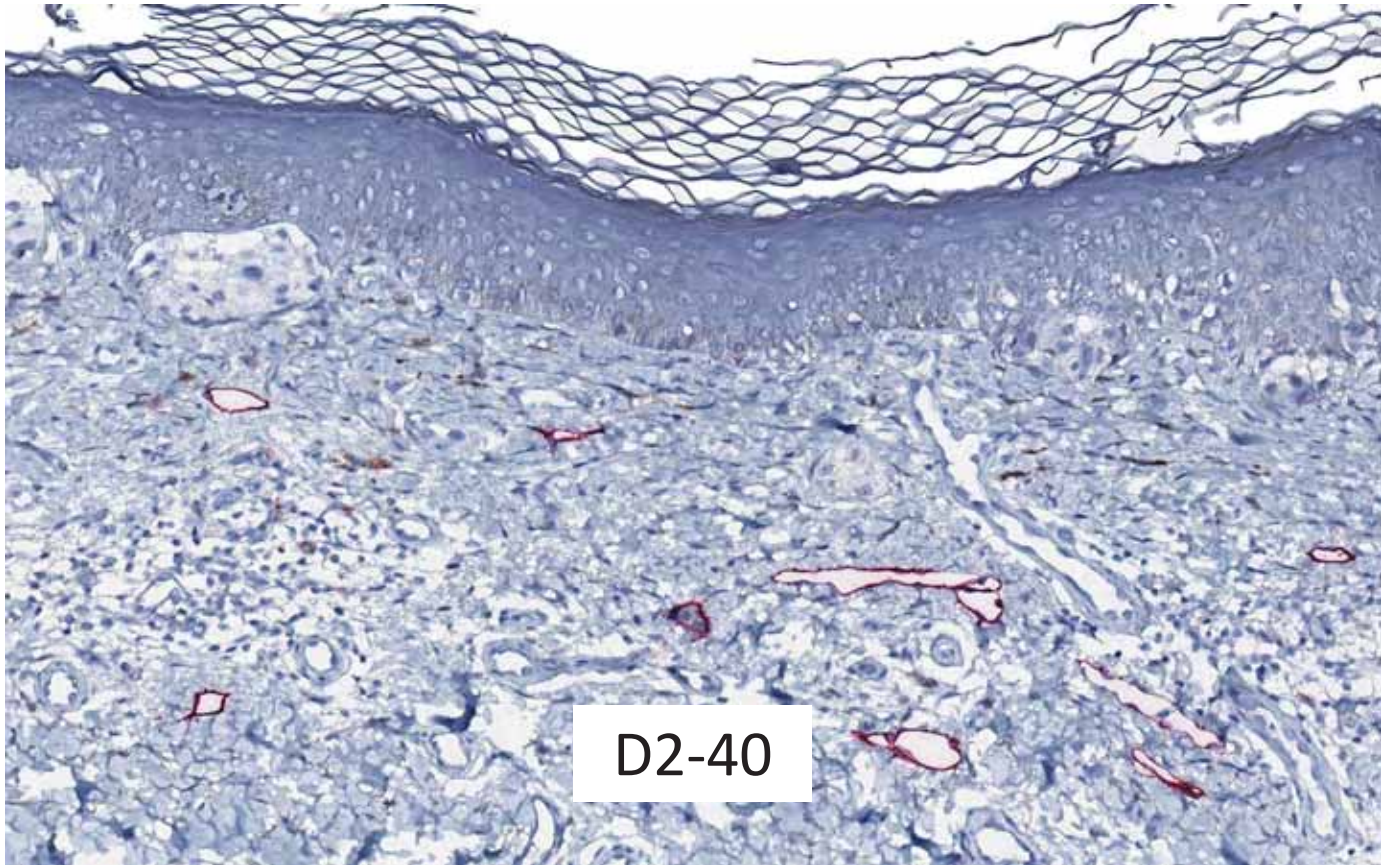


β-catenin

PRKAR1A

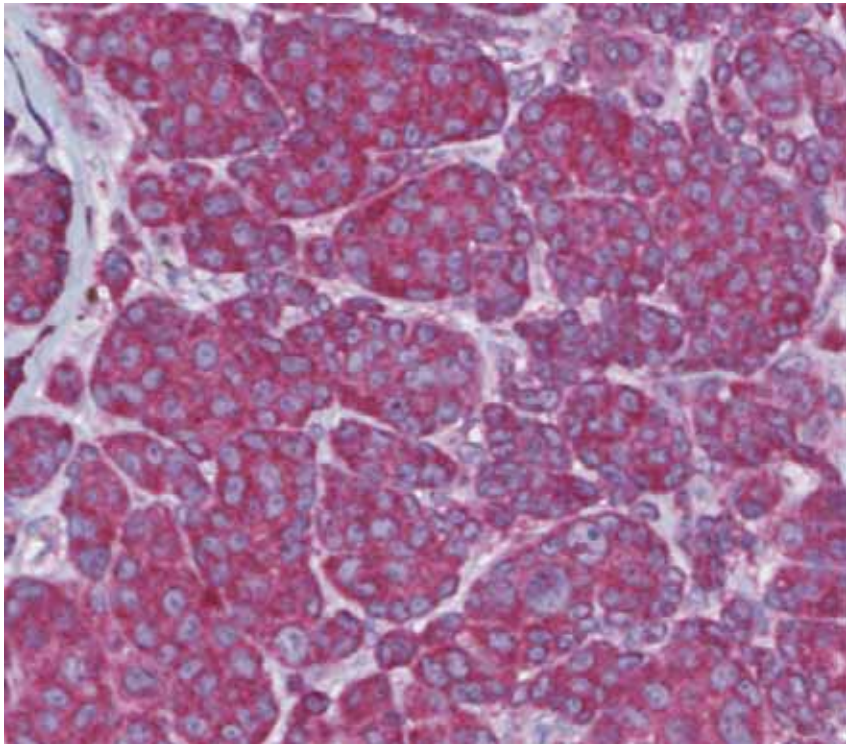
Immunohistochemistry
Other antibodies (DD, etc)

- …
- PRAME
- D2-40
- CD68
- …



Know your antibodies!
Which cellular compartment is supposed to be stained?

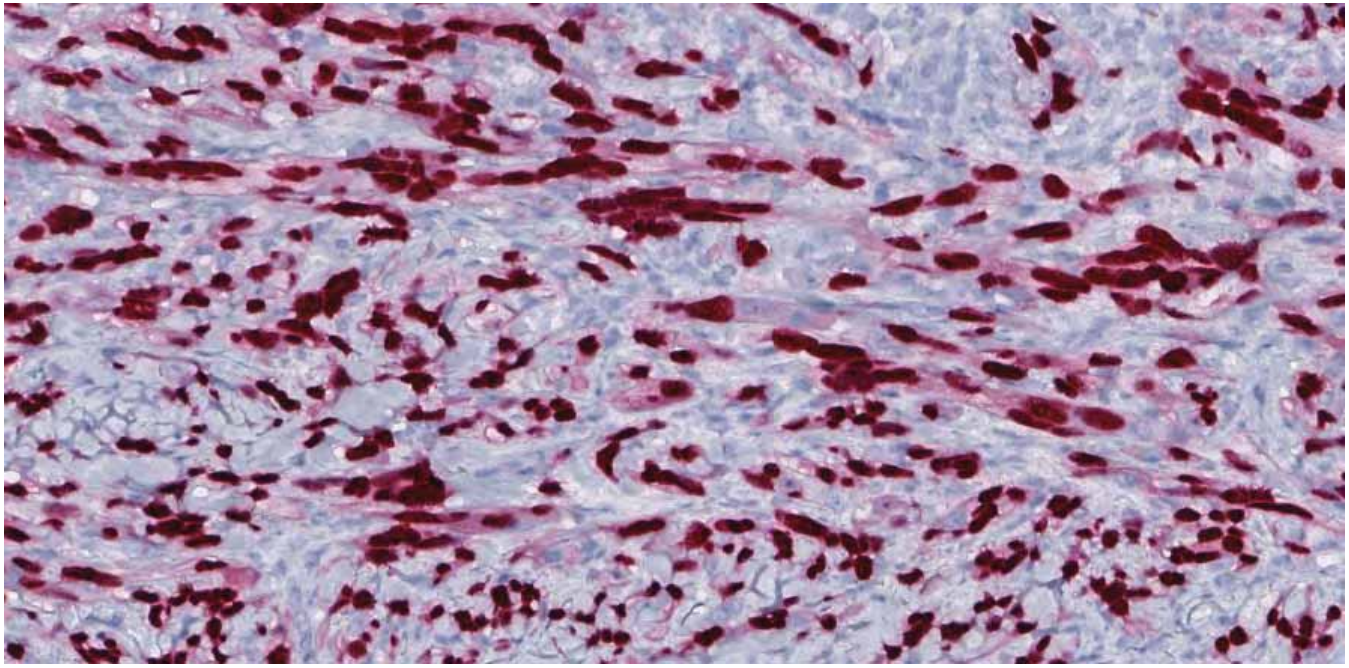
- Cytoplasm



BRAF V600E

Know your antibodies!
Which cellular compartment is supposed to be stained?

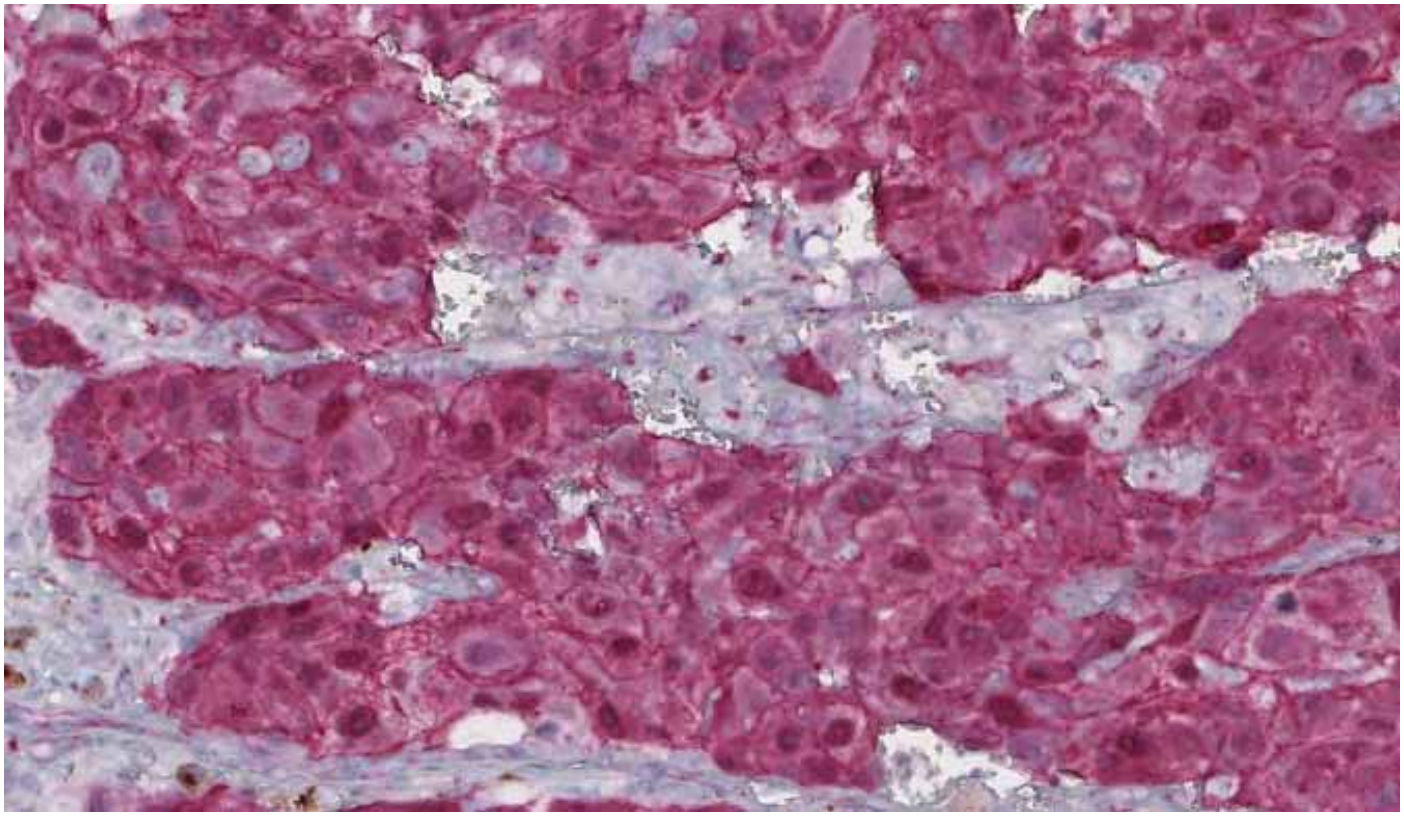
- Cytoplasm
- Nuclei



Sox 10

Know your antibodies!
Which cellular compartment is supposed to be stained?

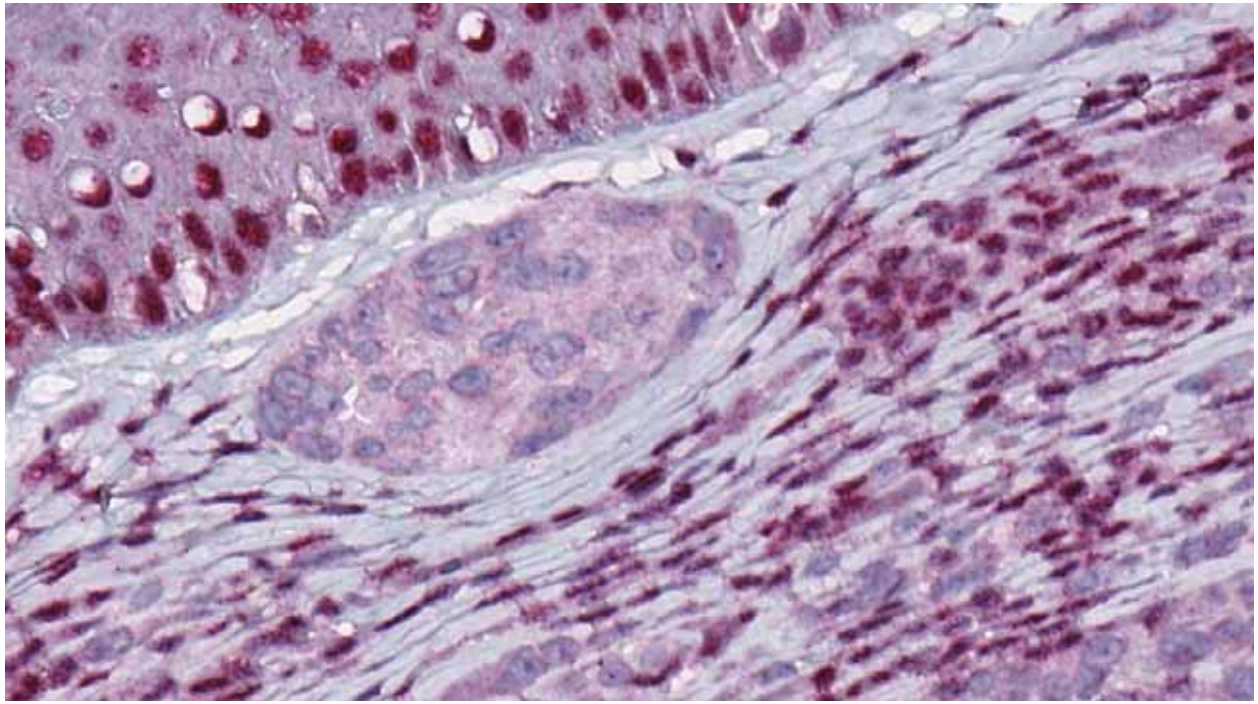
- Cytoplasm
- Nuclei
- Cell membrane



Beta-catenine

Know your antibodies!
Which cellular compartment is supposed to be stained?

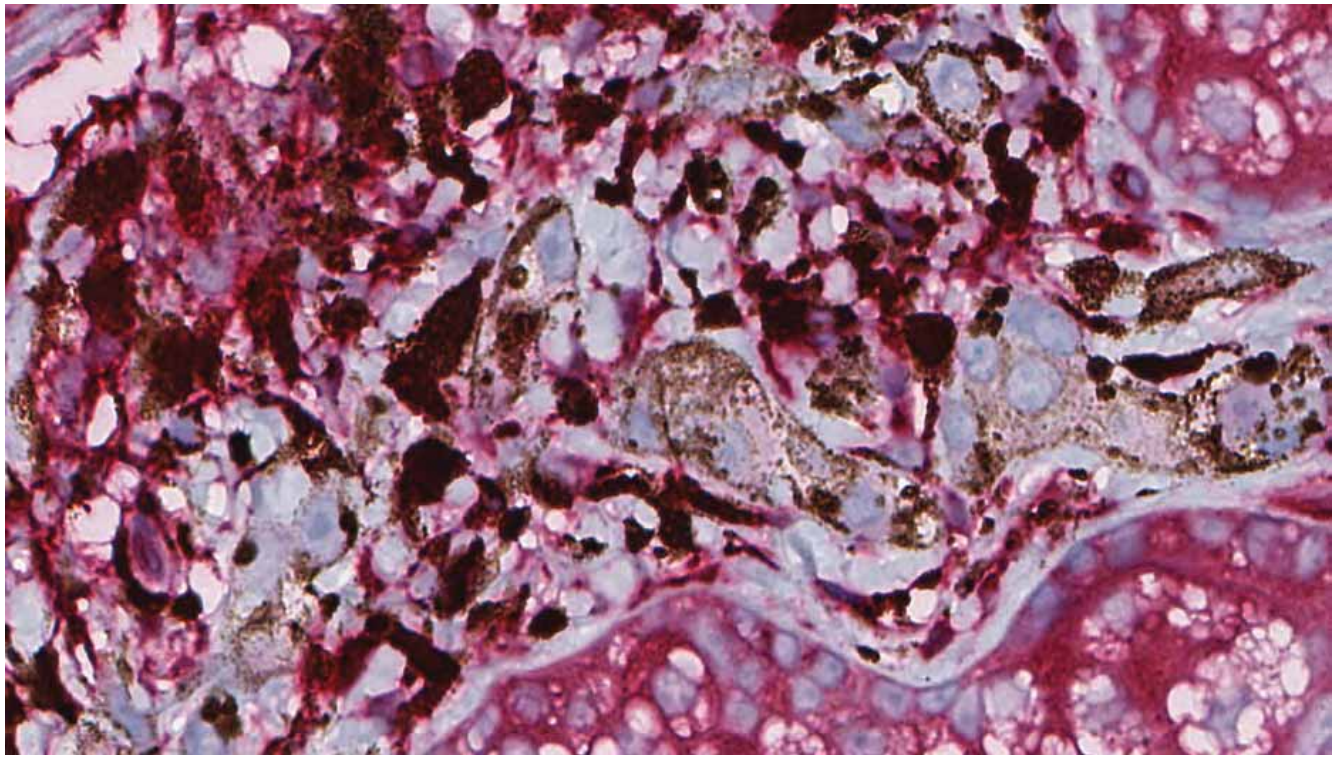
- Cytoplasm
- Nuclei
- Cell membrane
- Nuclear loss



BAP1

Know your antibodies!
Which cellular compartment is supposed to be stained?

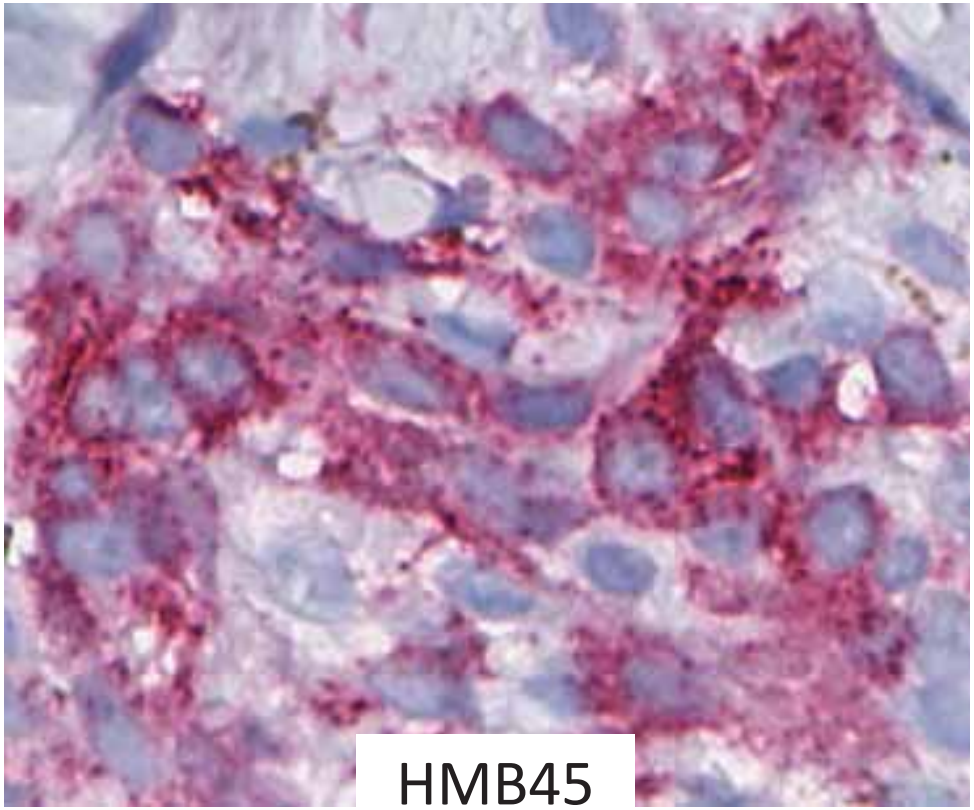
- Cytoplasm
- Nuclei
- Cell membrane
- Nuclear loss
- Cytoplasmic loss



PRKAR1A

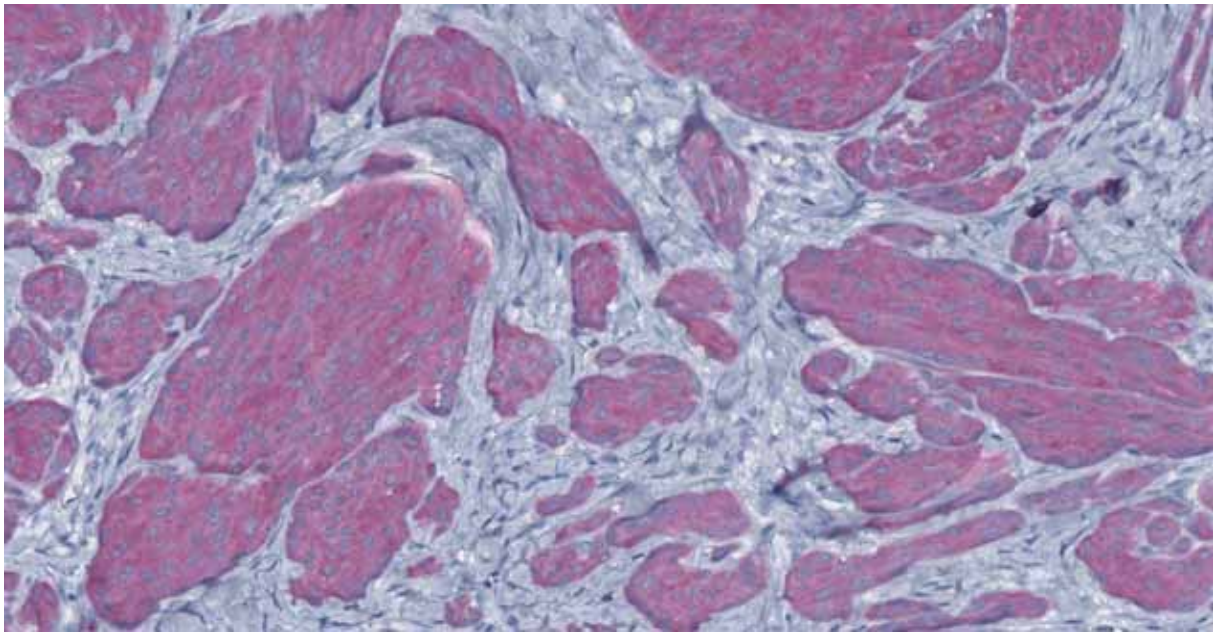
Know your antibodies!
Type of staining

- Granular



Know your antibodies!
Type of staining

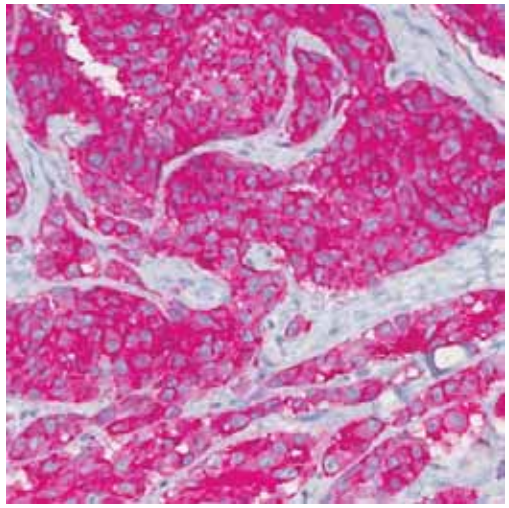
- Granular
- Diffuse +



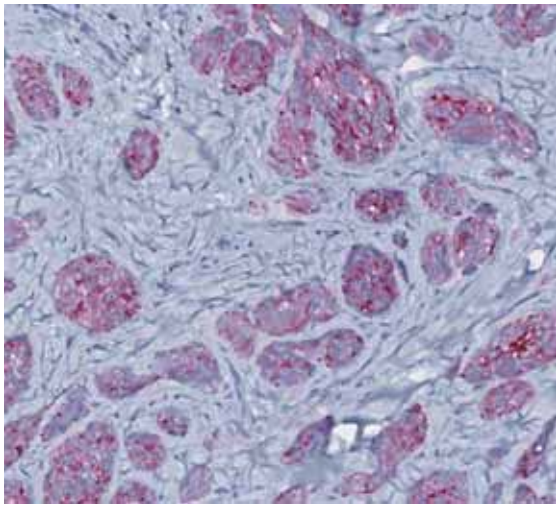
ALK

Fusion specific IHC (ALK, ROS1, Pan-TRK)

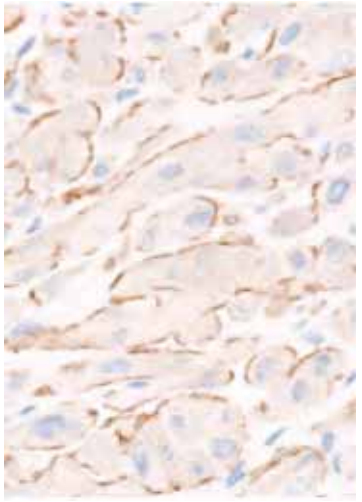
- Confirm the overexpression of the RTK not the fusion *per se*
- Signal location depends on 5’ partner



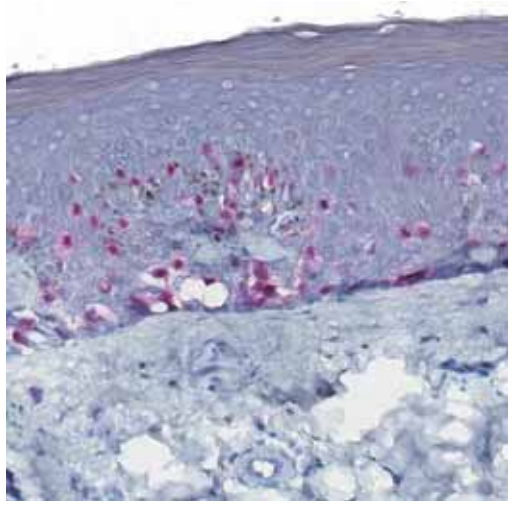
Cytoplasmic diffuse



Cytoplasmic heterogeneous



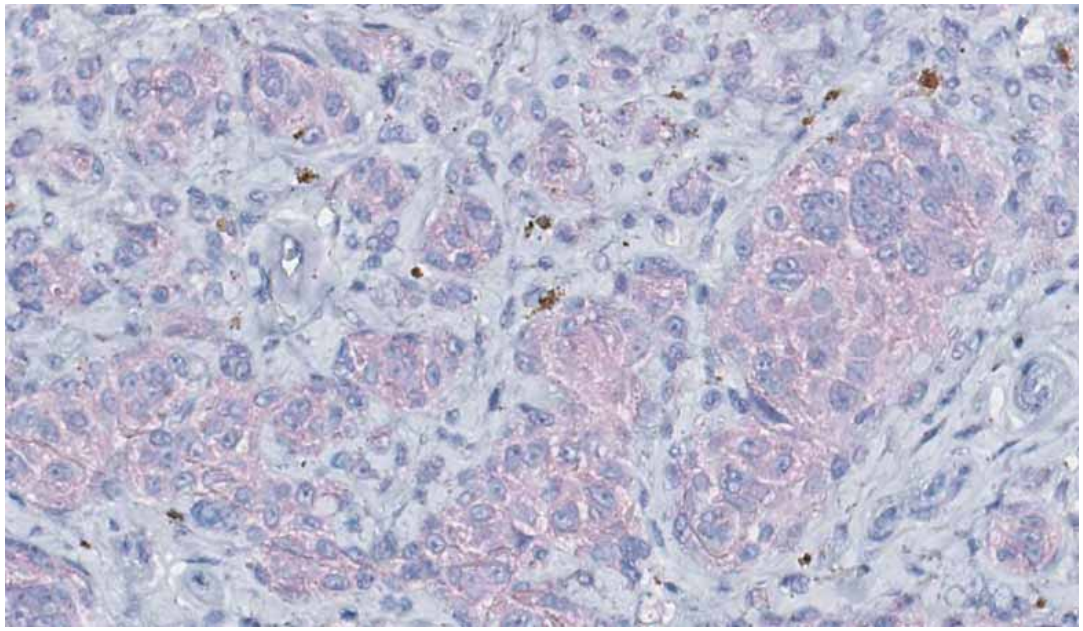
Membranous



Nuclear

Know your antibodies!
Expected intensity of staining

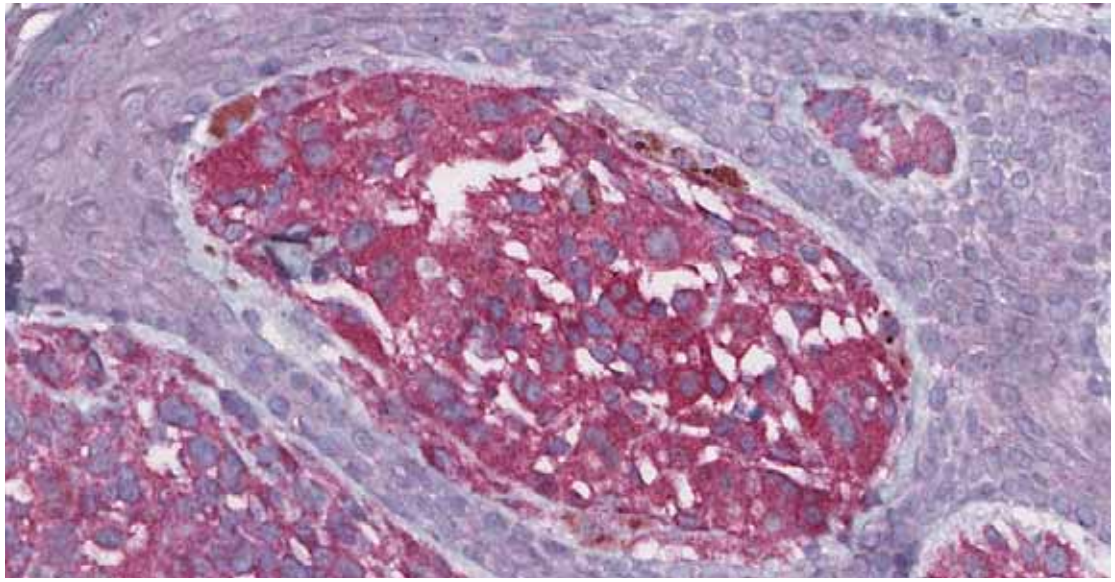
- Weak stain with negative background



NRAS Q61R

Know your antibodies!
Expected intensity of staining

- Weak stain with negative background
- Strong with background positivity



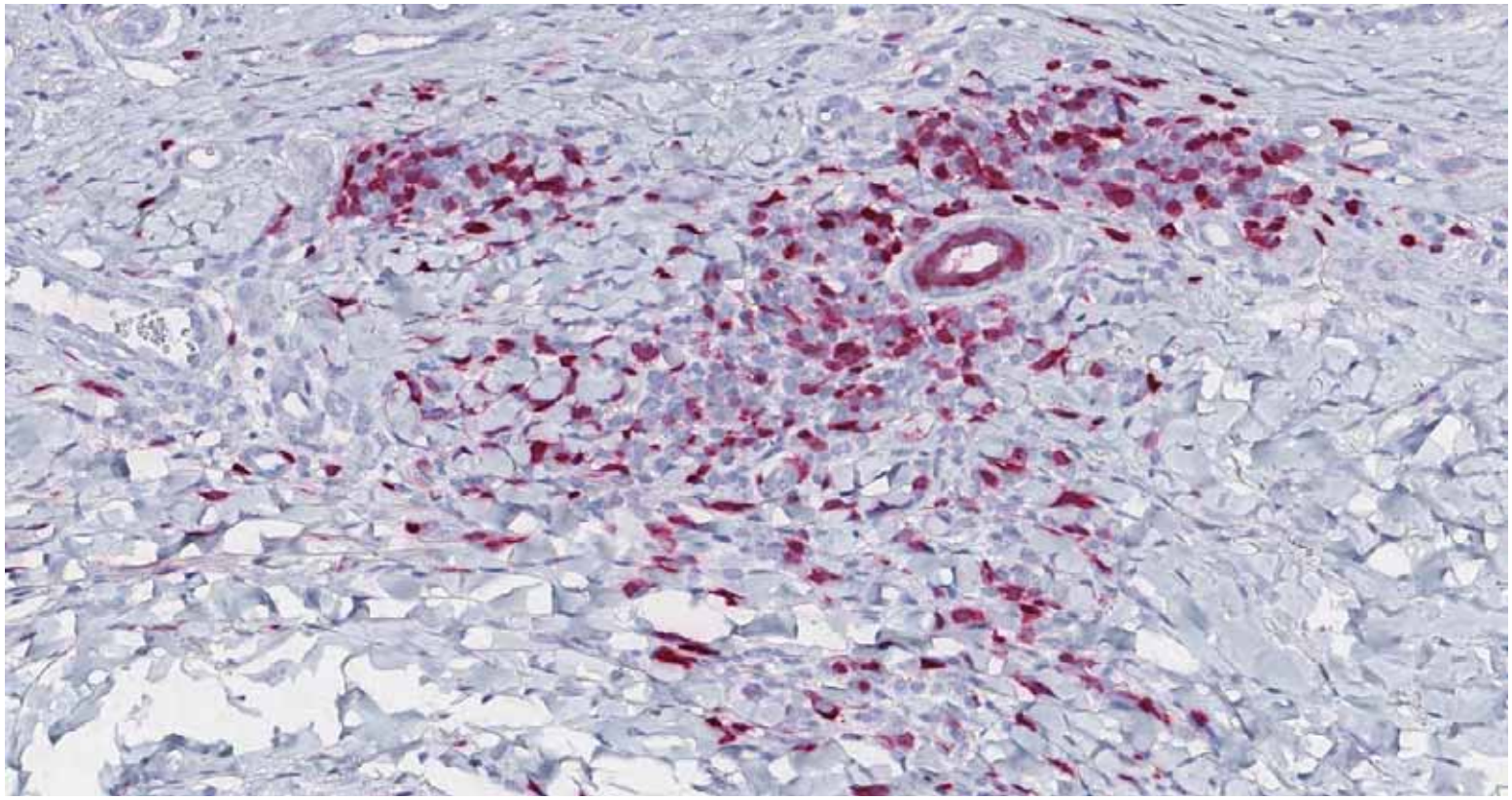
BRAF V600E

Know your antibodies!
Staining pattern

- Diffuse
- Top-heavy gradient
- Inverted gradient (bottom-heavy)
- Mosaic of positive and negative cells (Checkerboard)
- Clonal positivity/loss
- Differential expression between components (Strong/mild positivity)
- Heterogeneous

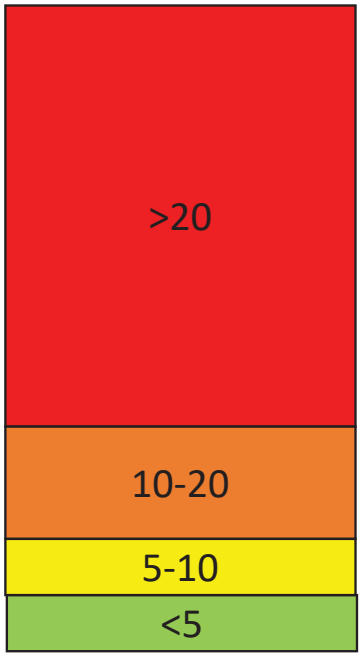
Know your antibodies!
Internal controls (otherwise external)

- p16



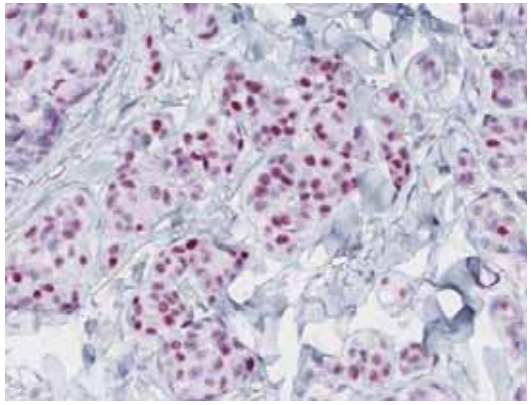
Know your antibodies!
ki-67/MIB1 Thresholds

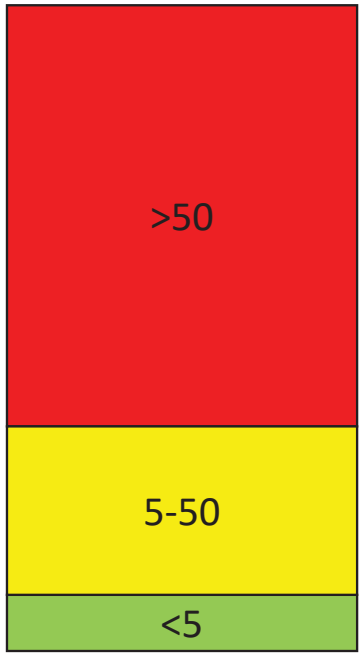
- Dermal melanocytes



Know your antibodies!
p53 thresholds

- Dermal melanocytes





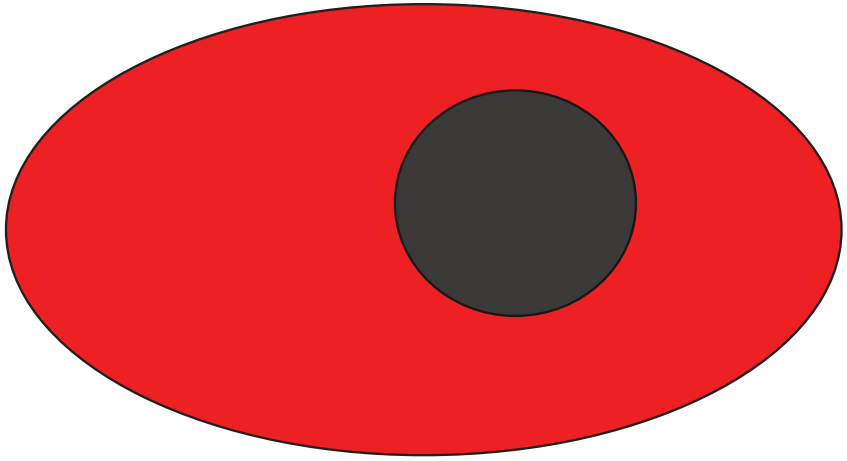
Know your antibodies!
Technique crititism is mandatory

Do not interpret IHC at all costs :

- Region of interest missing (too many recuts or scratched)
- Technical imperfections

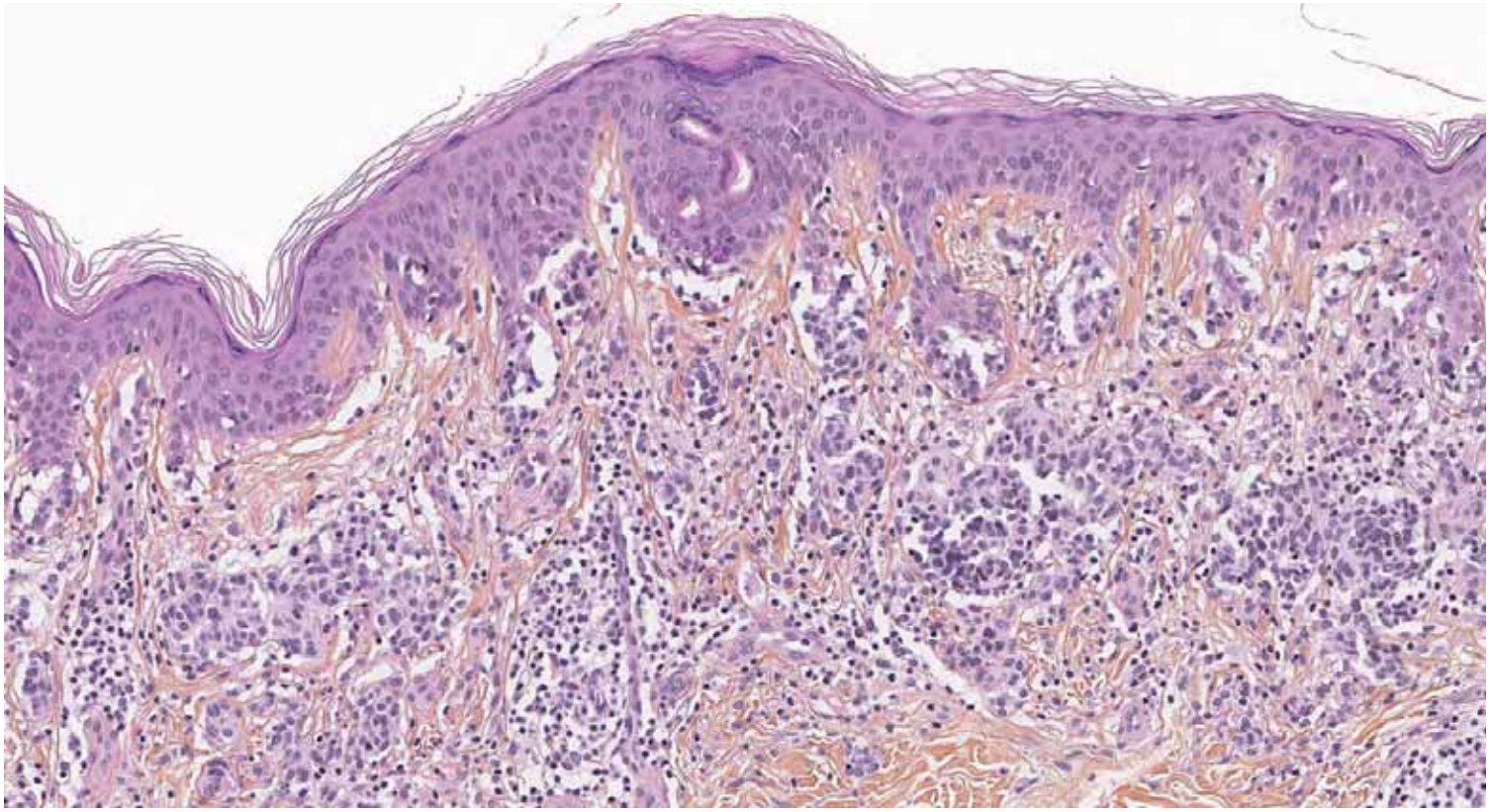
Double-staining (expl: ki-67/MelanA)

Cytoplasmic stain in red

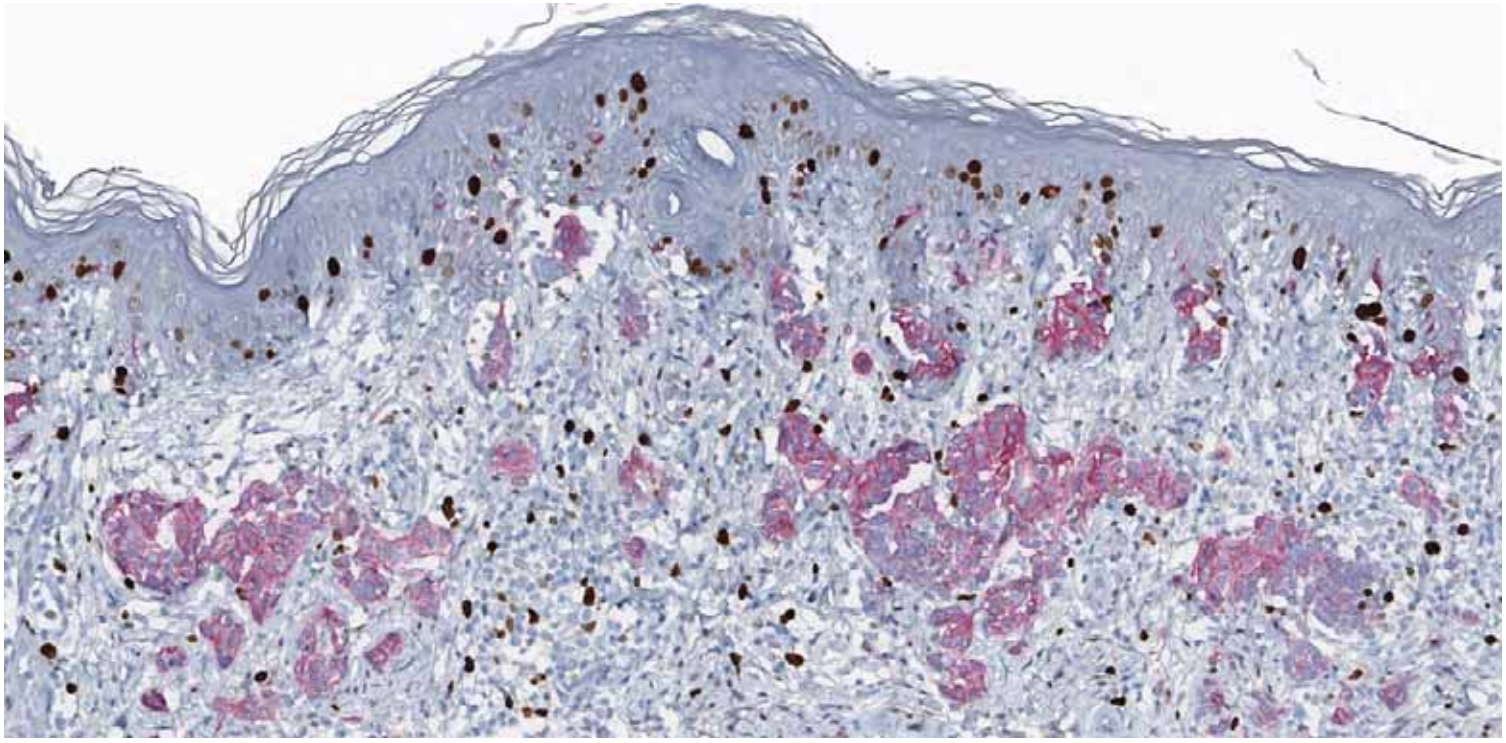


Nuclear stain in brown

ki-67/MelanA double-staining



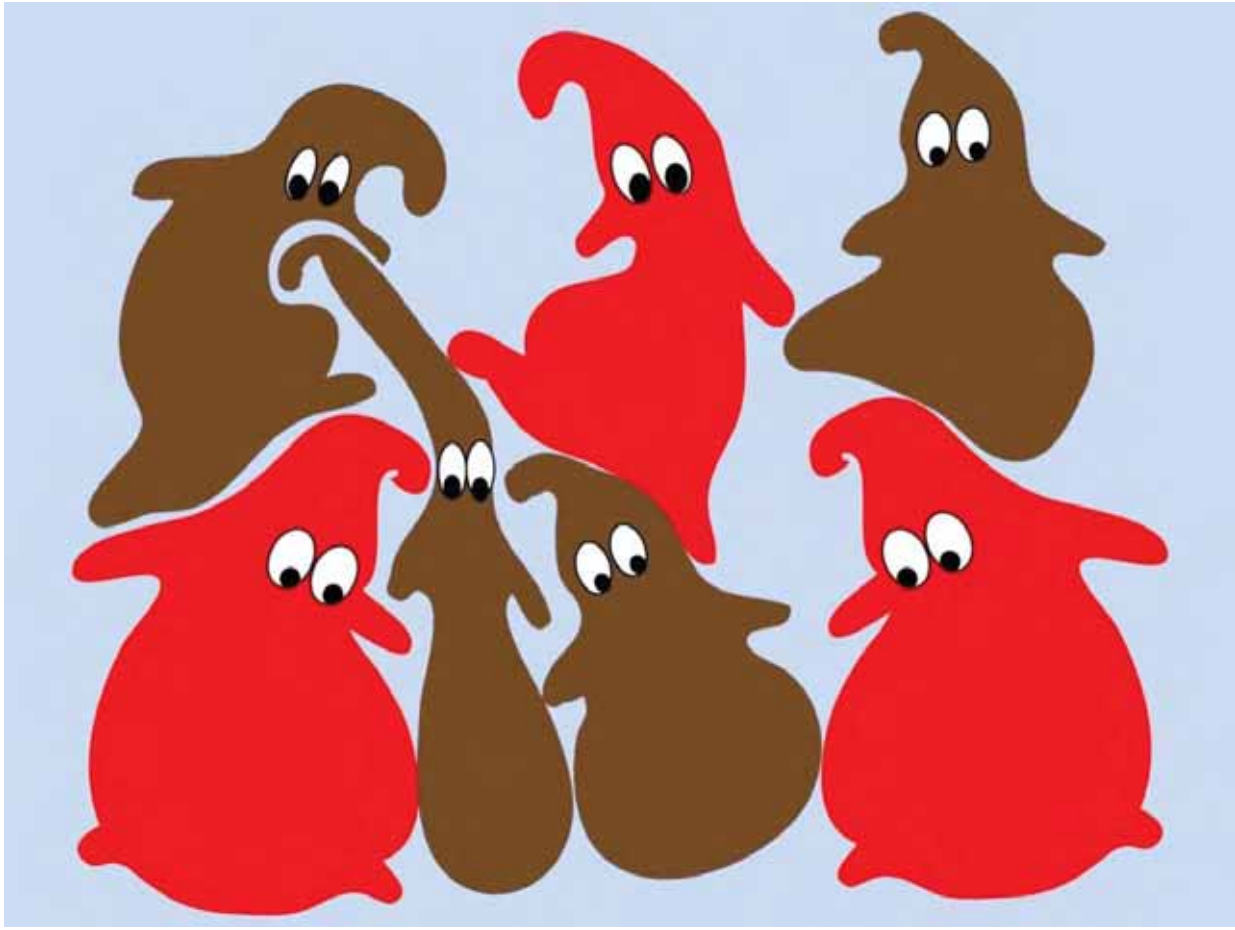
ki-67/MelanA double-staining



Inflammatory/stimulated nevus

Immunohistochemistry

- Use a red chromogene



Let’s take a deep dive into a few antibodies

- MelanA
- Sox10
- HMB45
- P16
- S100P

MelanA

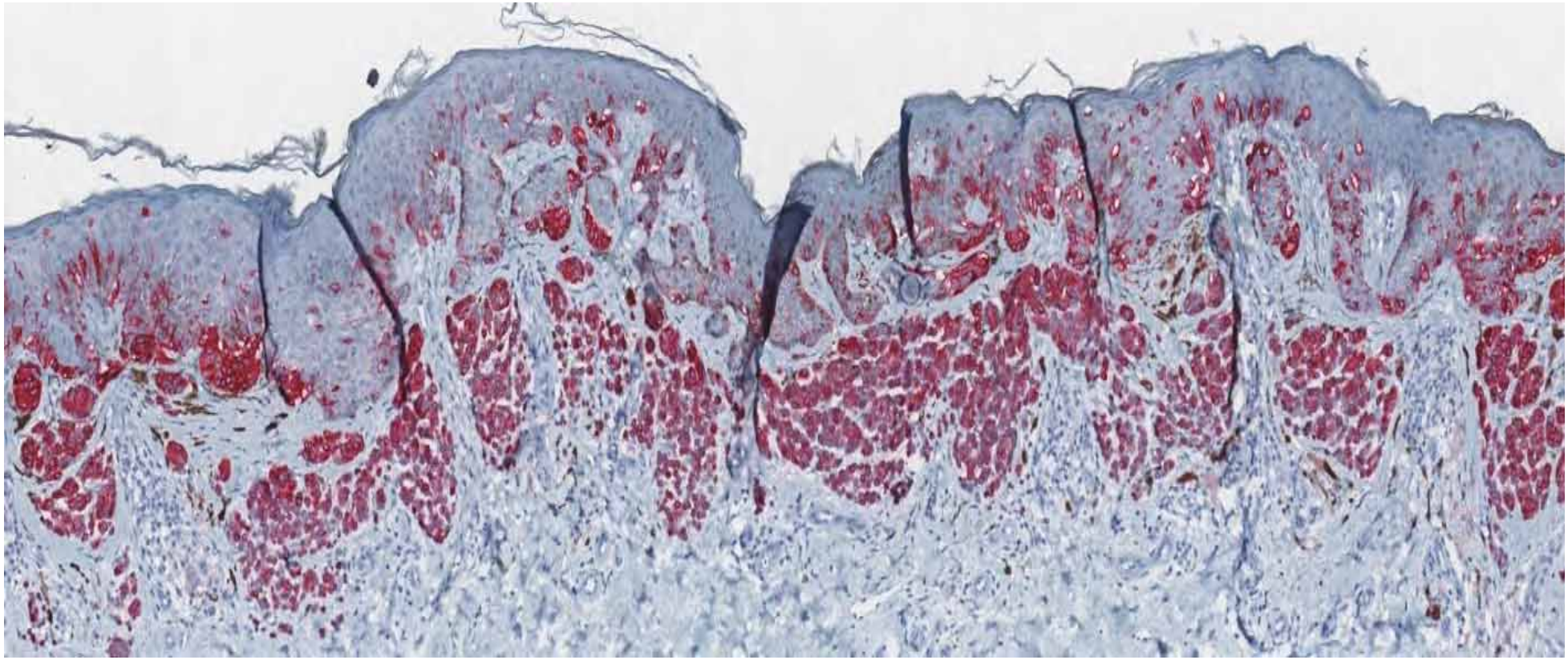
- Coded by *MLANA* gene (Chromosome 9p24)
- Involved in melanosome biogenesis by ensuring the stability of GPR143. Plays a vital role in the expression, stability, trafficking, and processing of melanocyte protein PMEL, which is critical to the formation of stage II melanosomes.
- Cytoplasmic stain
- Stains all normal melanocytes (internal control)

MelanA Tasks
Confirm and visualise melanocytic lineage

- All nevi
- All melanoma
- Sentinel lymph node /metastatic melanoma

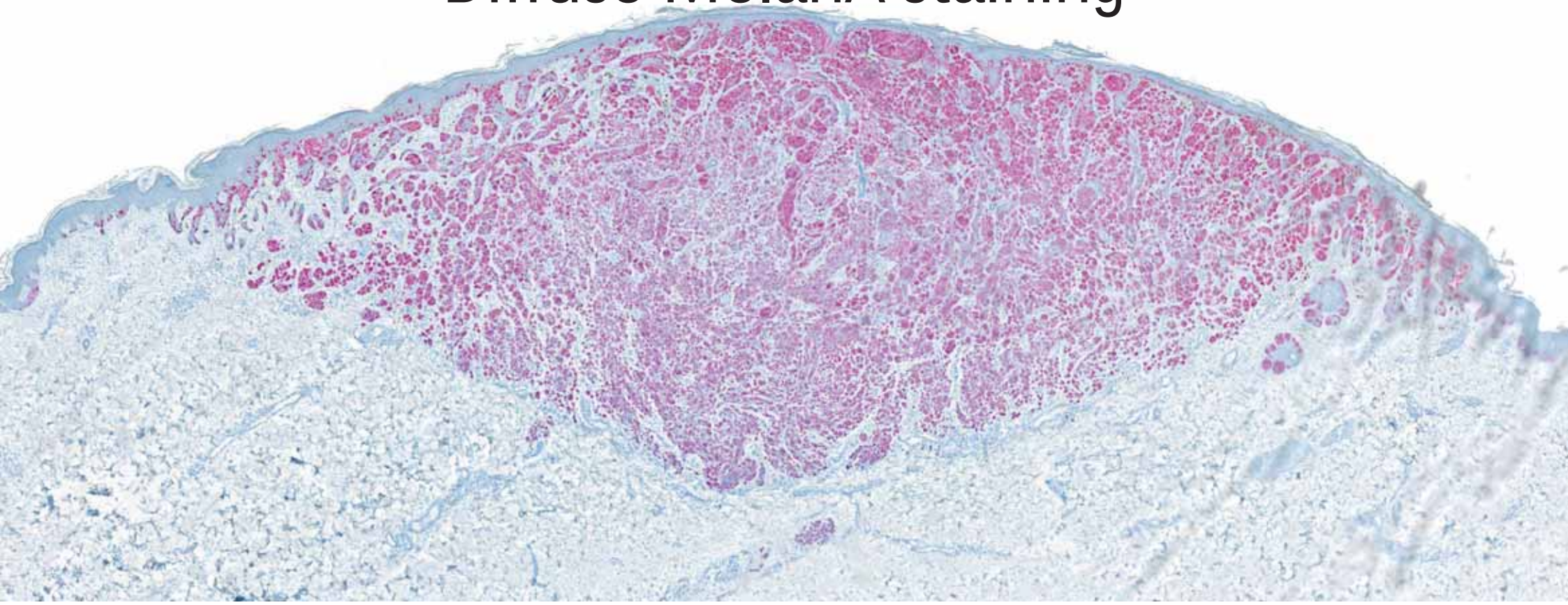
Common Nevus

Common nevus
Diffuse MelanA staining



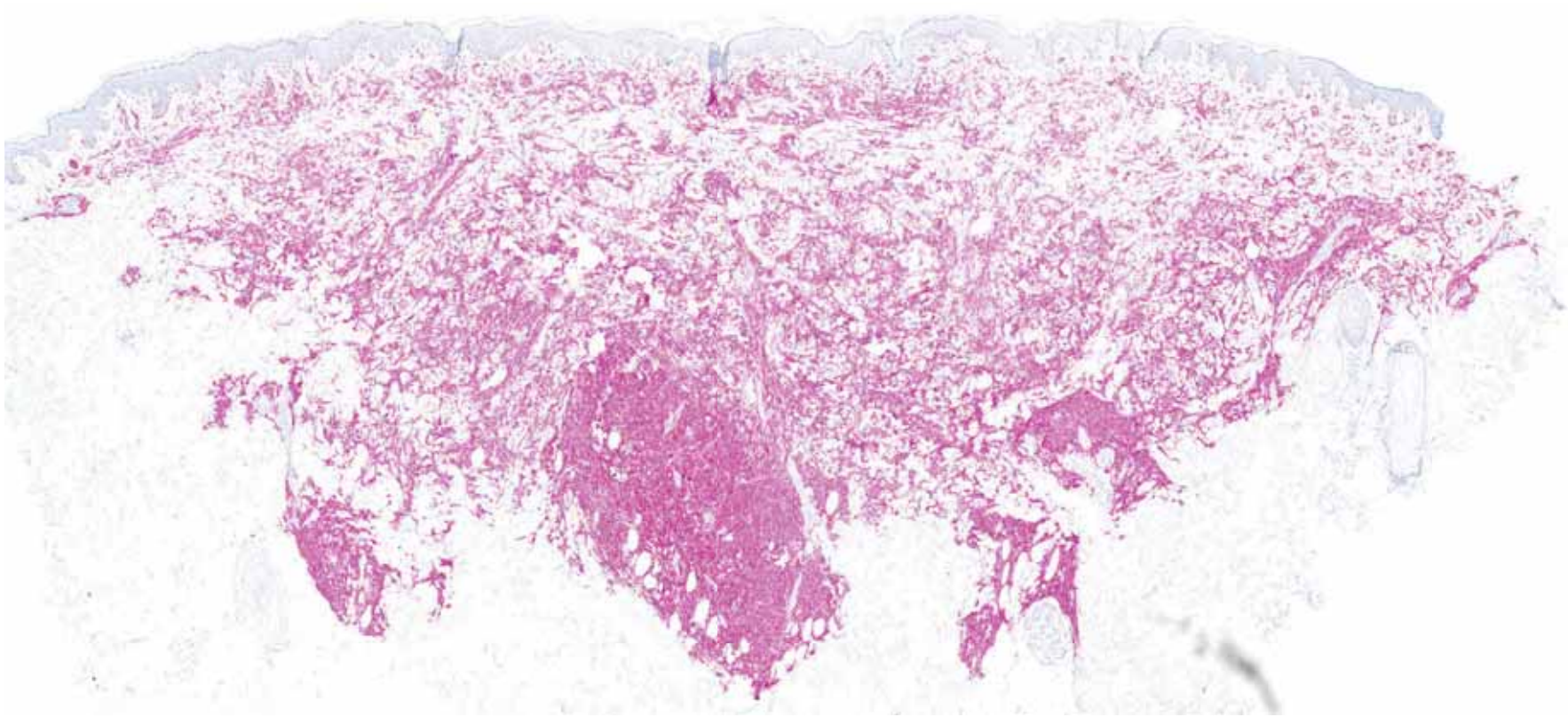
Spitz Nevus

Spitz nevus
Diffuse MelanA staining



Dendritic/Cellular blue nevus

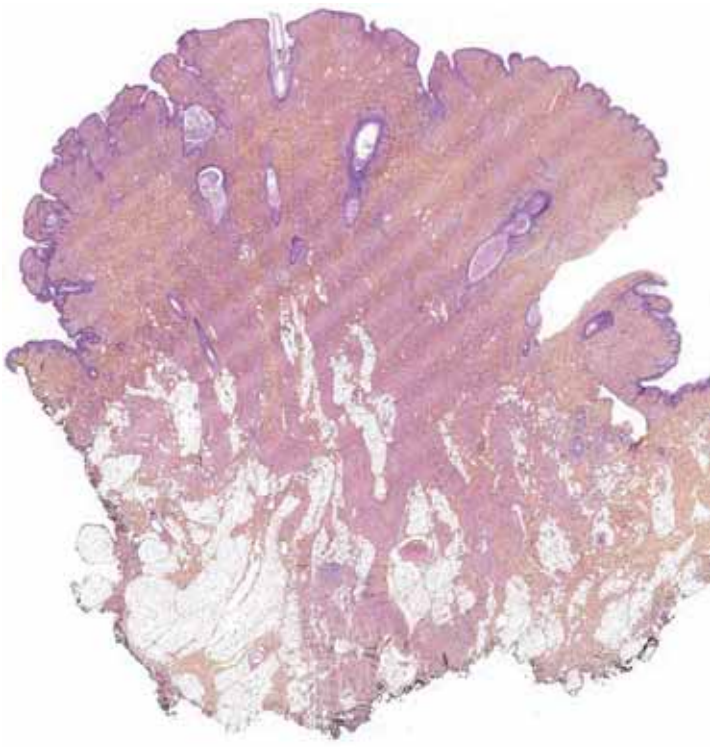
Blue nevus / CBN
Diffuse MelanA staining

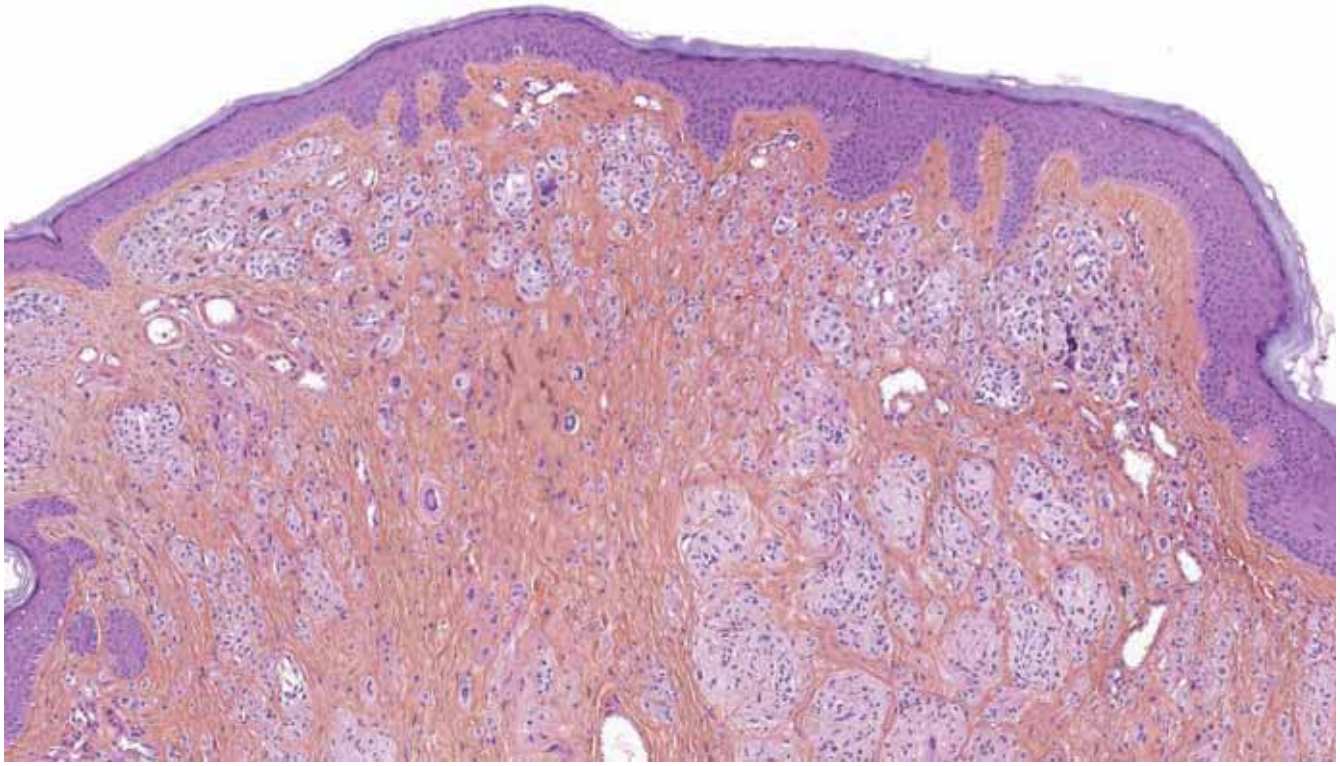


Expected melanA loss in nevi

- Type C melanocytes
- Nevi with a dermal myxoid component/MMYSTAR
- Desmoplastic nevus

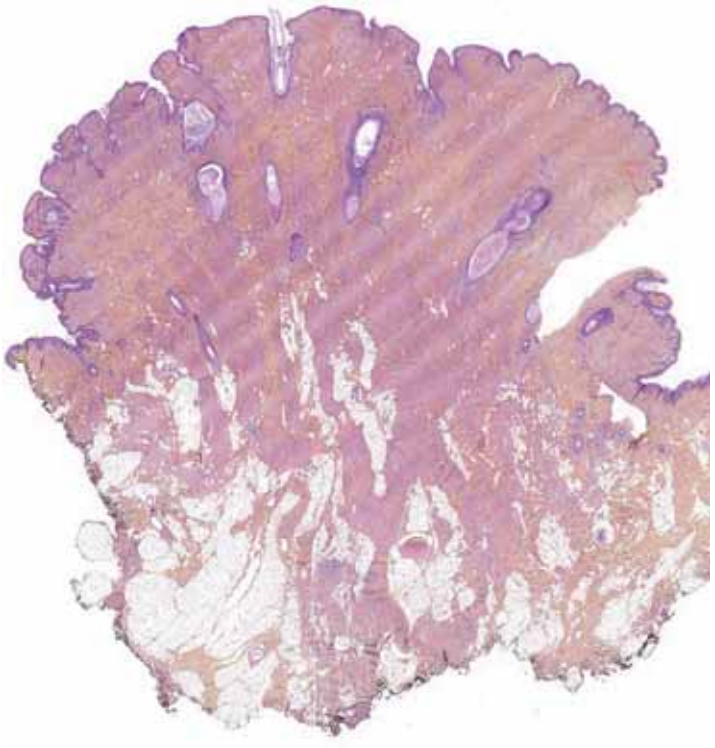
Expected melanA loss
Type C melanocytes

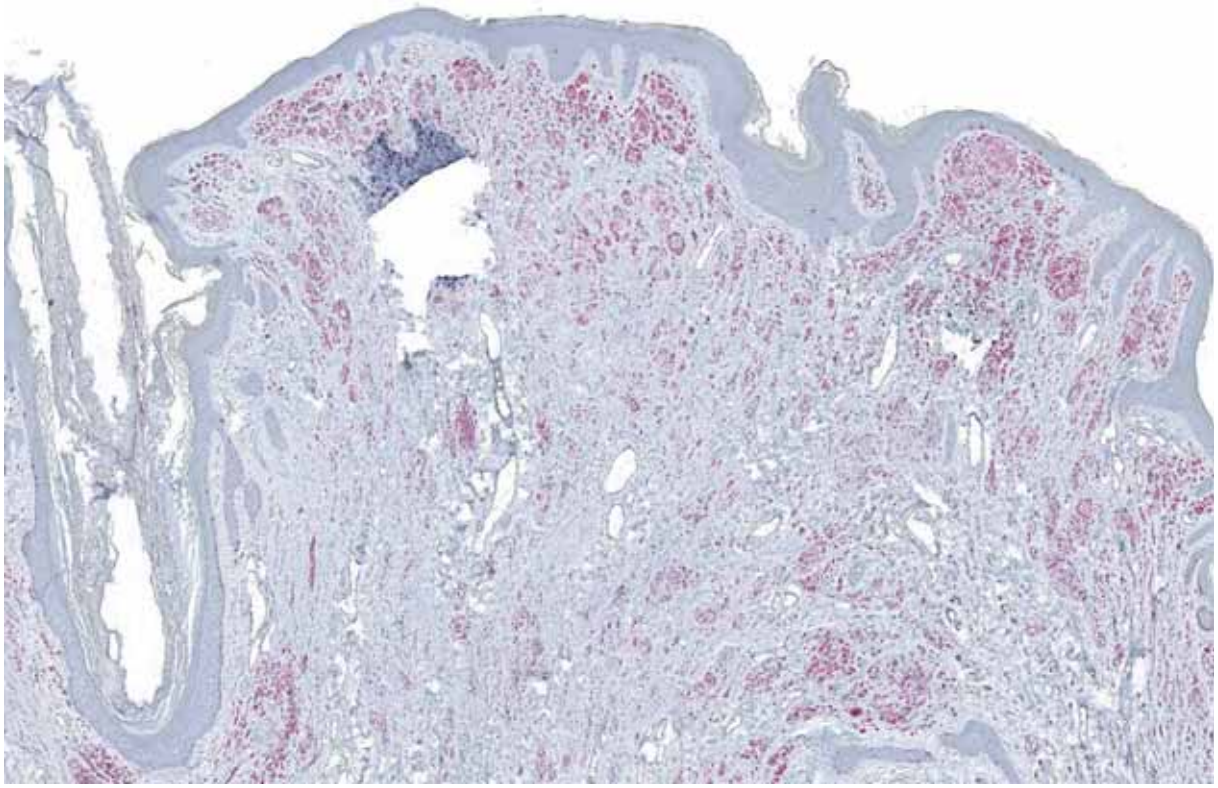




Expected melanA loss

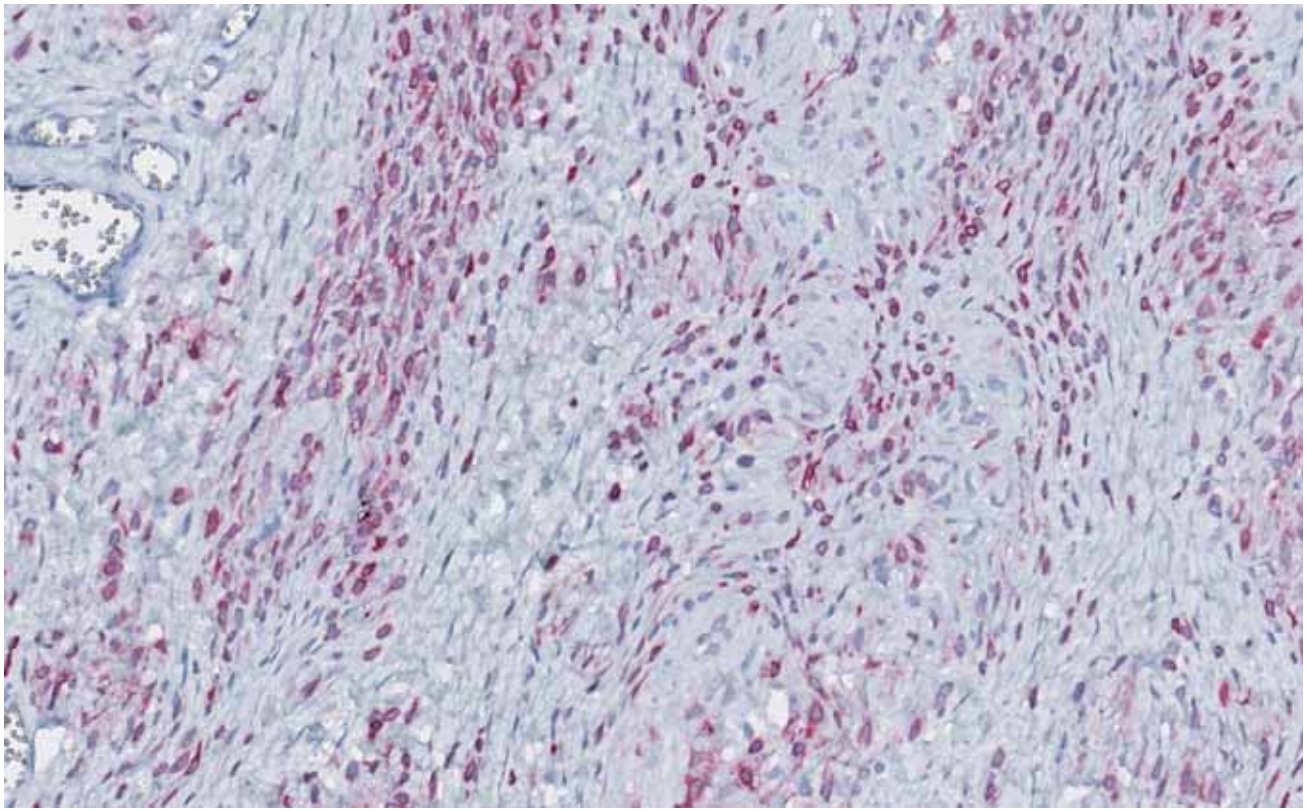
- Type C melanocytes



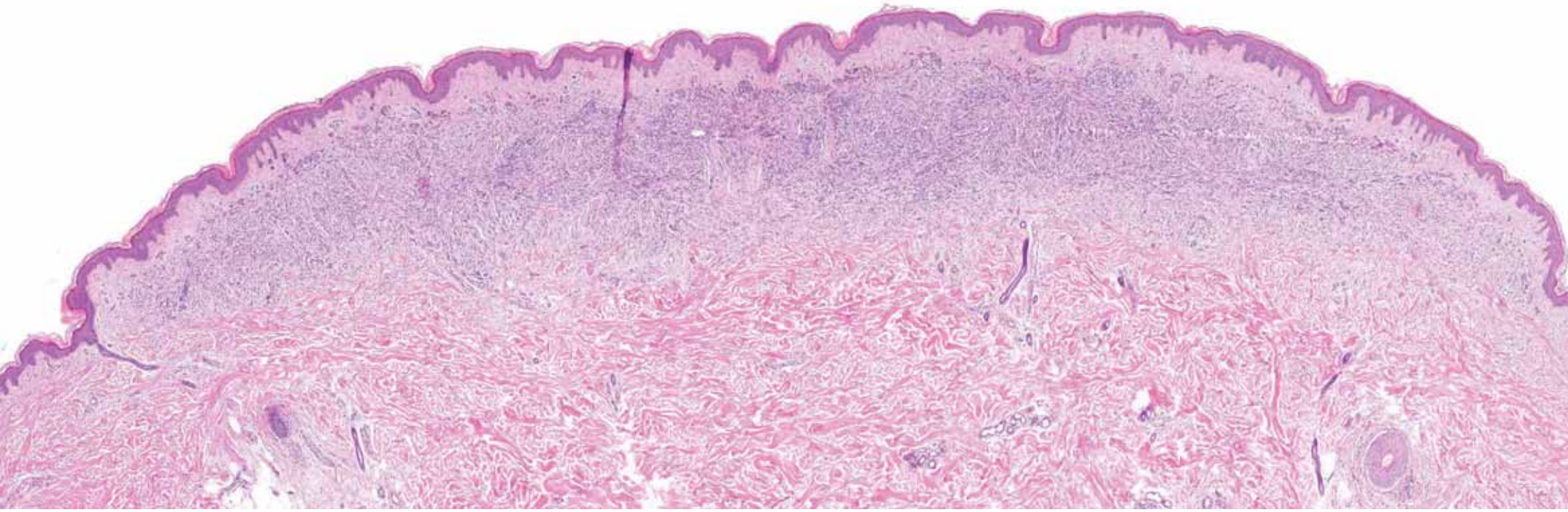


Expected melanA loss

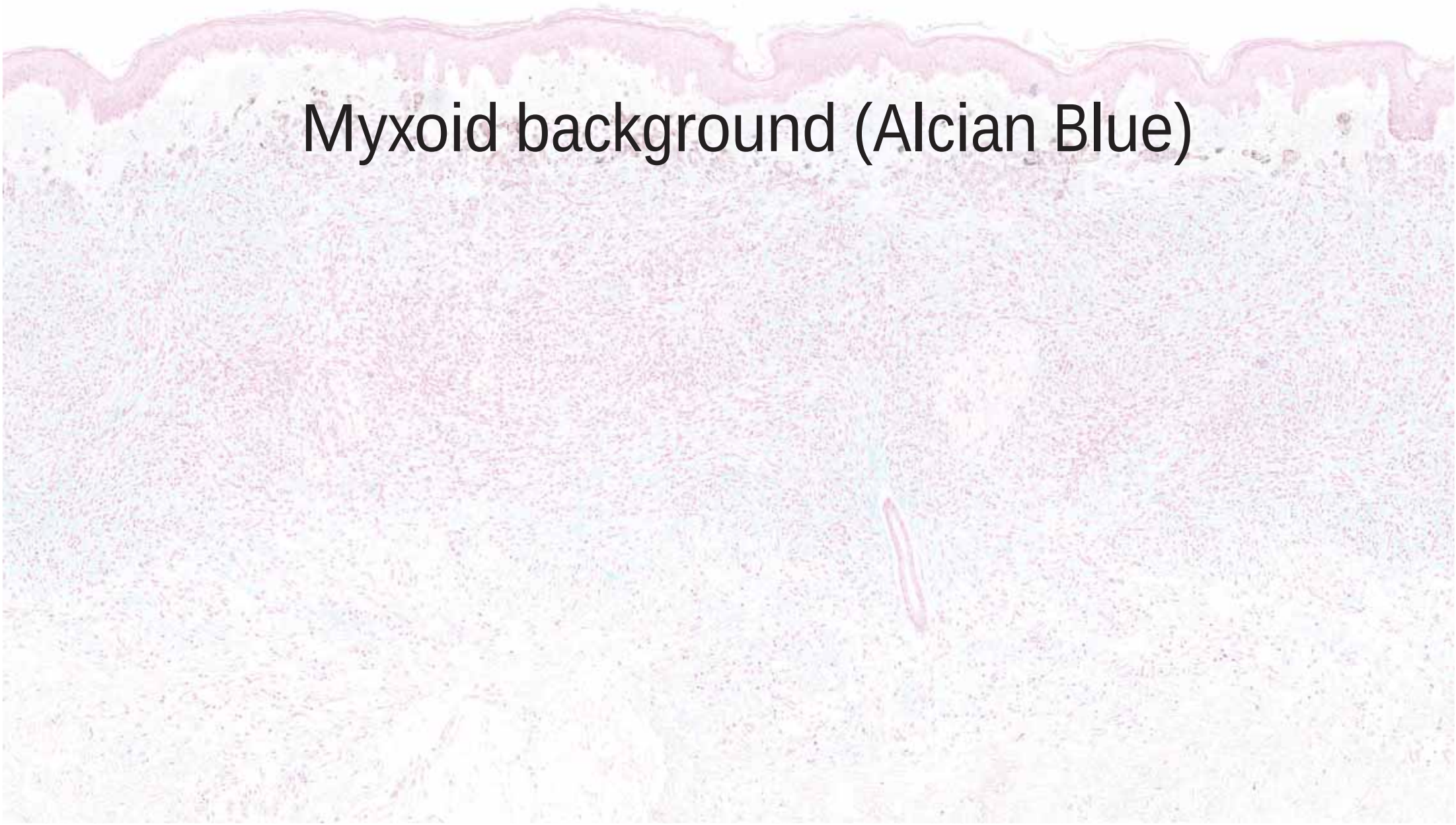
- Type C melanocytes



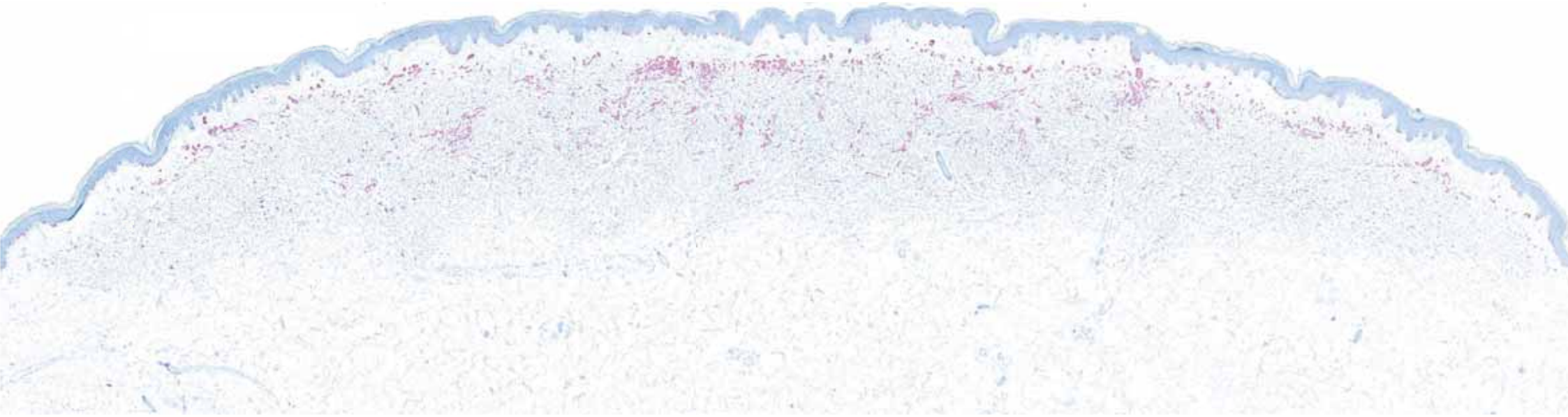
Expected melanA loss
F42 Thigh; Myxoid nevus NRAS p.Q61K

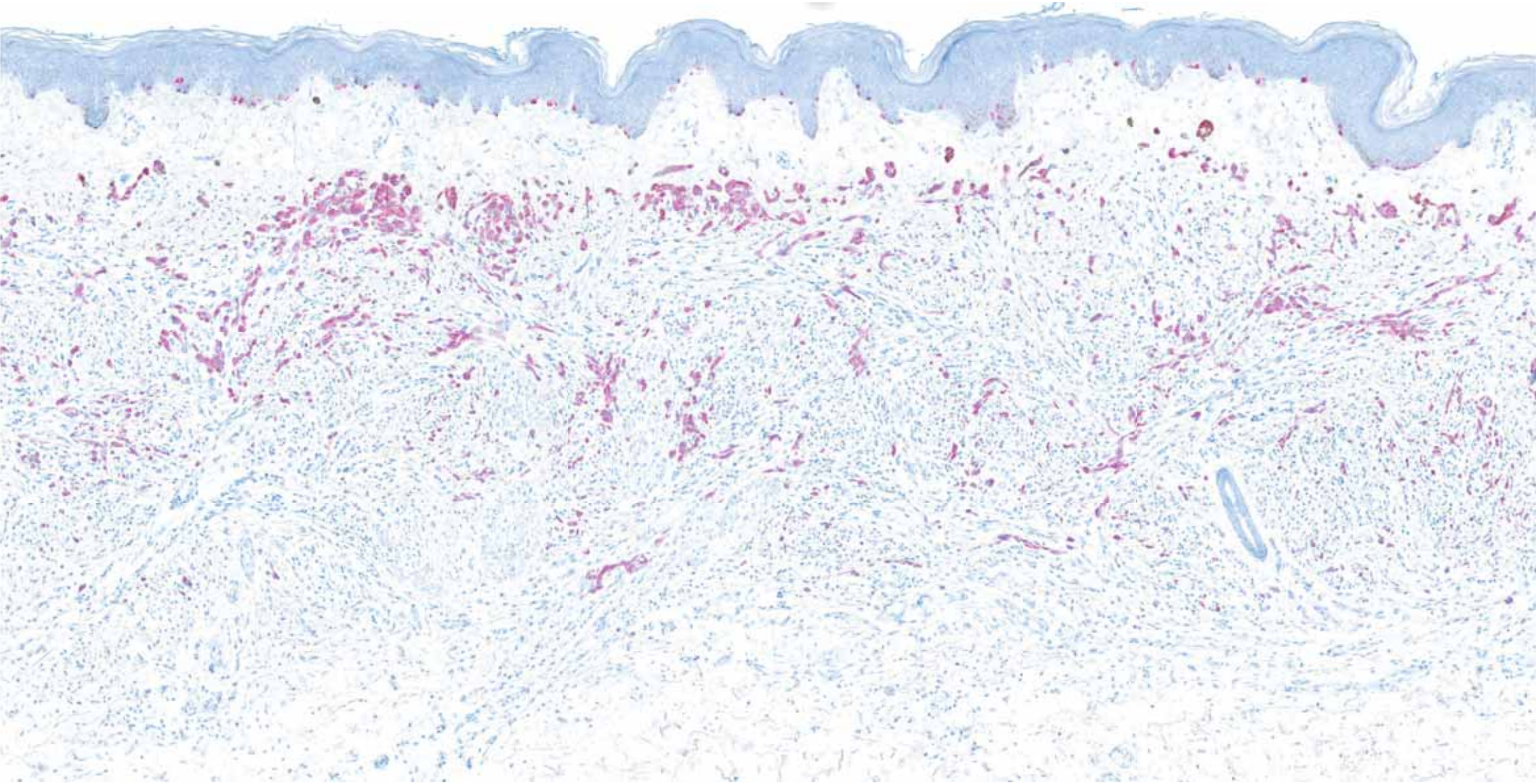


Myxoid background (Alcian Blue)



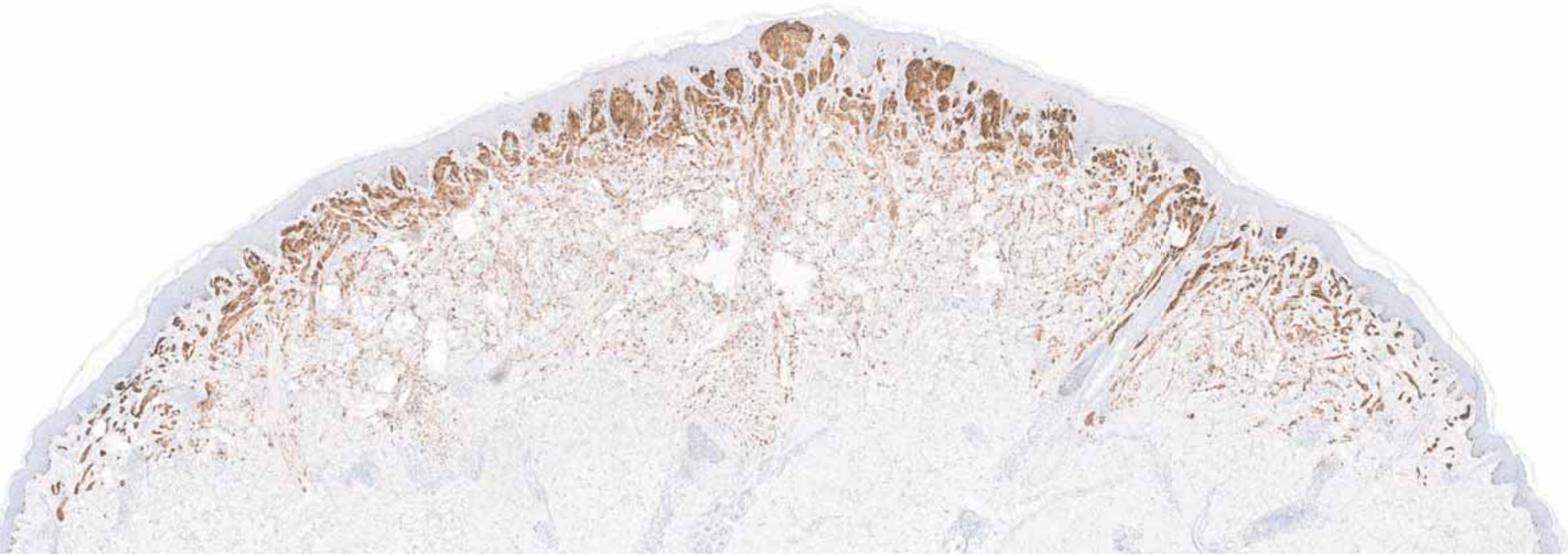
Expected melanA loss
myxoid background



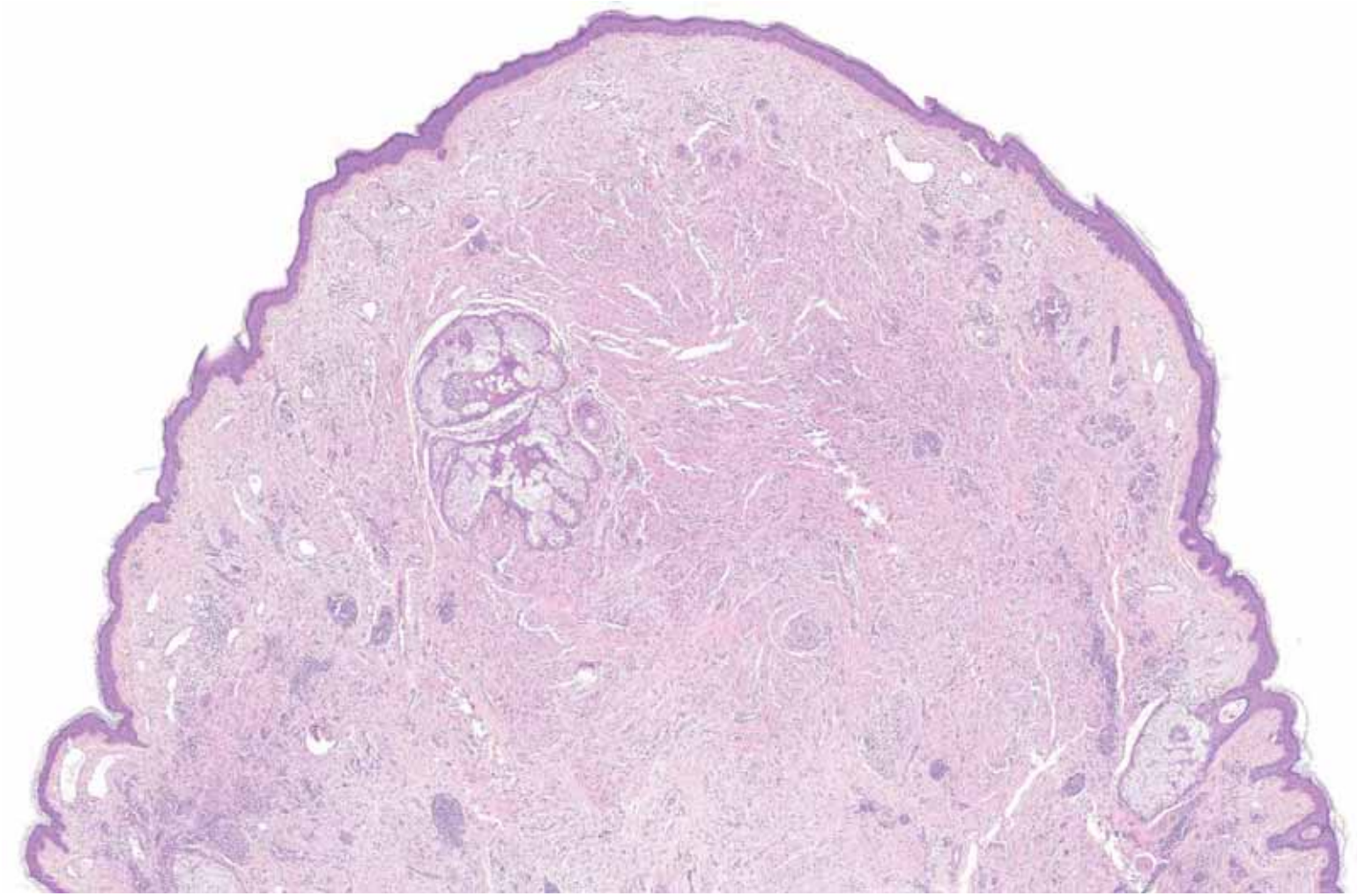


Desmoplastic nevus

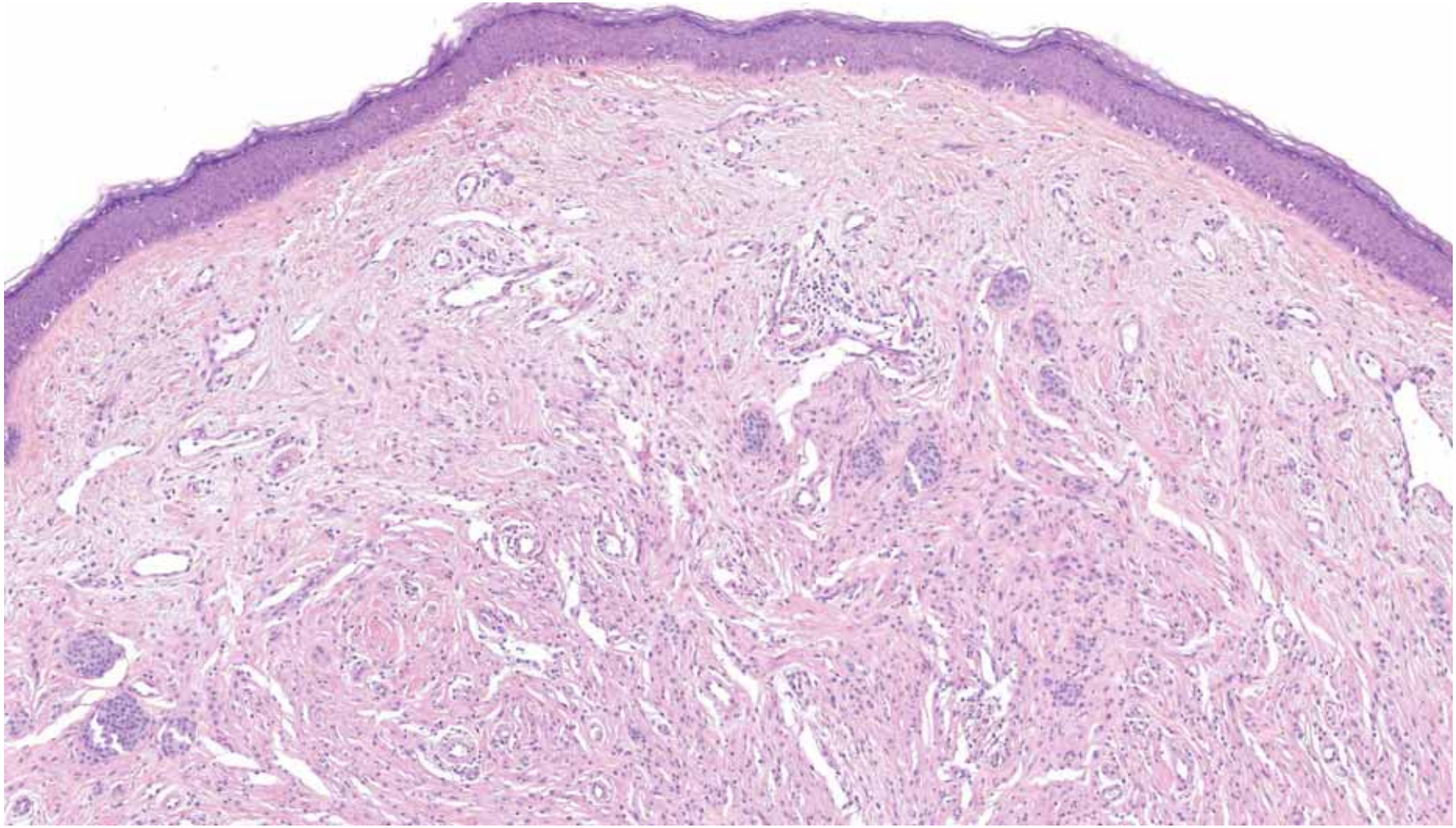
MelanA is often top heavy



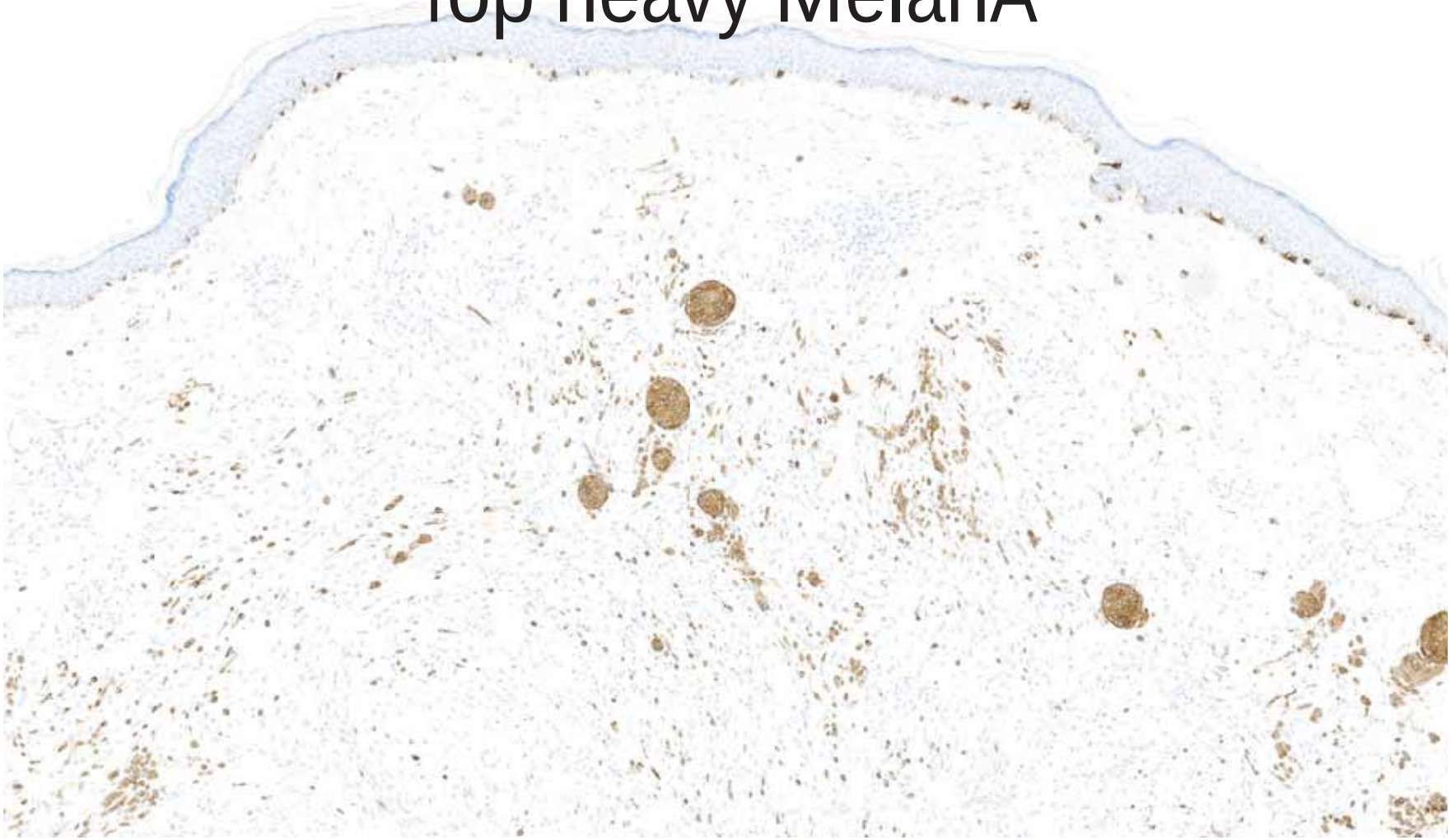
Fibrous involuting nevus (BRAF V600E)



Fibrous involuting nevus (BRAF V600E)



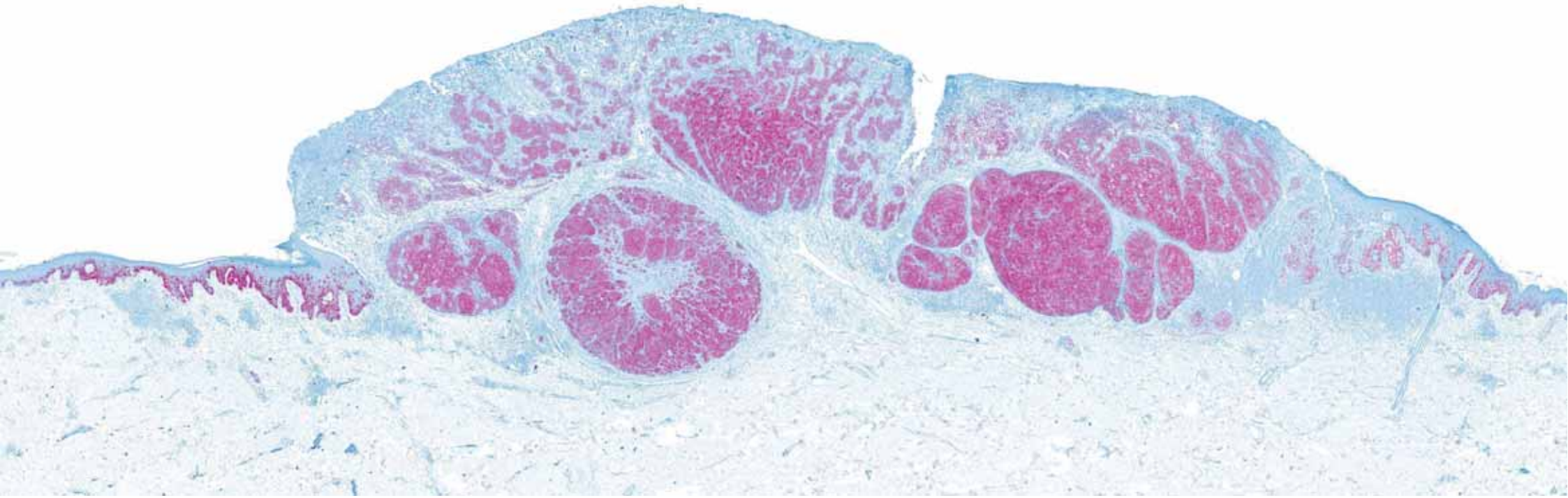
Fibrous involuting nevus (BRAF V600E)
Top heavy MelanA



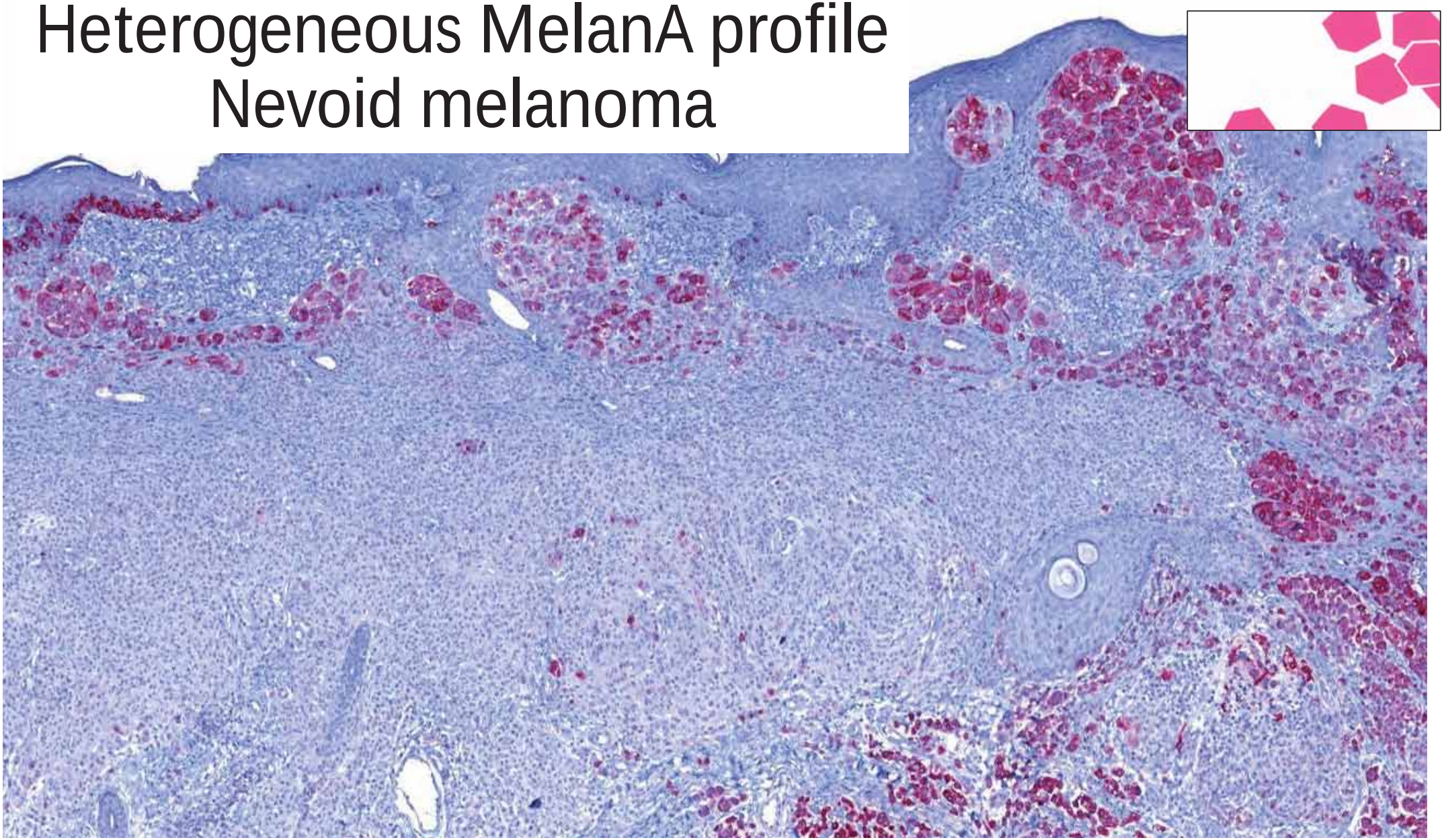
Expected melanA loss in melanoma

- Inconstant, usually the junctional component stays positive
- Nevoid melanoma (patchy positivity)
- Melanomas with spindled component / Desmoplastic Melanoma
- Melanomas with a dermal myxoid component

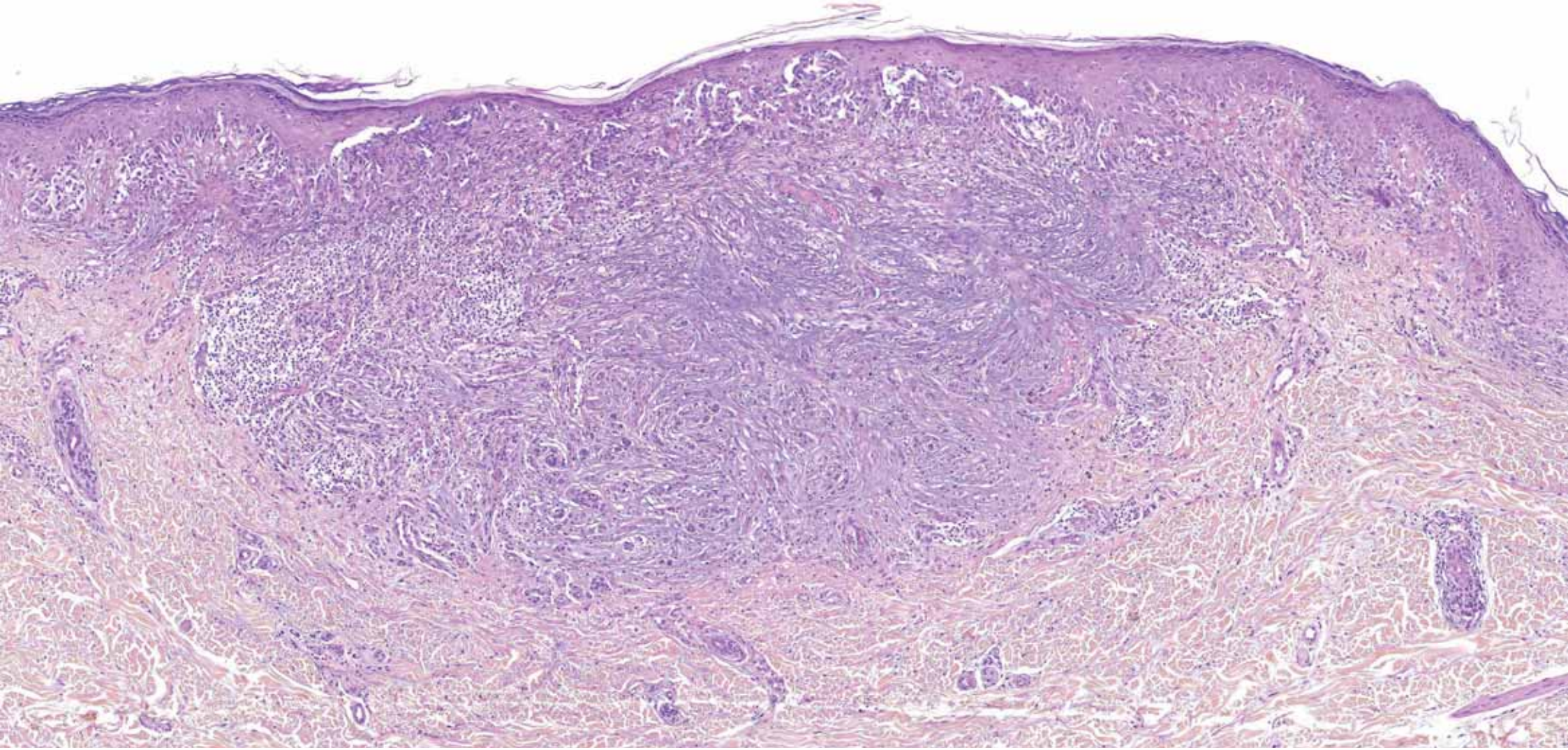
Diffuse MelanA in common melanoma



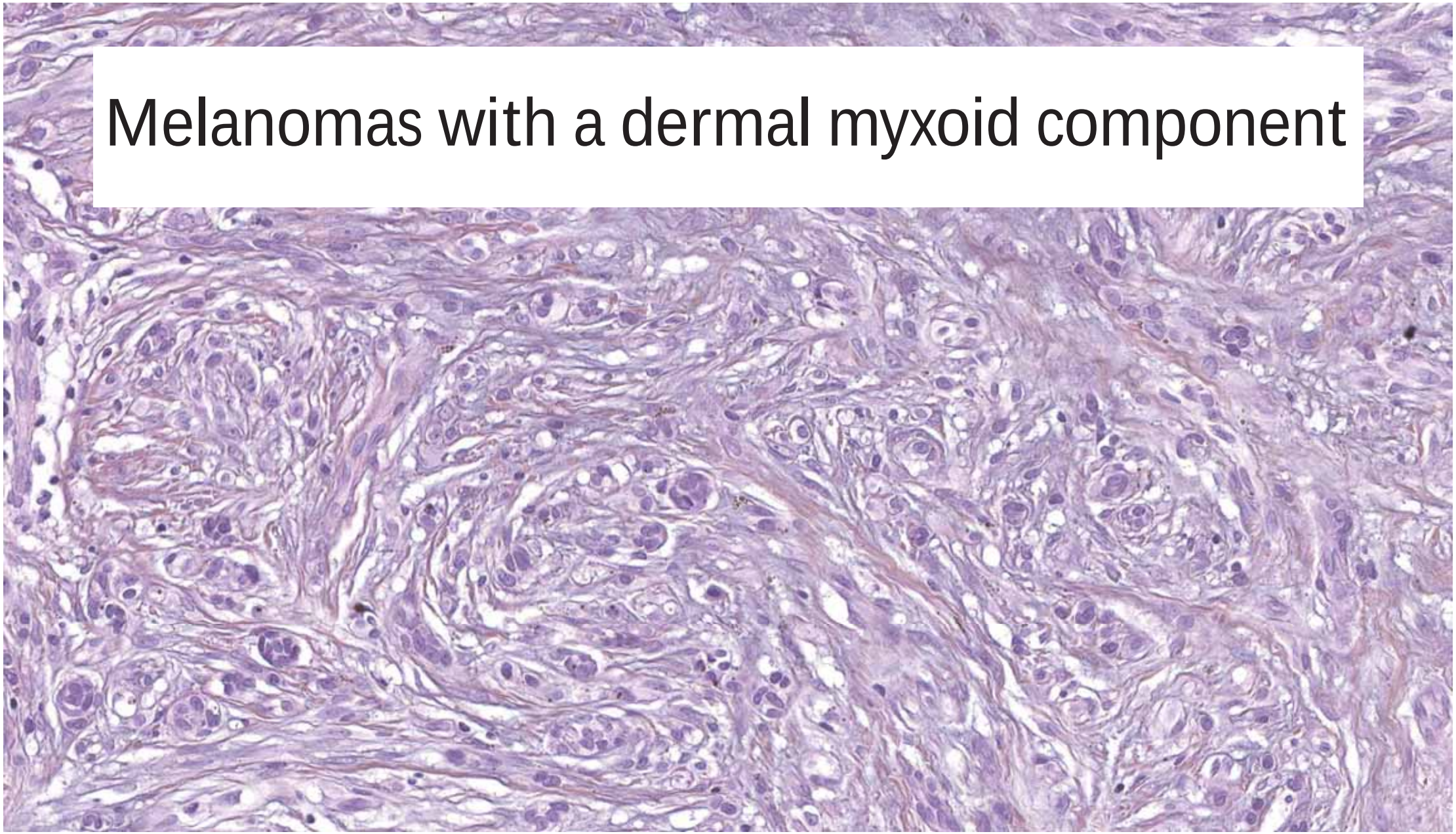
Heterogeneous MelanA profile
Nevoid melanoma

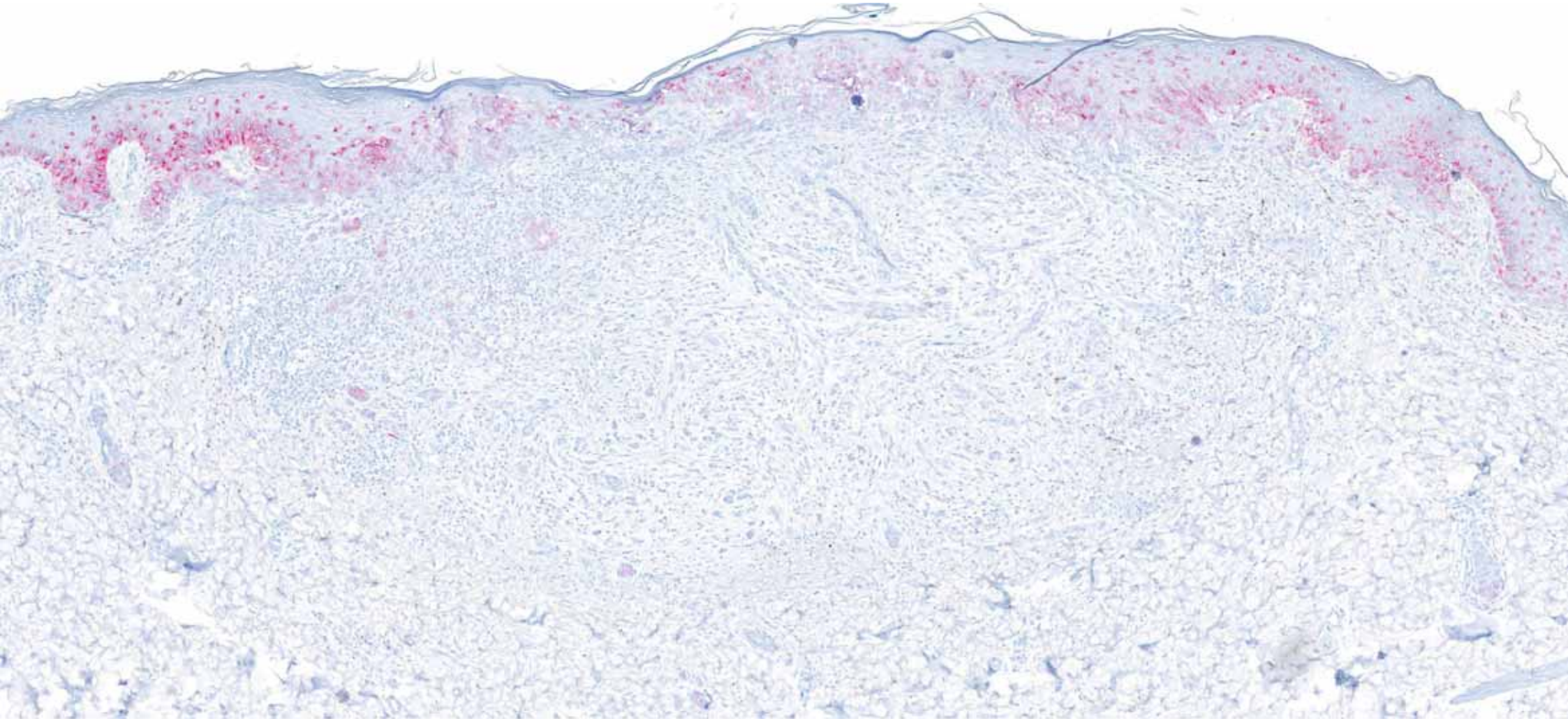


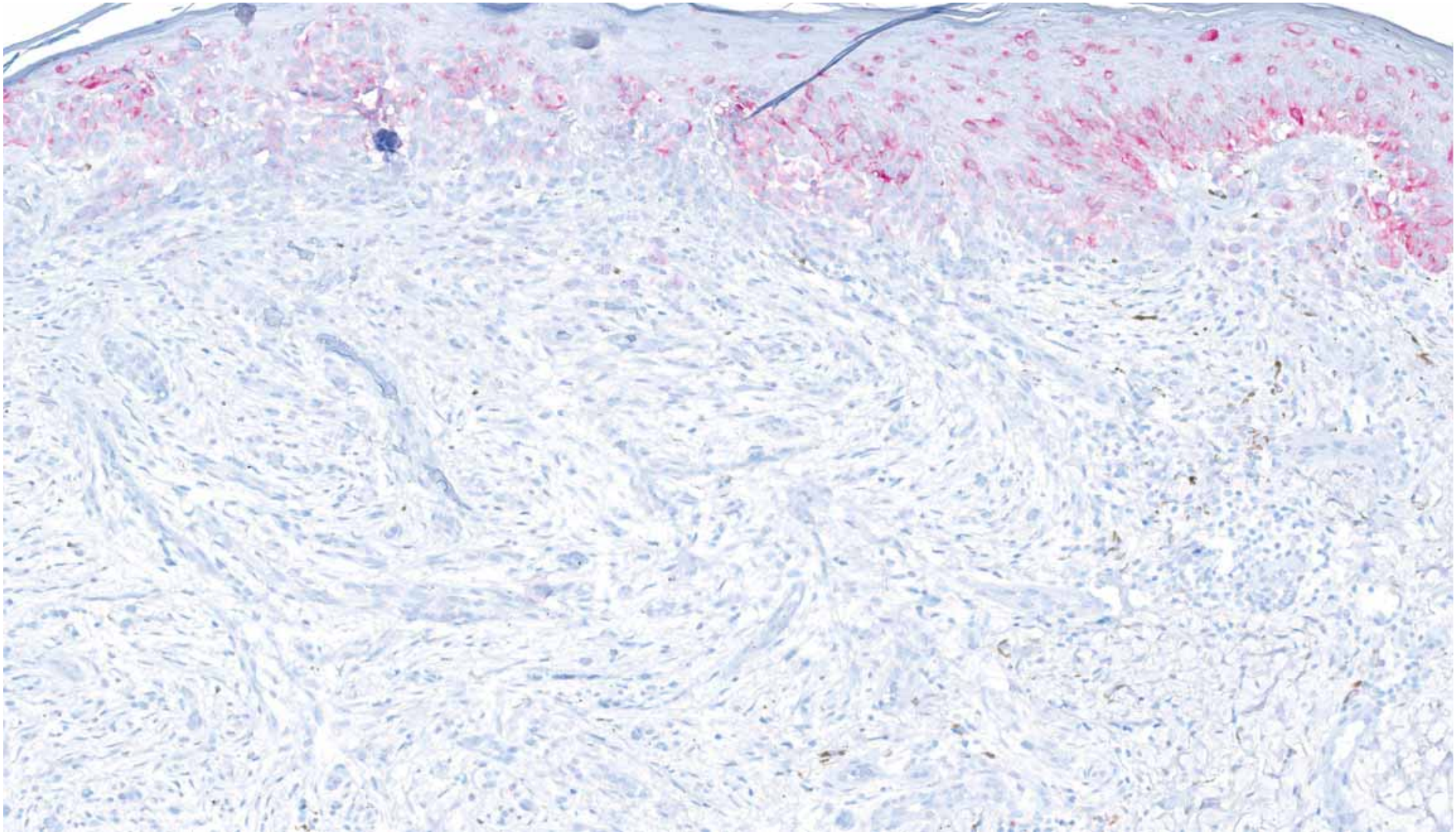
Melanomas with a dermal myxoid component



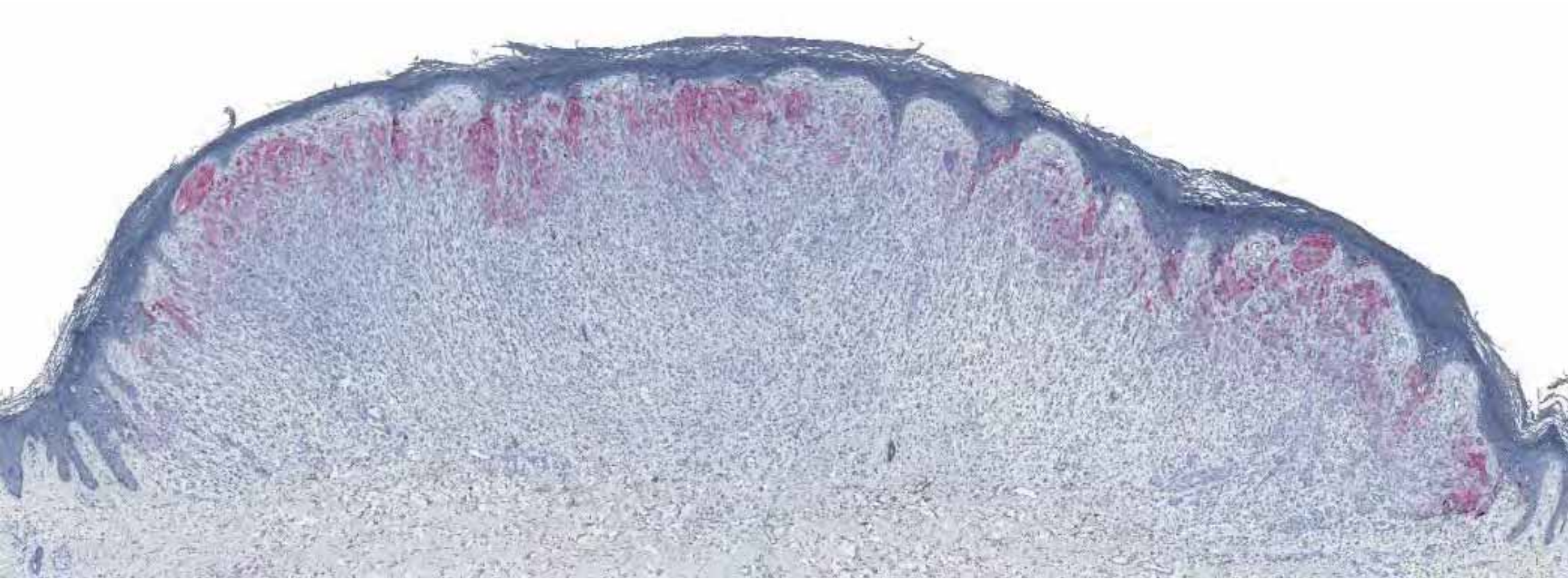
Melanomas with a dermal myxoid component



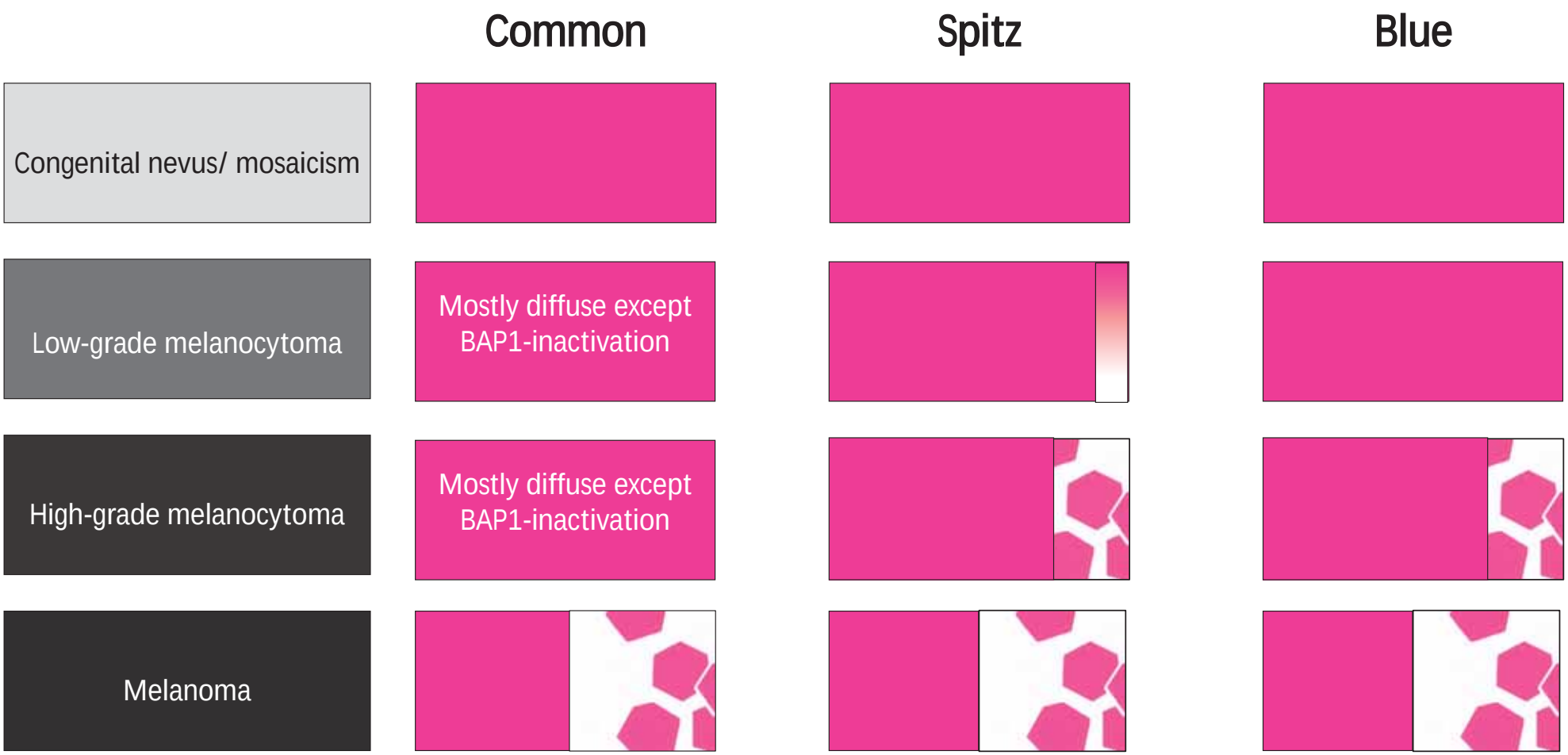




Top-heavy MelanA in nevoid melanoma



Melan A synoptic IHC expression chart

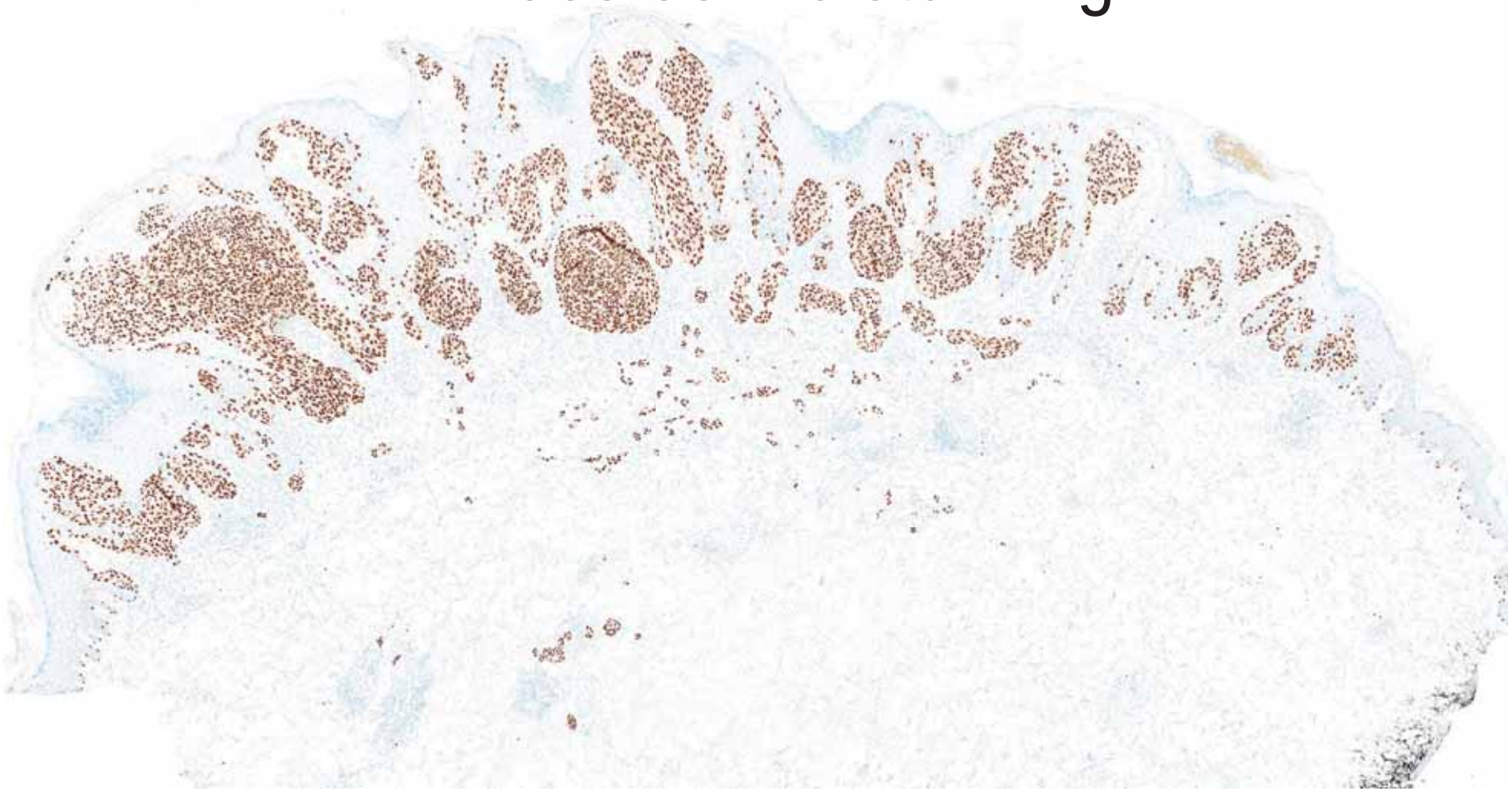


Transcription factor Sox10

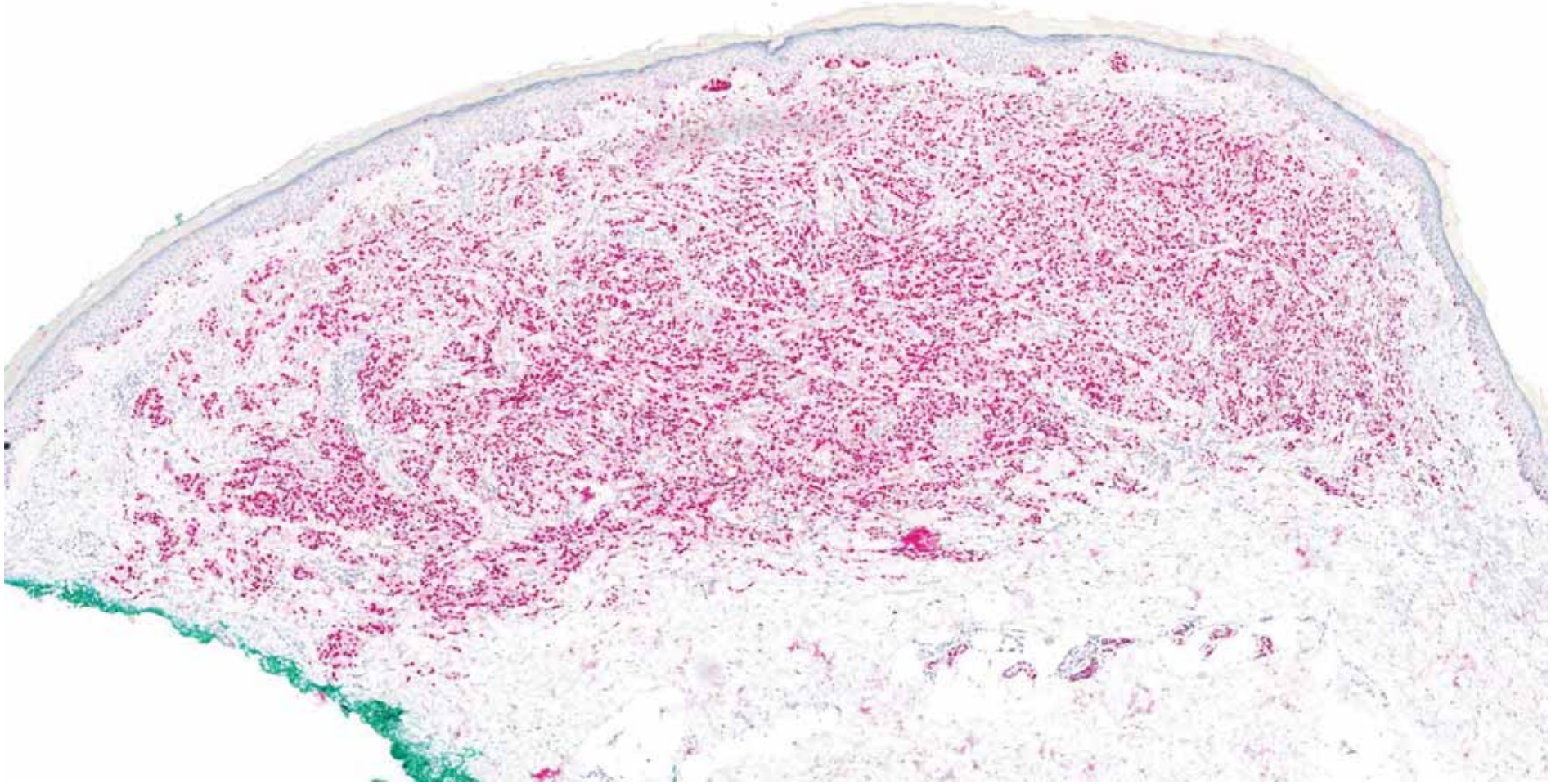
- Coded by *SOX10* gene : SRY (Sex Determining Region Y)-Box 10 (Chr.22)
- Involved in neural crest differentiation, olidendrocyte specification and differentiation
- Transcriptional activator of MiTF
- Stains all normal melanocytes (nuclear stain)
- Internal controls : normal melanocytes and nerves

Spitz Nevus

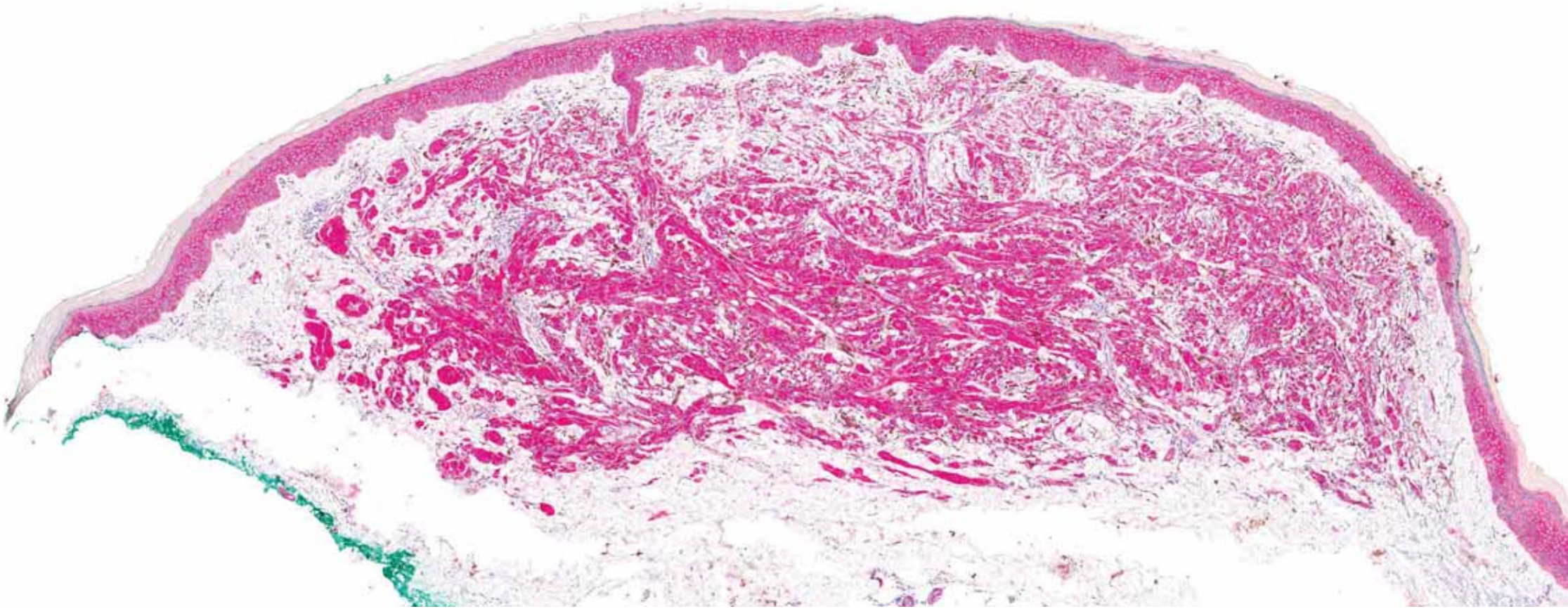
Spitz nevus
Diffuse Sox10 staining



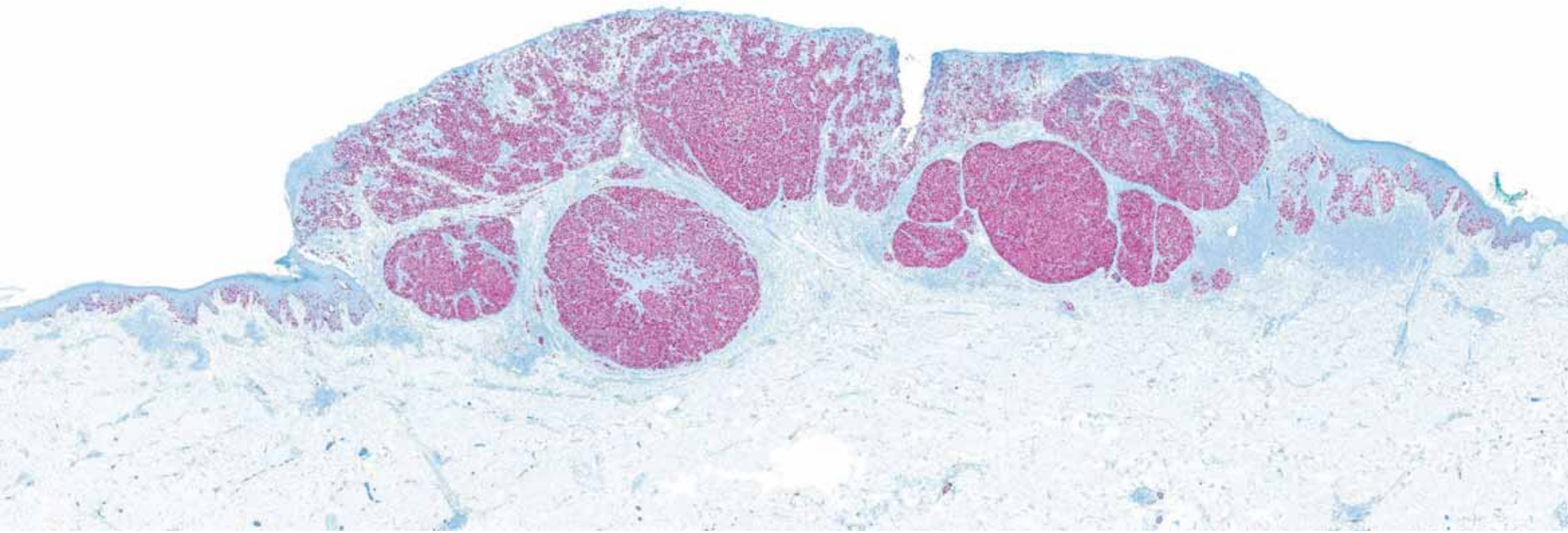
WNT-activated melanocytoma
Sox10 is positive but doesnt outline architecture



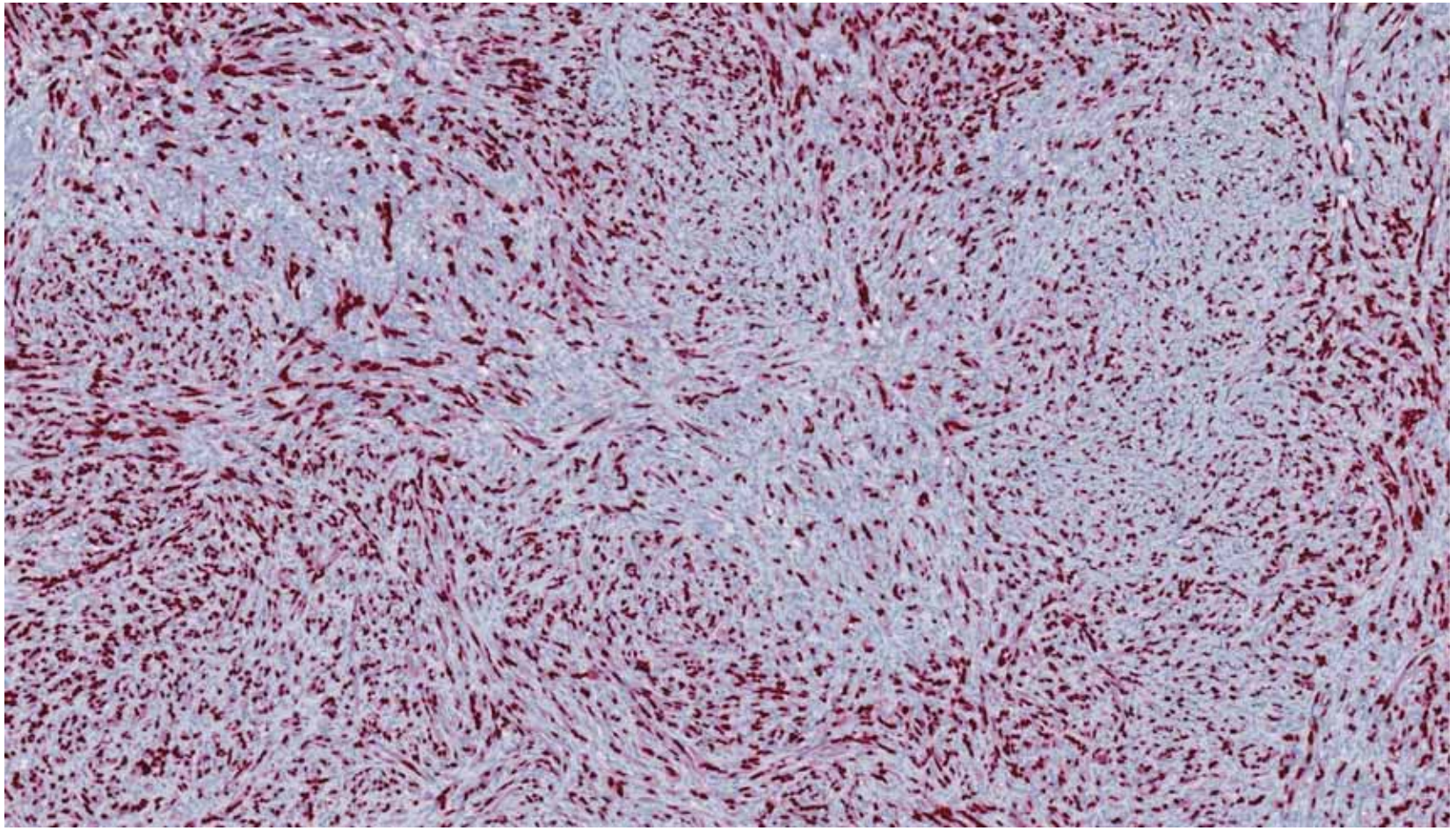
Beta-catenine outlines architecture in WNT-activated melanocytoma



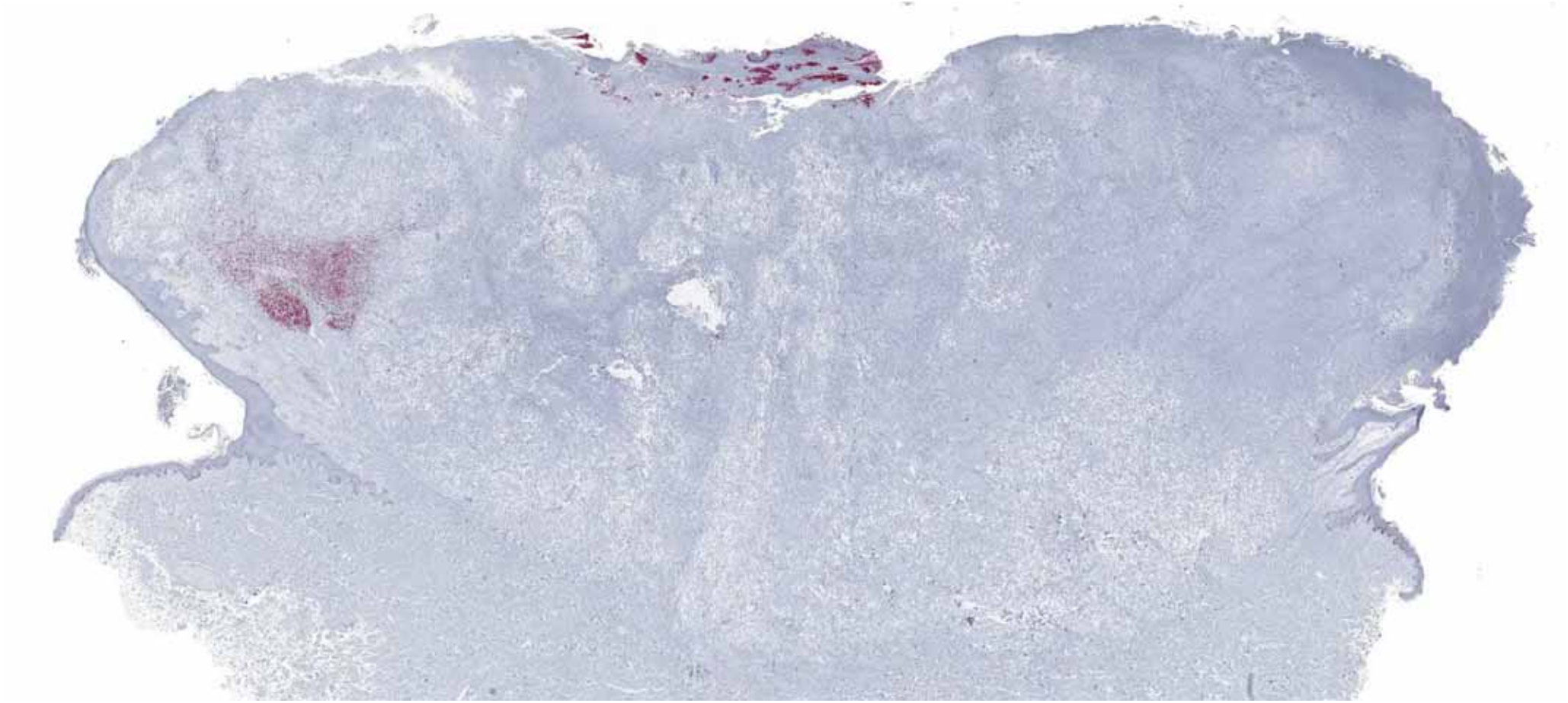
Sox10 in common melanomas



Sox10 is helpful (mandatory?) in the diagnosis of Desmoplastic Melanoma



Sox10 is diffusely positive in melanocytic tumors except dedifferenciated melanoma



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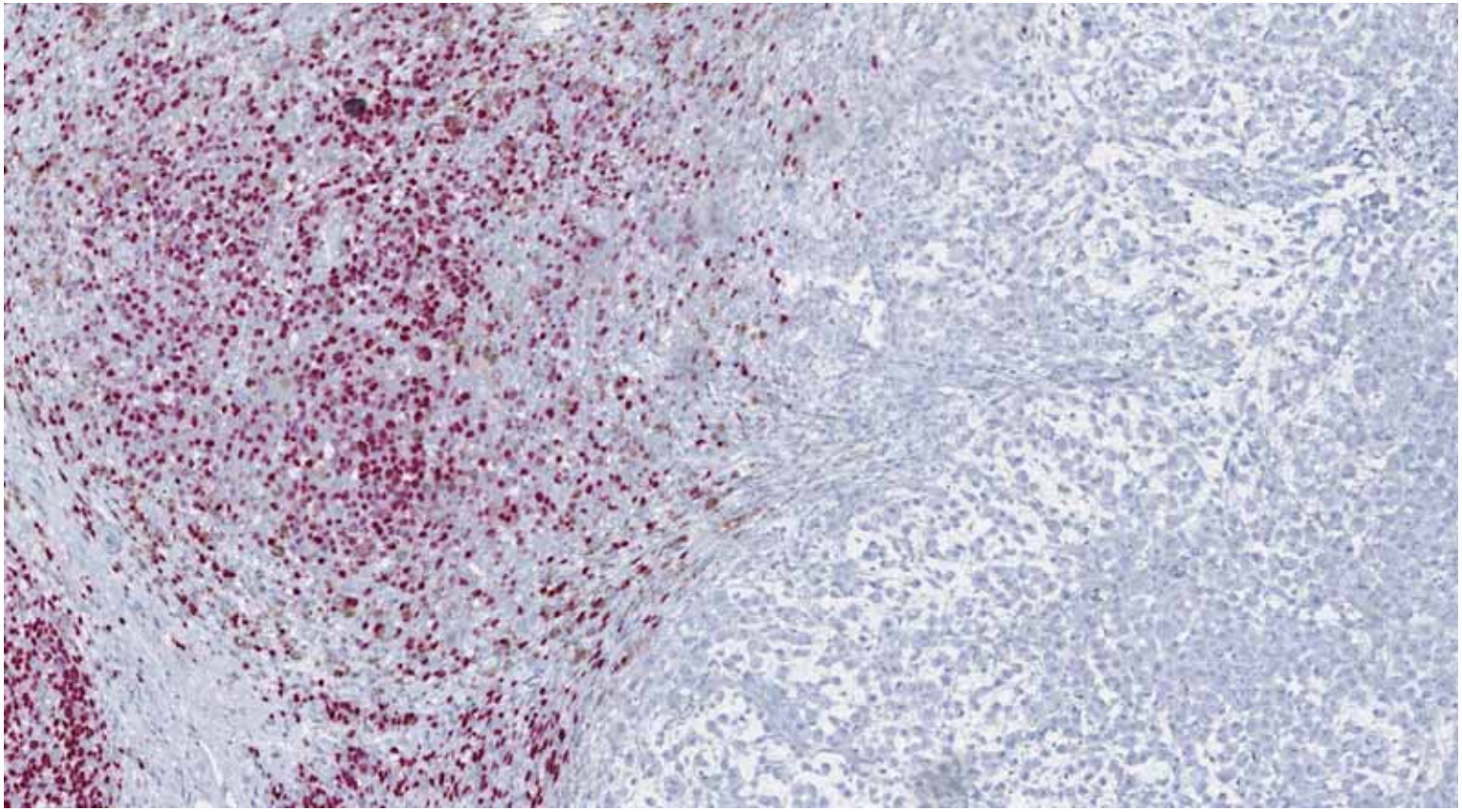
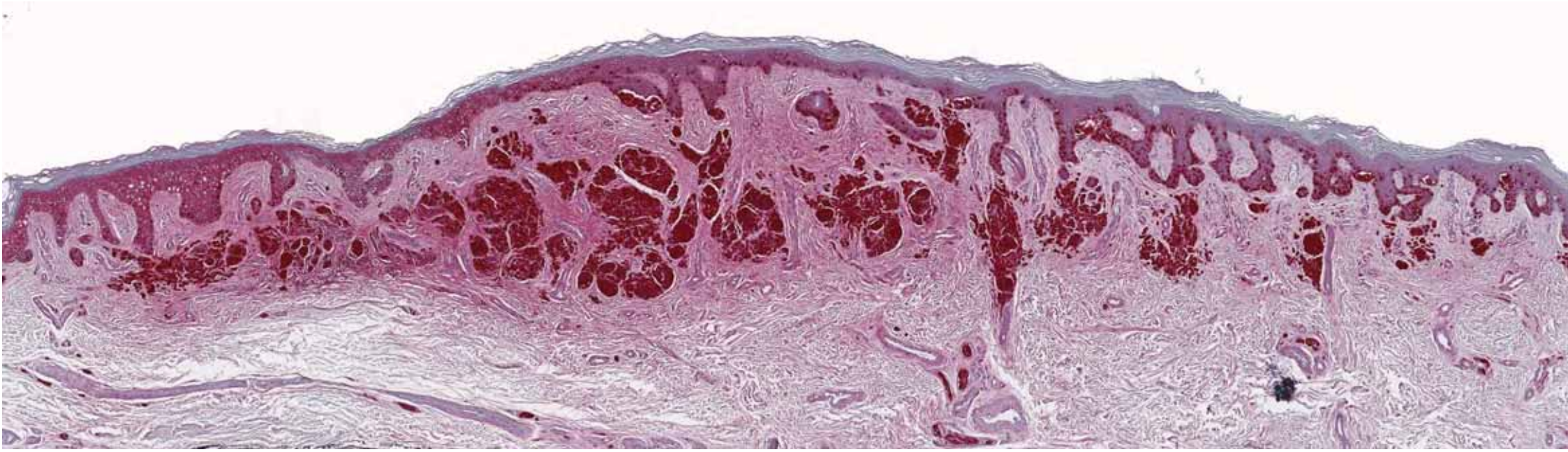


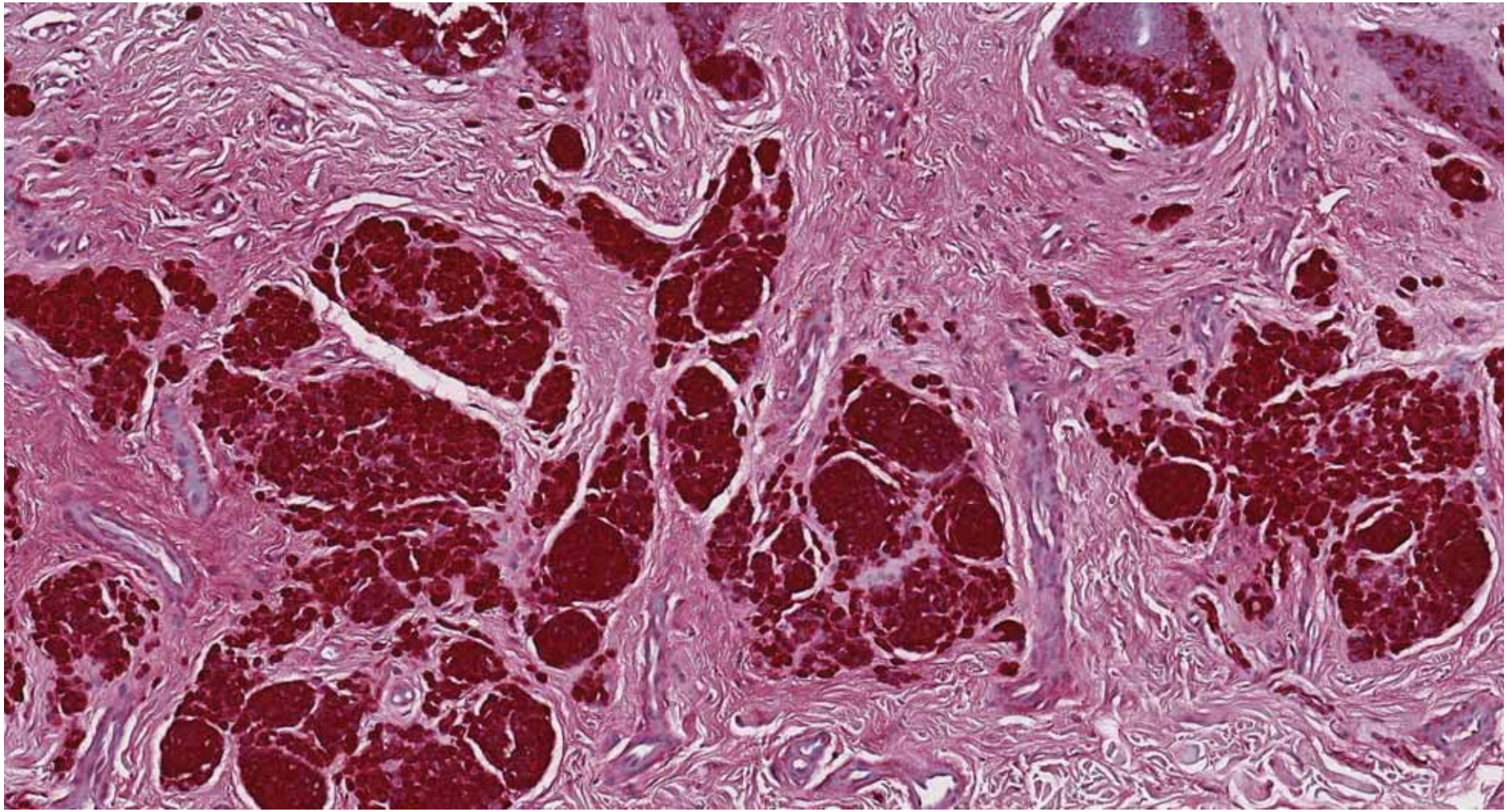
S100B Protein

- Coded by *S100B* gene (Chr 21) part of a family of S100 proteins
- Small zinc and calcium binding protein
- Multiple functions including melanocyte proliferation
- Cytoplasmic and nuclear staining of melanocytes
- Extra-cellular activity
- Internal controls : nerves, adipocytes, Langerhans cells,…

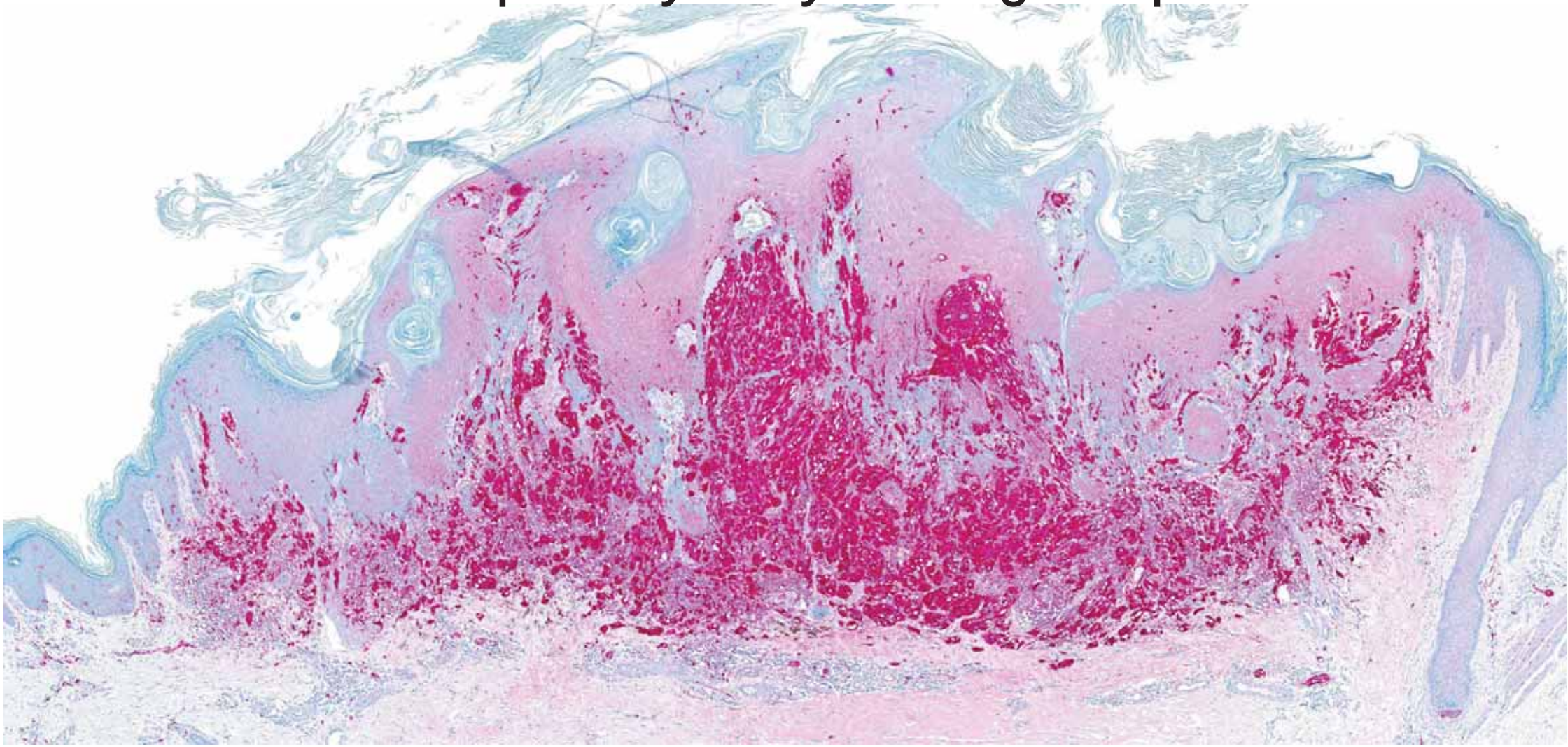
S100 in Common nevus



S100 in Common nevus



S100 is frequently very strong in Spitz nevi



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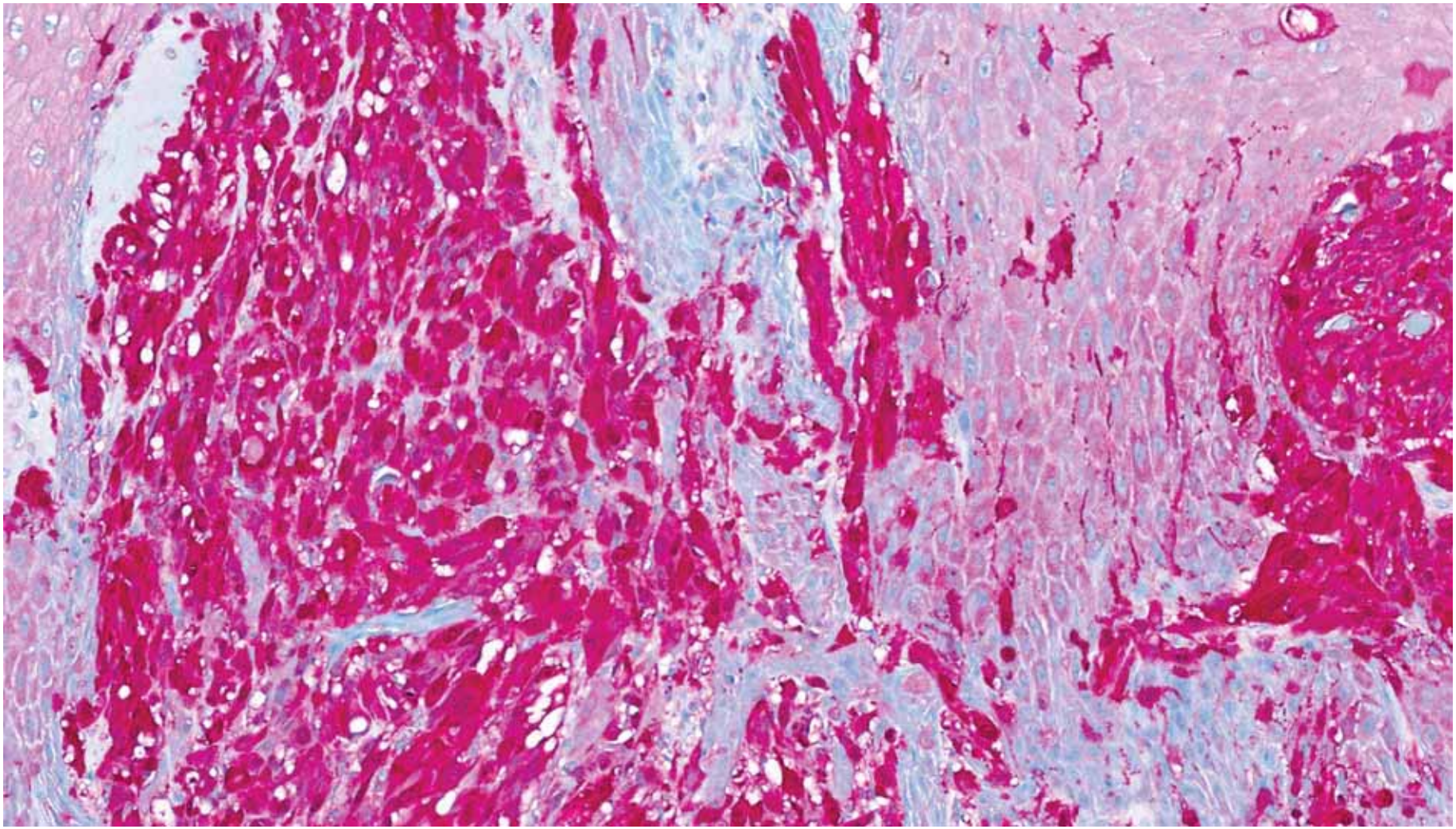
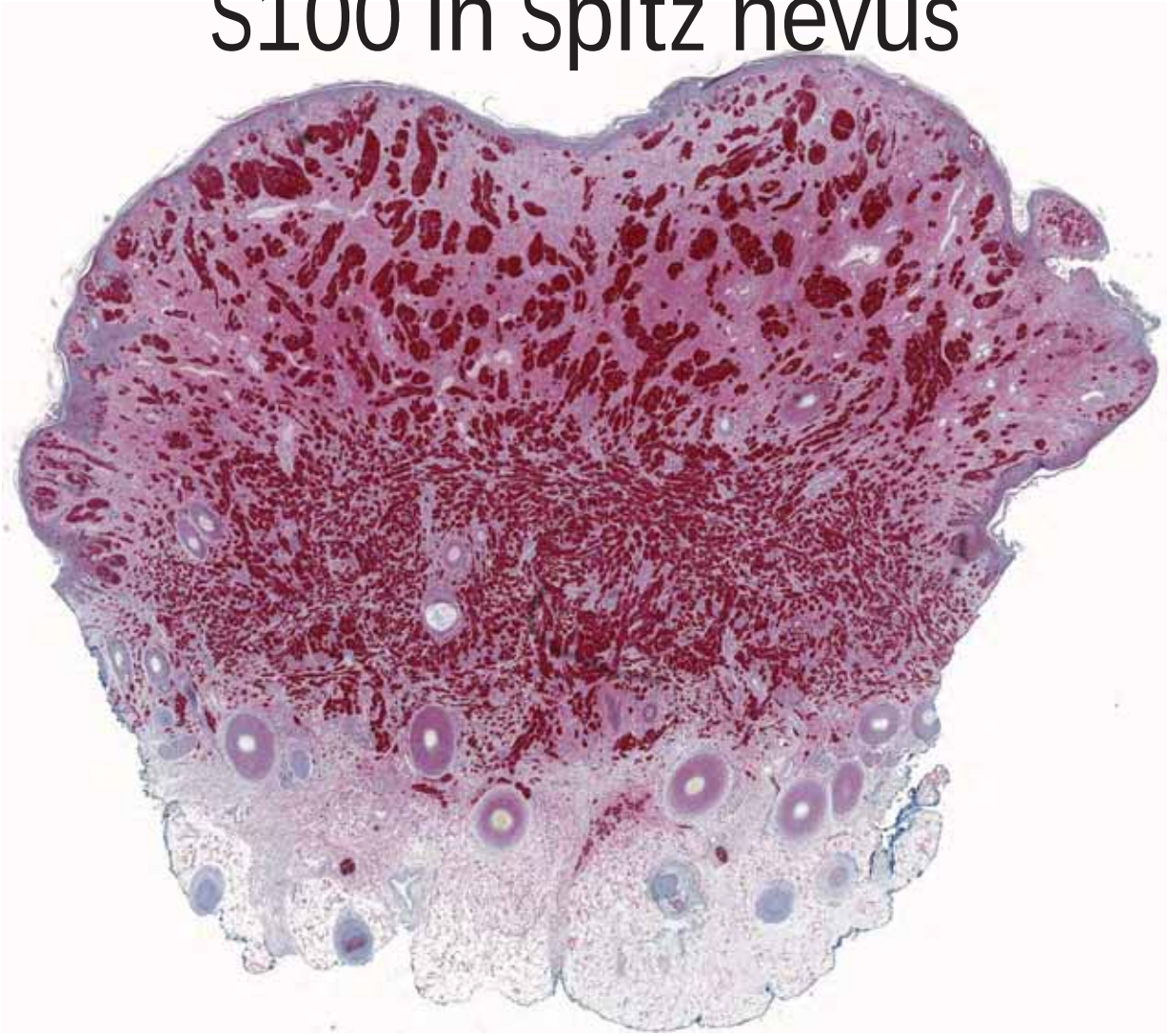
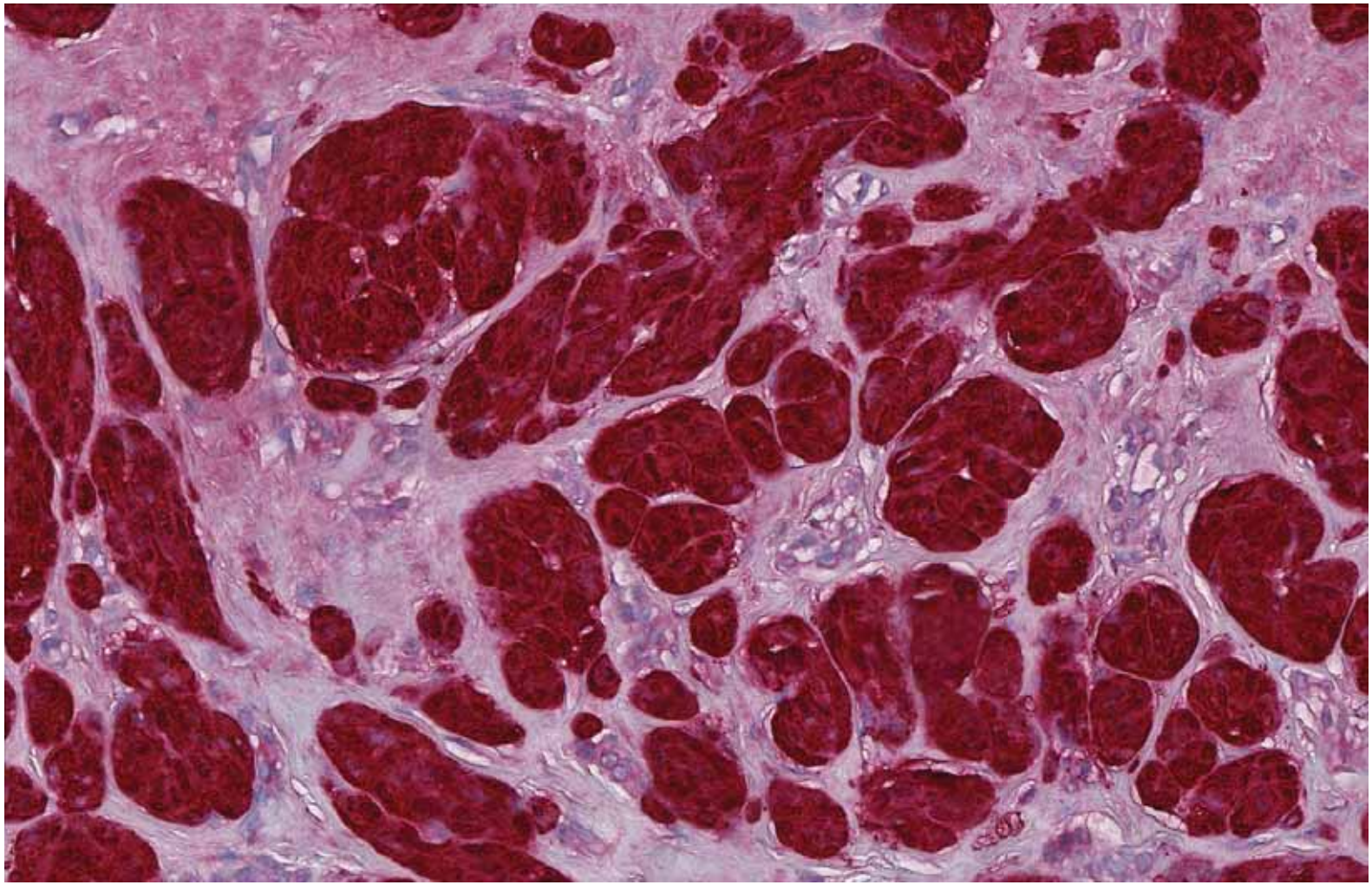


S100 in Spitz nevus



S100 in Spitz nevus



S100P is a screening tool for Blue nevi

ADVANCES IN
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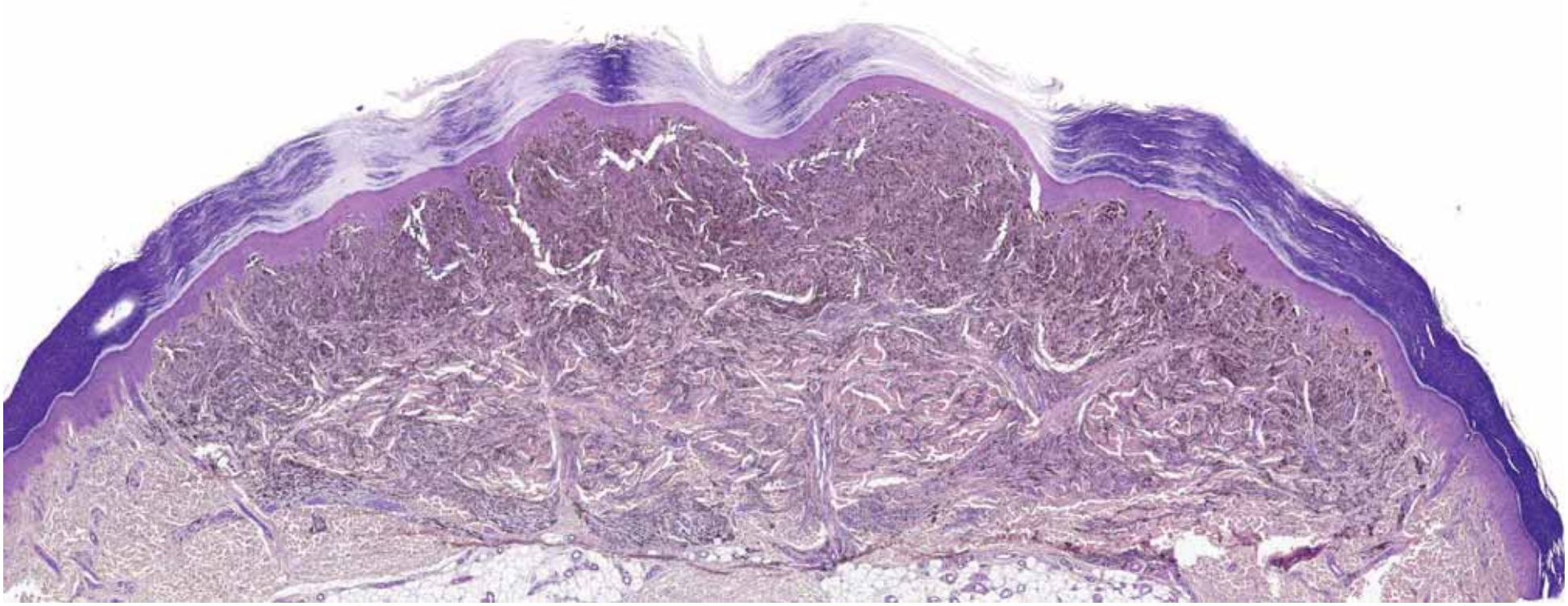
NEWS IN BRIEF

S-100 protein negative blue nevi and cellular blue nevi: potential diagnostic pitfalls

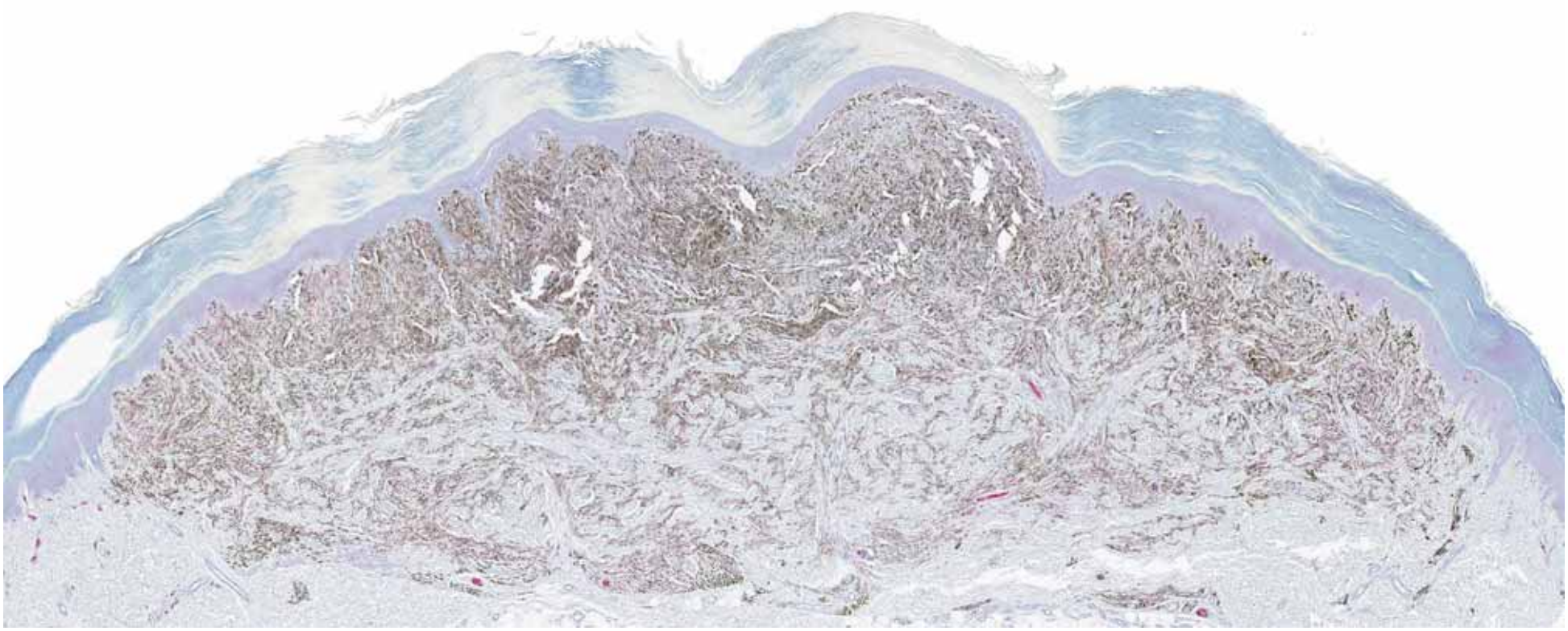
Folpe, Andrew L.

Advances in Anatomic Pathology 8(2):p 114-115, March 2001.

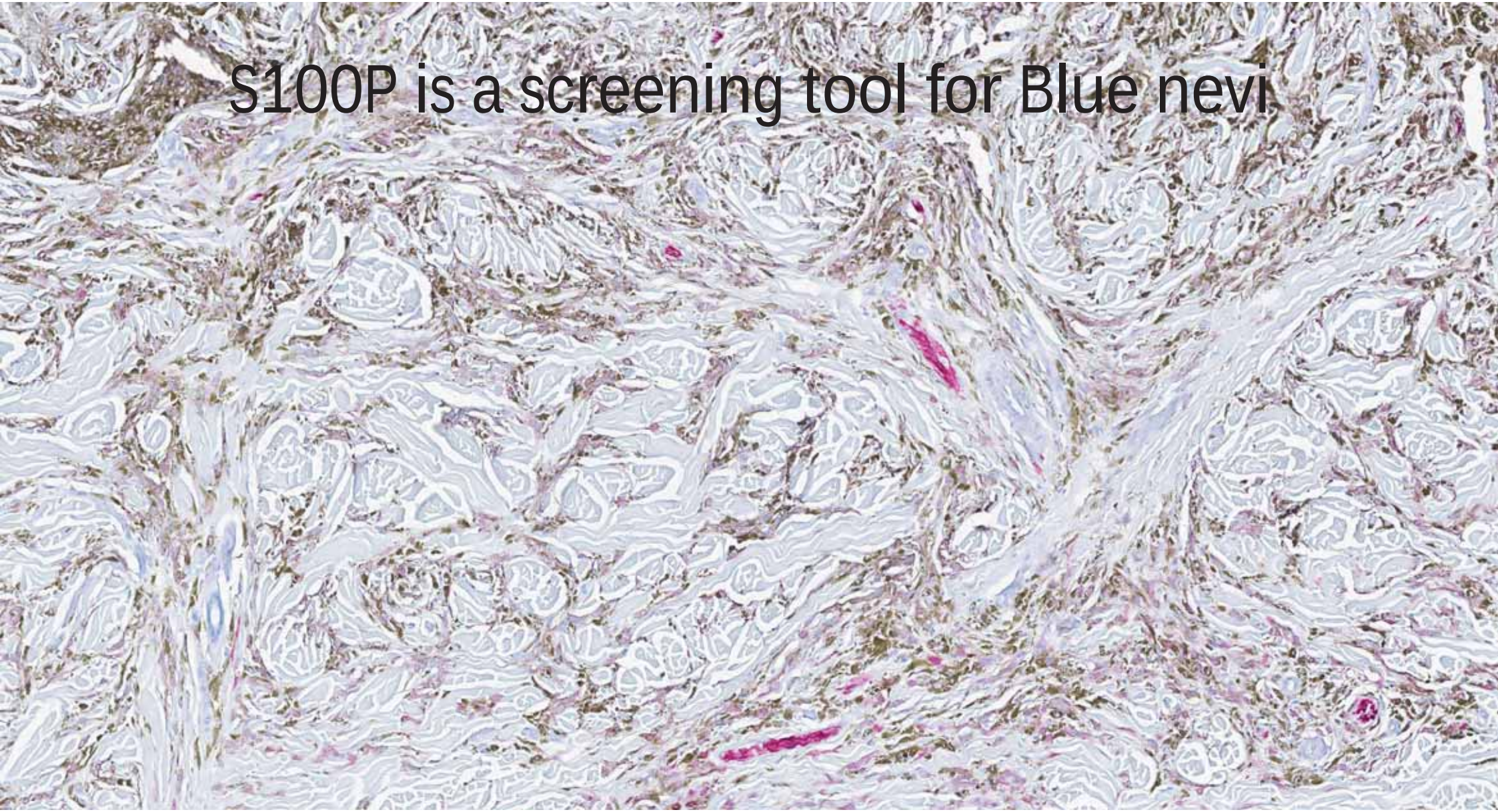
S100P is a screening tool for Blue nevi



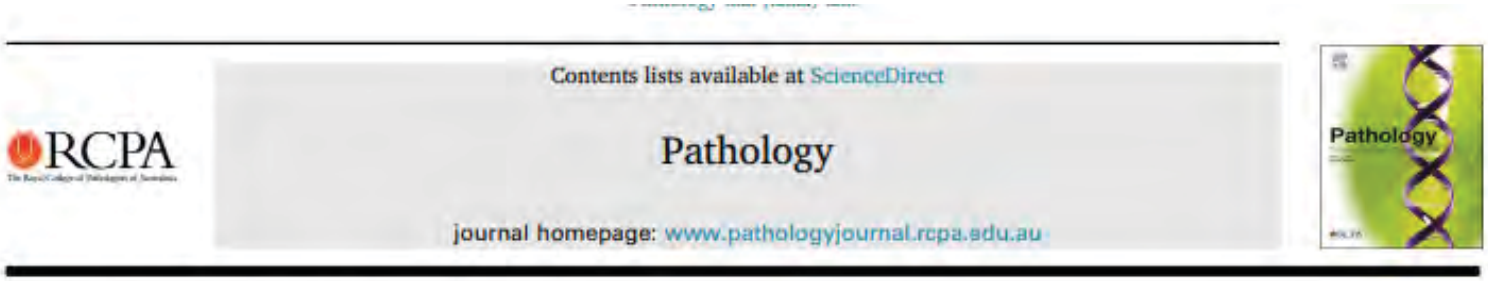
S100P is a screening tool for Blue nevi



S100P is a screening tool for Blue nevi



Loss of S100 expression in the superficial band is a good diagnostic sign of PKC-fused blue nevus



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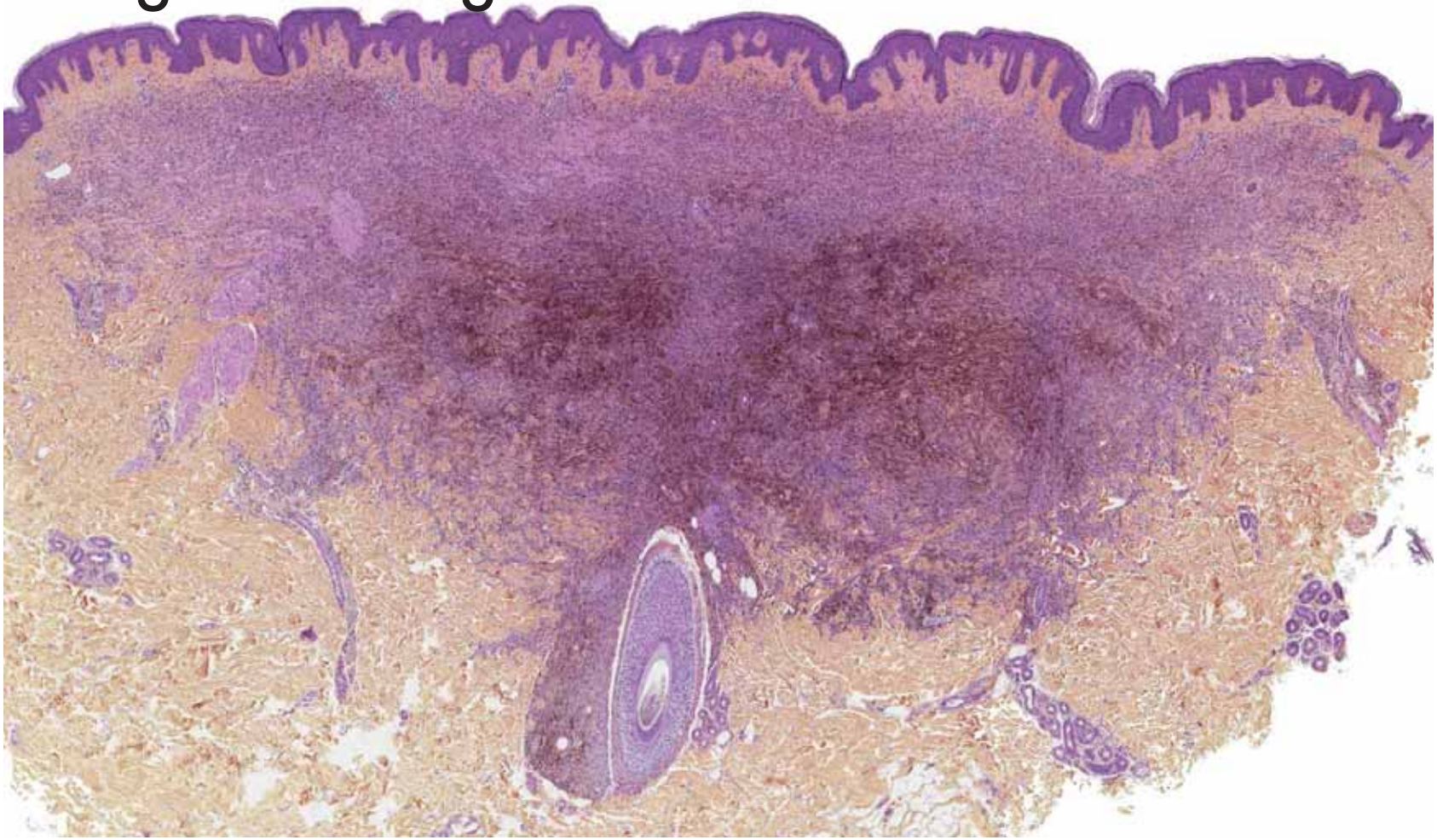
ANATOMICAL PATHOLOGY

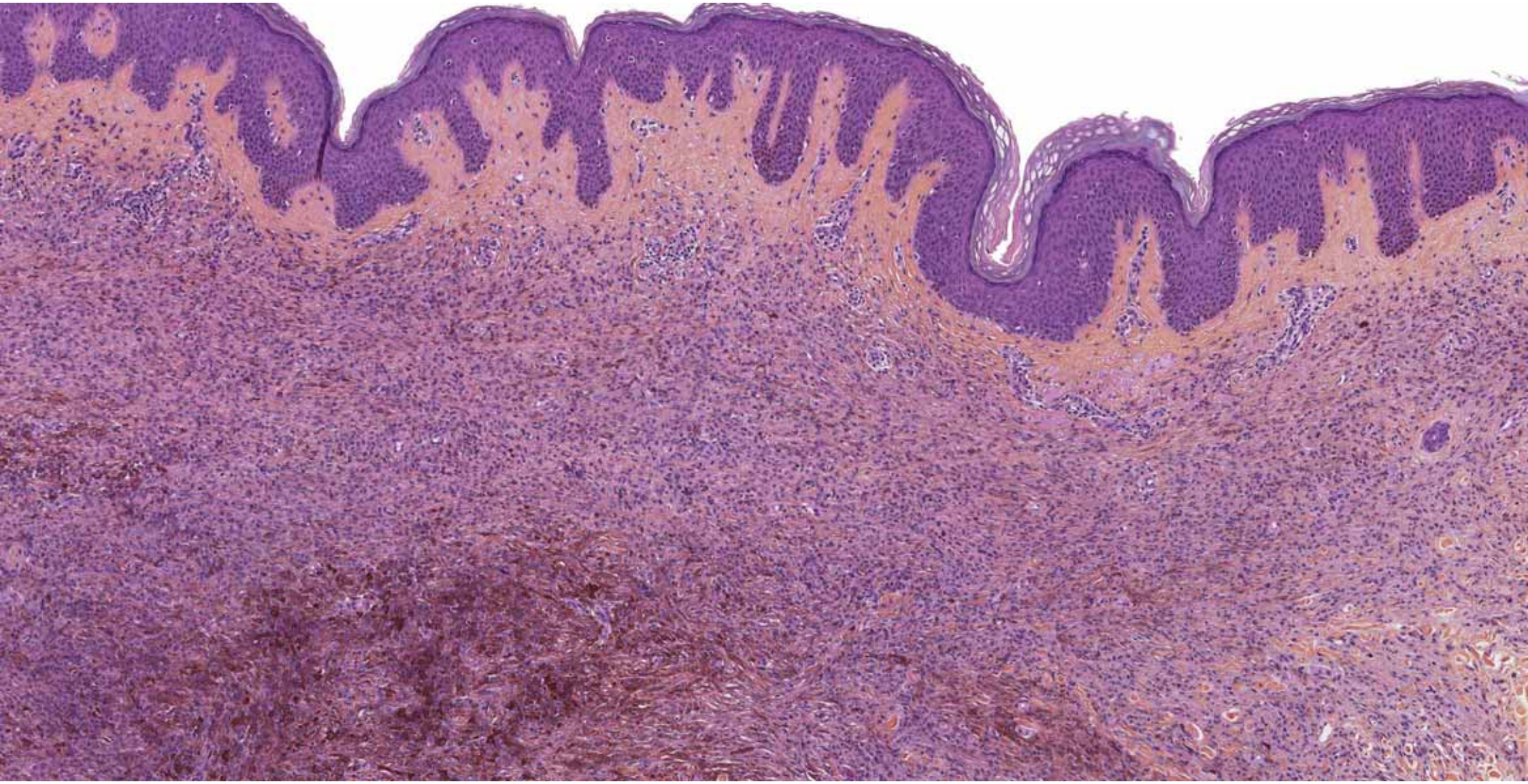
S100 protein expression in PKC-fused blue naevi, cellular blue naevi and PRKAR1A-inactivated pigmented epithelioid melanocytomas

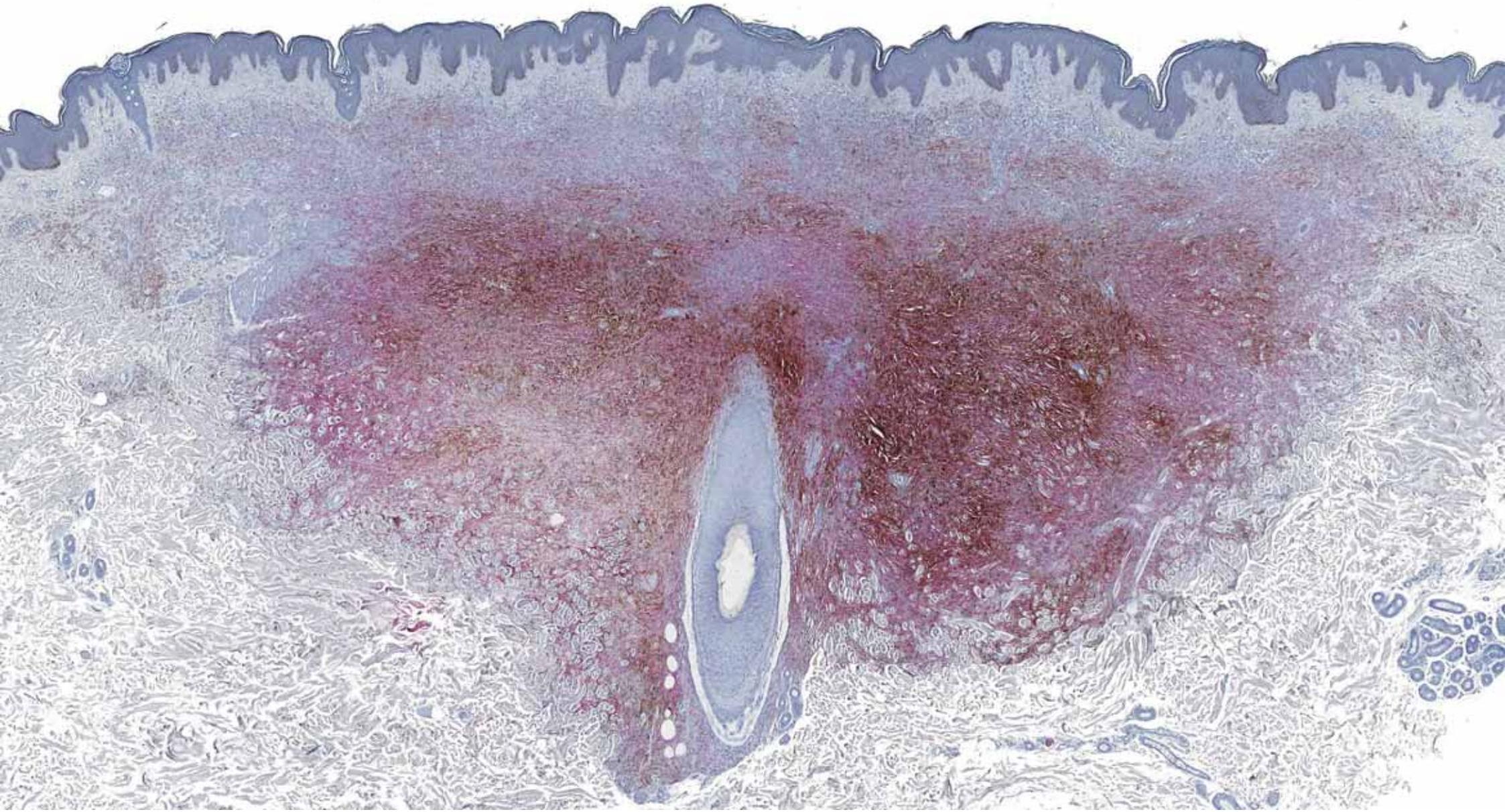
Pauline Hayenne [1], Daniel Pissaloux [1,2], Franck Tirode [1,2], Arnaud de la Fouchardiere [1,2,*]

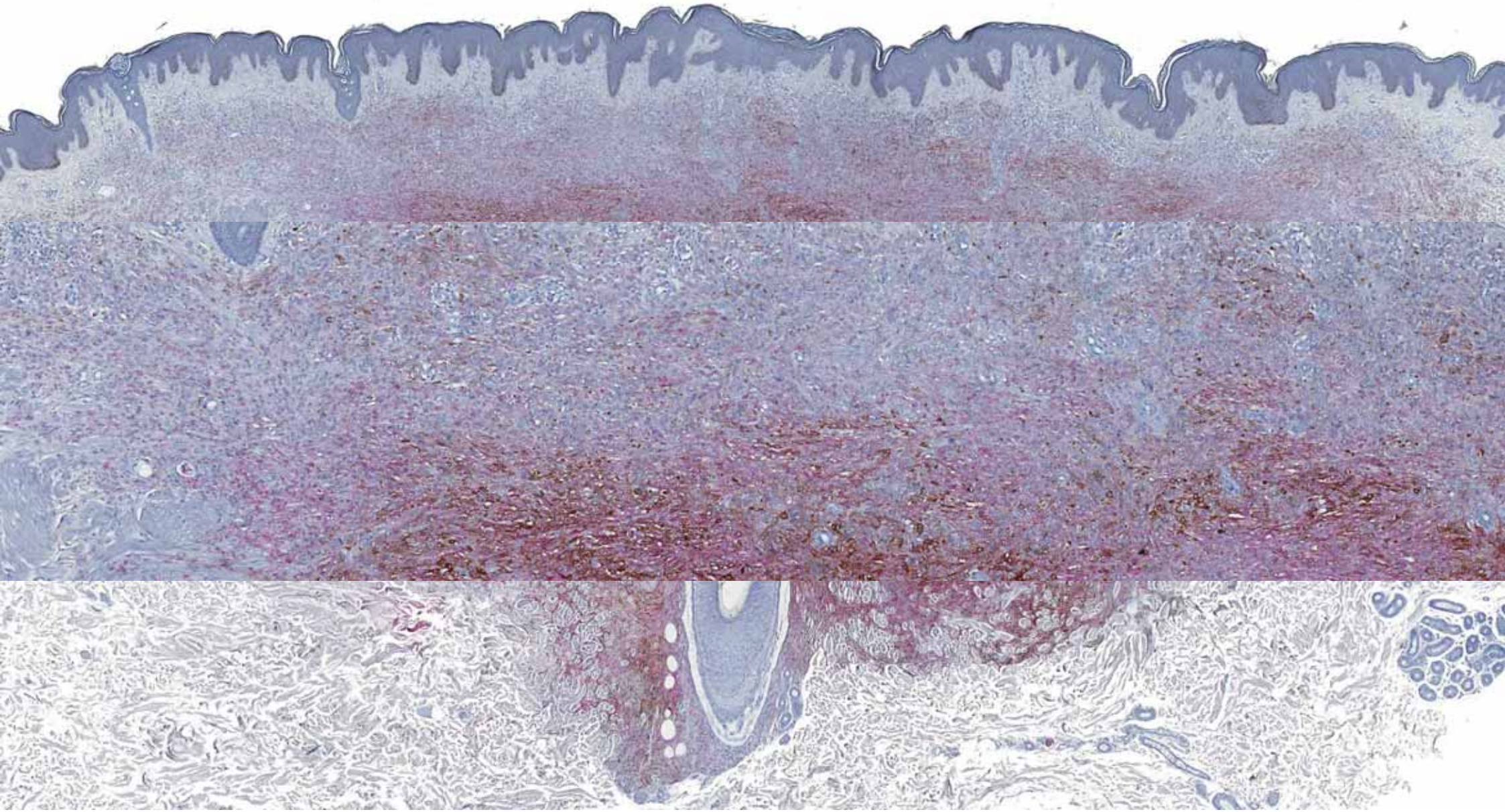
[1] *Department de Biopathologie, Centre Léon Bérard, Lyon, France*
[2] *Université de Lyon, Université Claude Bernard Lyon 1, INSERM 1052, CNRS 5286, Centre Léon Bérard, Cancer Research Center of Lyon, Equipe Labellisée Ligue contre le Cancer, Lyon, France*

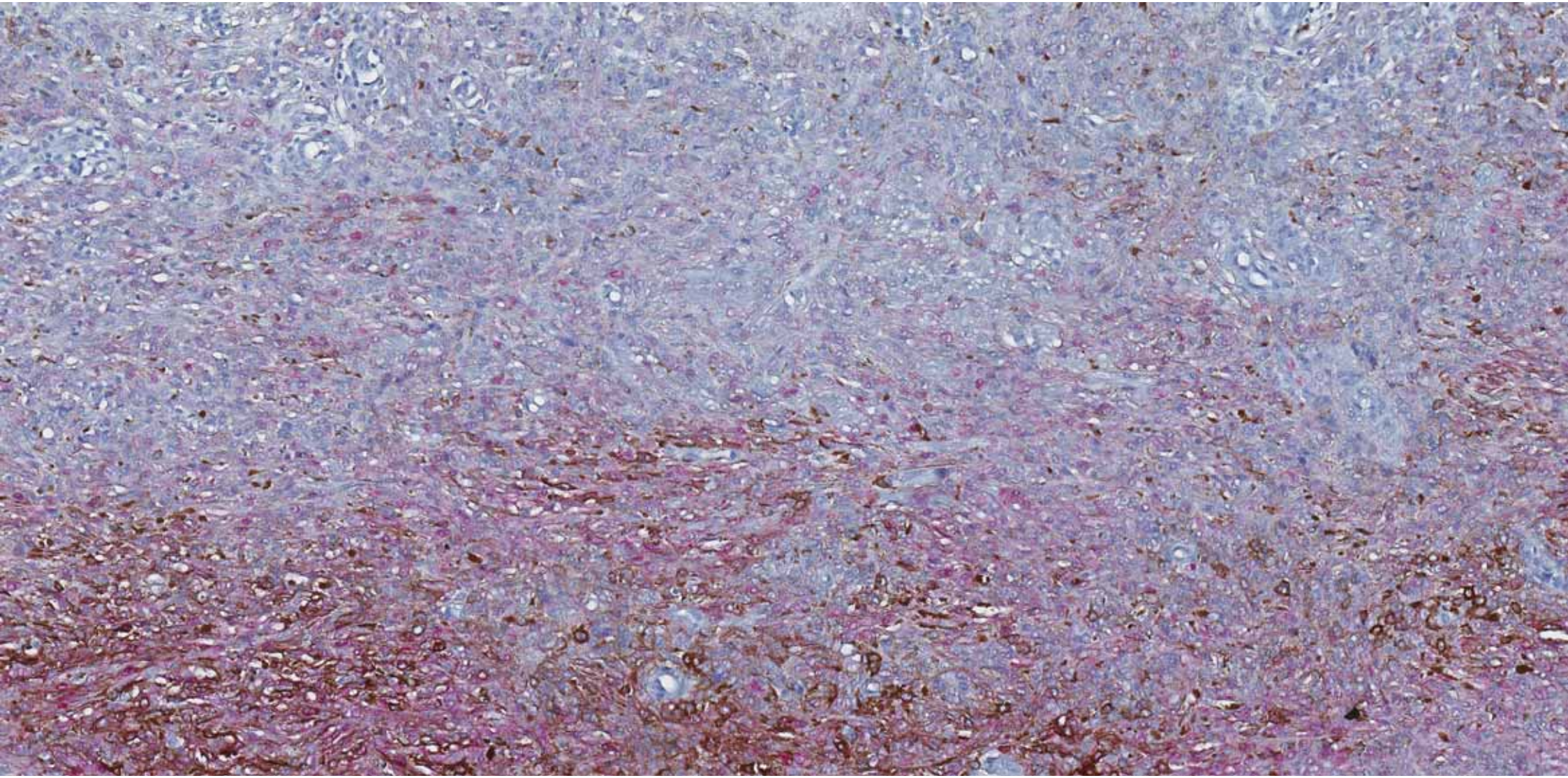
Loss of S100 in the superficial band is a good diagnostic sign of PKC-fused blue nevus



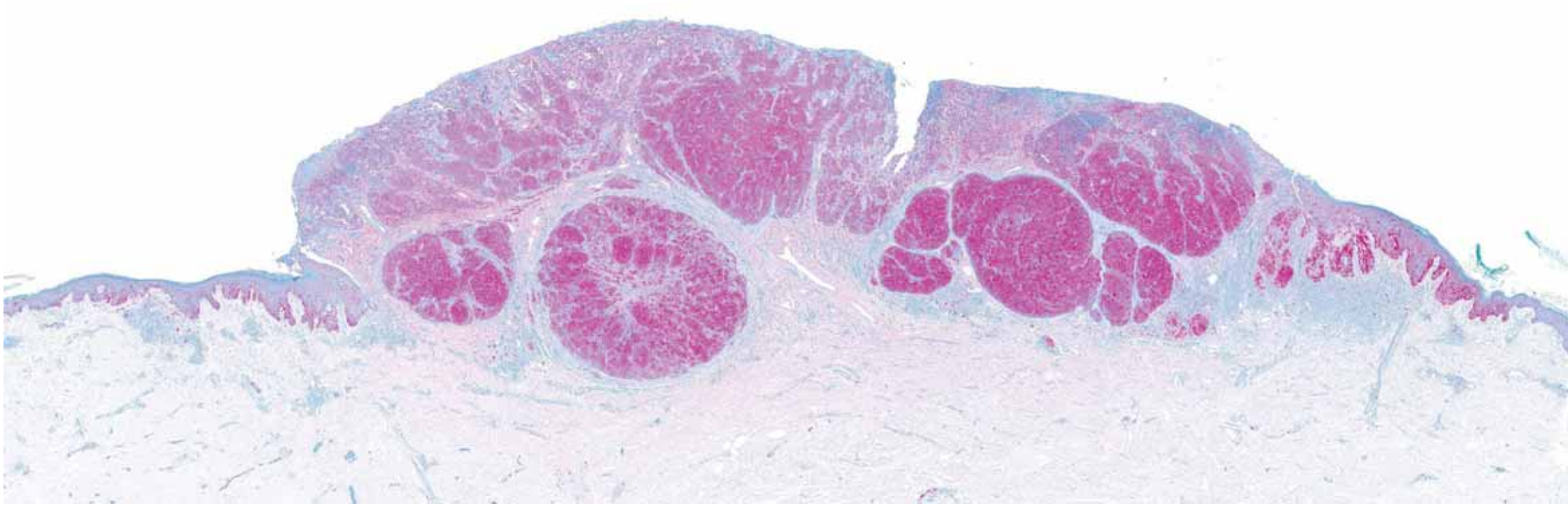




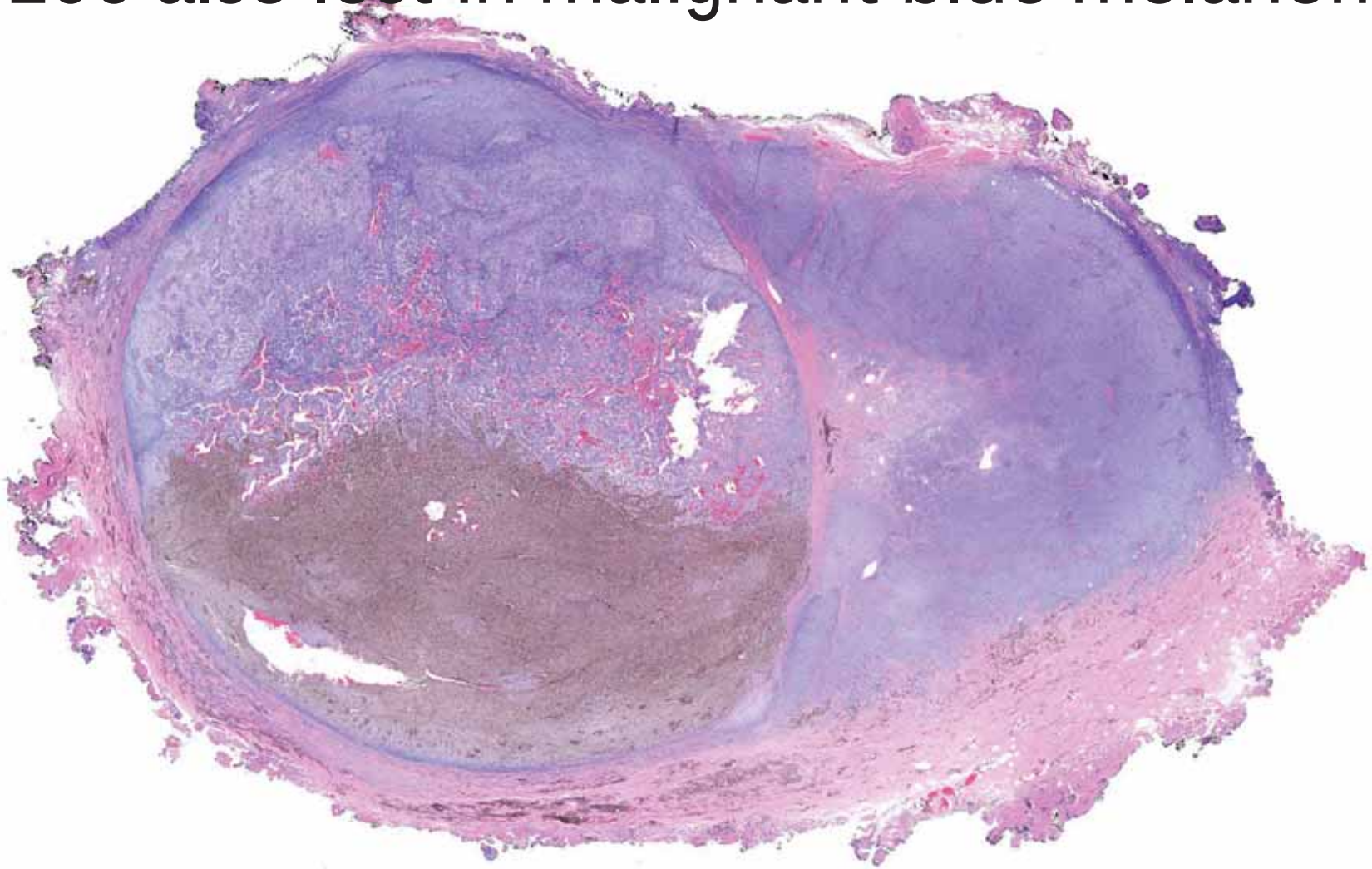




S100 is usually positive in invasive melanoma



Is S100 also lost in malignant blue melanoma?



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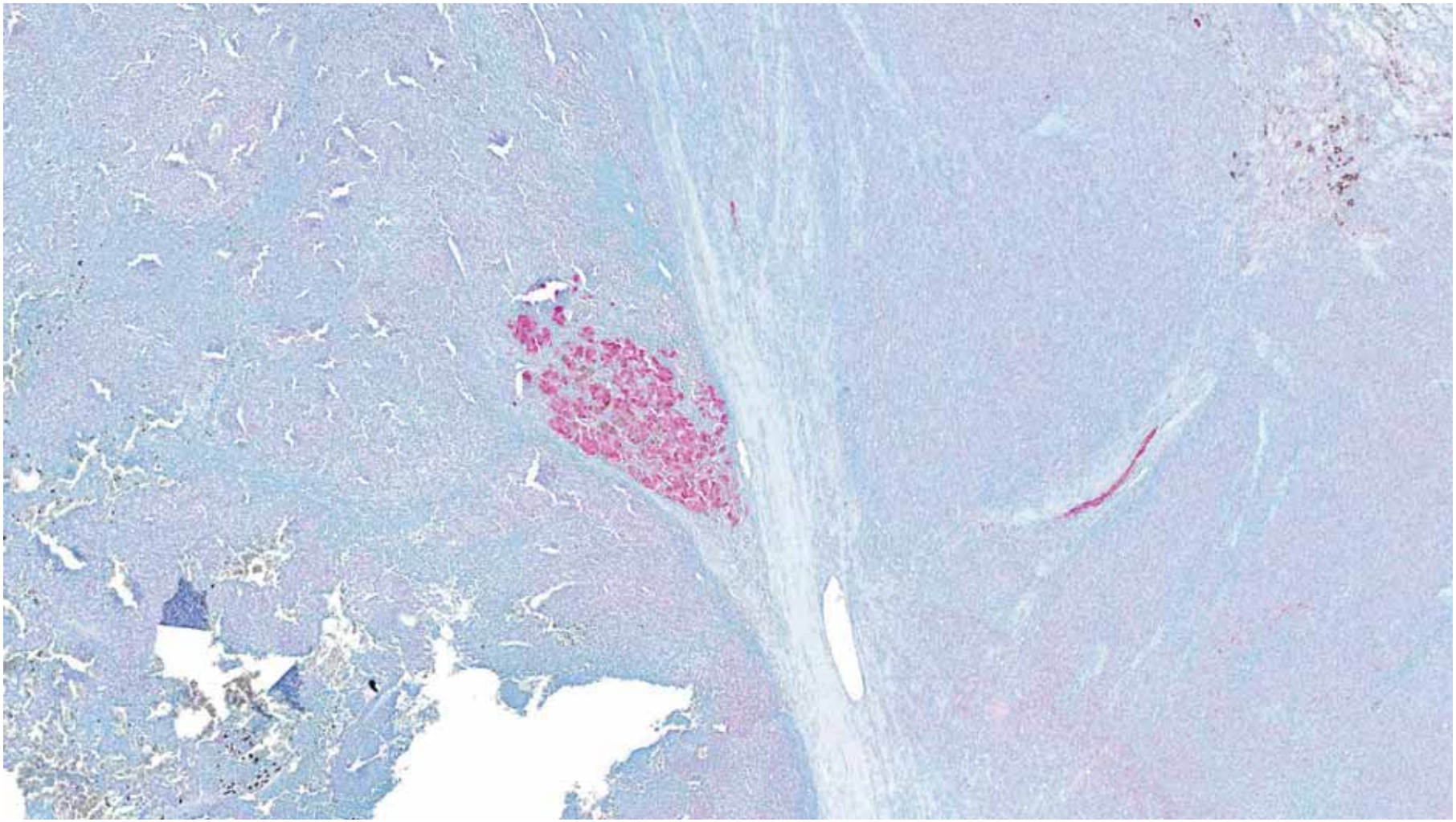
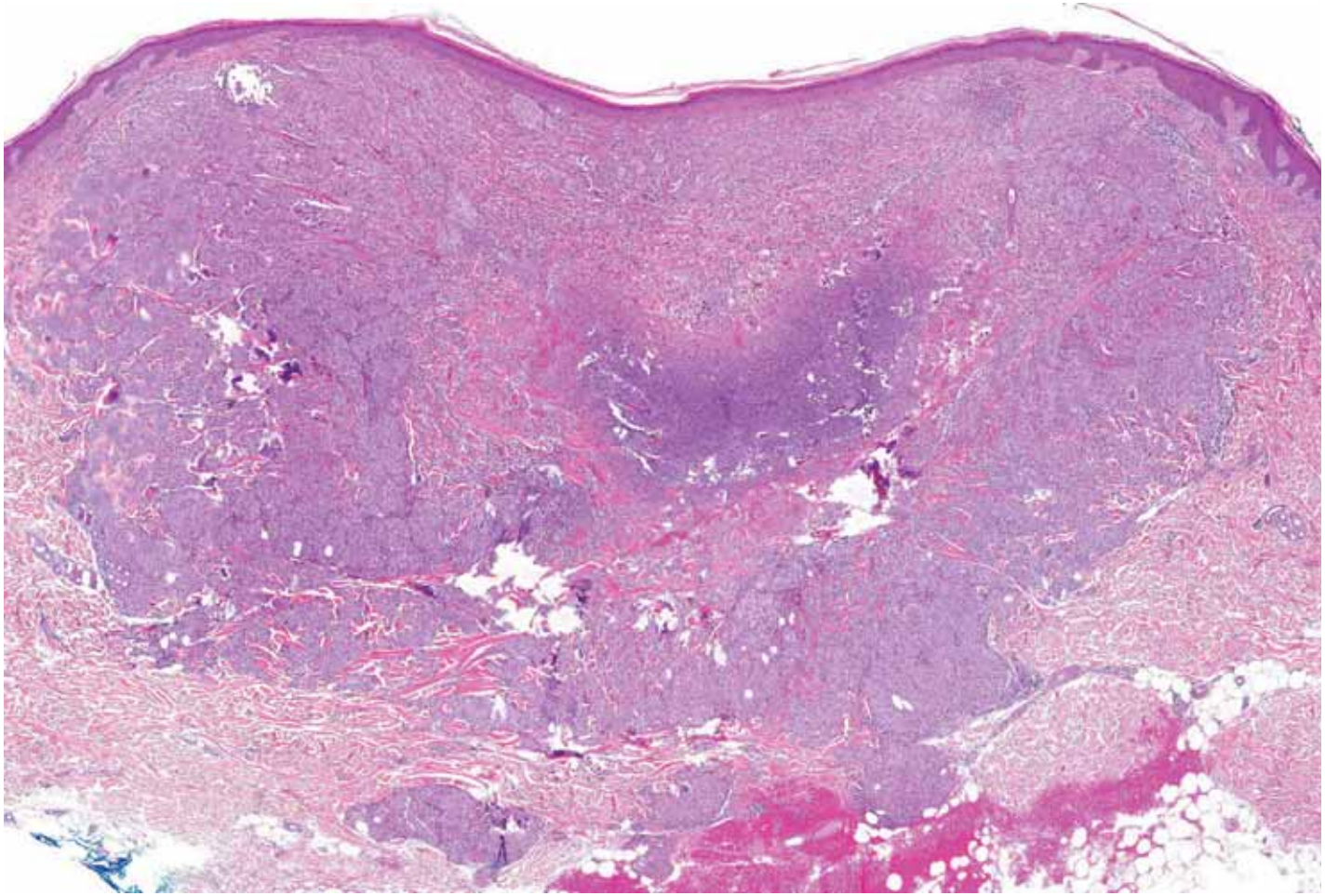


Is S100 loss restricted to Blue melanomas in non-undifferenciated melanomas?



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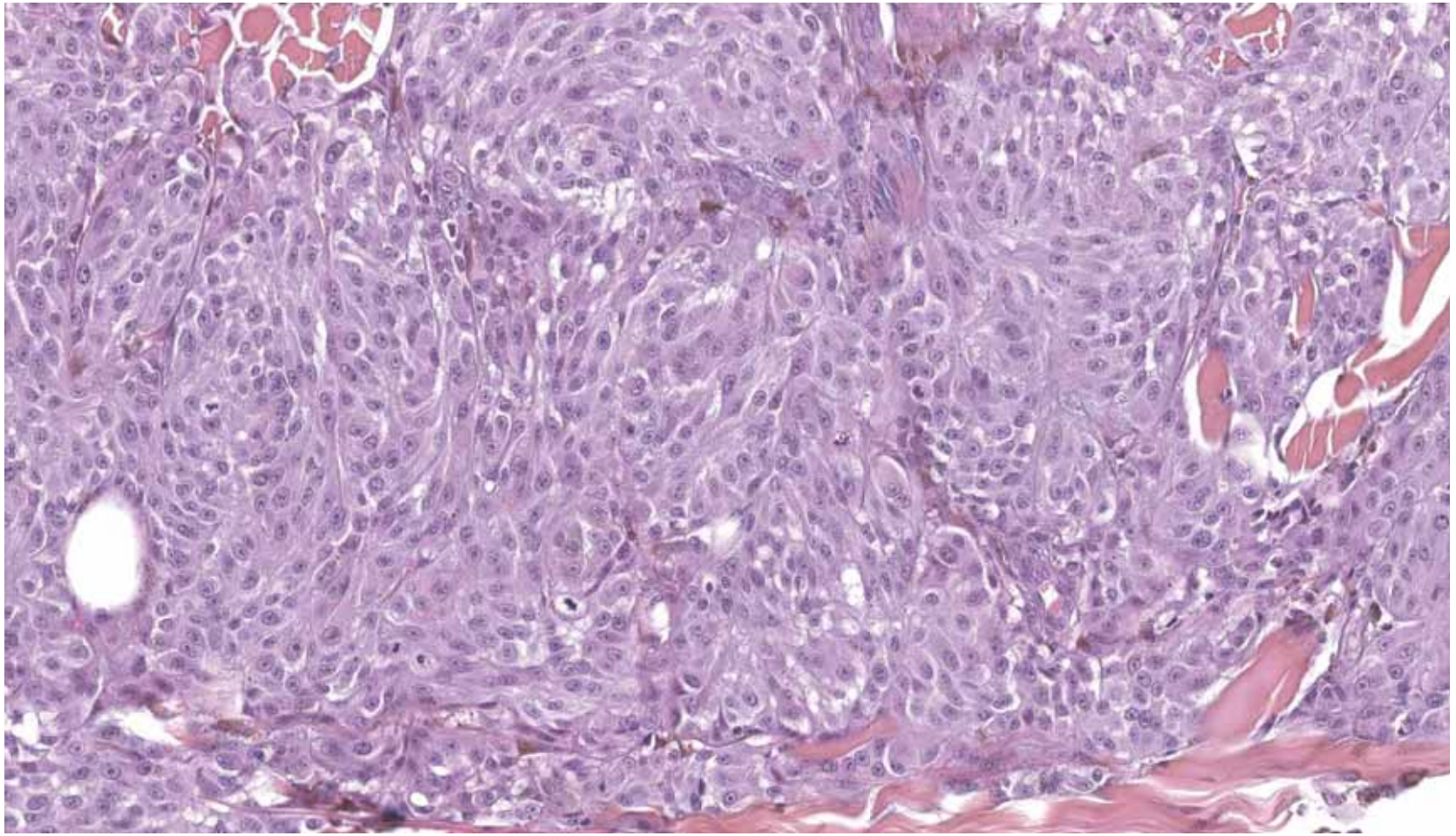

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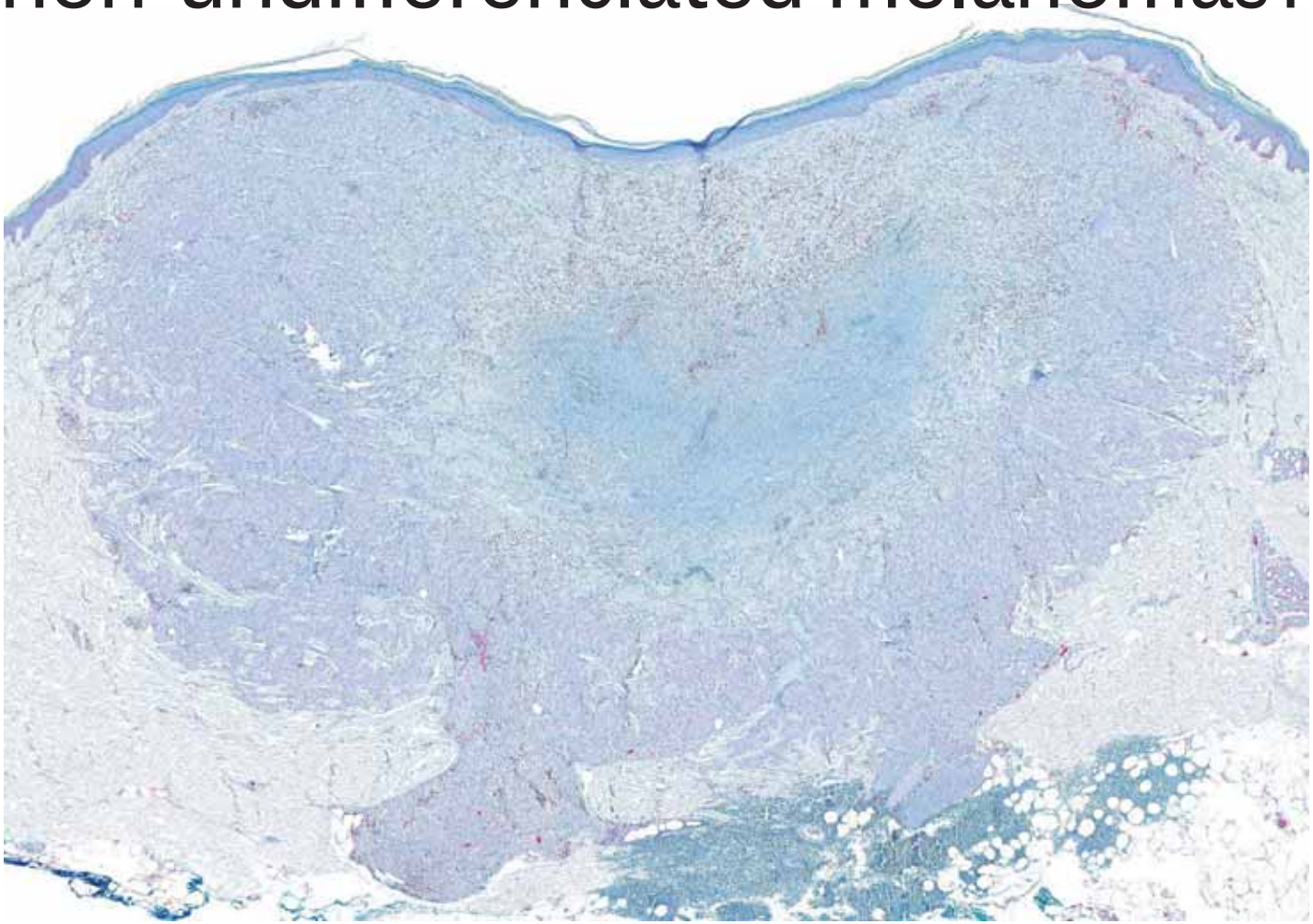
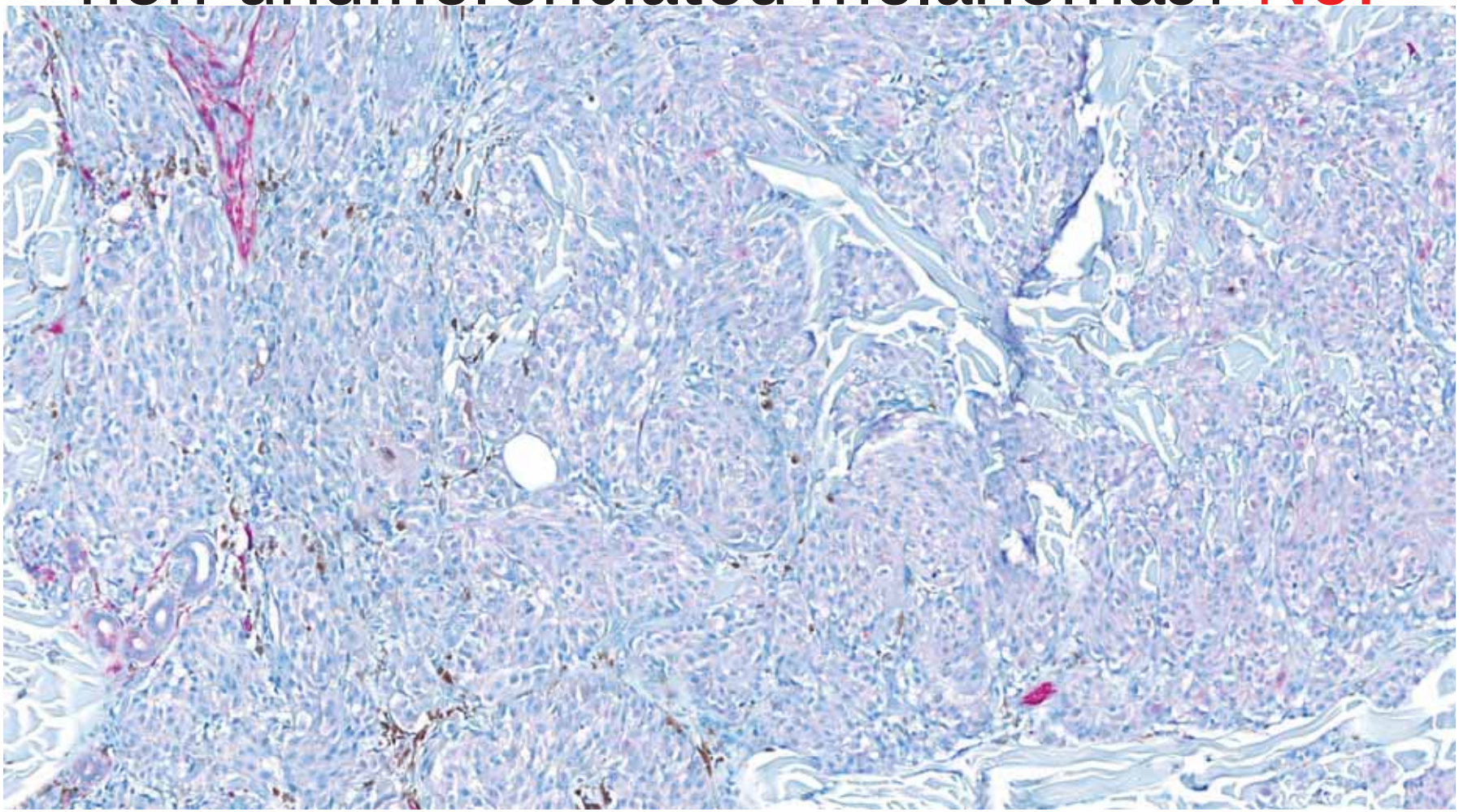
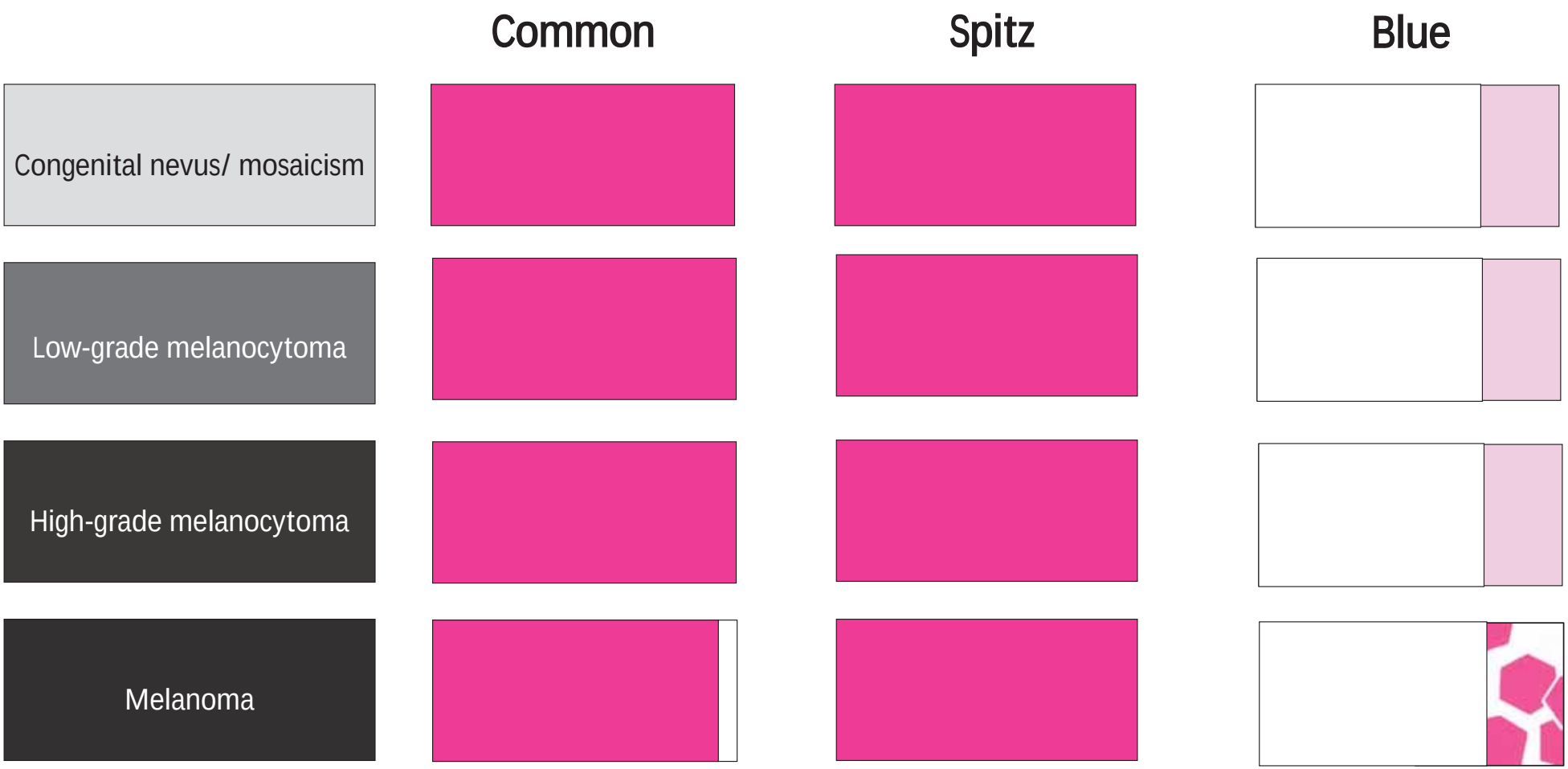


Is S100 loss restricted to Blue melanomas in non-undifferenciated melanomas? No!

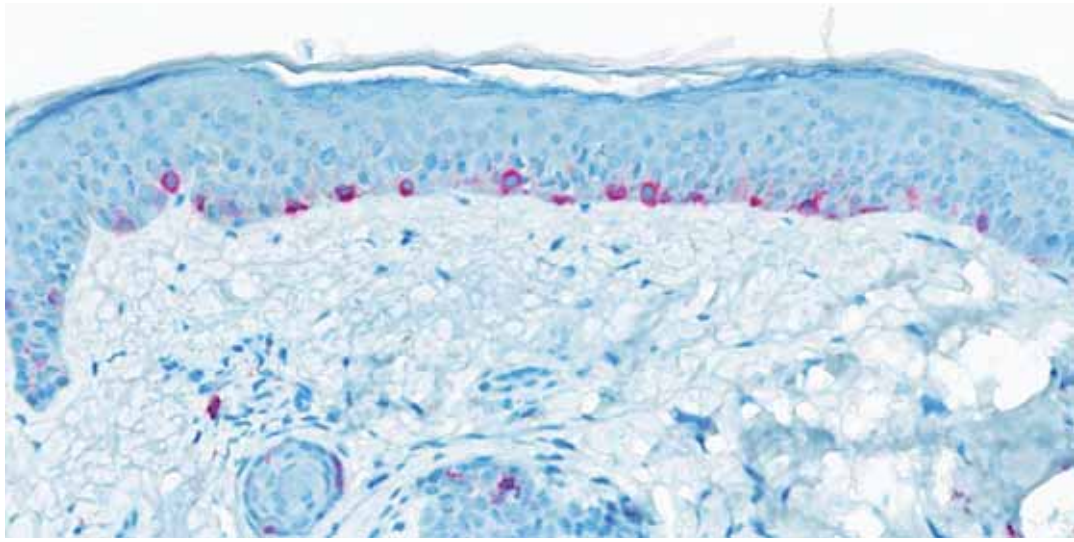


S100P synoptic IHC expression chart

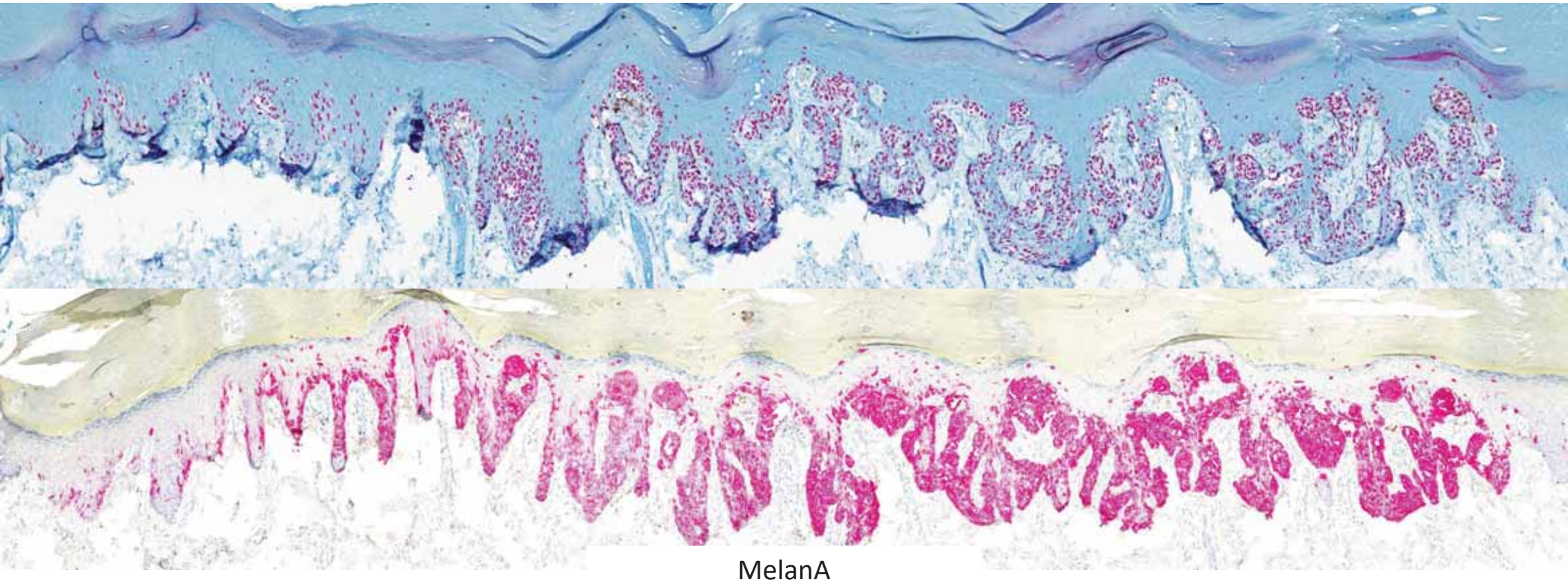


S100P/SOX10/MelanA
Help visualize melanocyte distribution

- Normal unit of melanisation
- Assessing melanocytic distribution



Sox10 is diffusely positive in melanocytic tumors
Acral Lentiginous Melanoma



MelanA

Punch biopsy on the face in a setting of a clinical suspicion of lentigo maligna

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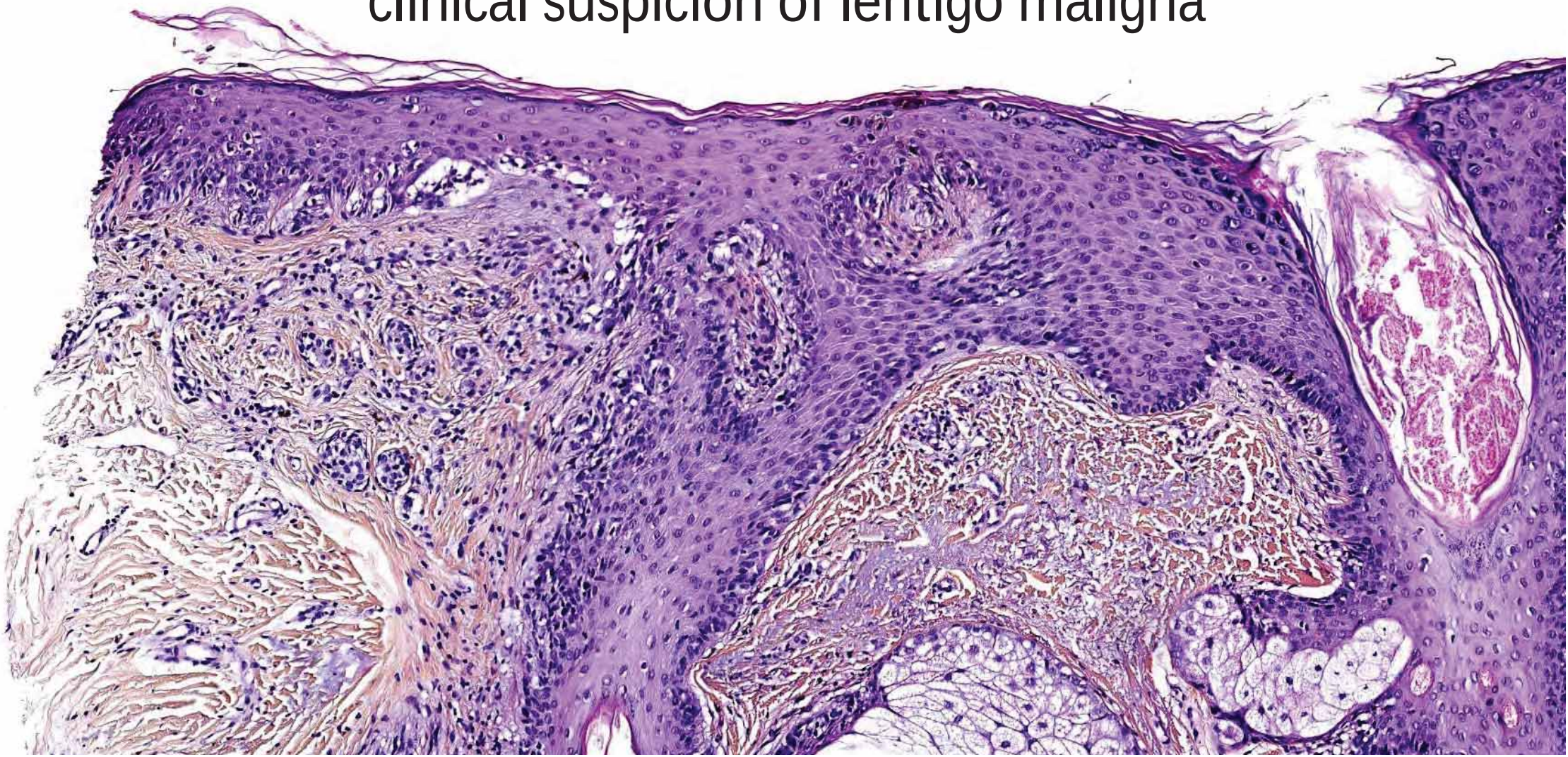
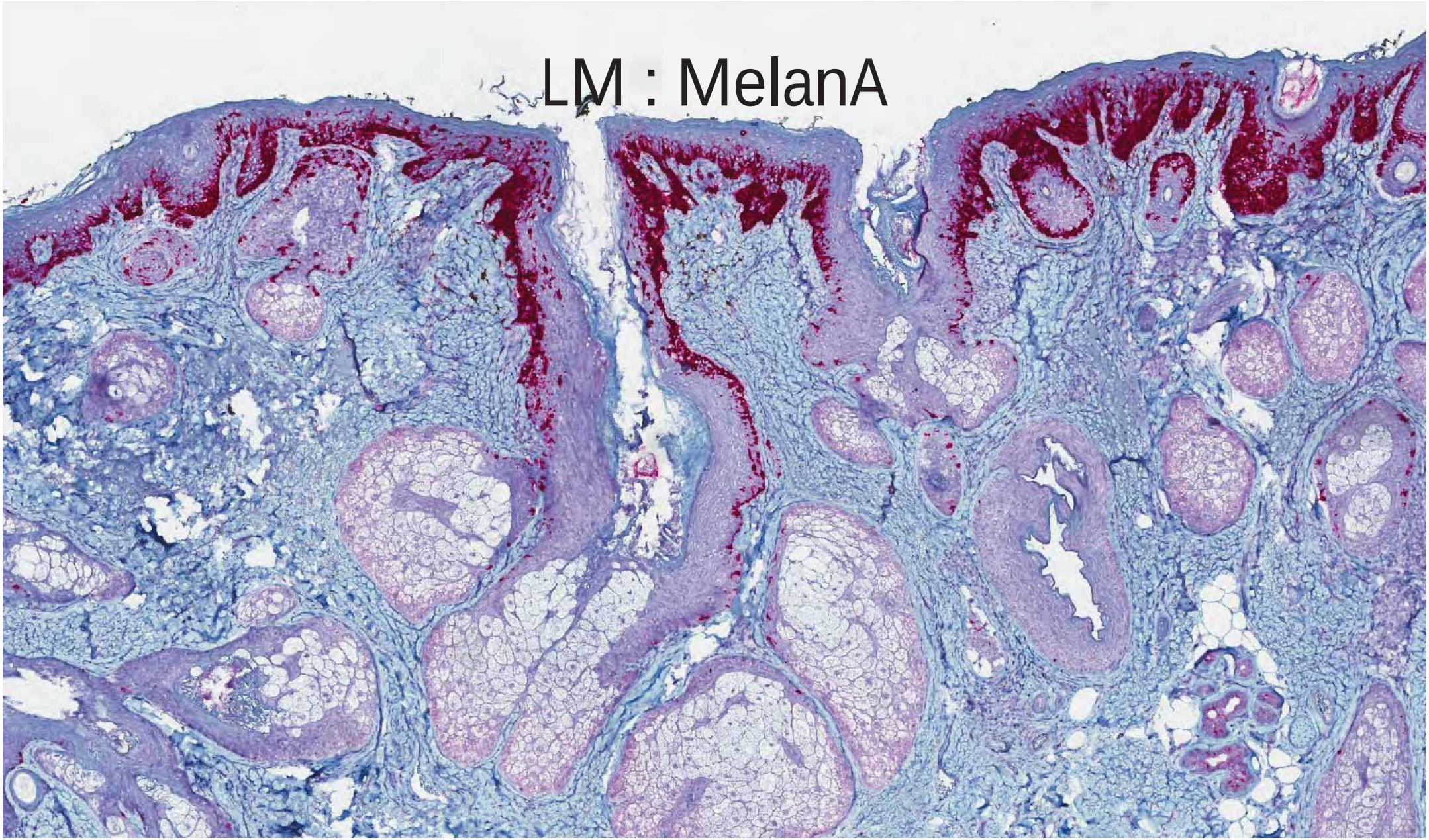
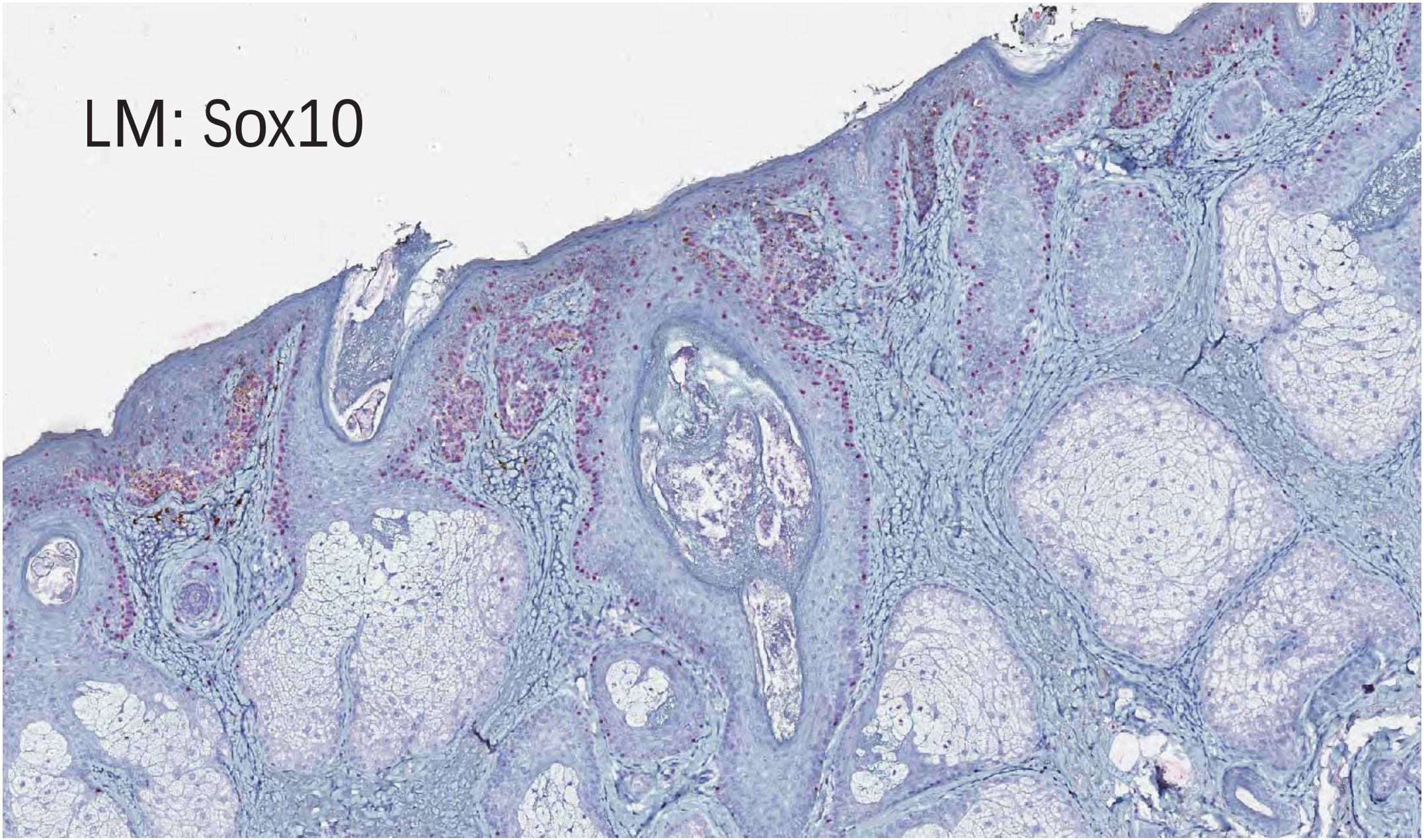


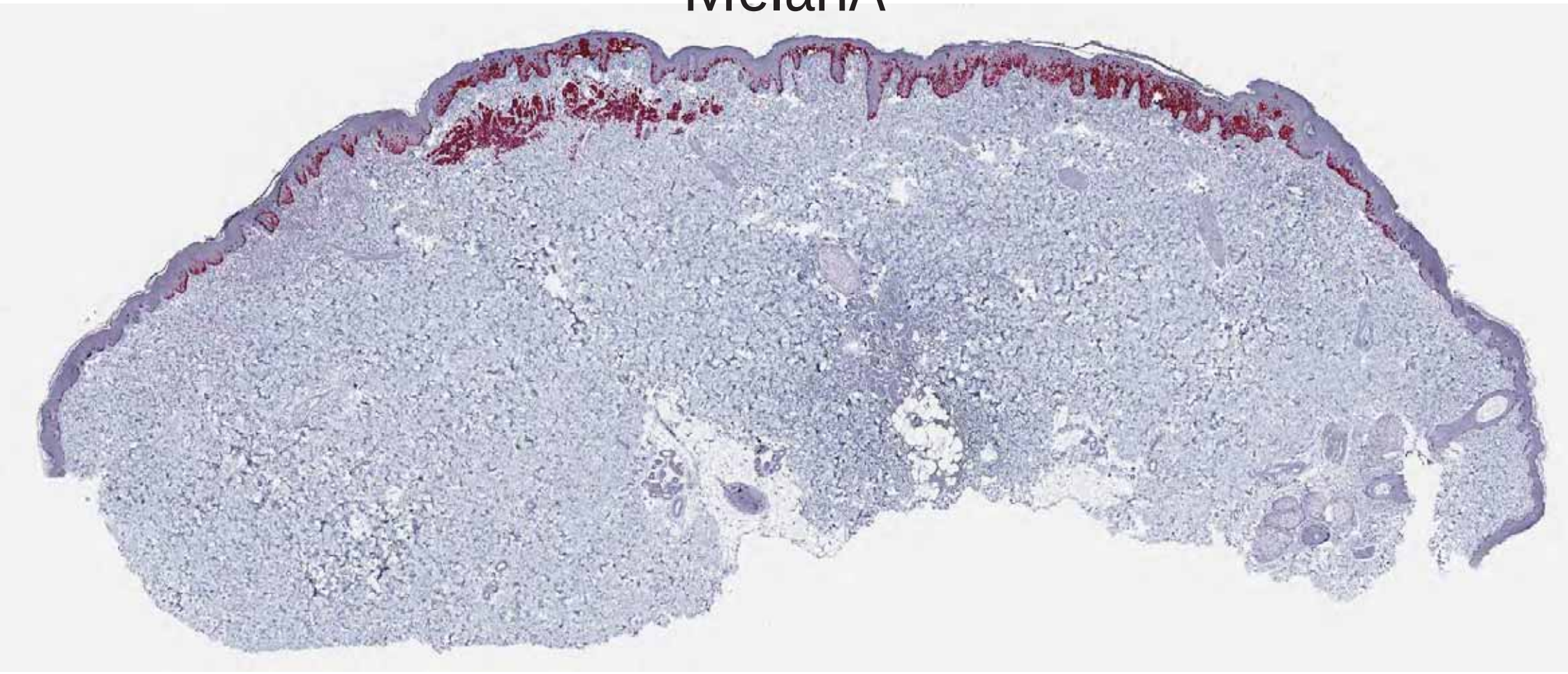
LM : MelanA



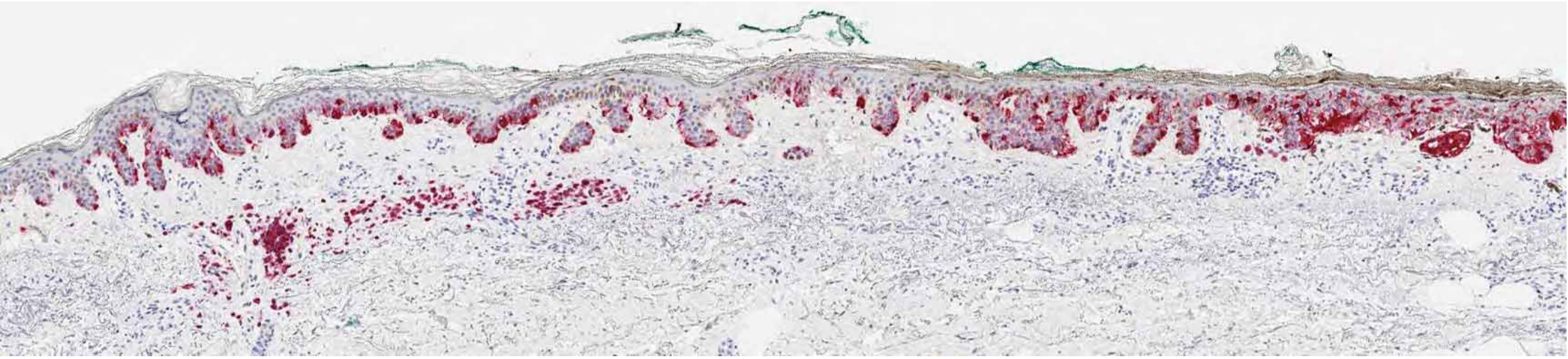
LM: Sox10



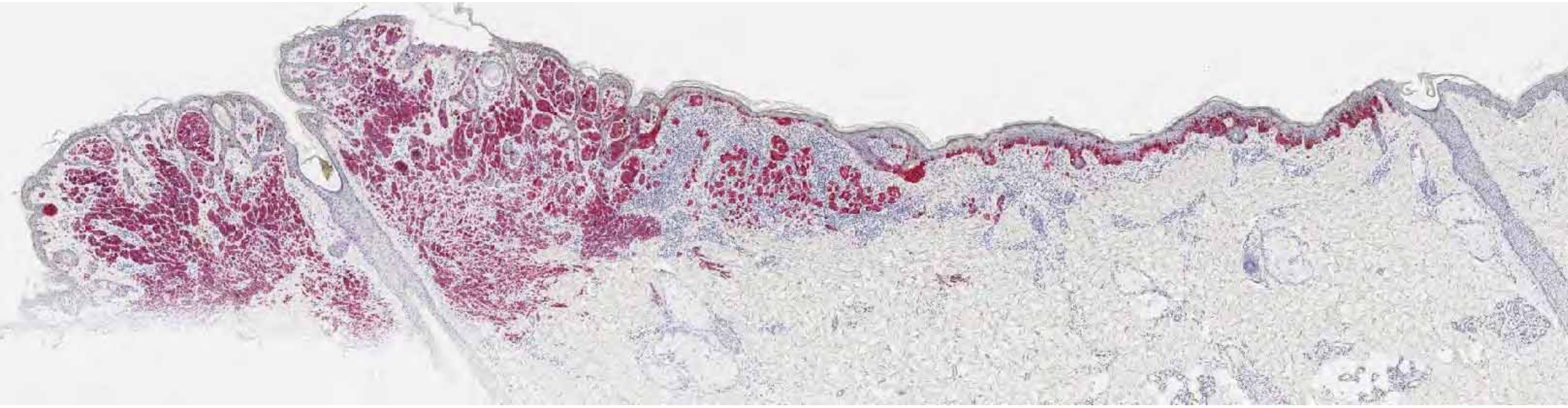
Asymetrical melanocytic distribution «chicane/zig-zag» architecture MelanA



Asymetrical melanocytic distribution «chicane/zig-zag» architecture MelanA

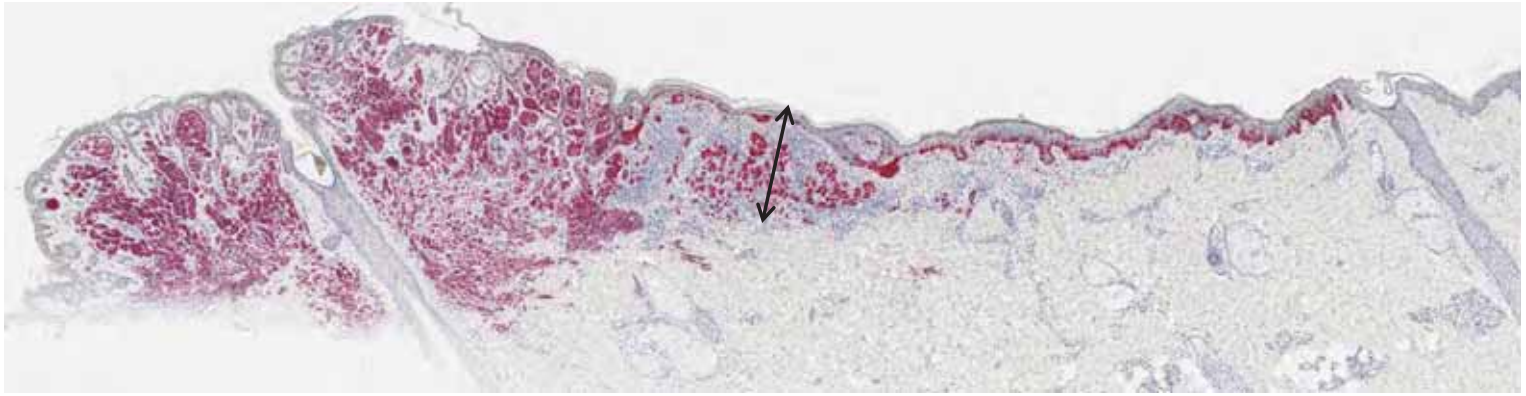


Asymetrical melanocytic distribution «chicane/zig-zag» architecture MelanA



Asymetric melanocytic distribution

Breslow assessment

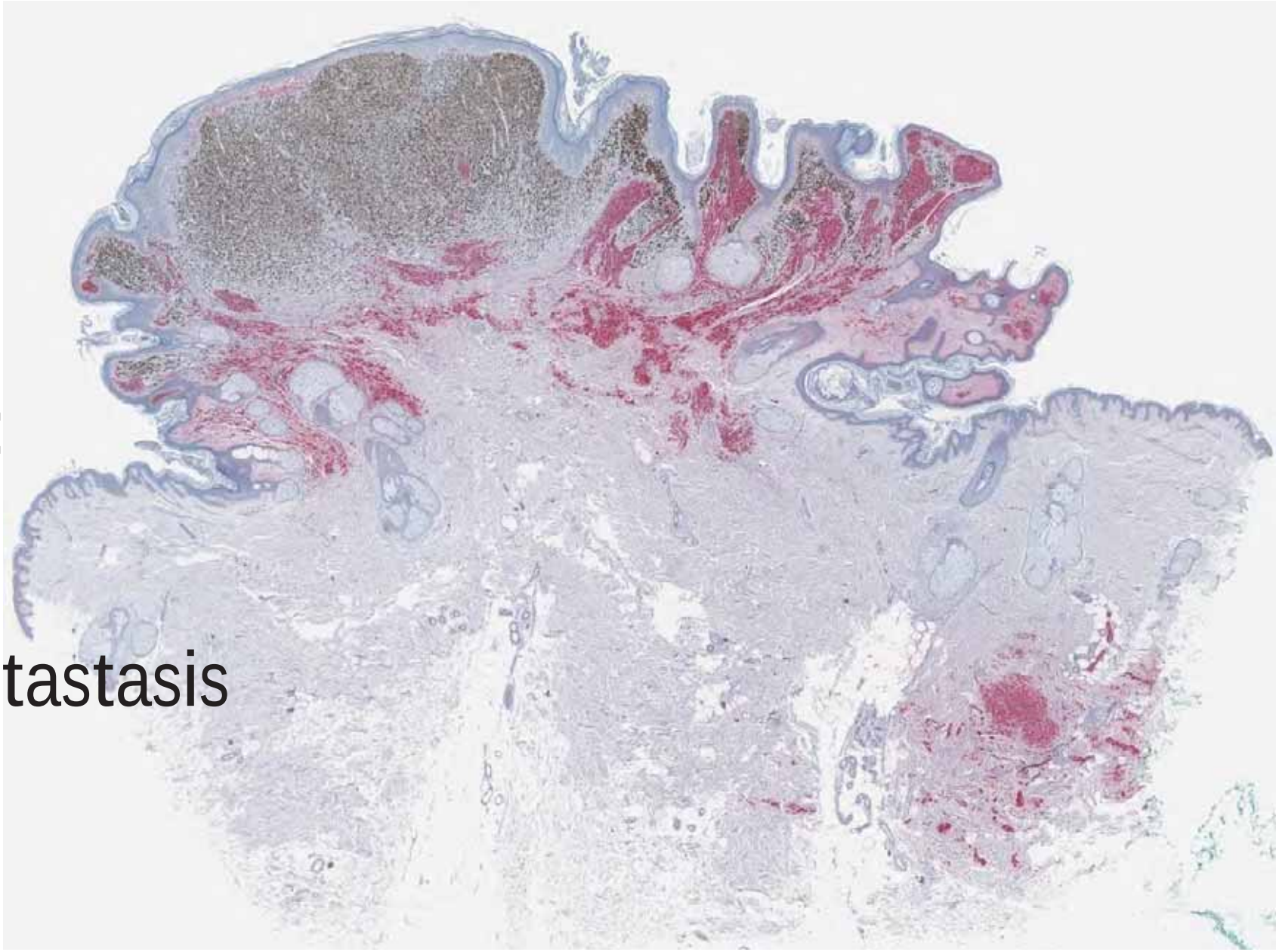


MelanA

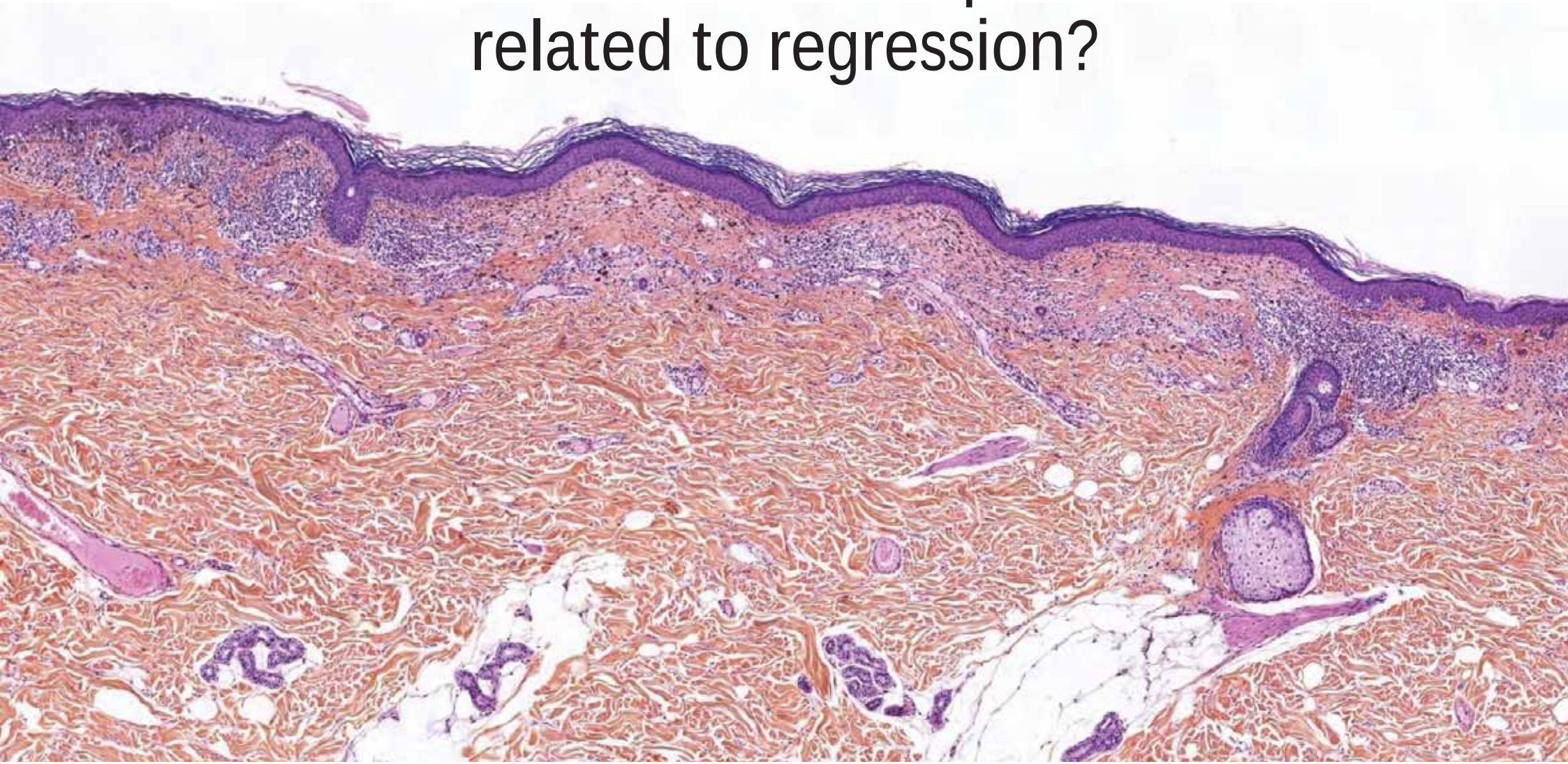
Melanoma ex-nevus

Melan-A highlighting:

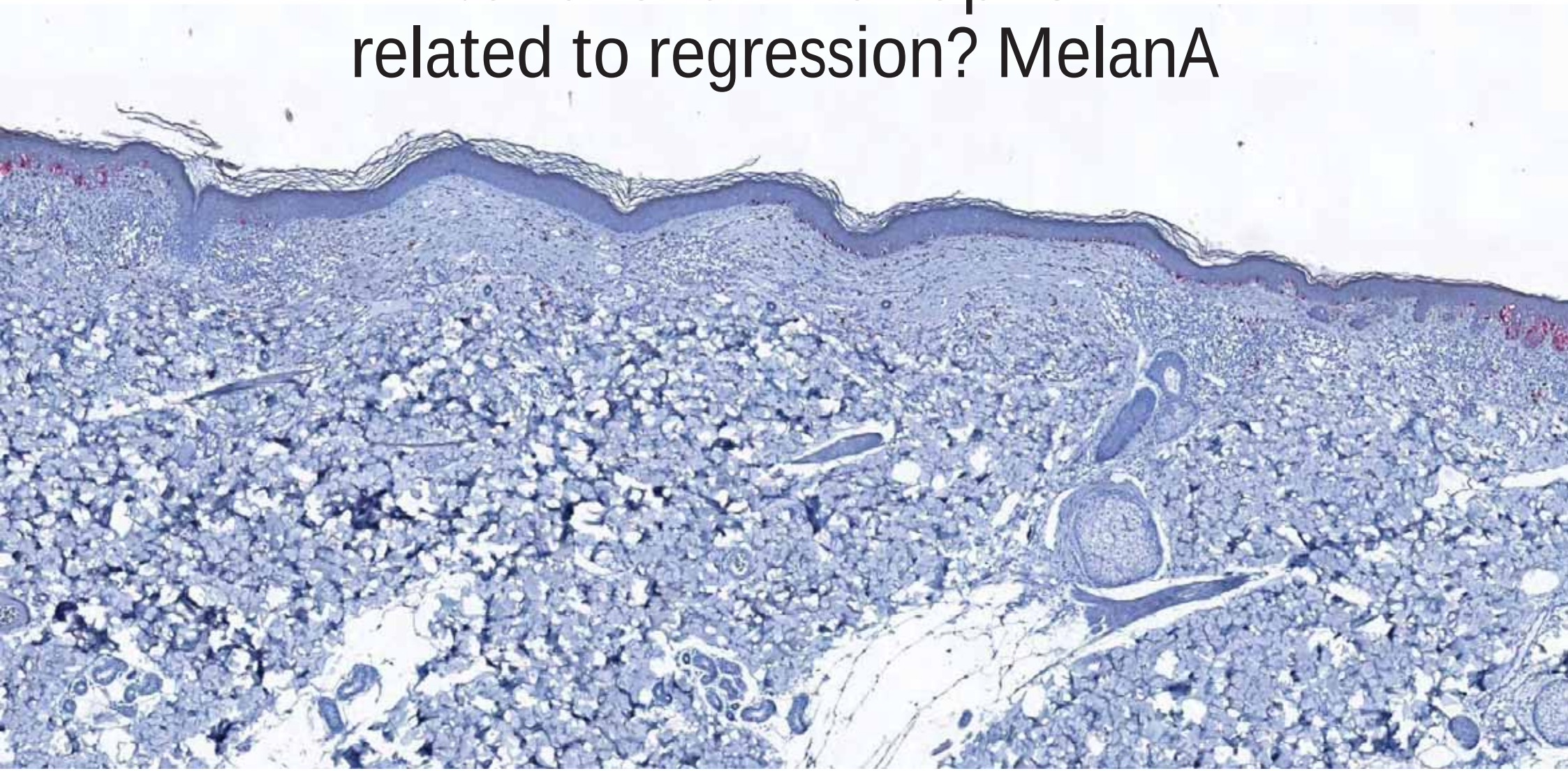
- Regression
- Distant metastasis



Junctional interruption related to regression?



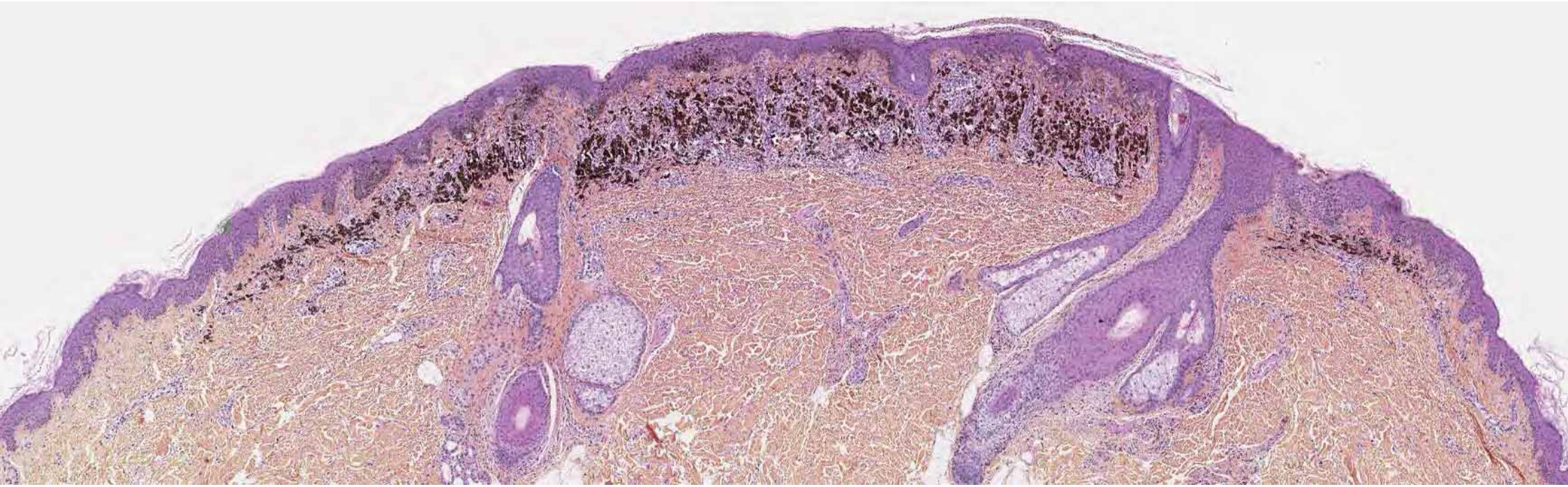
Junctional interruption related to regression? MelanA



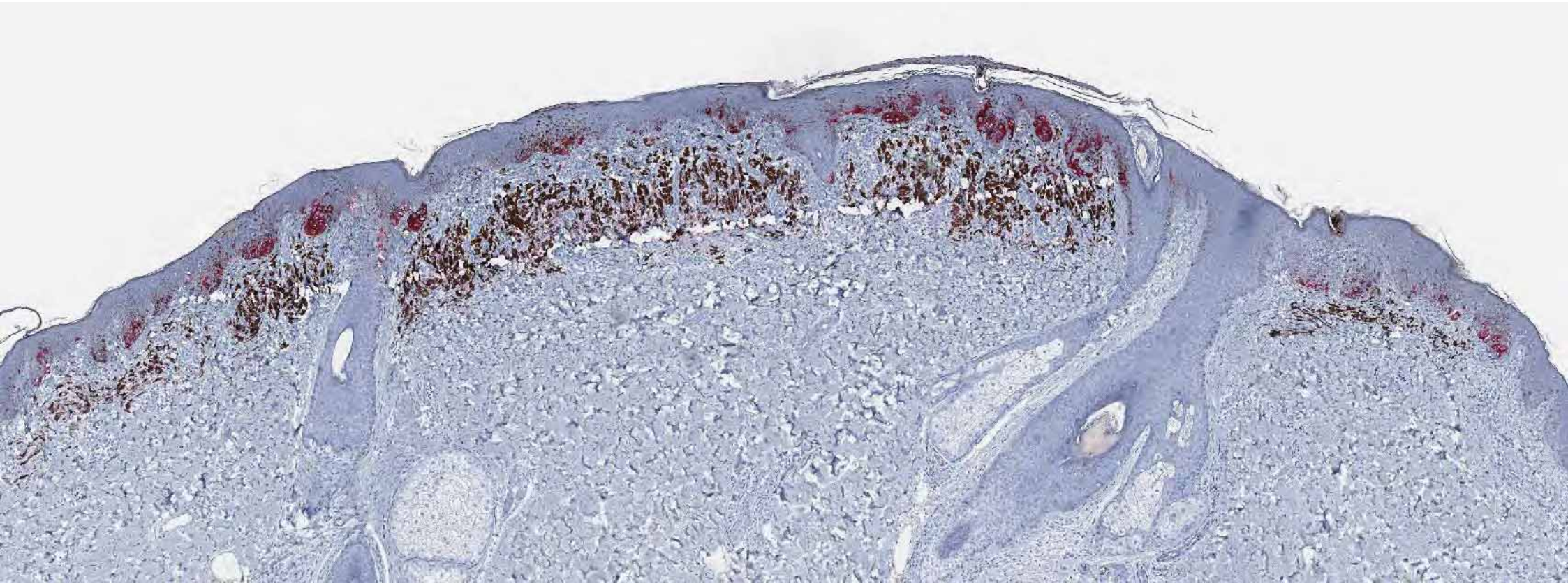
S100P/SOX10/MelanA
Help visualize melanocyte distribution

- Normal unit of melanisation
- Assessing melanocytic distribution
- Heavily pigmented tumours

Hyperpigmented lesion : density assessment

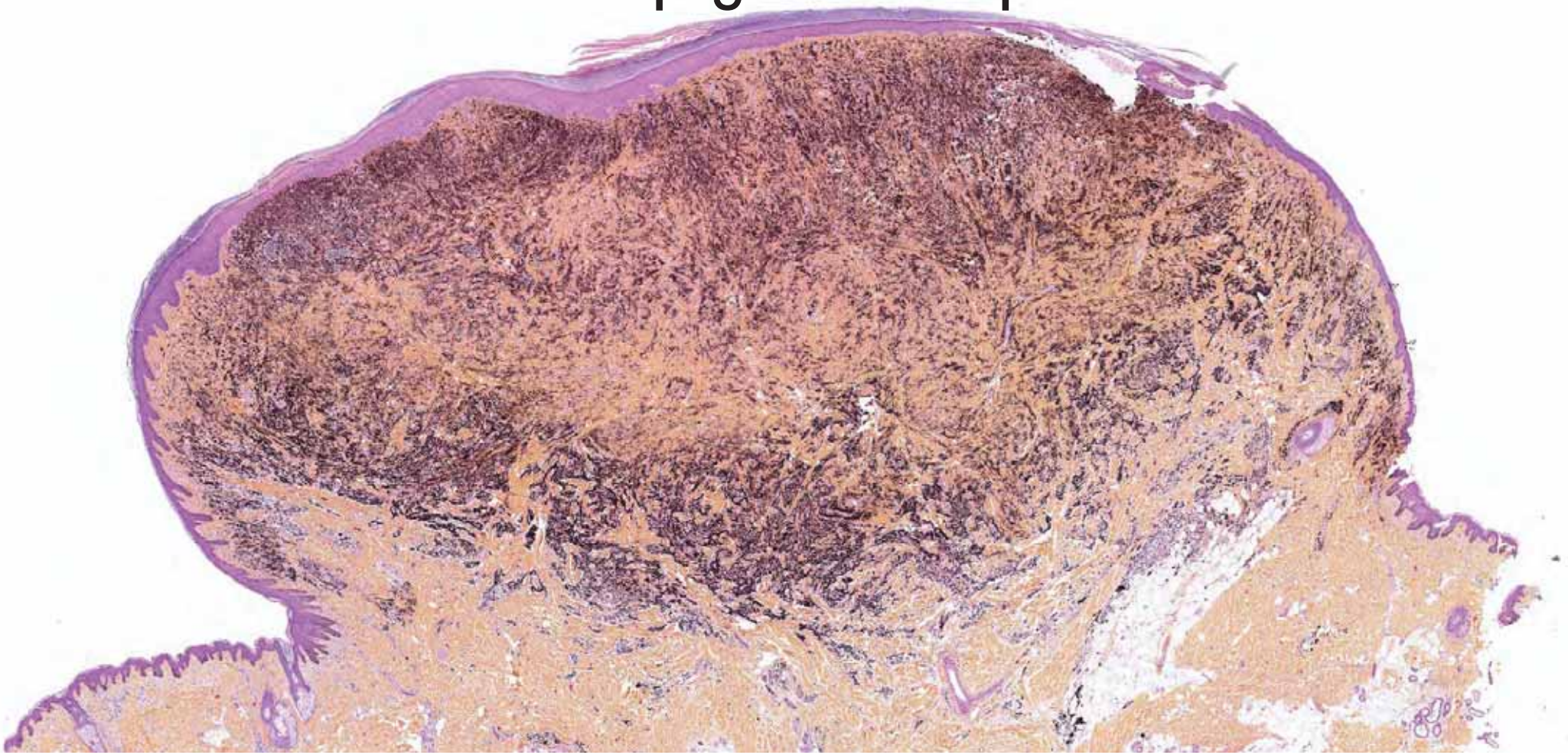


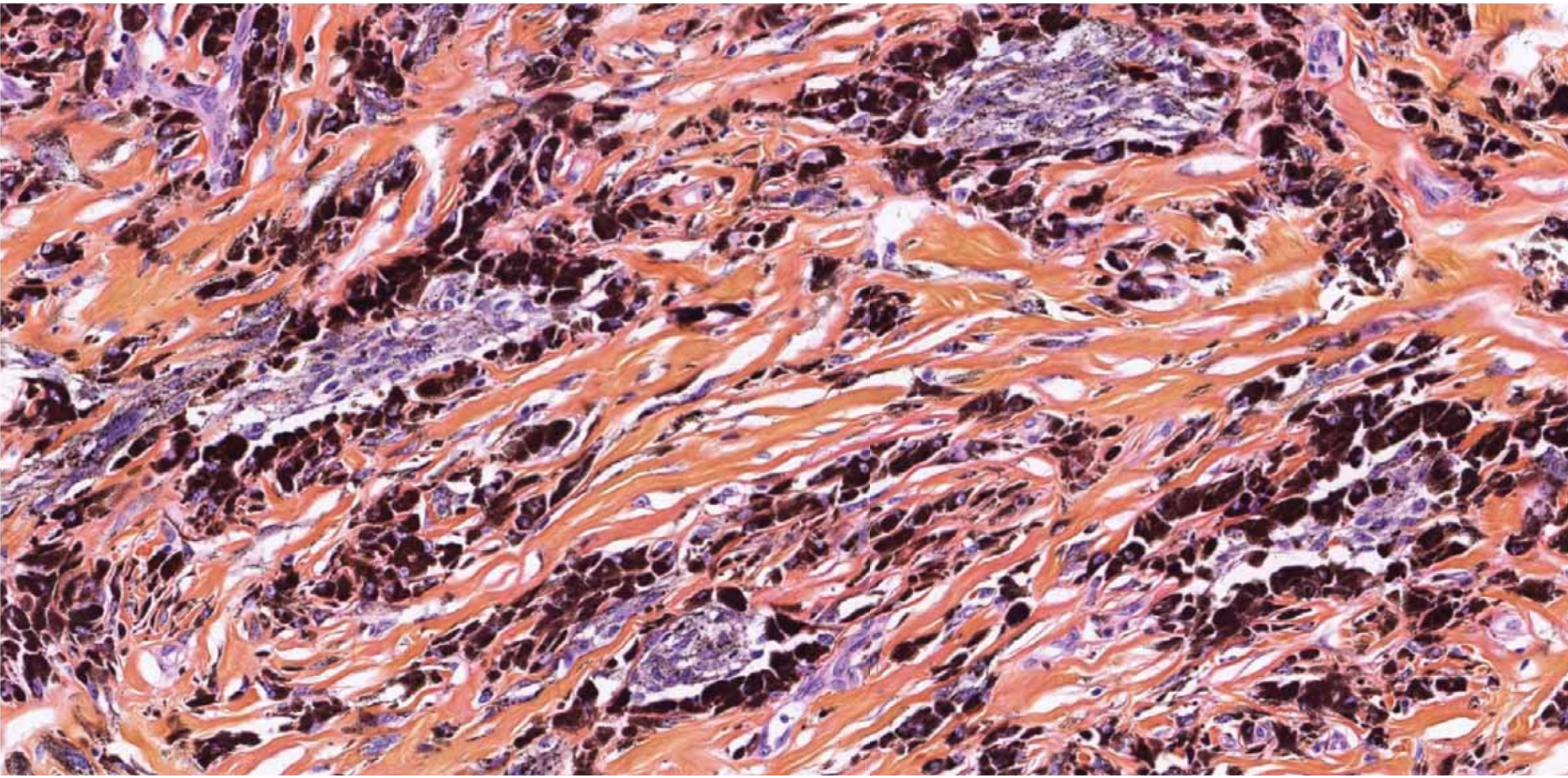
Hyperpigmented lesion : density assessment



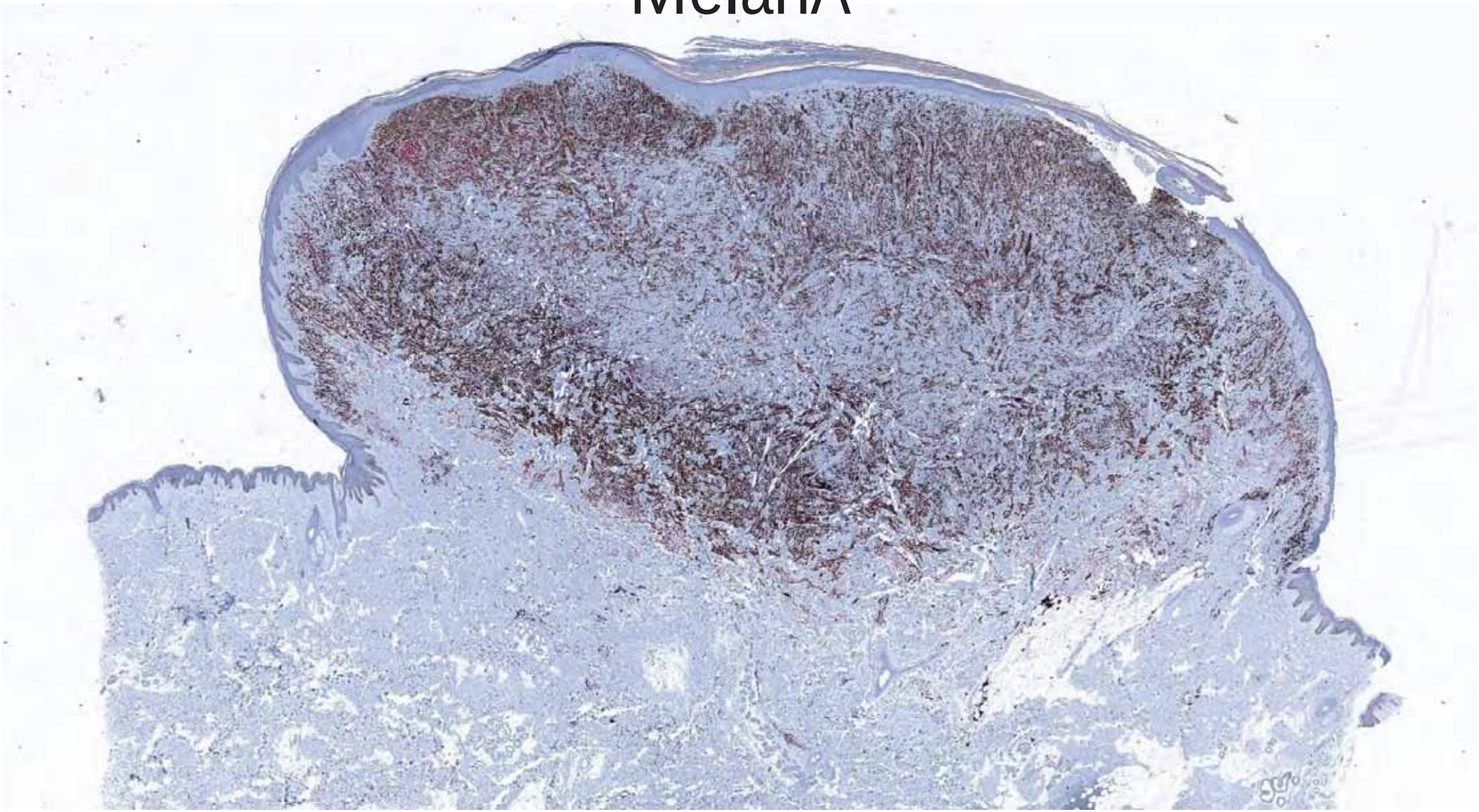
Low density, regularly distributed junctionnal nests of melanocytes: benign lesion

Melanocytes can be hard to see when too much pigment is present

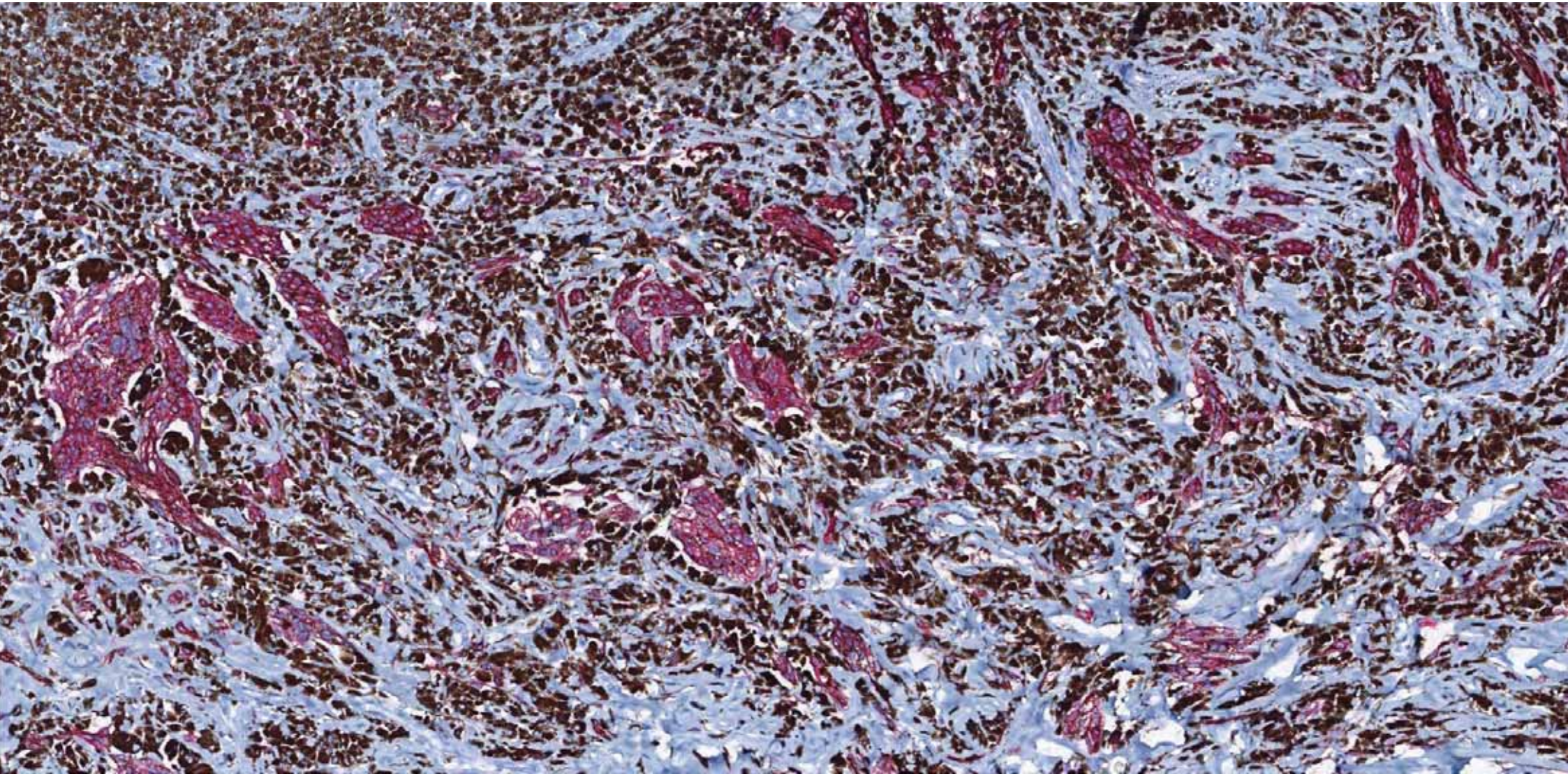




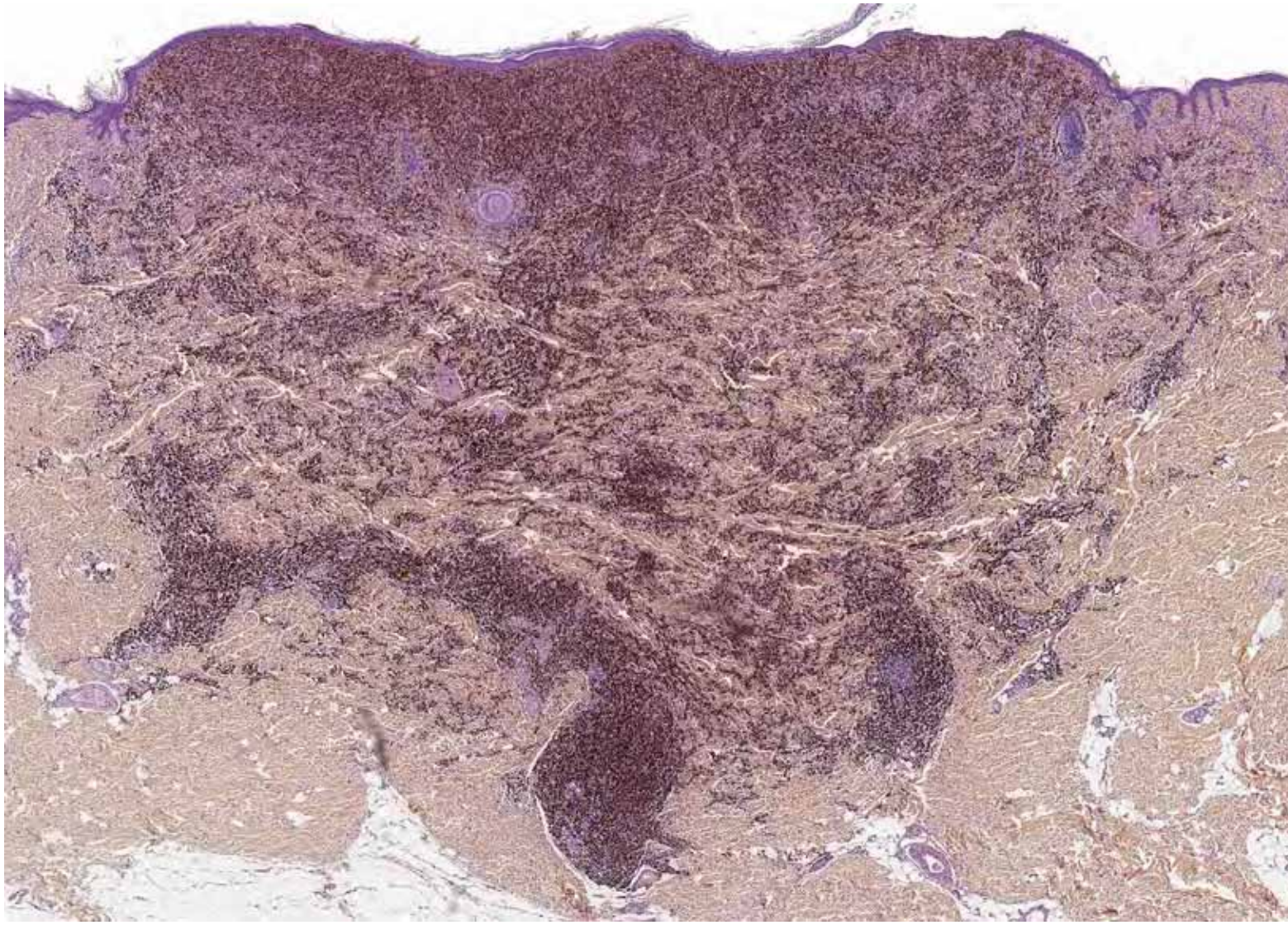
MelanA

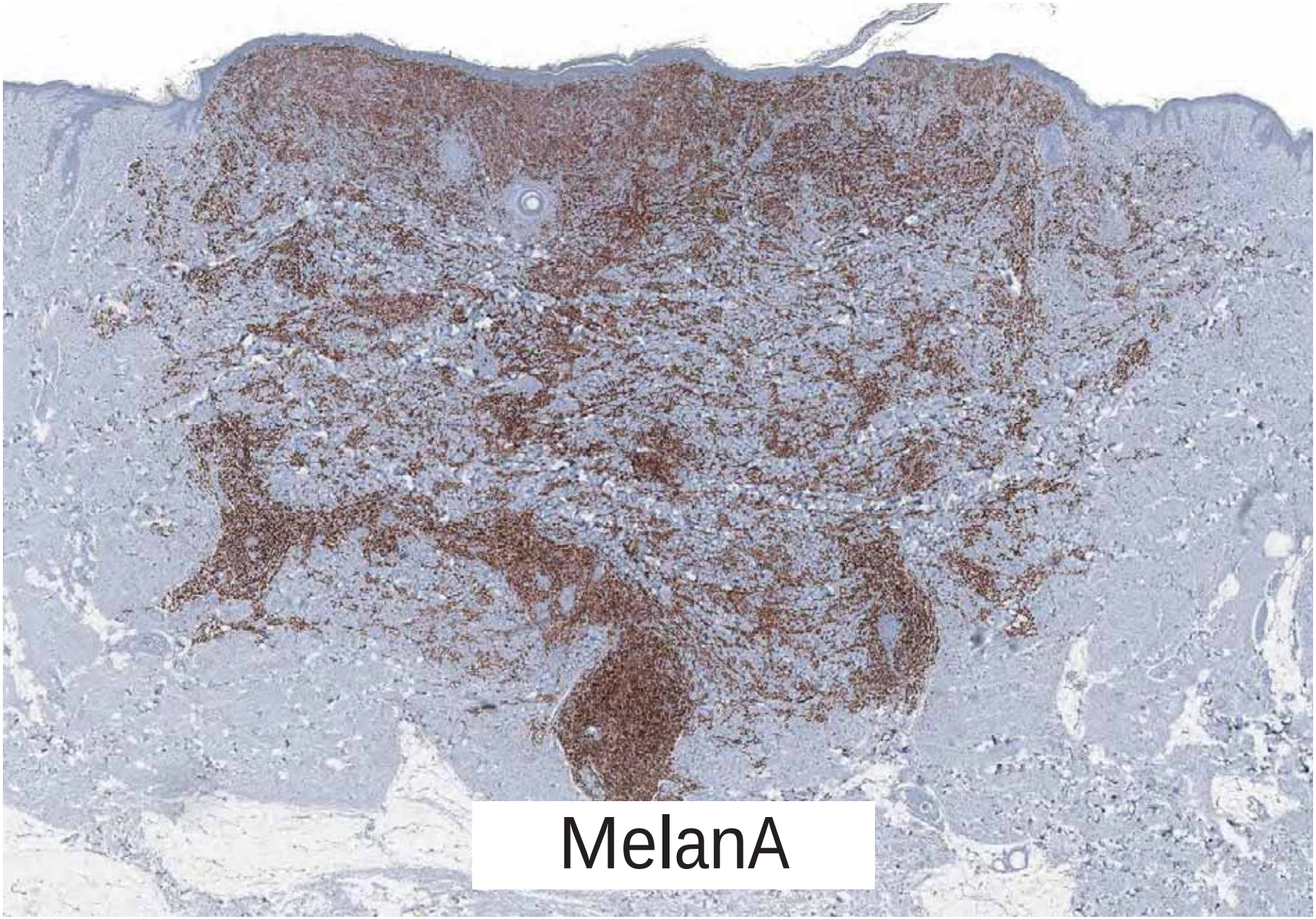


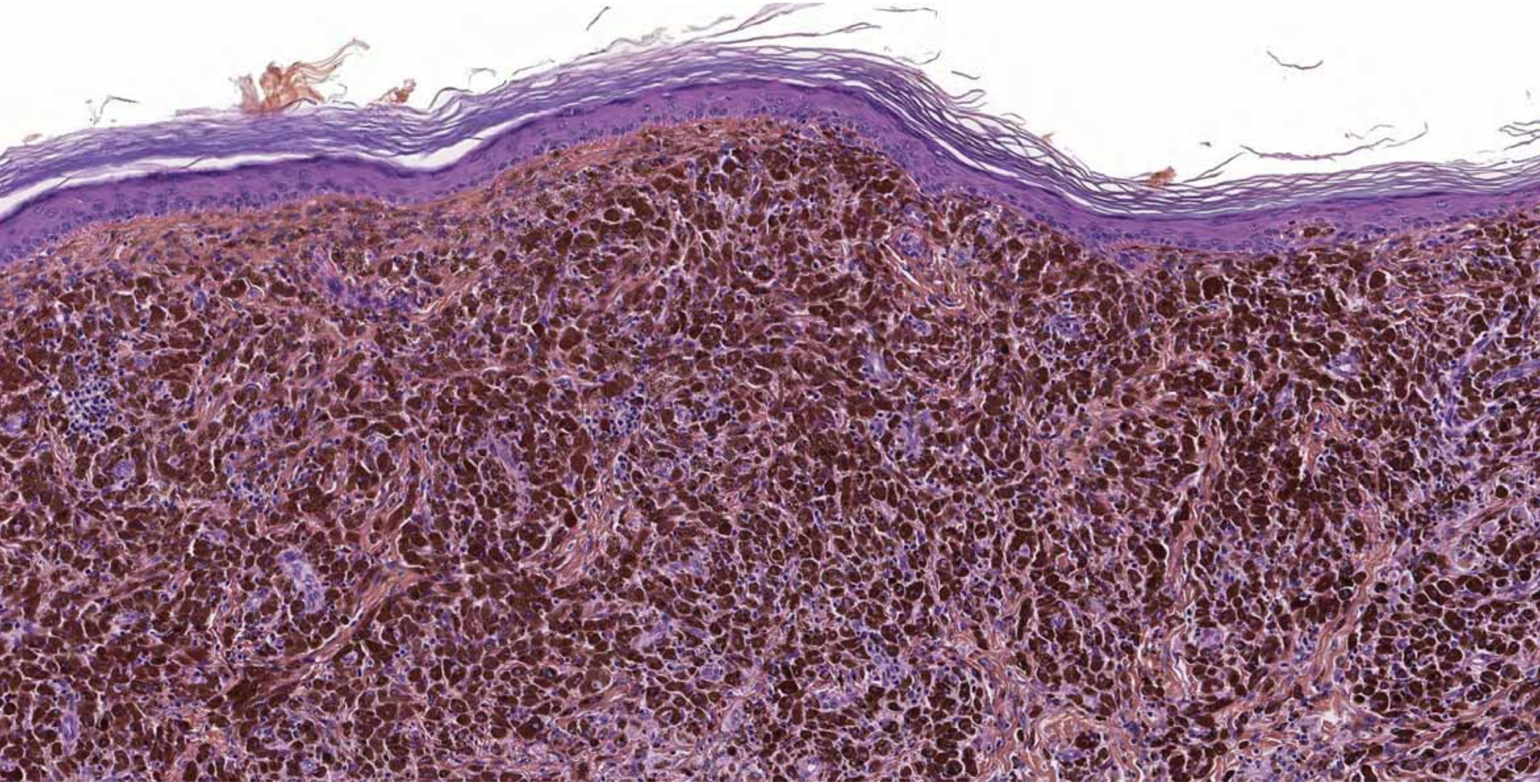
MelanA



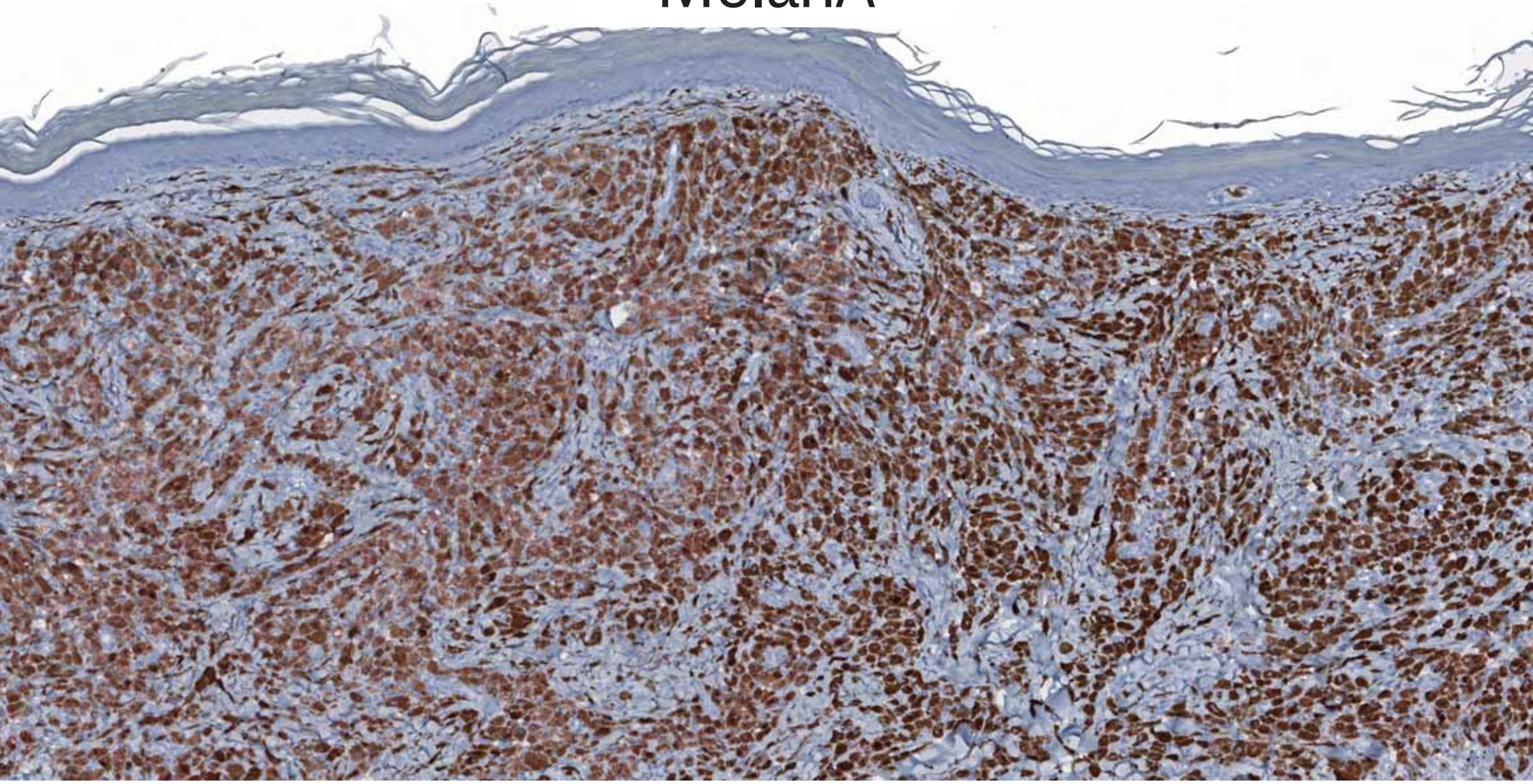
Fully regressive melanoma «Melanophage tumor»




MelanA



MelanA

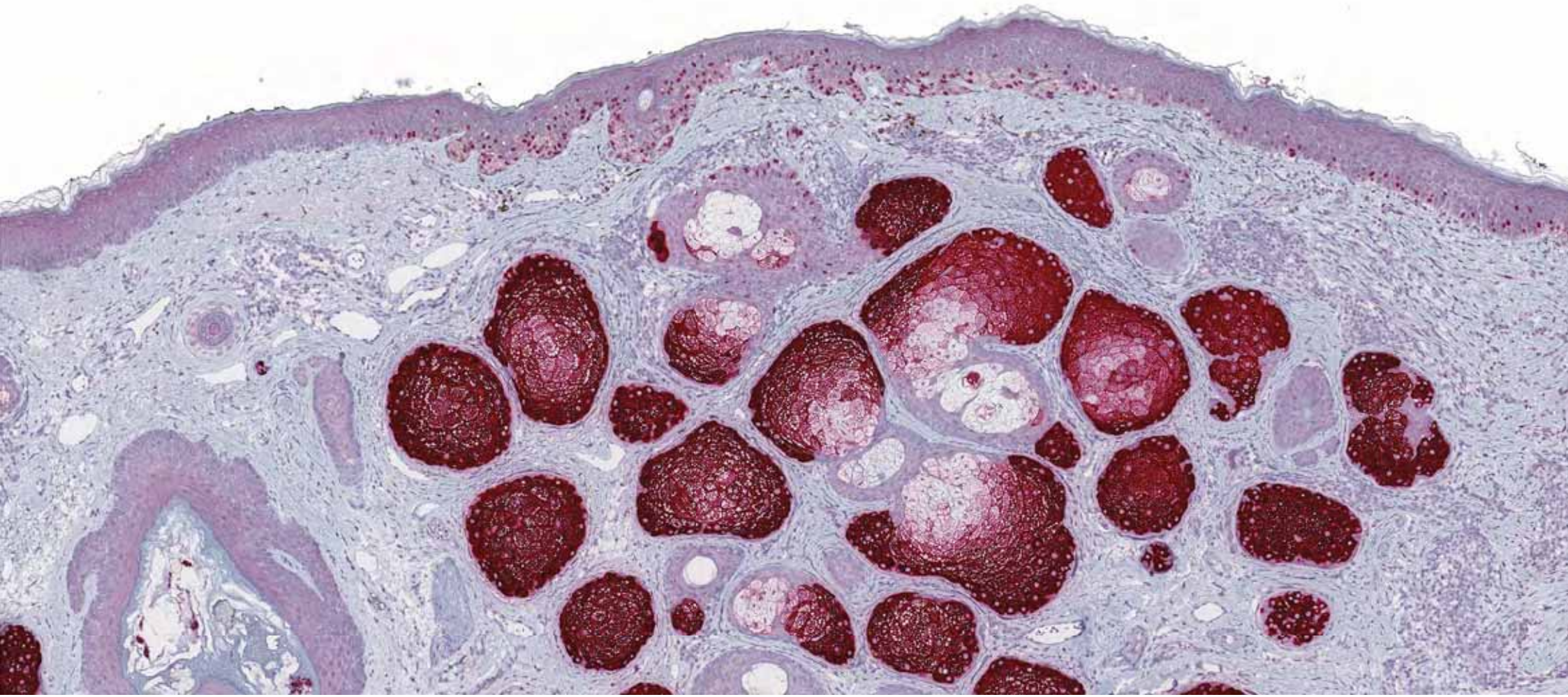


S100P/SOX10/MelanA
Help visualize melanocyte distributio

- Normal unit of melanisation
- Intra-epidermal ascension of cells
- Asymetric melanocytic distribution
- Heavily pigmented tumours
- Margin assessment
- Lympho-vascular invasion
- SLN

} Outside scope of topic interest of time

PRAME expression in LM



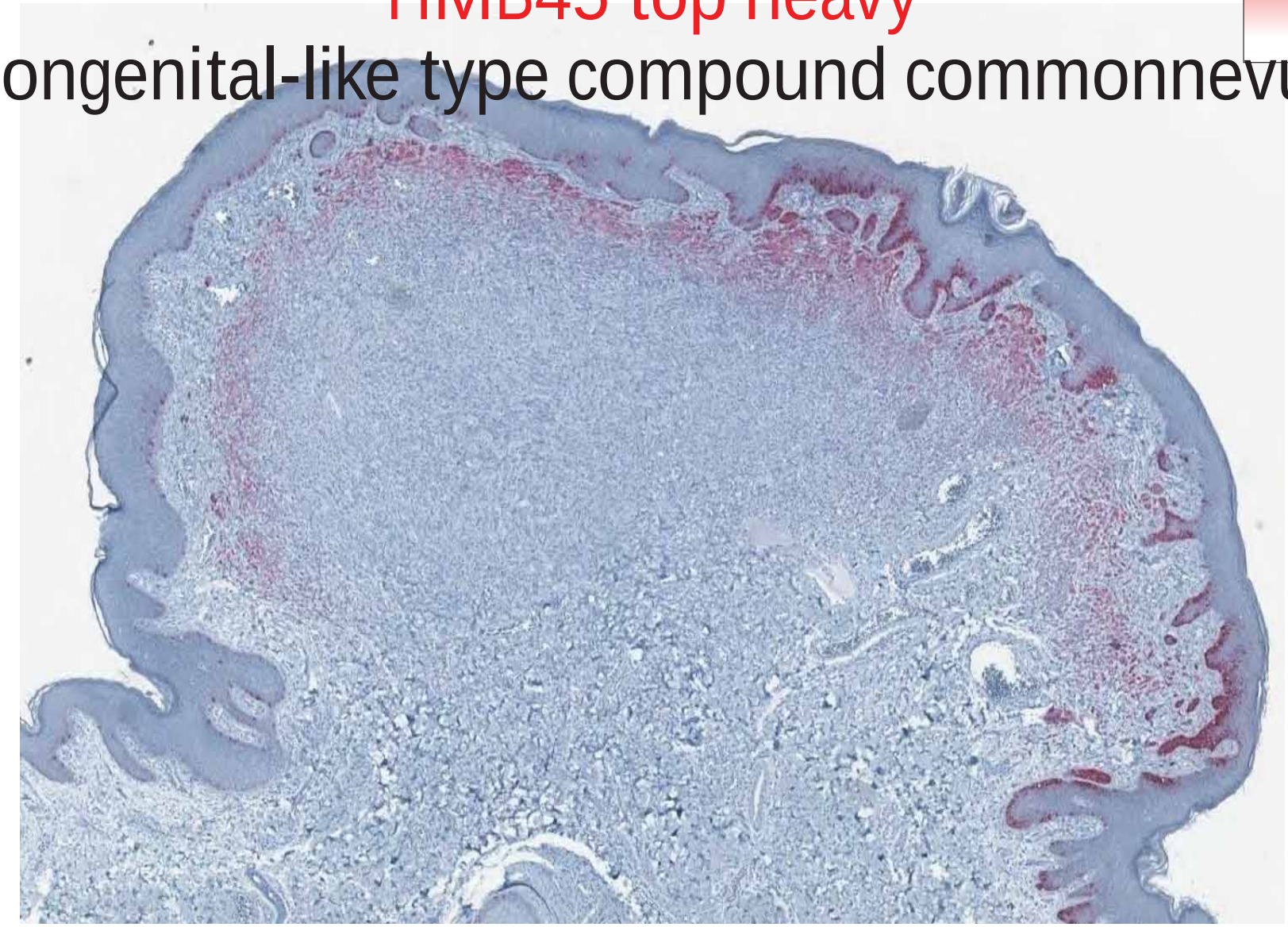
HMB45
(Human Melanoma Black 45)

- Coded by *PMEL* gene (*GP100*) Chromosome 12q13.2
- Melanocyte-specific type I transmembrane glycoprotein enriched in melanosomes (transition from Stage I to Stage II melanosomes)
- Positive in "activated" non-tumoral melanocytes
- Cytoplasmic granular stain
- No internal controls

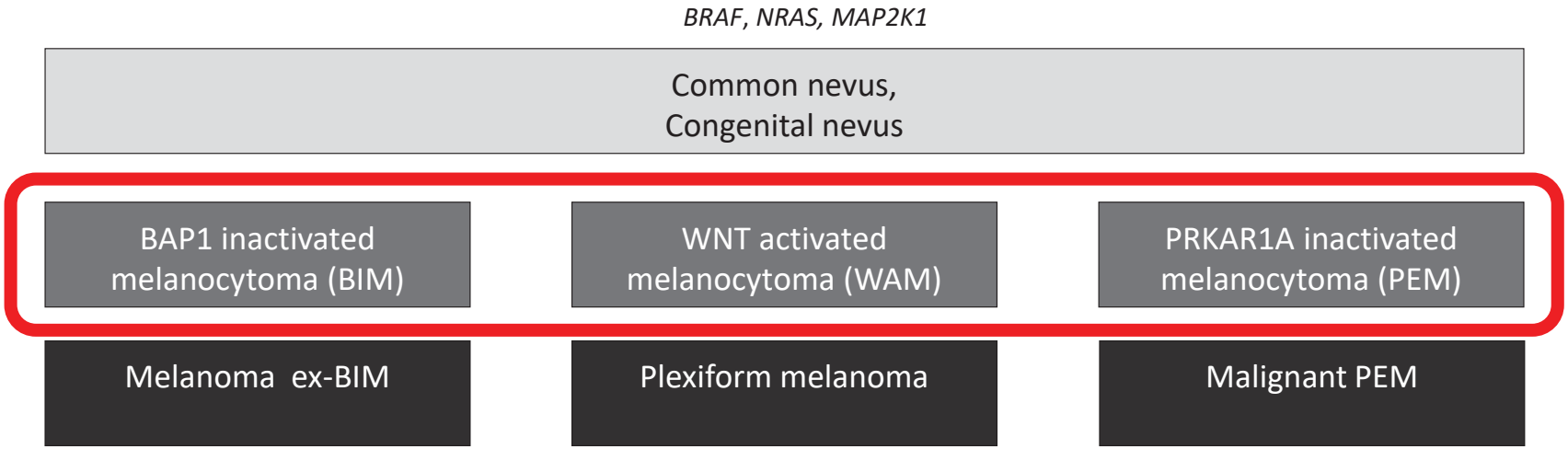
Using HMB45 in thin lesions (Breslow>1mm) is not cost-effective

HMB45 top heavy

Congenital-like type compound commonnevus

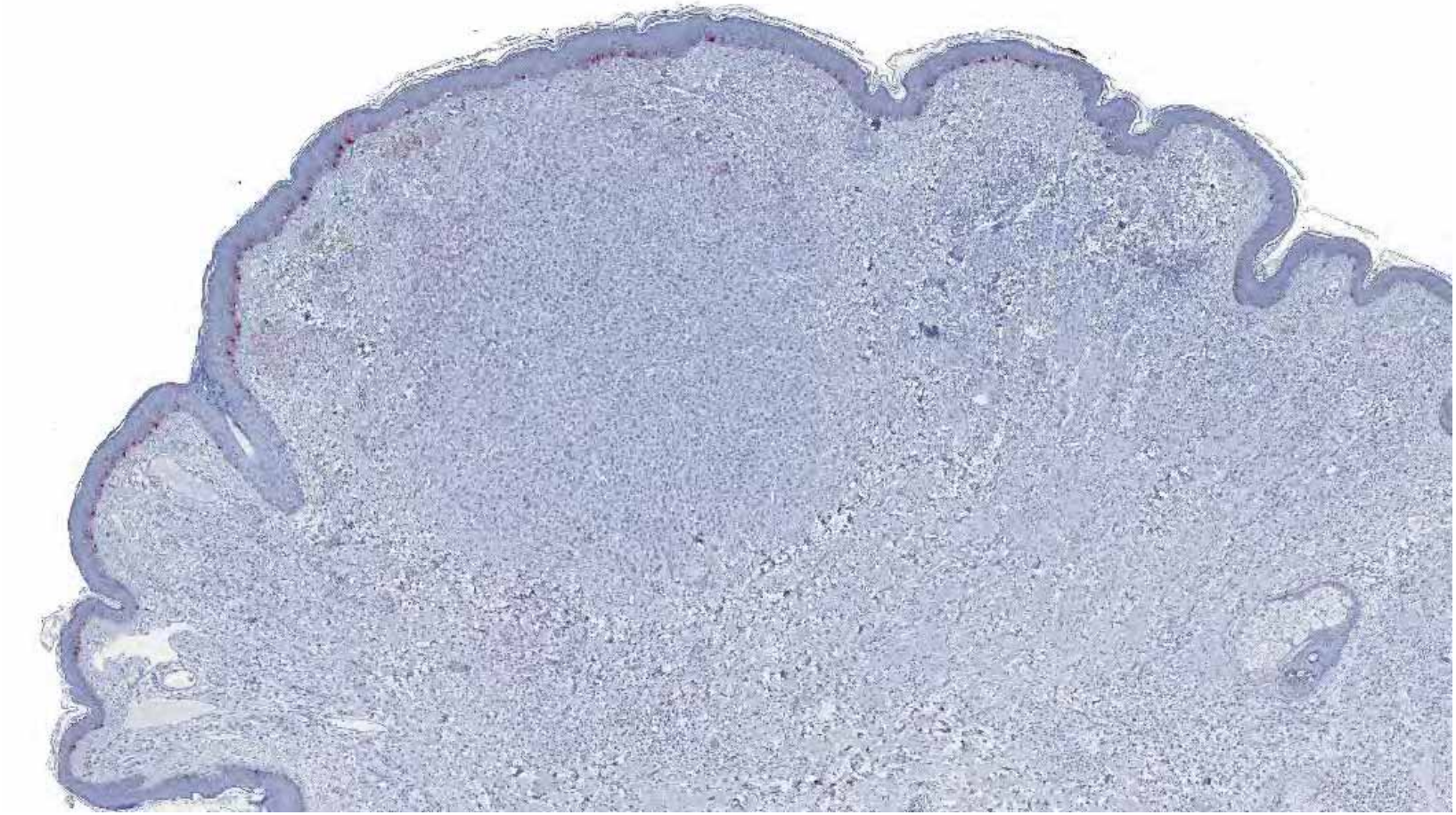


Secondary genetic alteration biphenotypic/clonal morphology



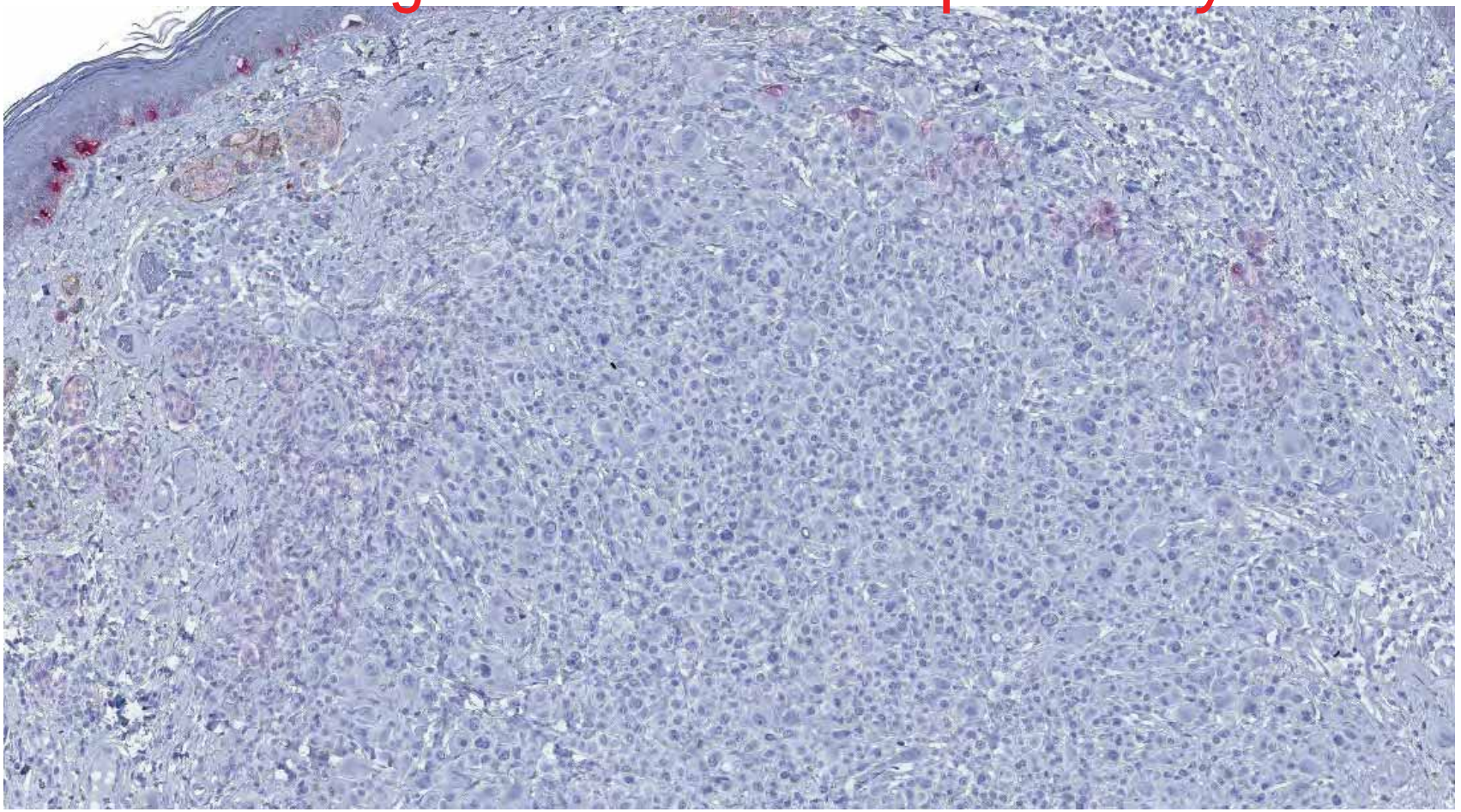
Combined common nevus /BAP1 inactivation melanocytoma

weak heterogeneous HMB45 positivity in clone

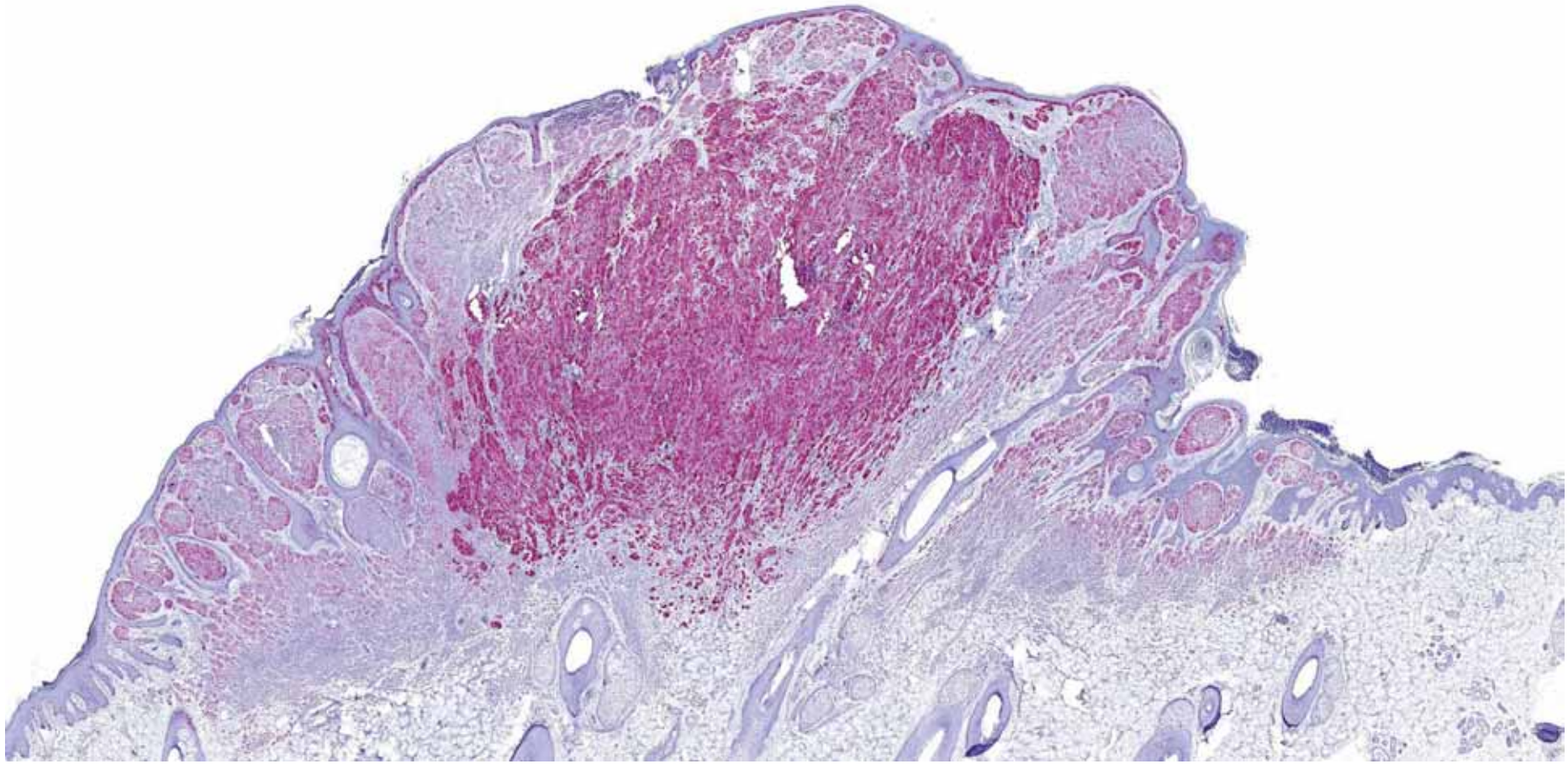


Combined common nevus /BAP1 inactivation melanocytoma

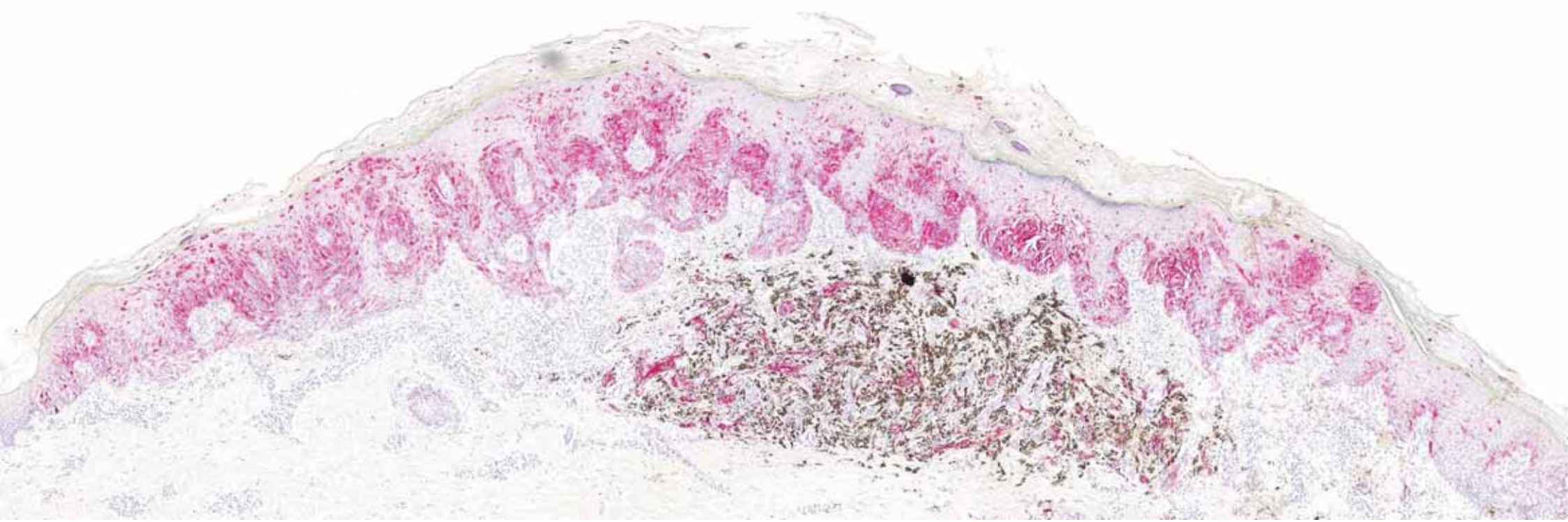
weak heterogeneous HMB45 positivity in clone



Combined common nevus
/WNT-activated/PEM/Clonal nevus
diffuse/heterogeneous HMB45 positivity in clone



Combined common nevus
/WNT activated/PEM/Clonal nevus
diffuse/heterogeneous HMB45 positivity in clone



Combined common nevus
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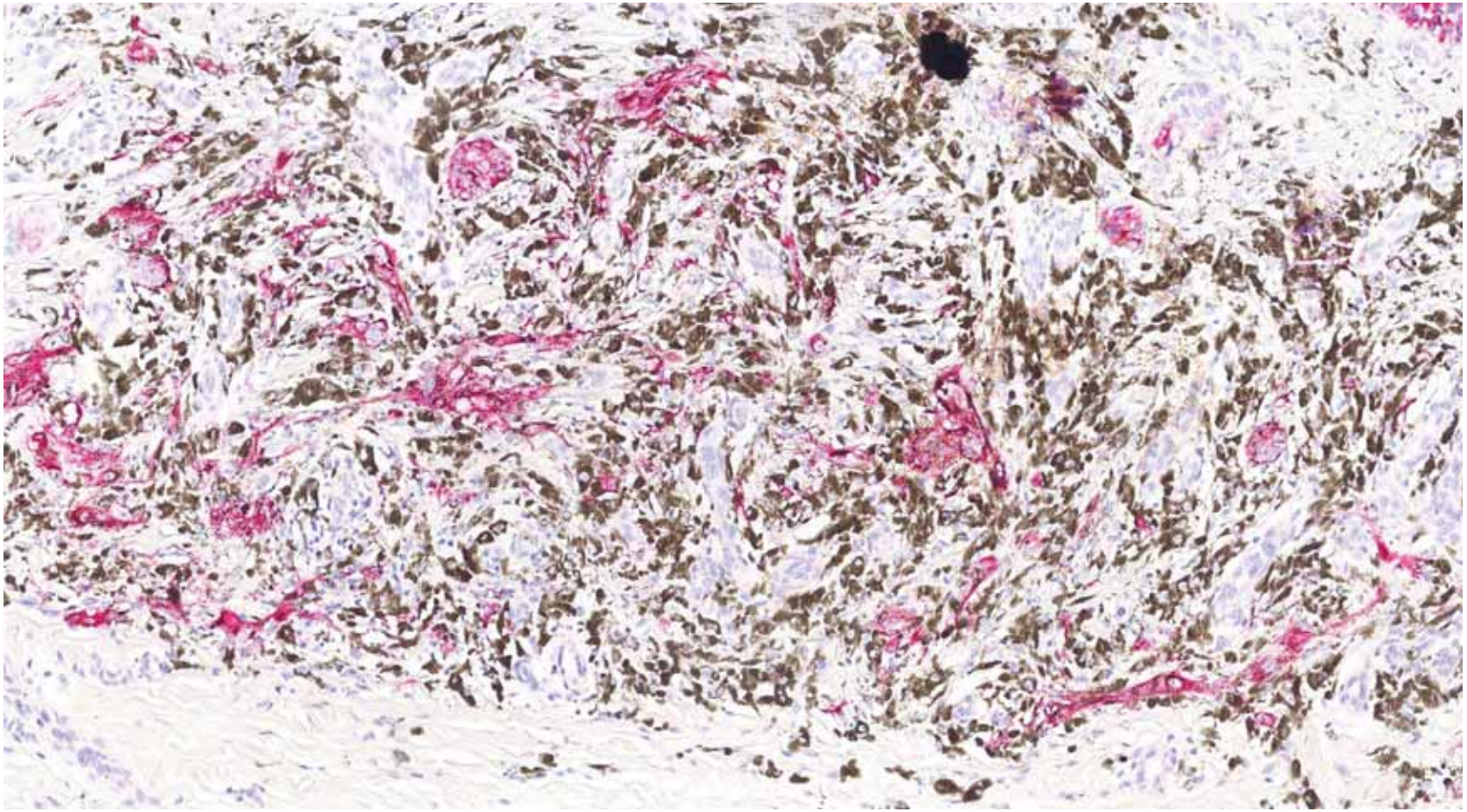

Combined common nevus
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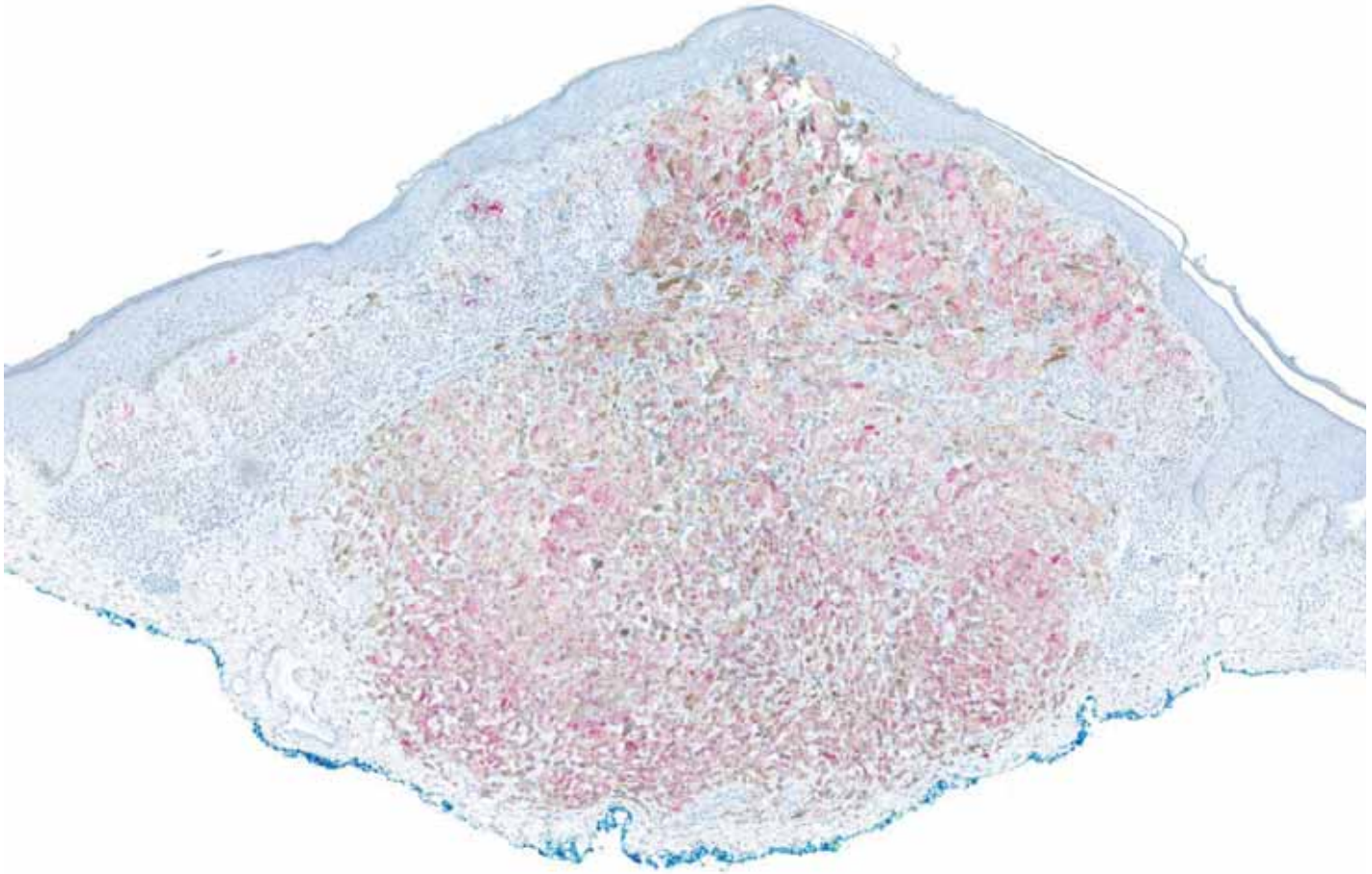
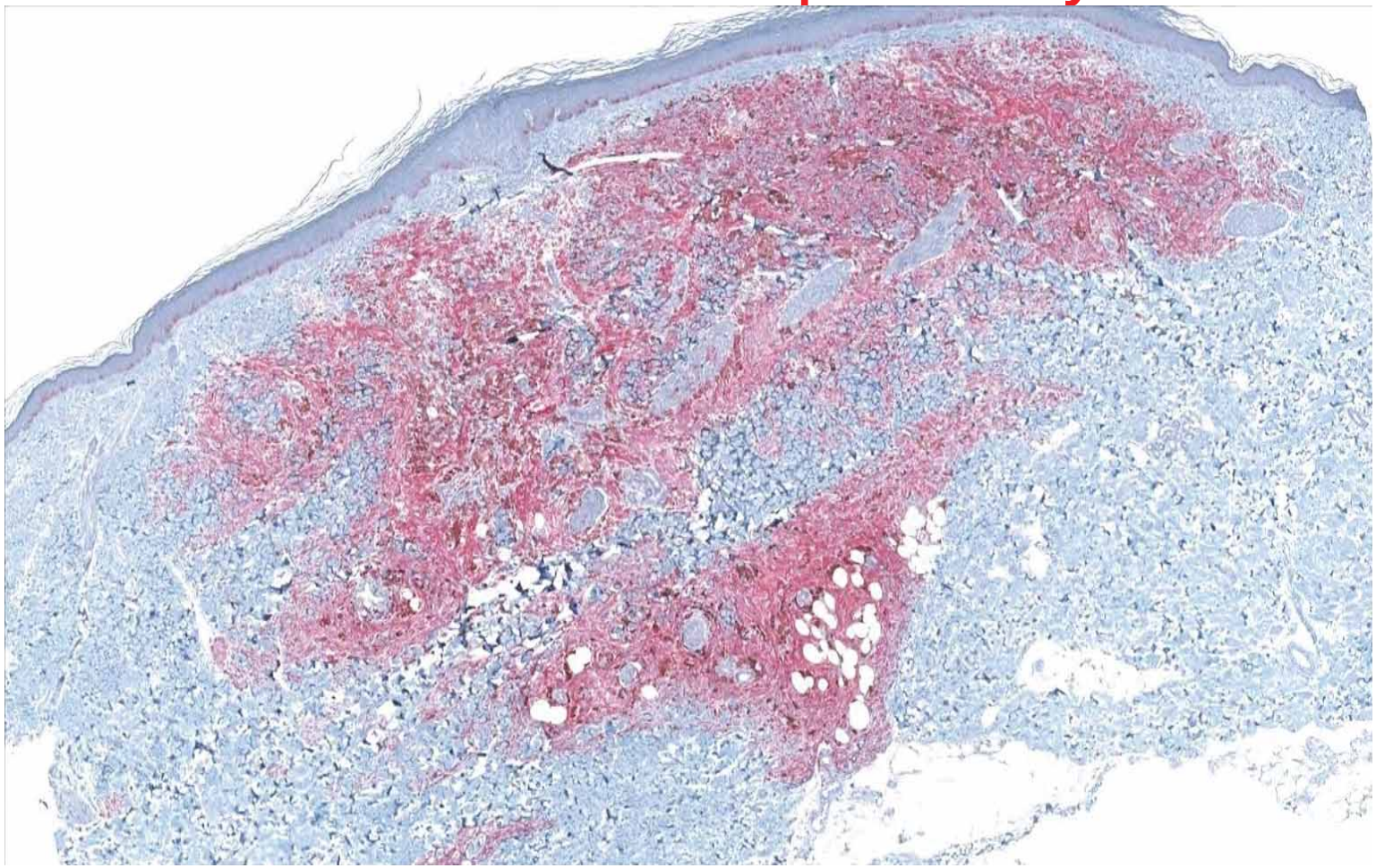
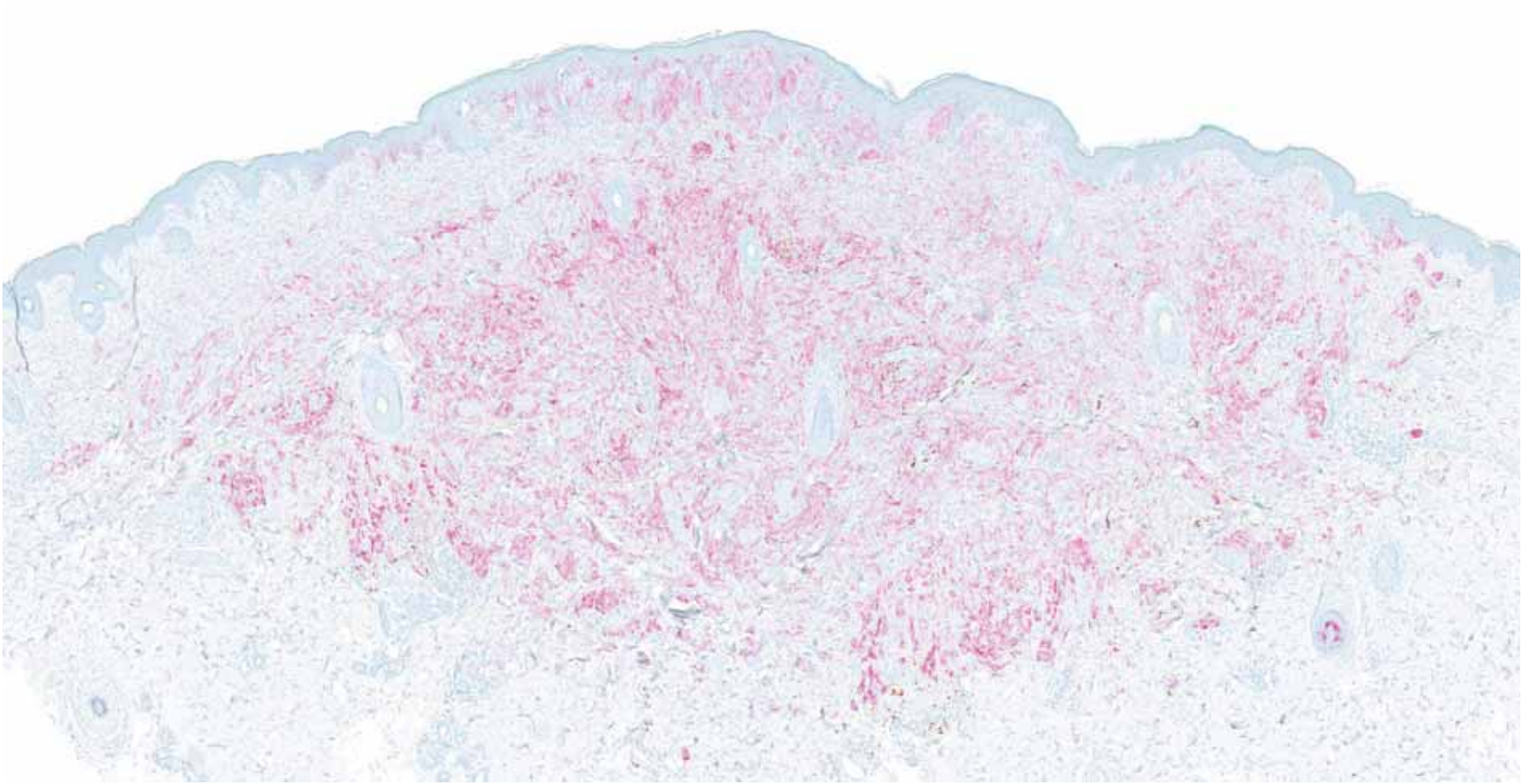
diffuse/heterogeneous HMB45 positivity in clone



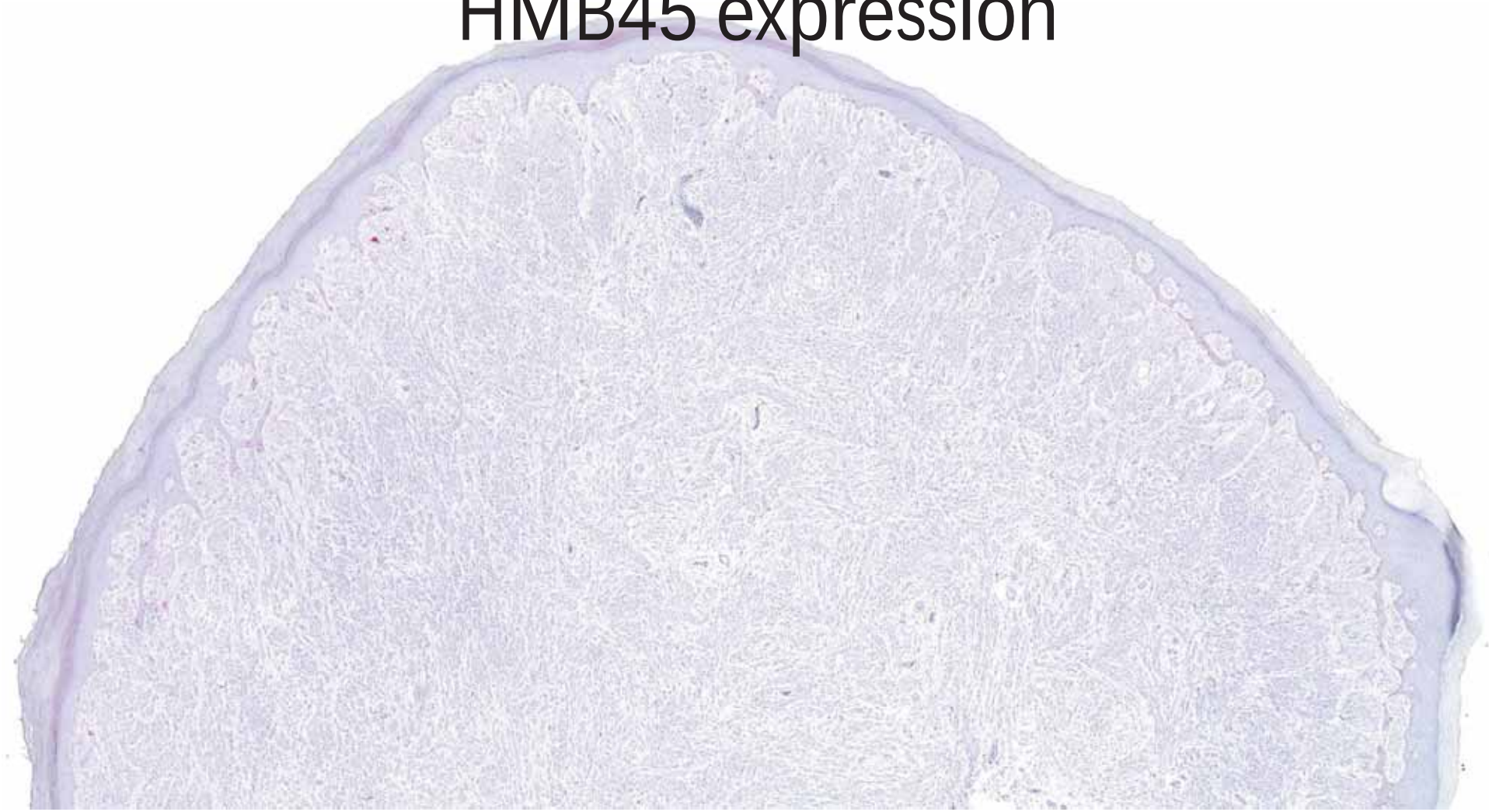
Non clonal (pure)
WNT activated melanocytoma (DPN/plexiform nevus)
HMB45 diffuse positivity



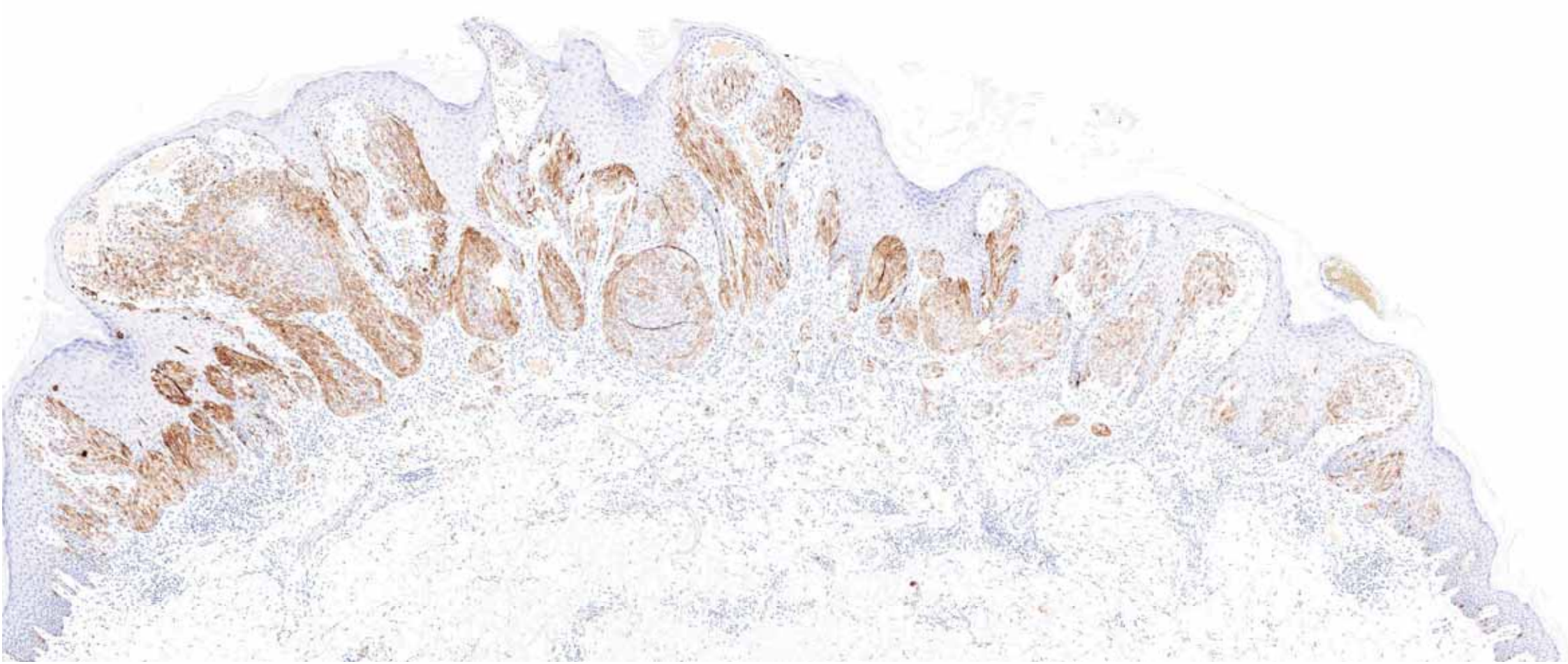
Non clonal (pure)
WNT activated melanocytoma (ex-DPN)
HMB45 heterogeneous positivity



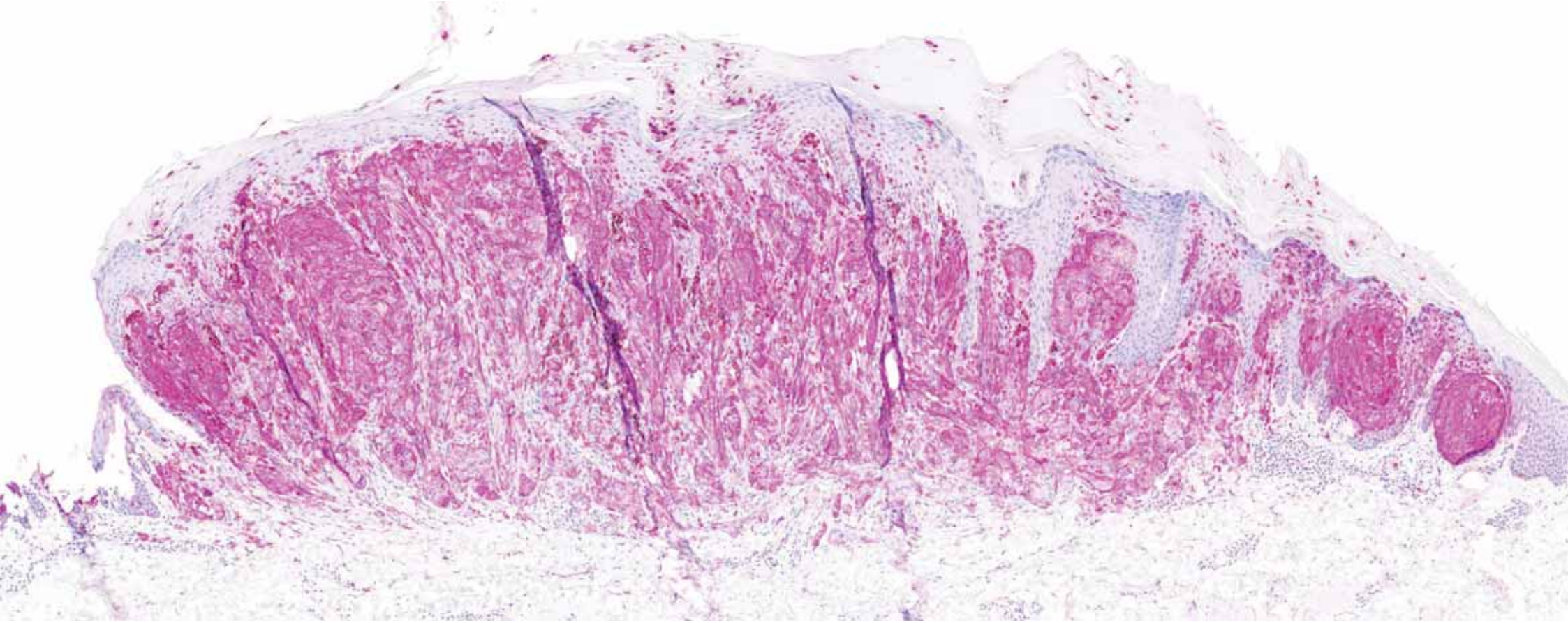
Compound Spitz nevi have variable HMB45 expression



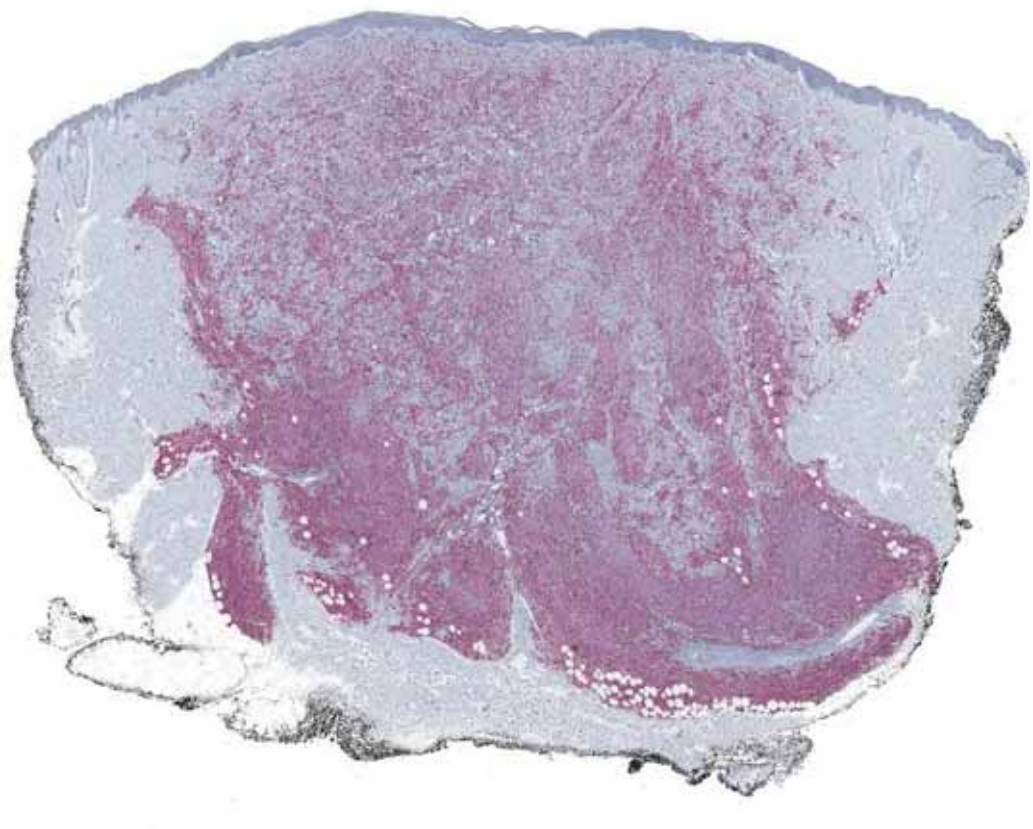
Compound Spitz nevi have variable HMB45 expression



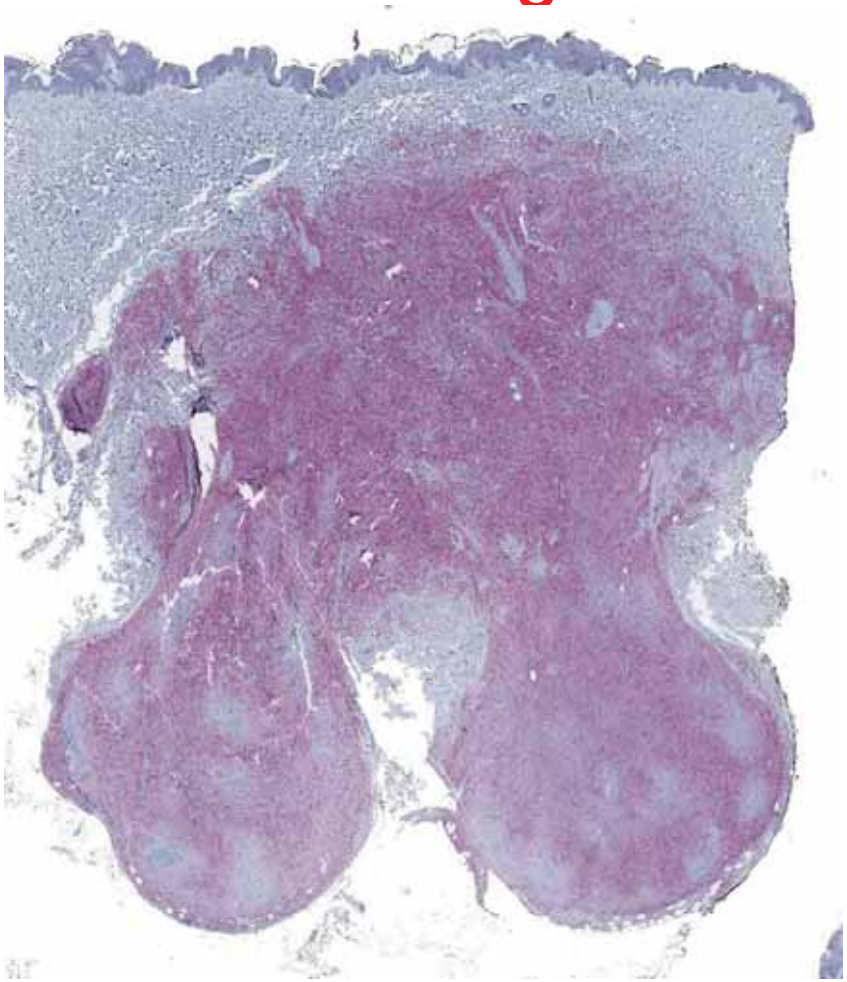
Compound Spitz nevi have variable HMB45 expression



Cellular blue nevus
HMB45 diffuse/heterogeneous positivity

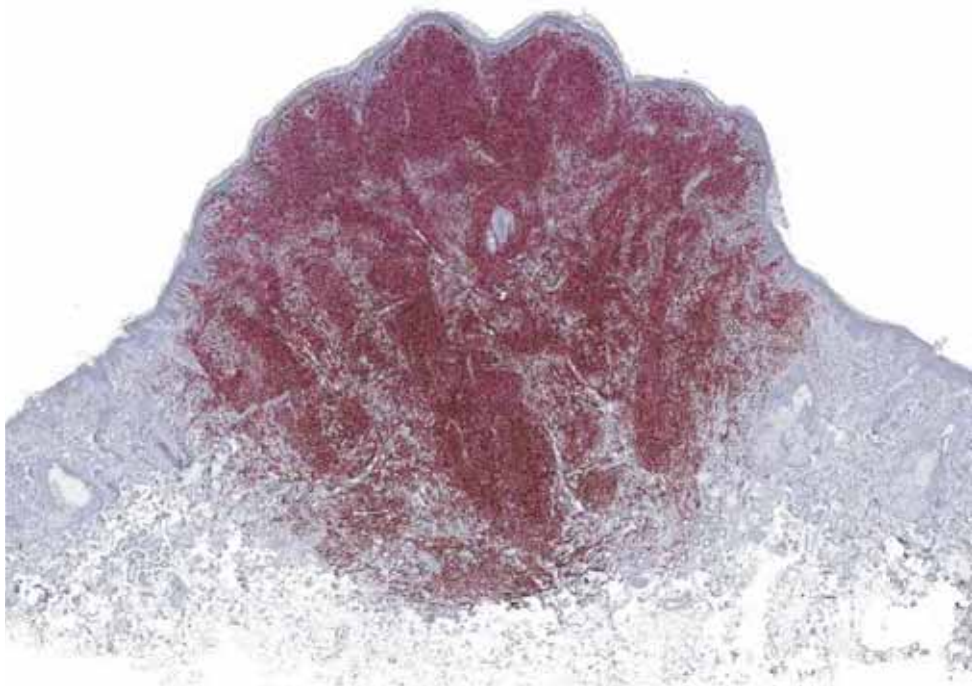


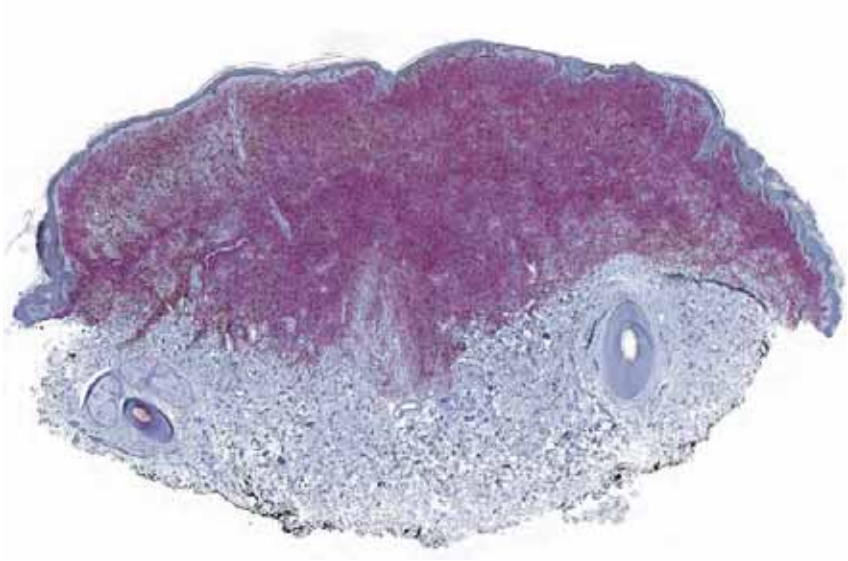
Cellular blue nevus
HMB45 diffuse/heterogeneous positivity



PKC gene fused blue nevus

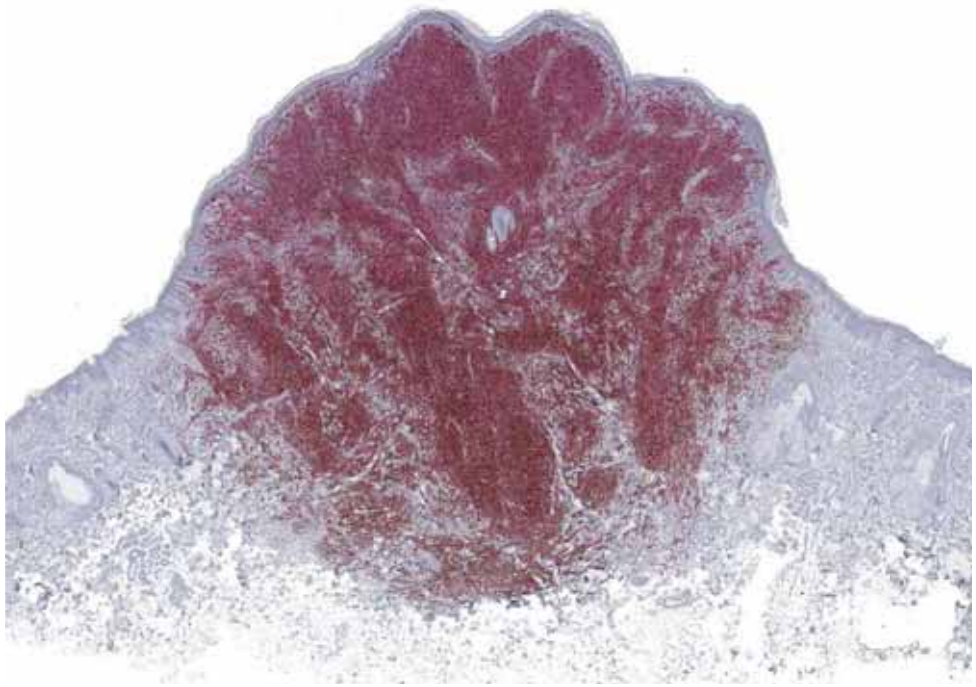
HMB45 diffuse/heterogeneous positivity

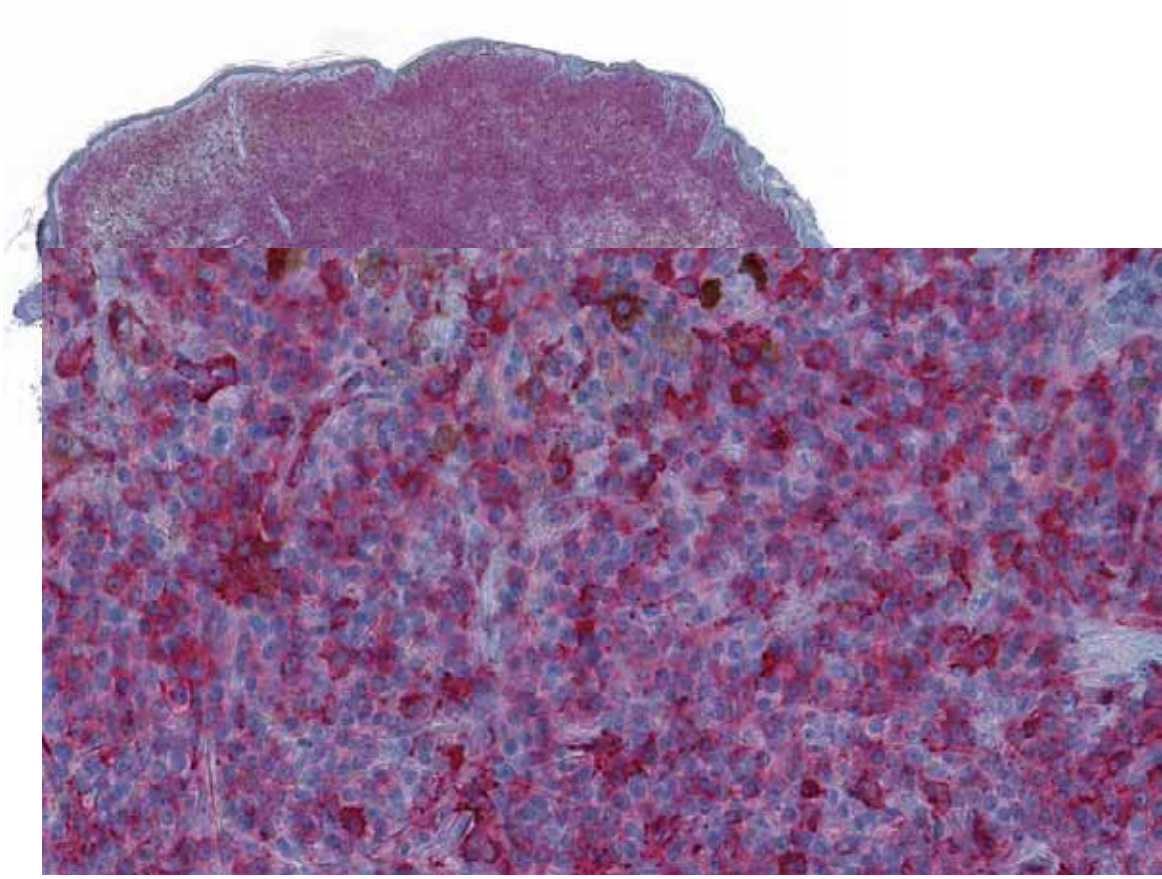




PKC gene fused blue nevus

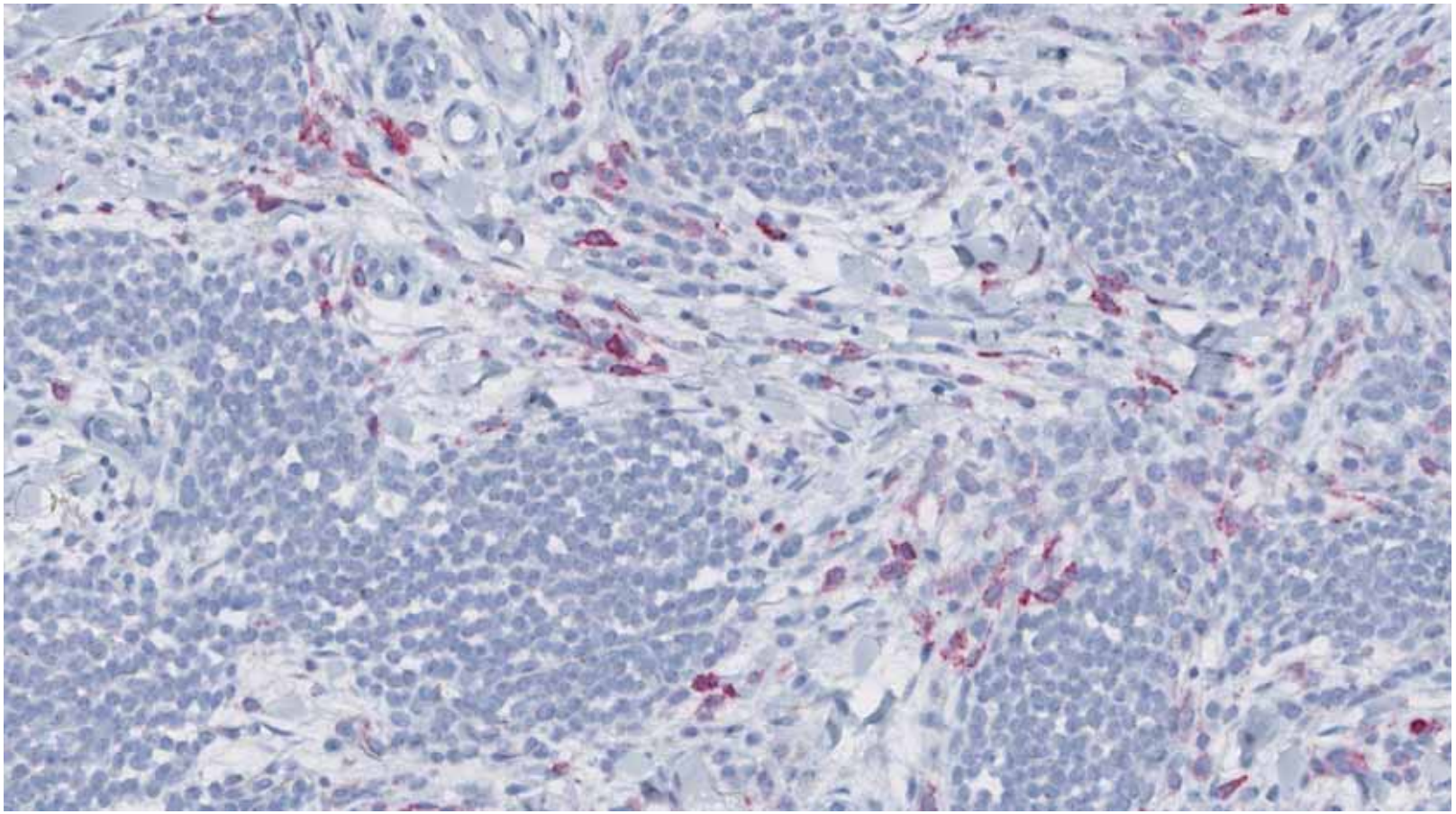
HMB45 diffuse/heterogeneous positivity



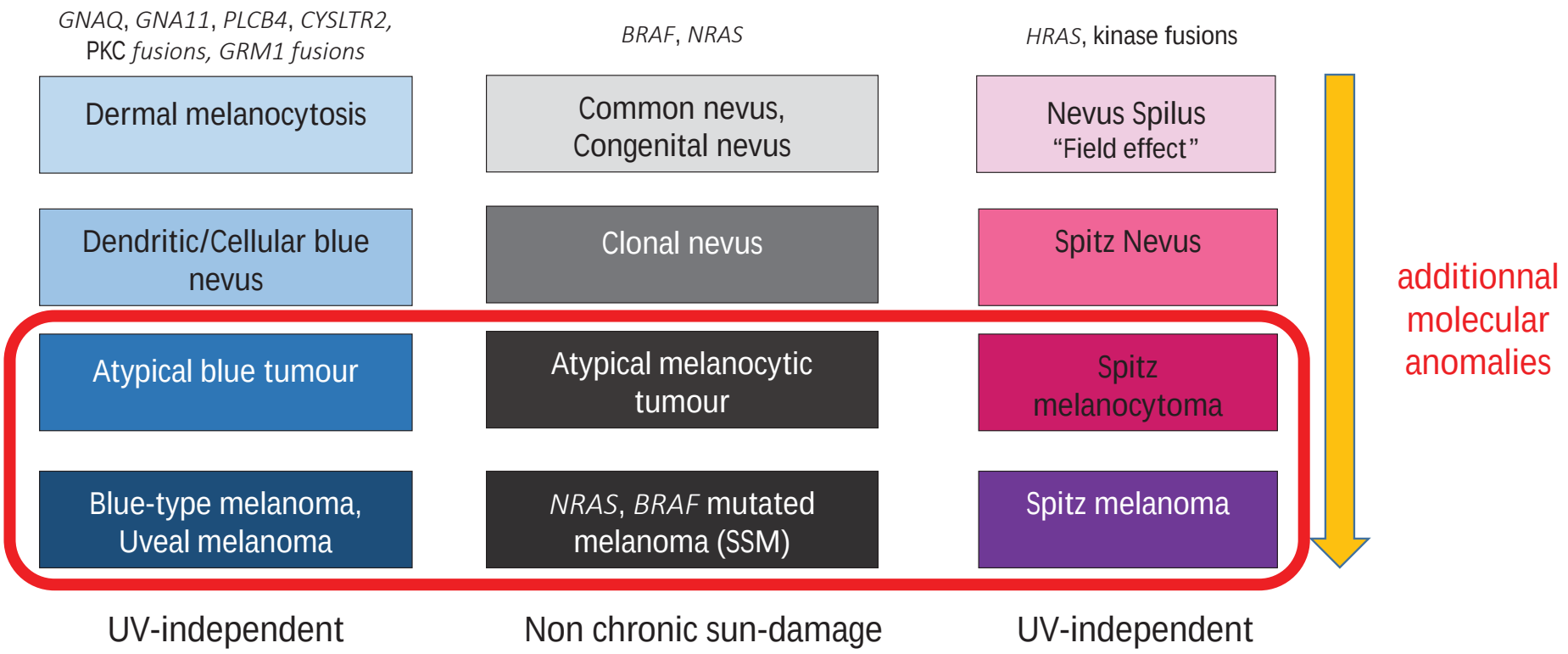


NRAS-IDH1 co-mutation melanocytoma

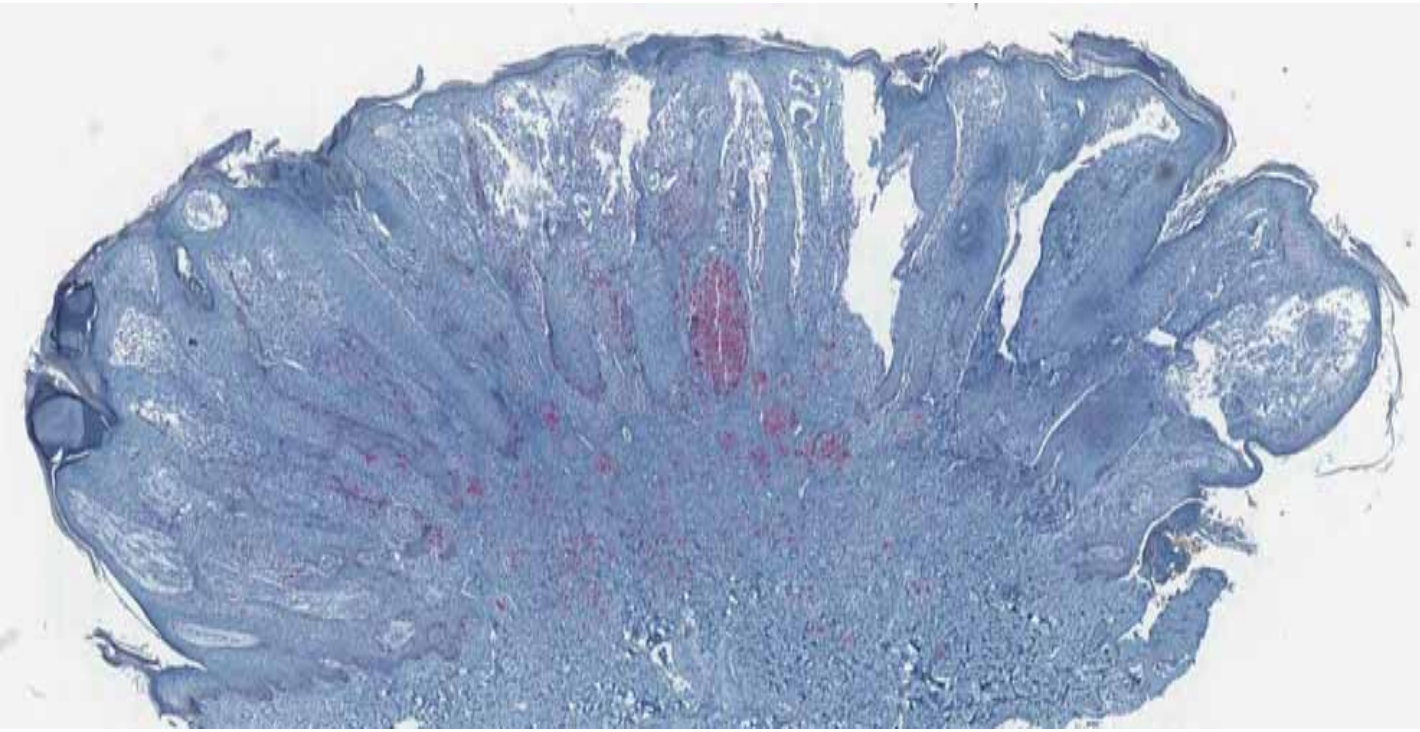
HMB45 positivity limited to spindled cells



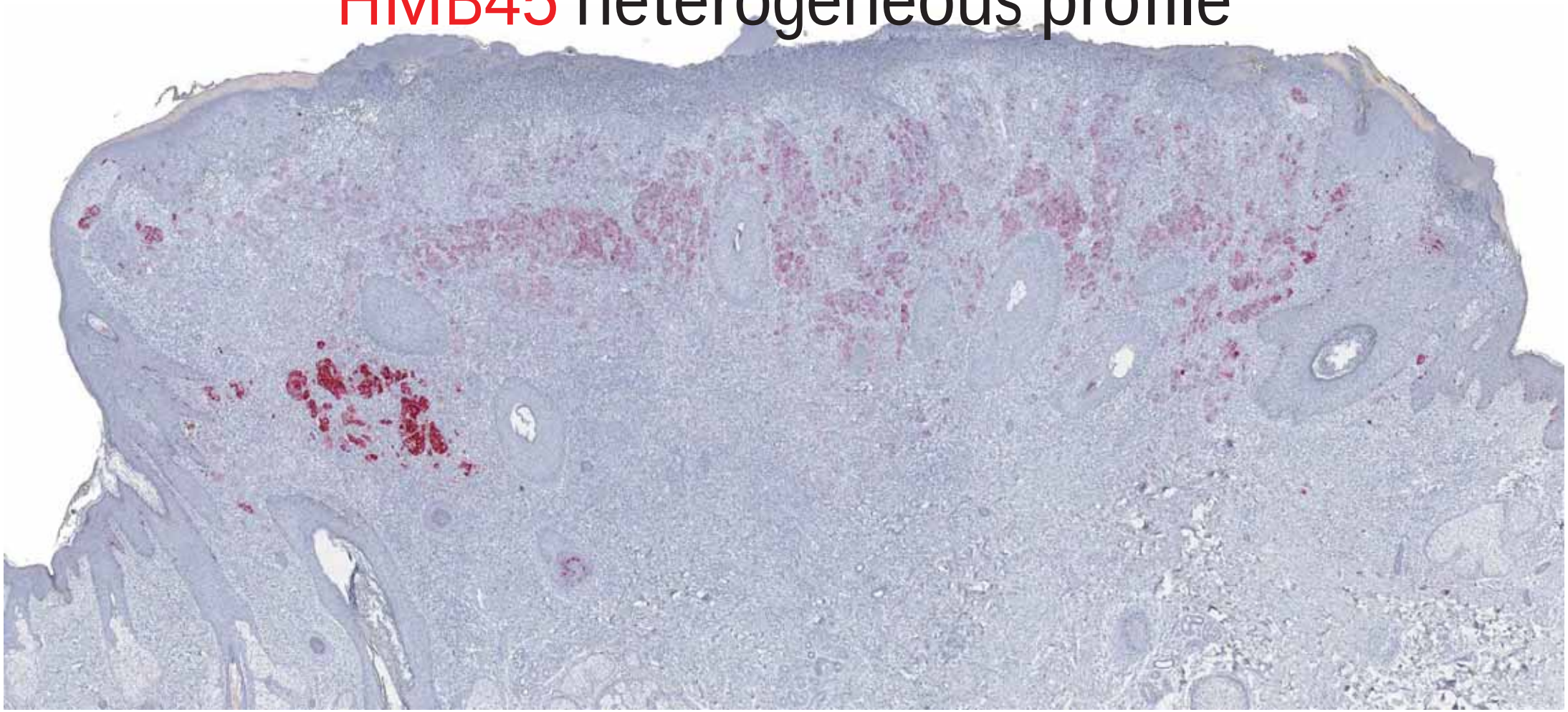
Integrative classification of melanocytic tumors
Nevus to melanoma groups



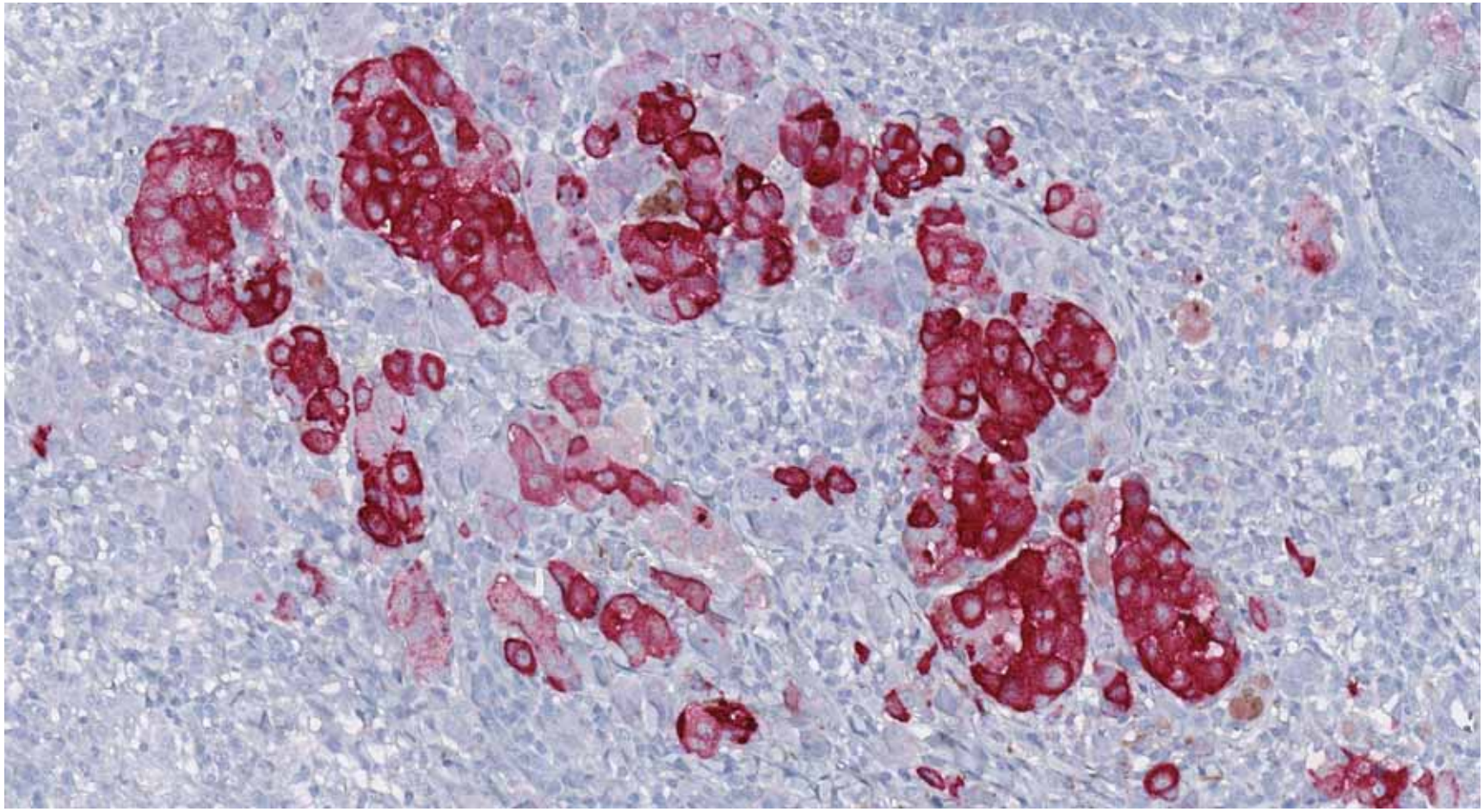
Atypical spitz nevus
HMB45 heterogeneous profile



Atypical spitz nevus MAP3K8 fusion
HMB45 heterogeneous profile

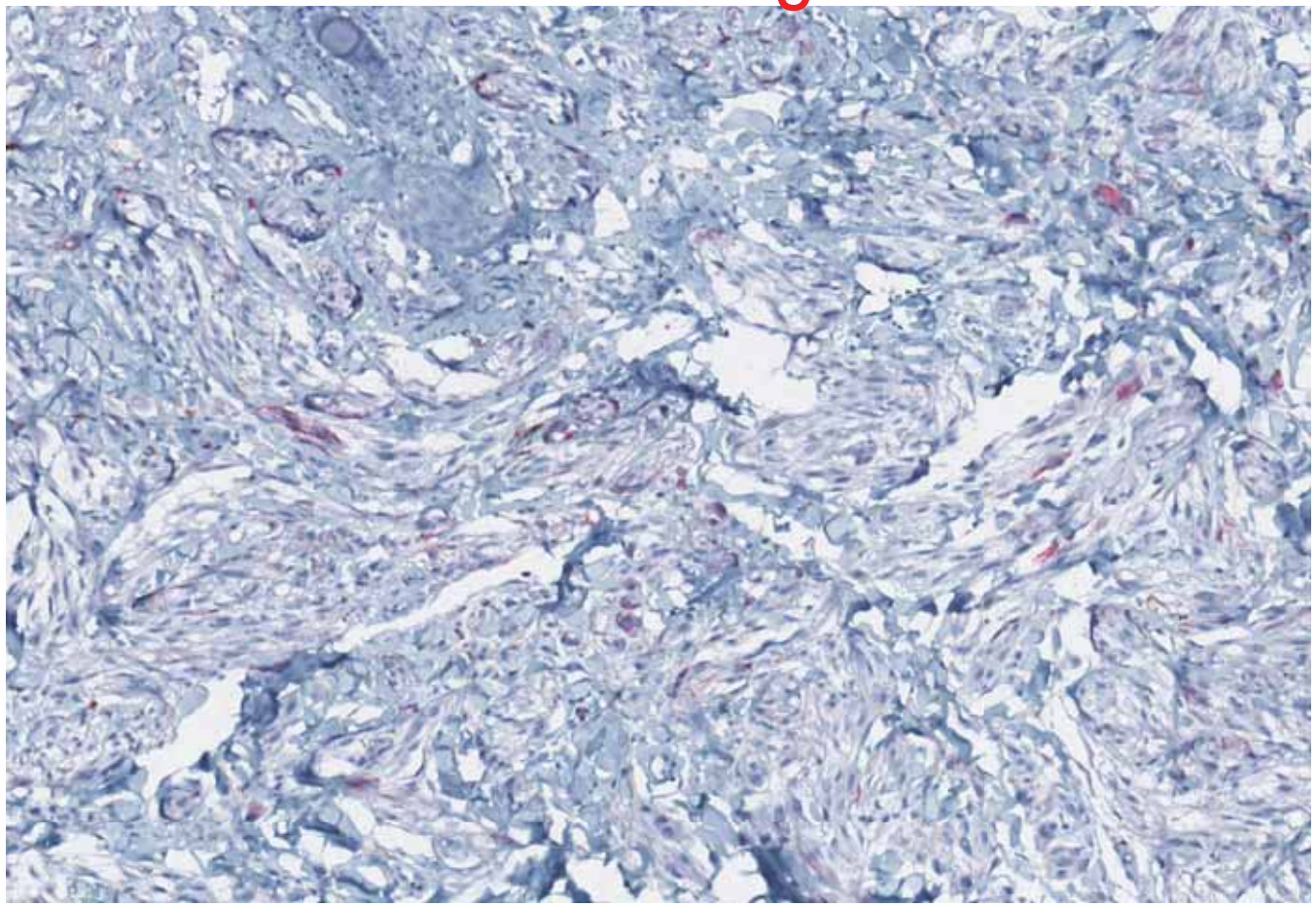


HMB45

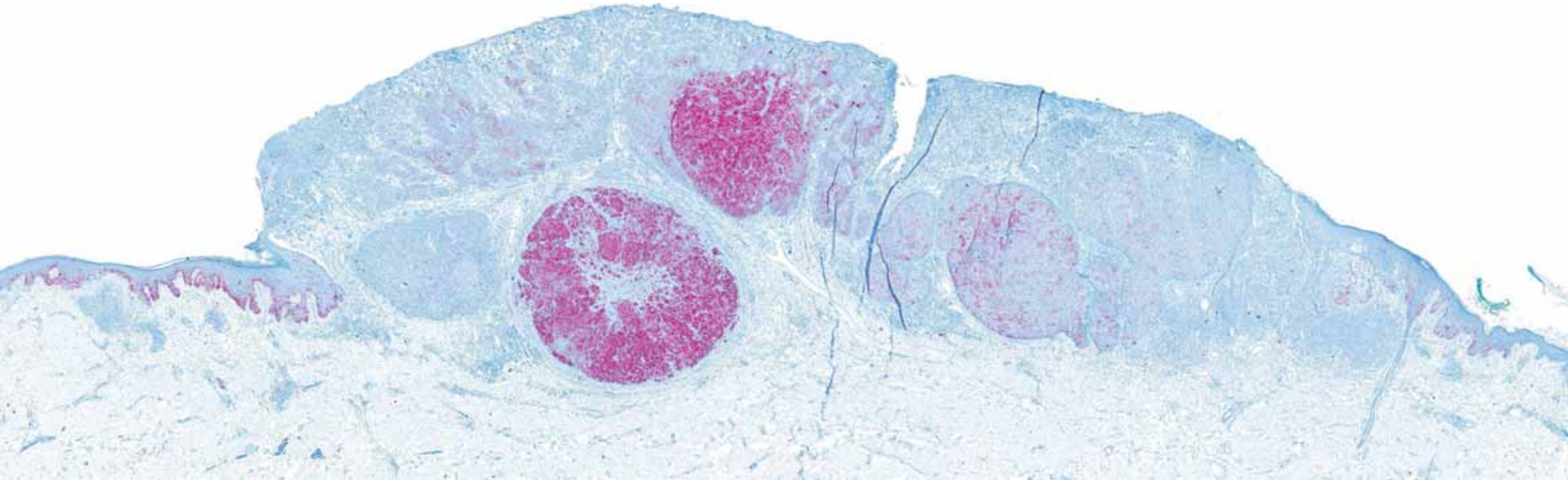


High grade WNT activated melanocytoma (ex-atypical DPN)

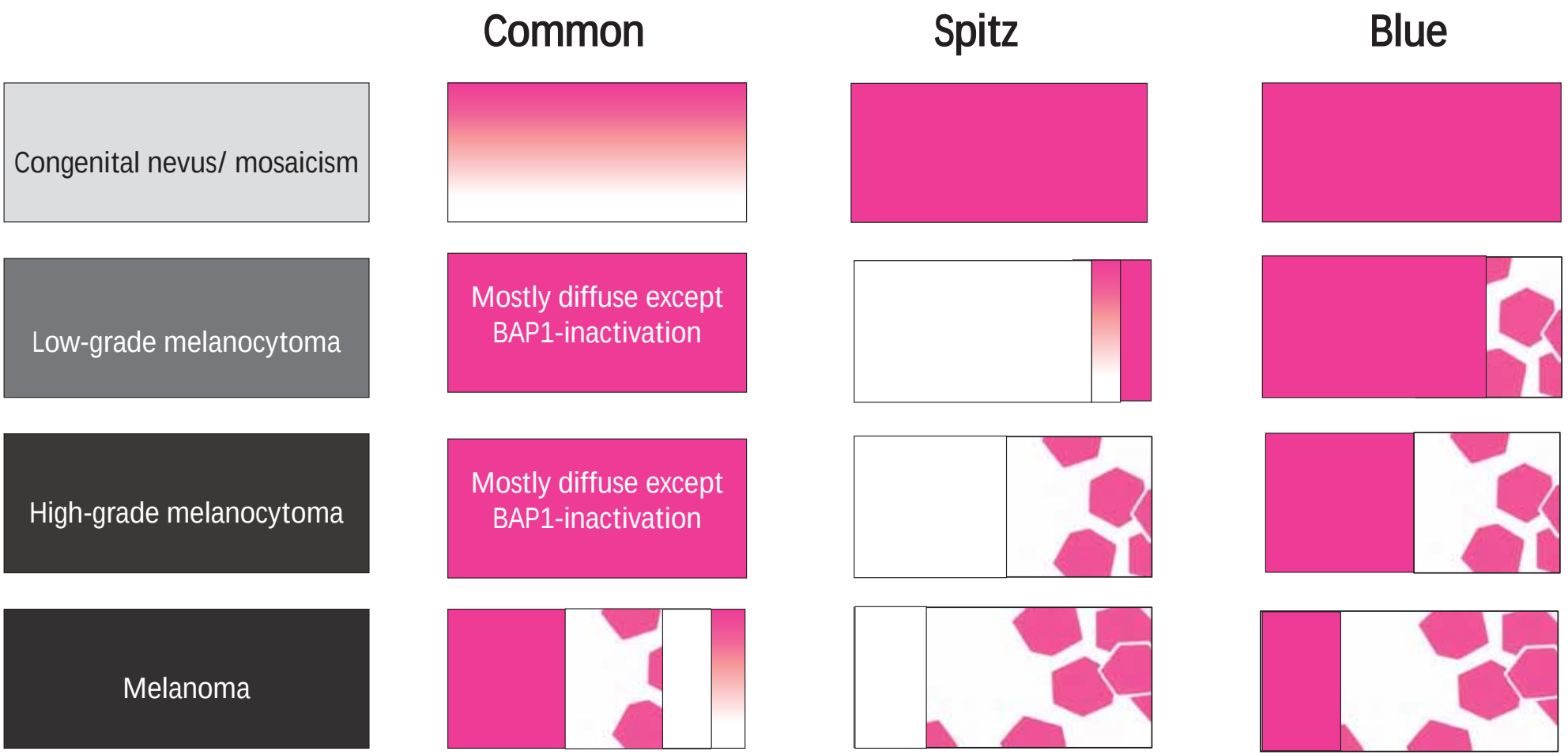
HMB45 heterogeneous



SSM melanoma
HMB45 heterogeneous profile

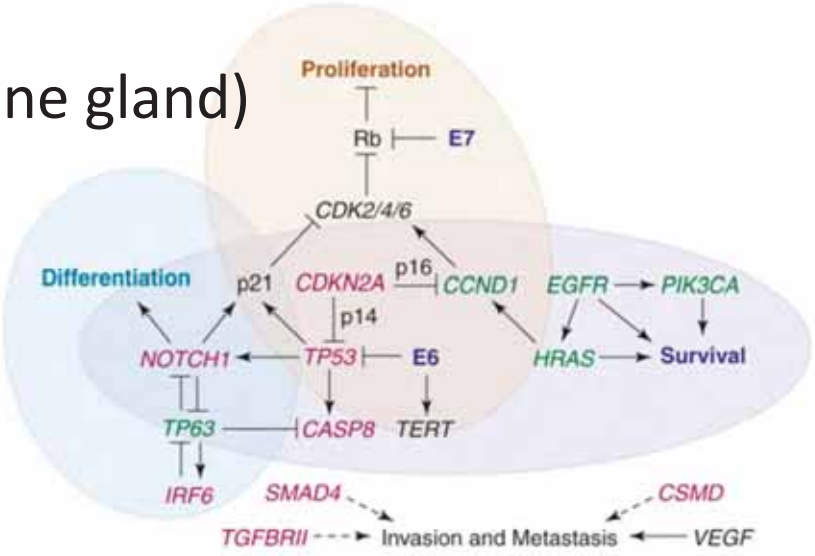


HMB45 synoptic IHC expression chart

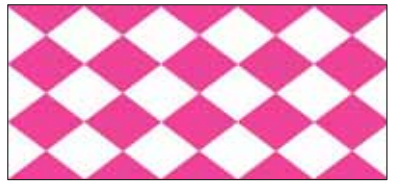


P16

- Coded by *CDKN2A* (Cyclin Dependent Kinase inhibitor 2A) Chr 9p21
- Tumor suppressor gene
- Heterogeneous stain in normal melanocytes
- Cytoplasmic and/or nuclear stain
- Sweat duct positive control (pars recta eccrine gland)

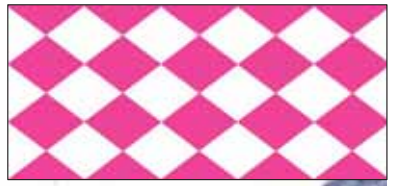


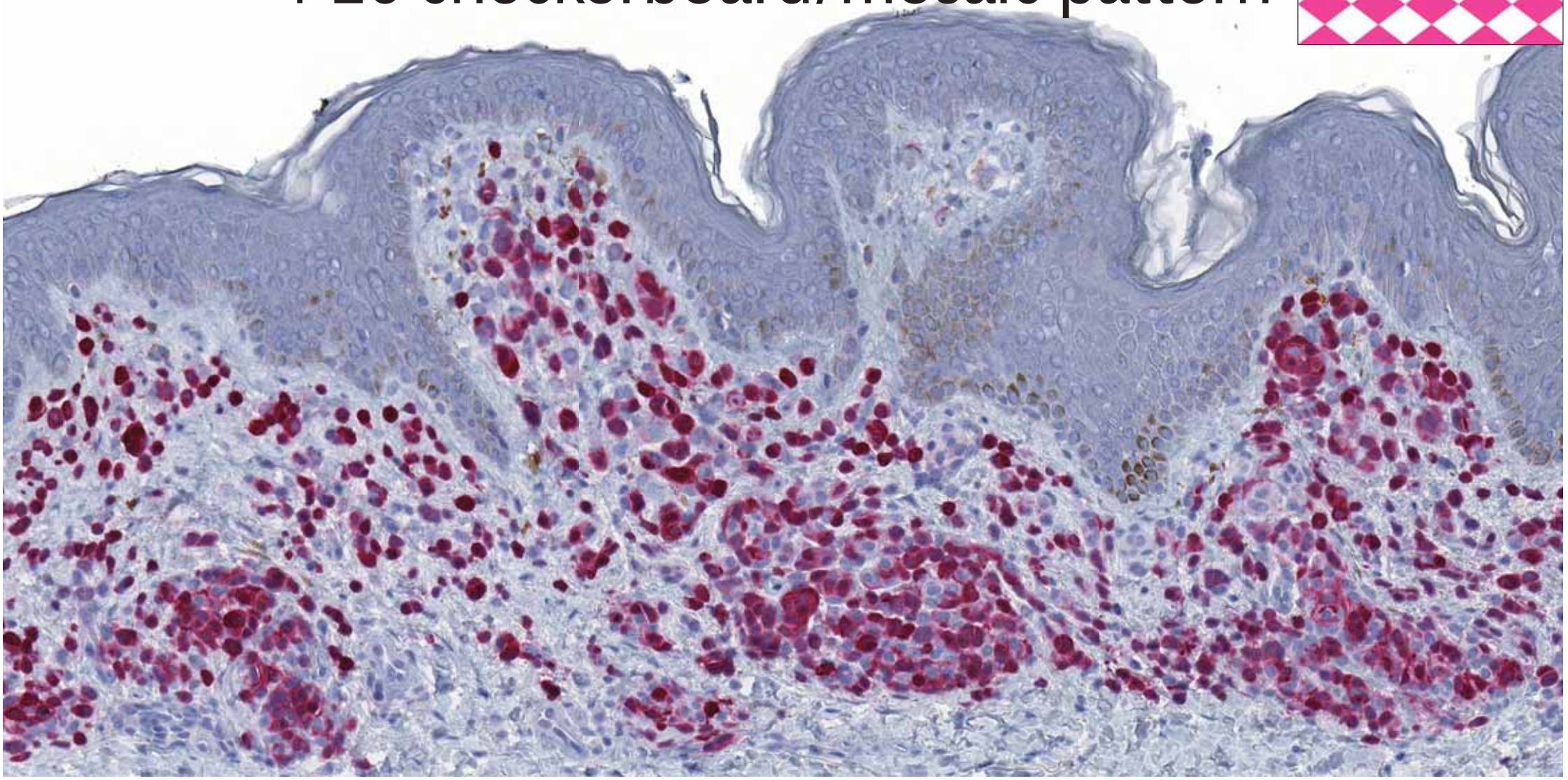
p16 staining patterns (useful even in thin lesions)



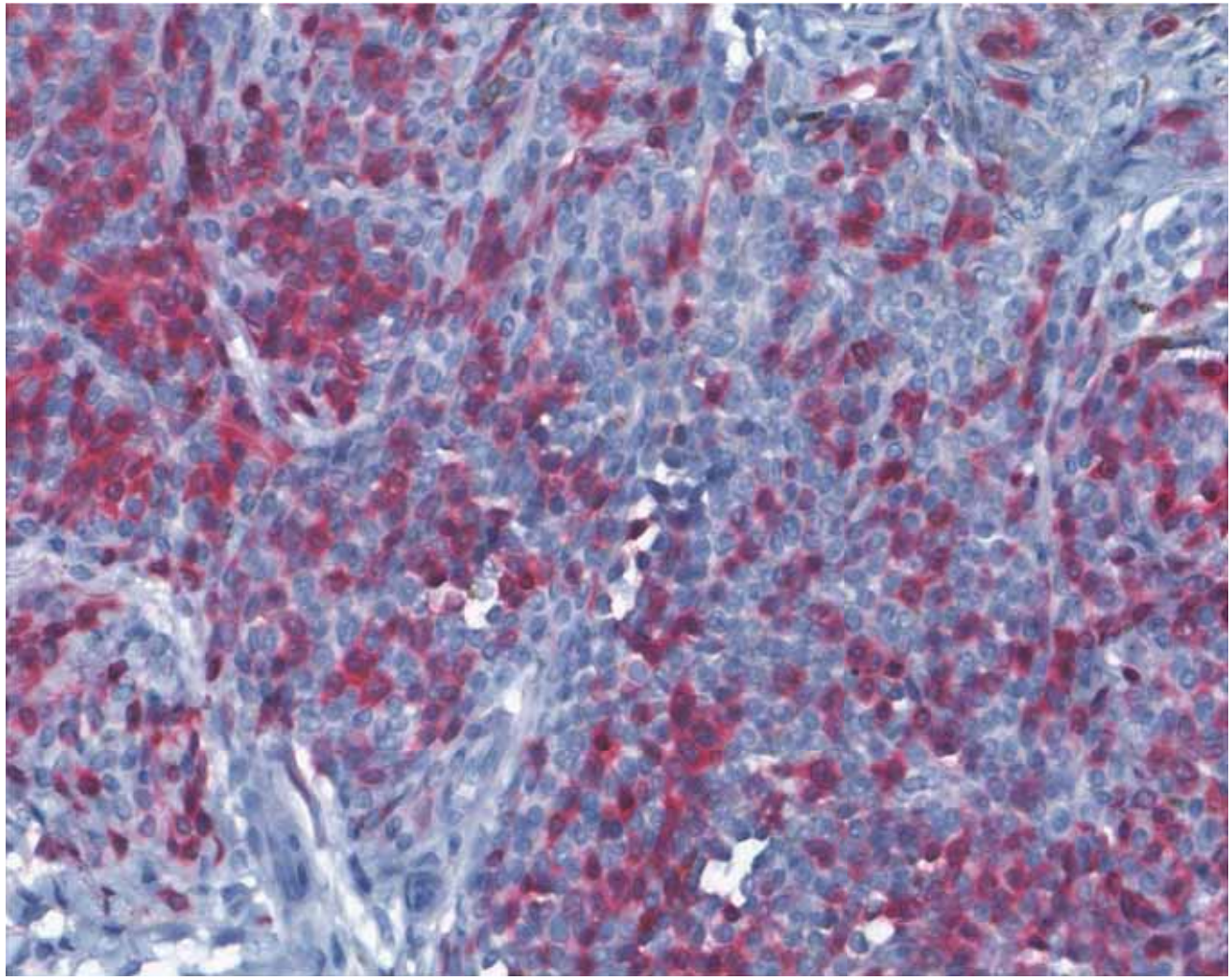
- Checkerboard stain

P16 checkerboard/mosaic pattern





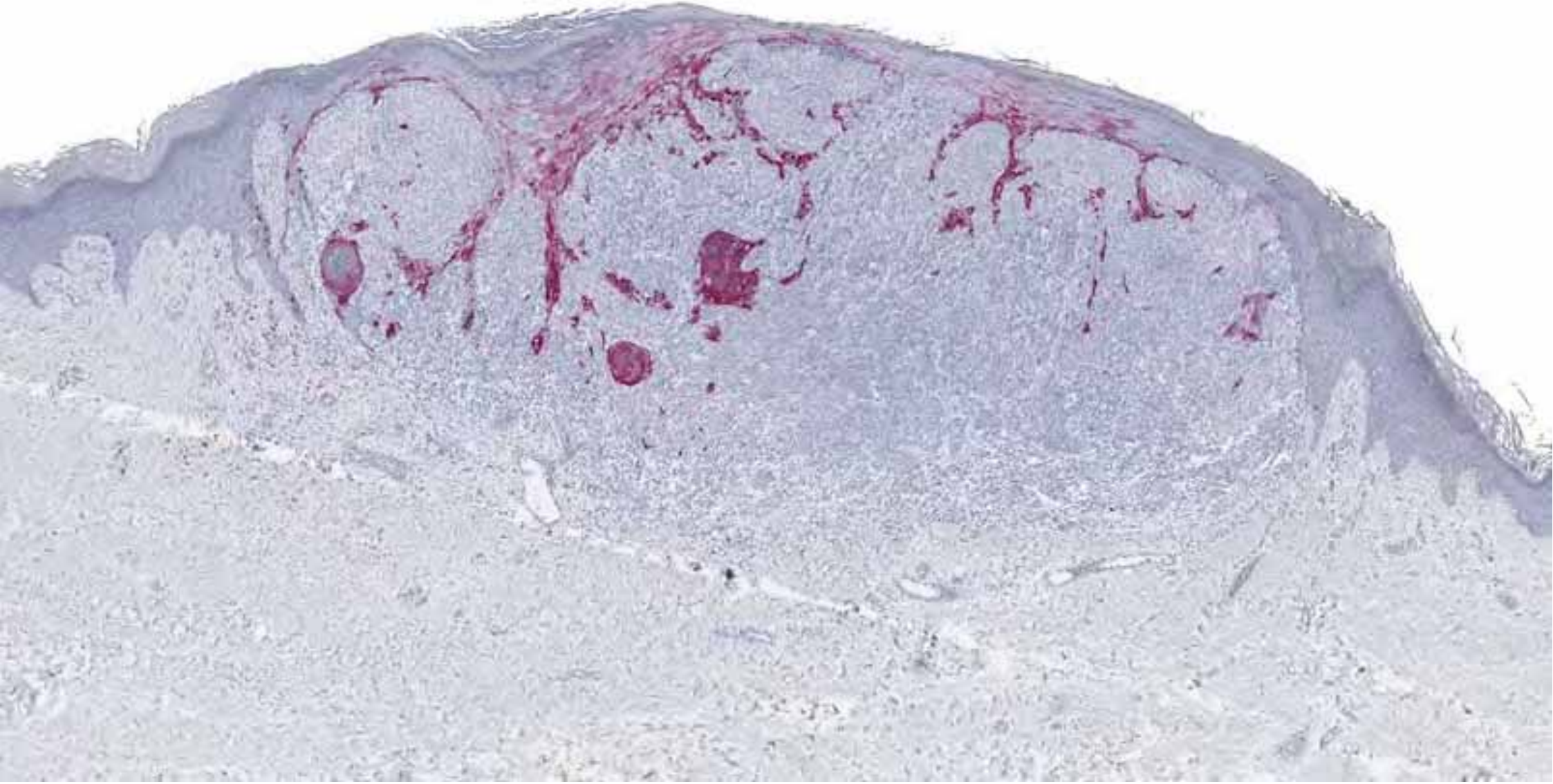
P16 checkerboard/mosaic pattern



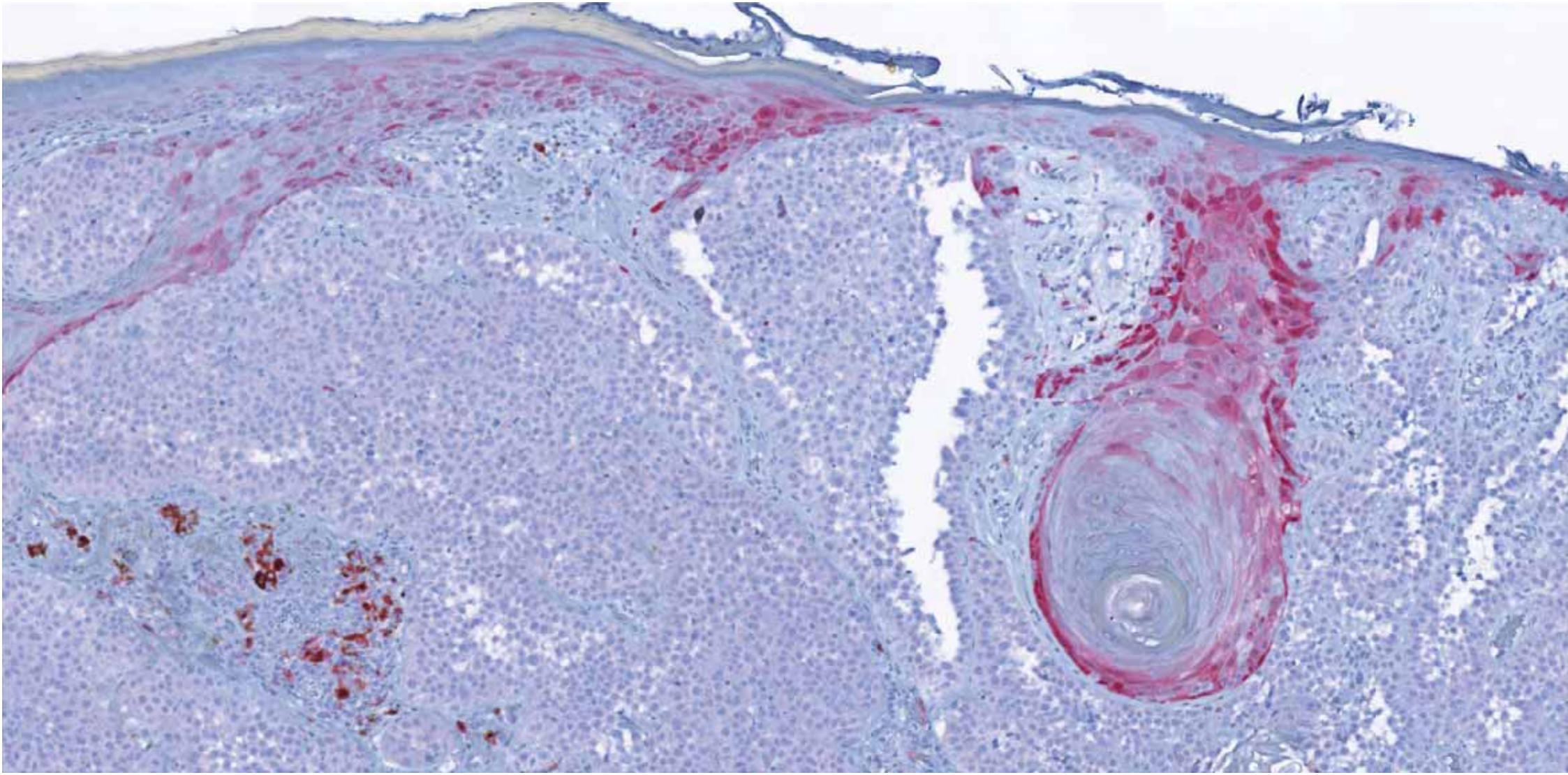
p16 staining patterns (useful even in thin lesions)

- Checkerboard stain
- Complete loss: check internal controls

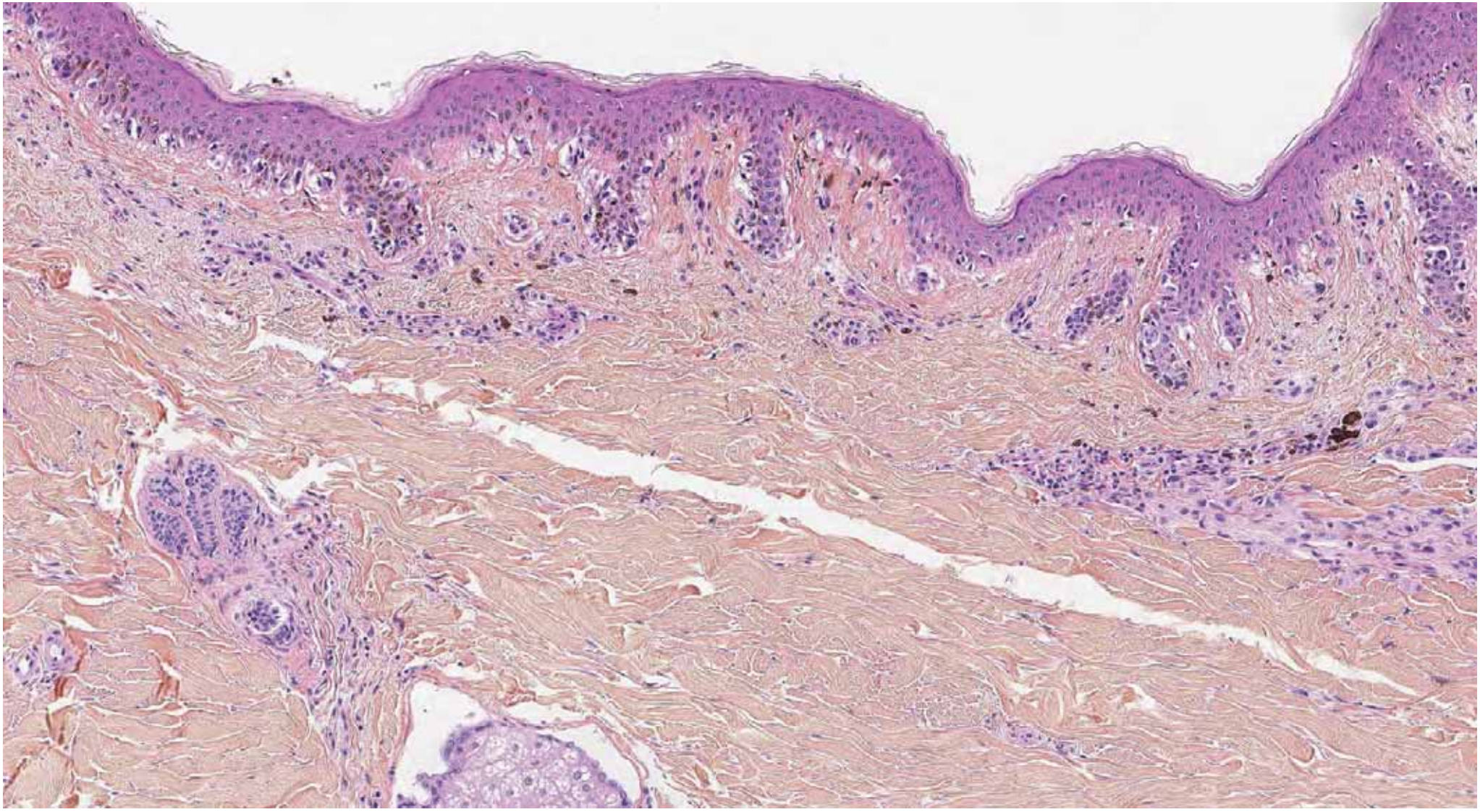
p16 complete loss

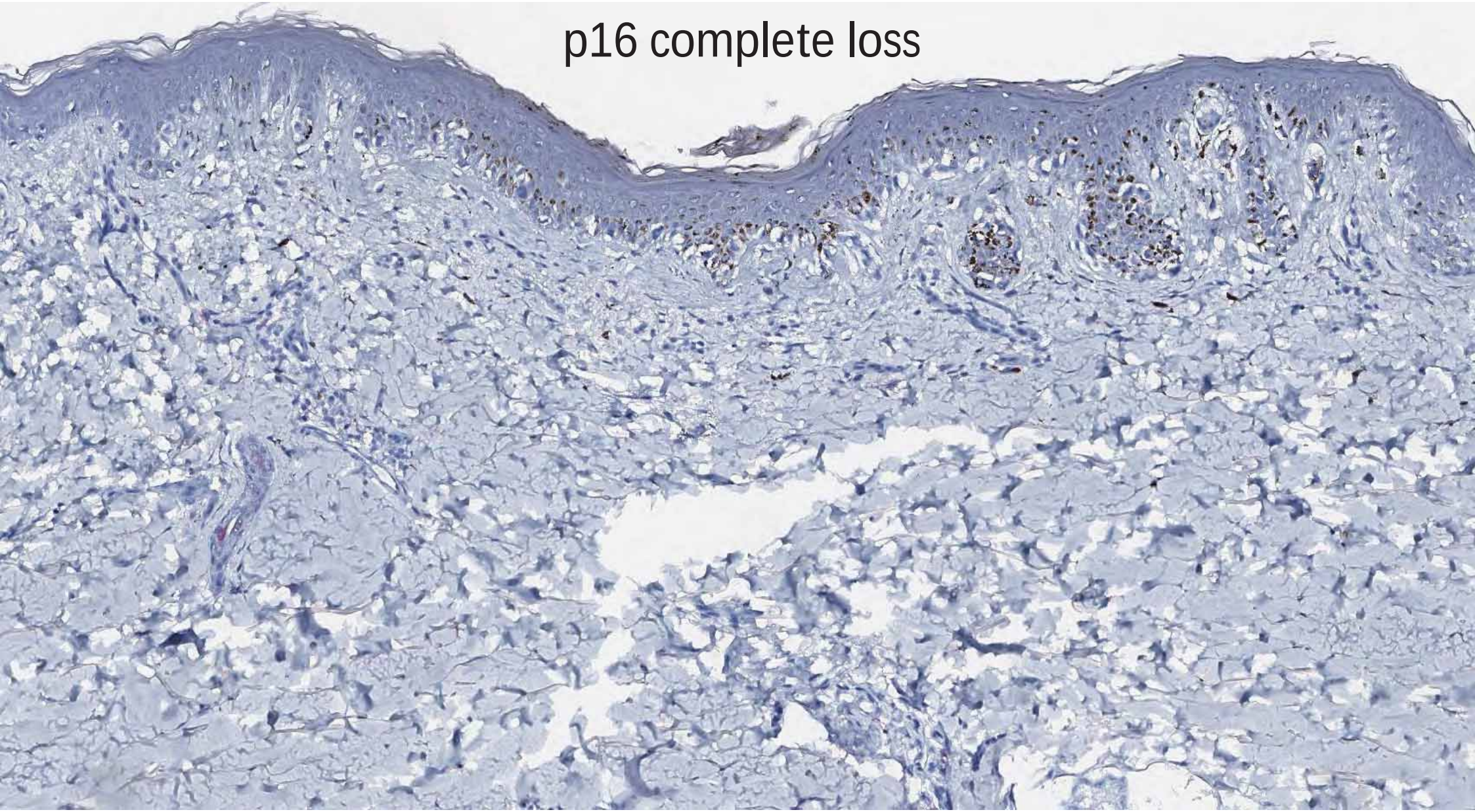


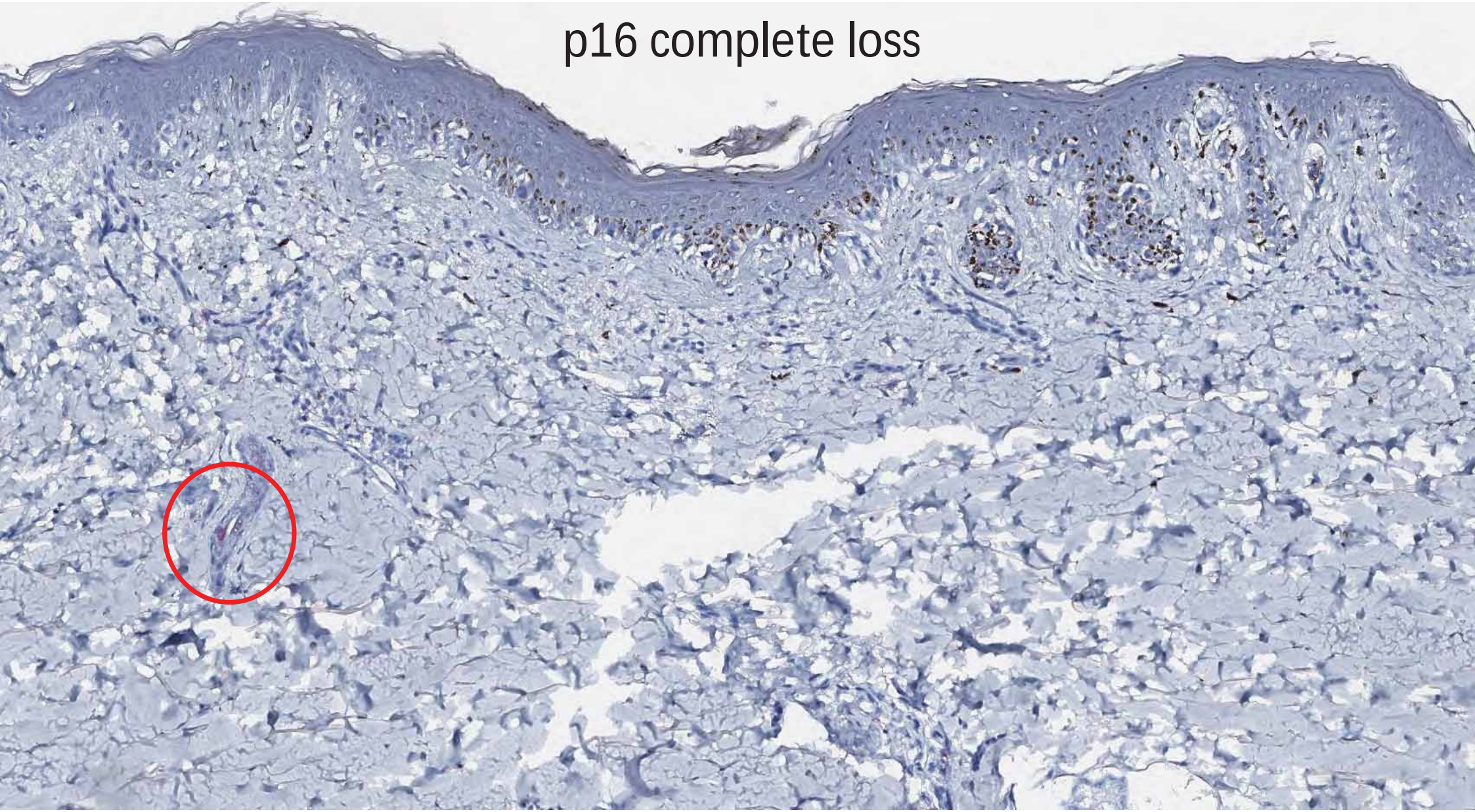
P16: Remodeling of the epidermis



Thin lesions




p16 complete loss

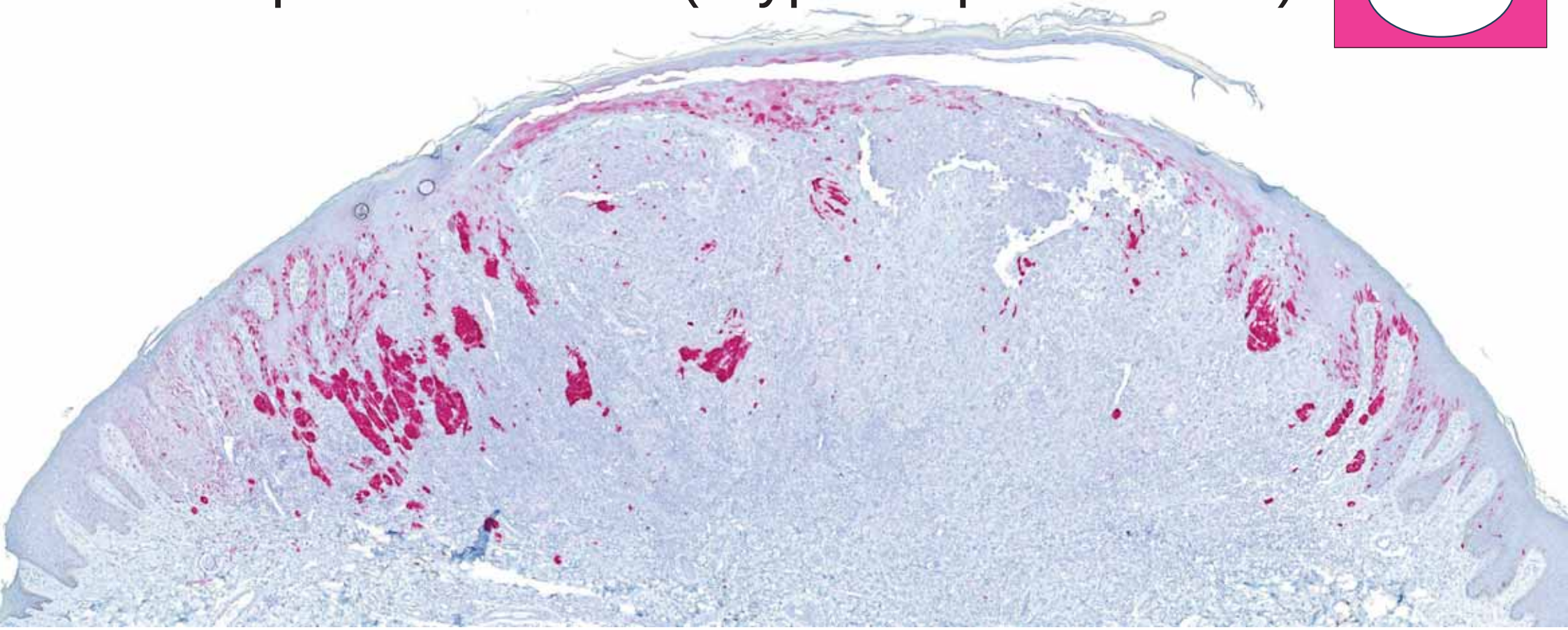

p16 complete loss

p16 staining patterns (useful even in thin lesions)

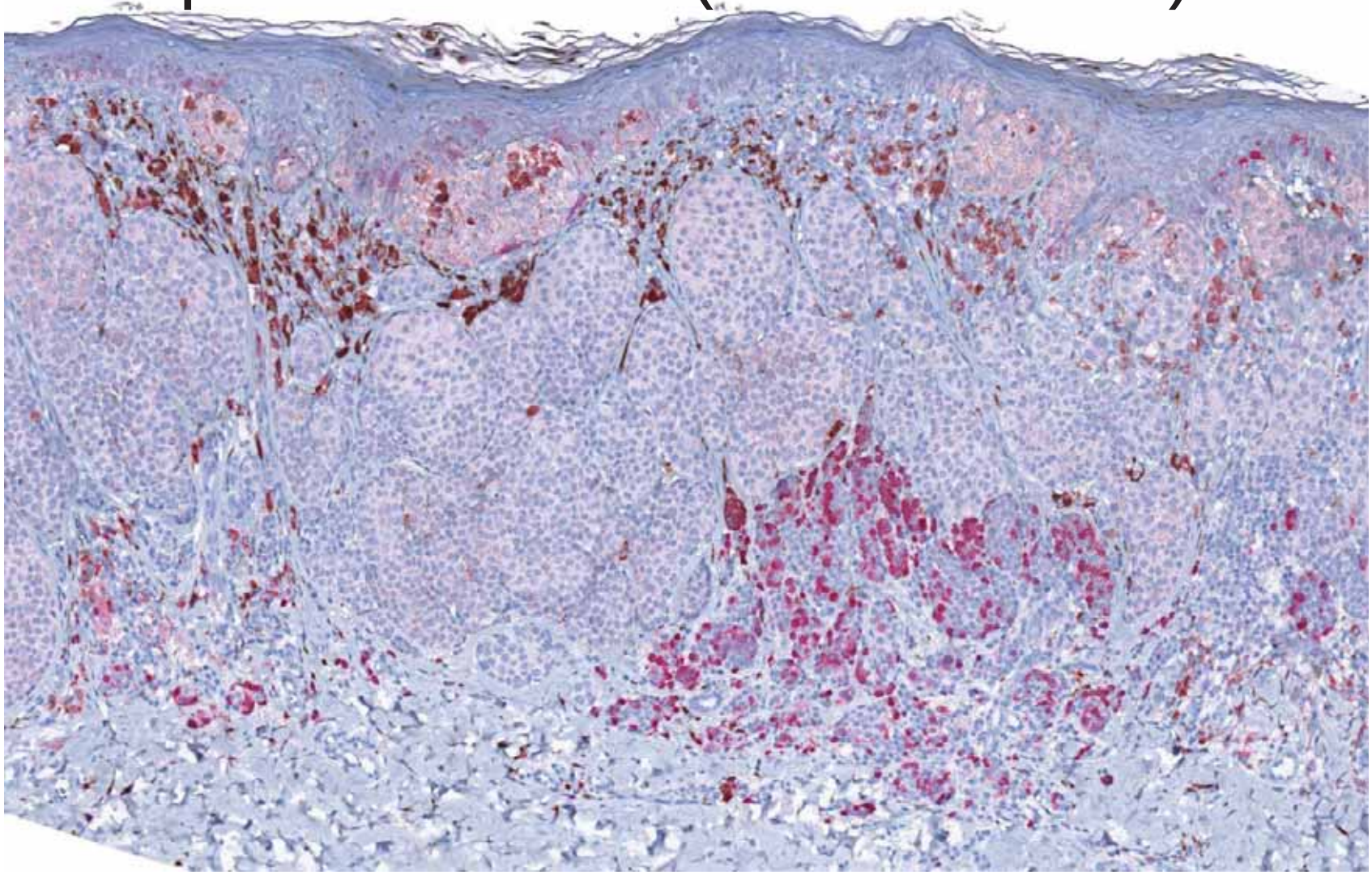
- Checkerboard stain
- Complete loss
- Clonal loss

Only an area of the tumor has lost p16 staining

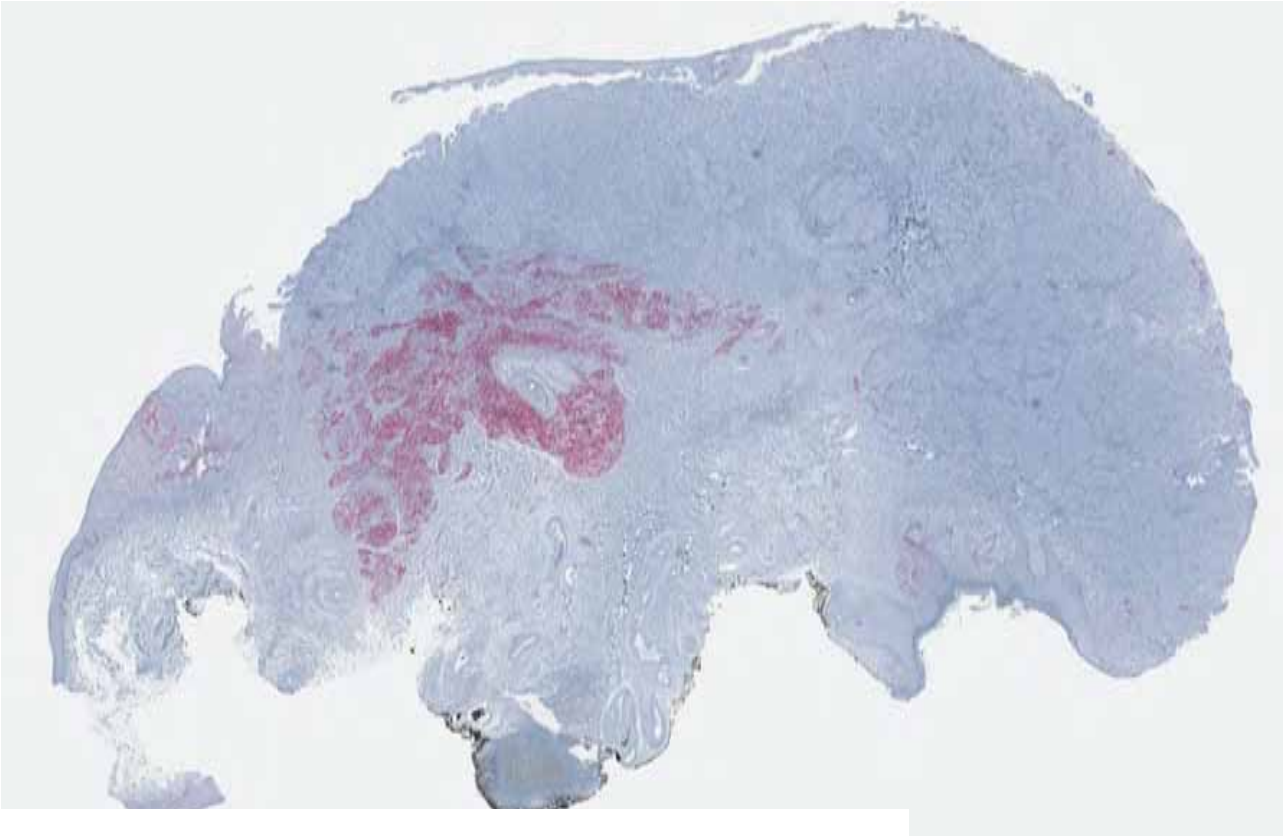
p16 clonal loss (Atypical Spitz Tumour)



p16 clonal loss (SSM ex nevus)



p16 clonal loss (SSM ex-nevus)
Can help Breslow assessment

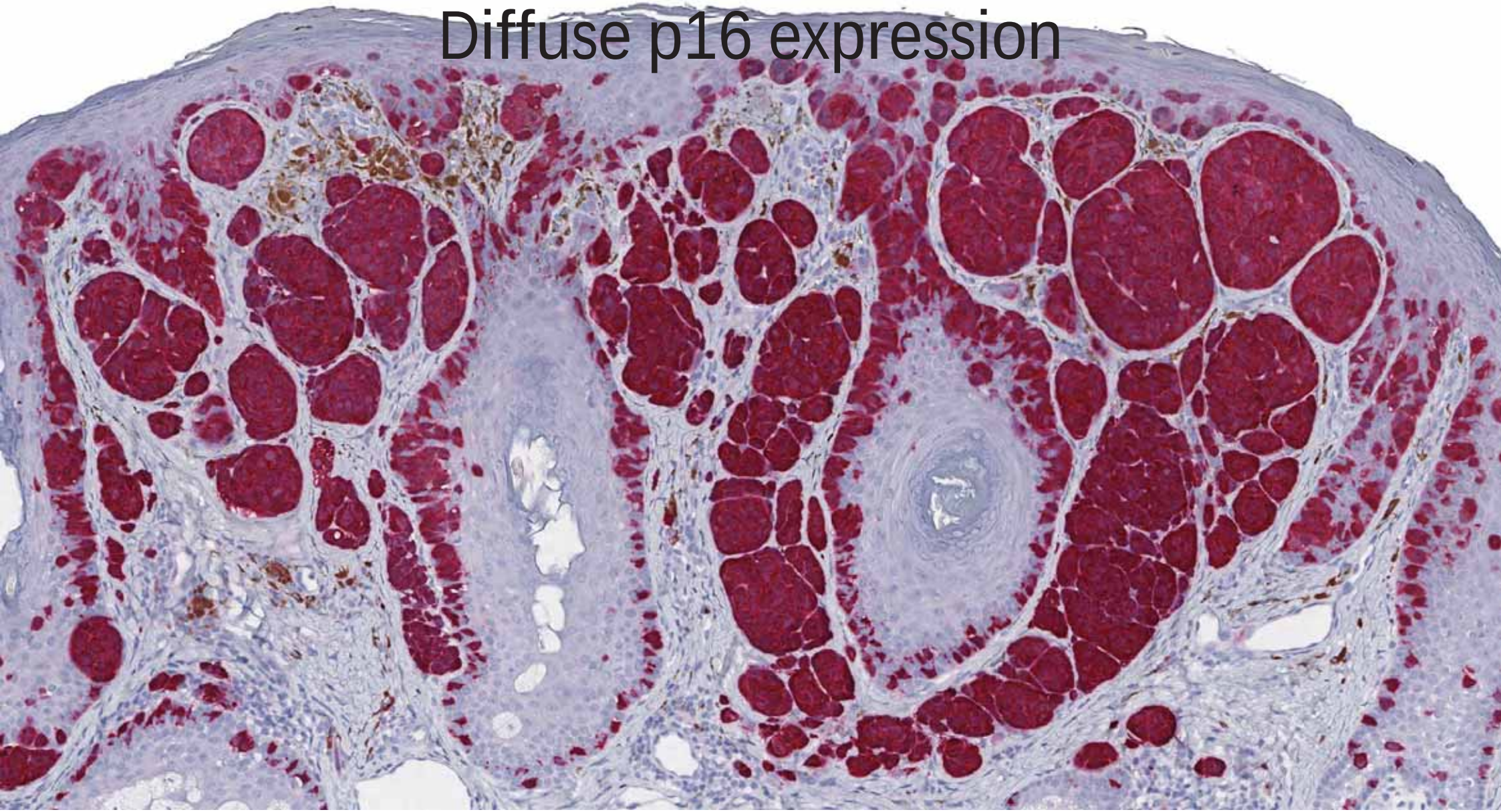


p16 clonal loss (Blue melanoma)

p16 patterns suggesting malignancy (useful even in thin lesions)

- Checkerboard stain
- Absence of staining (check internal controls)
- Clonal loss
- Melanoma ex-nevus staining pattern

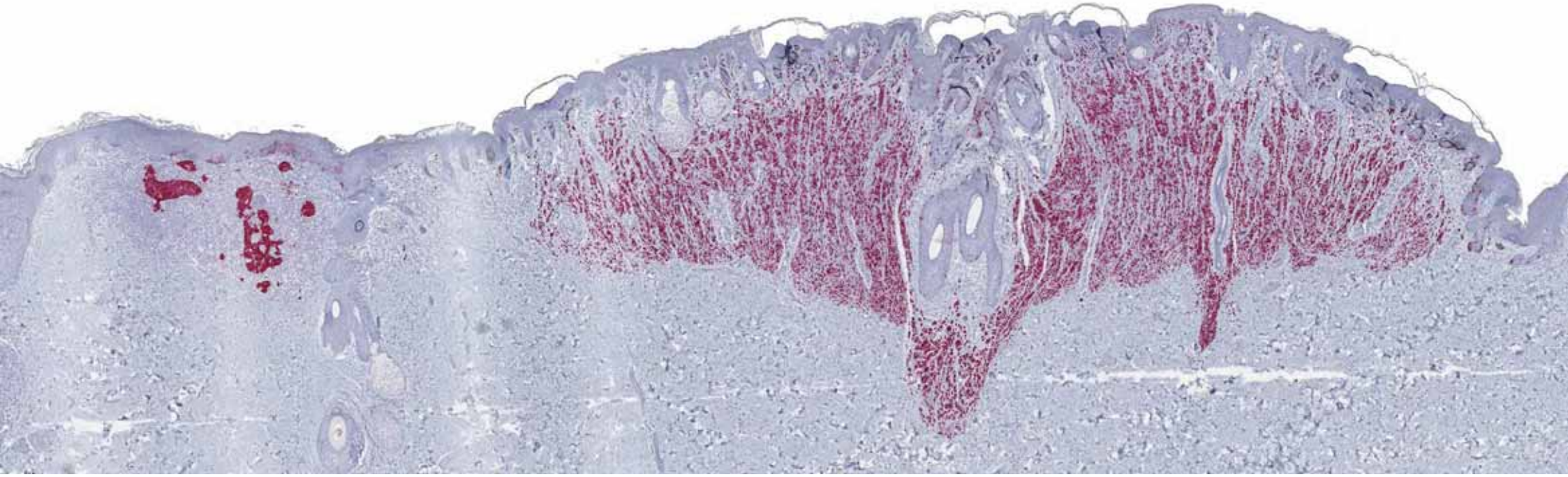
Diffuse p16 expression



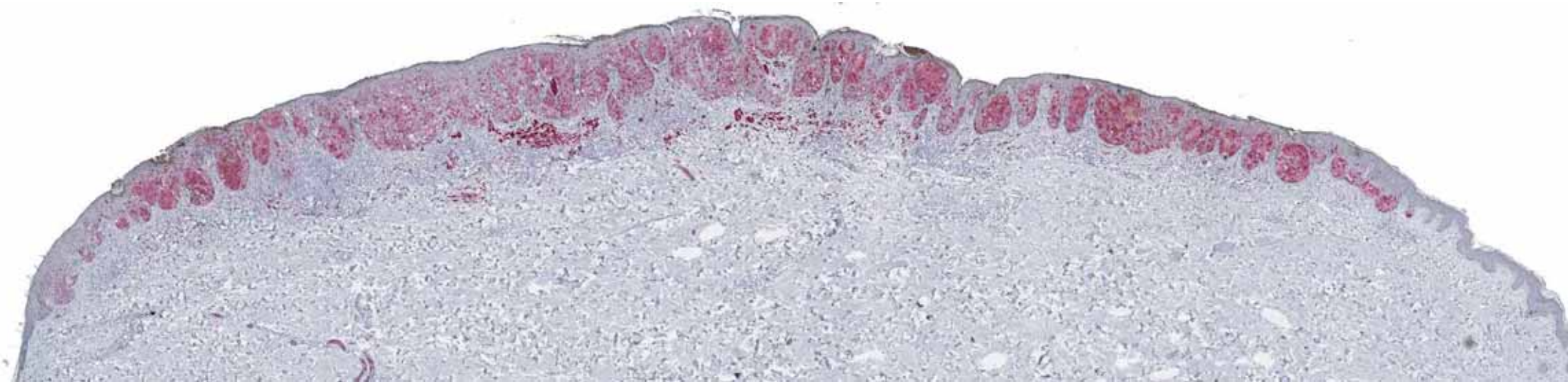
p16 staining patterns
unknown benign/malignant signification

- Checkerboard stain
- Absence of staining
- Clonal loss
- Diffuse positivity
- Melanoma ex-nevus staining pattern
- Different pattern in melanoma and nevus areas

Melanoma ex-nevus staining pattern

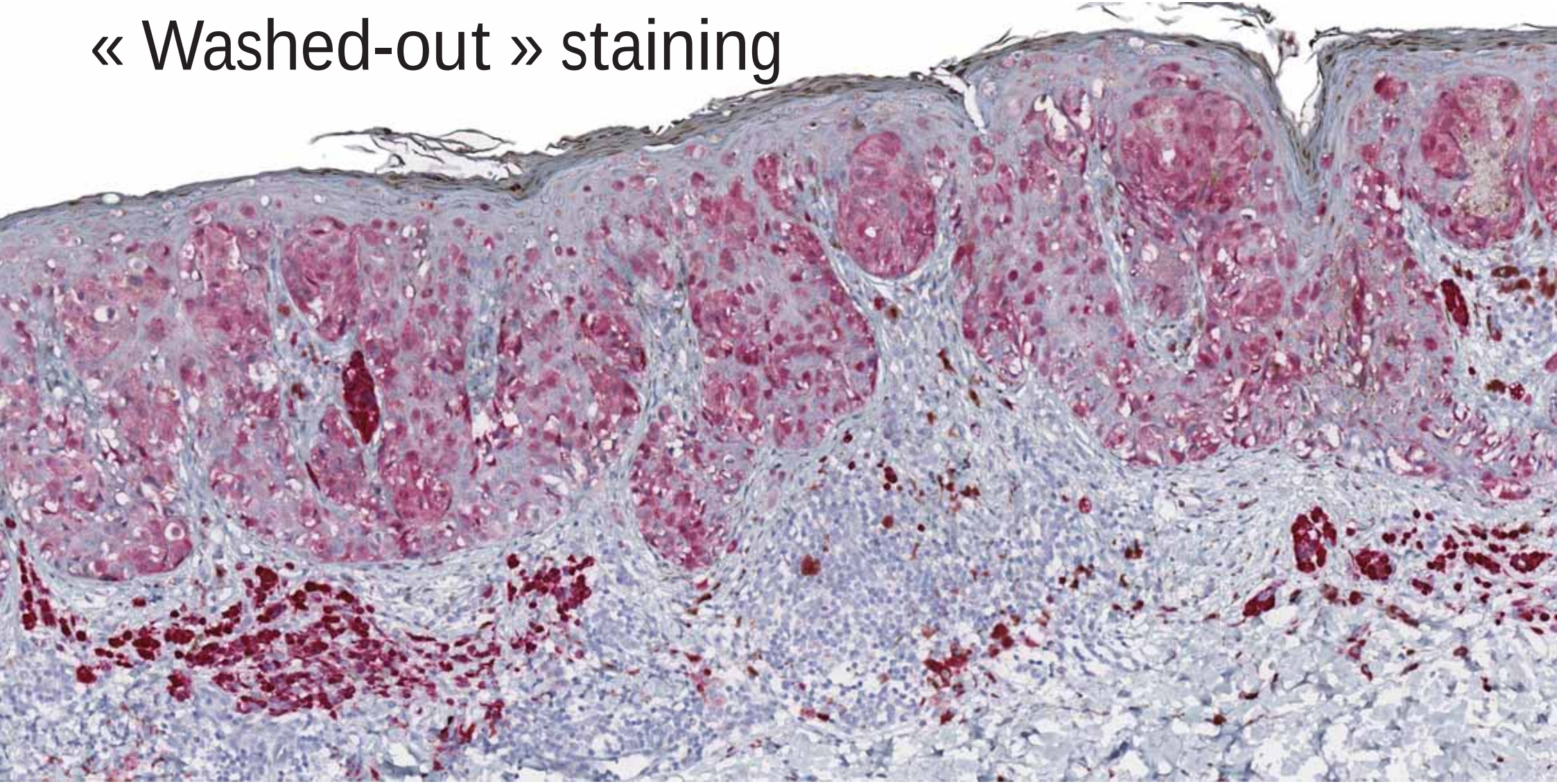


Melanoma ex-nevus staining pattern



Melanoma ex-nevus staining pattern

« Washed-out » staining



p16 staining patterns (useful even in thin lesions)

- Diffuse positivity
- Checkerboard stain
- Absence of staining
- Clonal loss
- Melanoma ex-nevus staining pattern

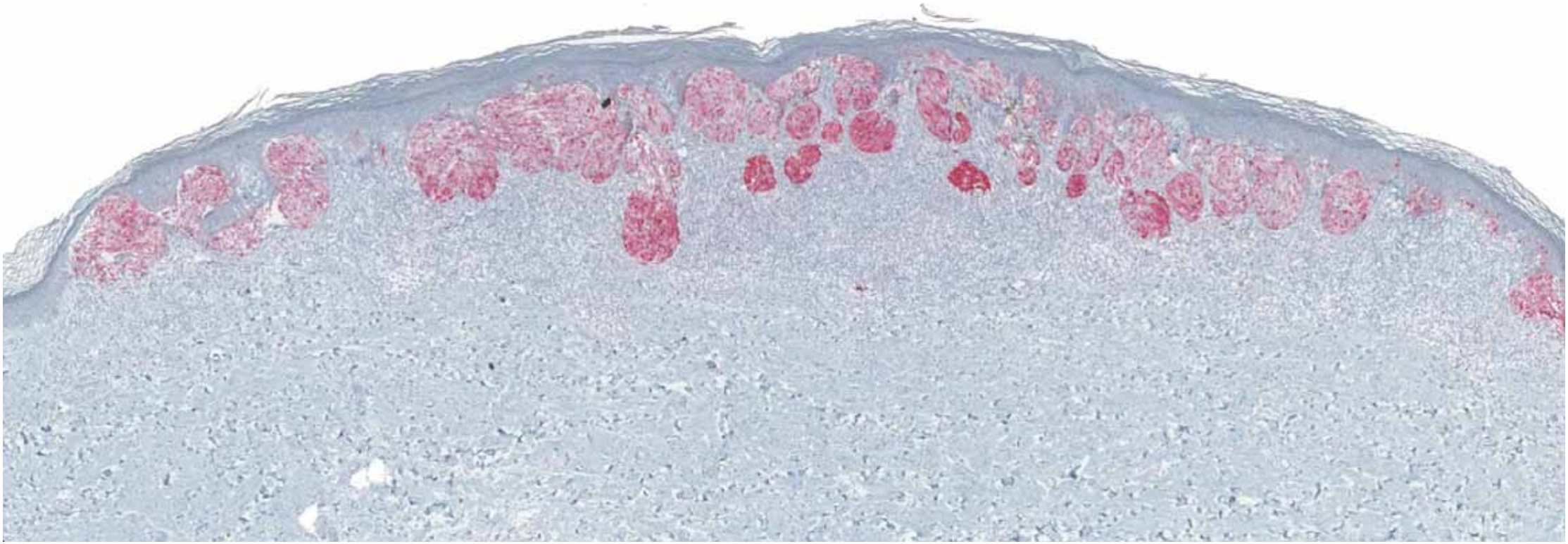
Different pattern in melanoma and nevus

p16 staining patterns (useful even in thin lesions)

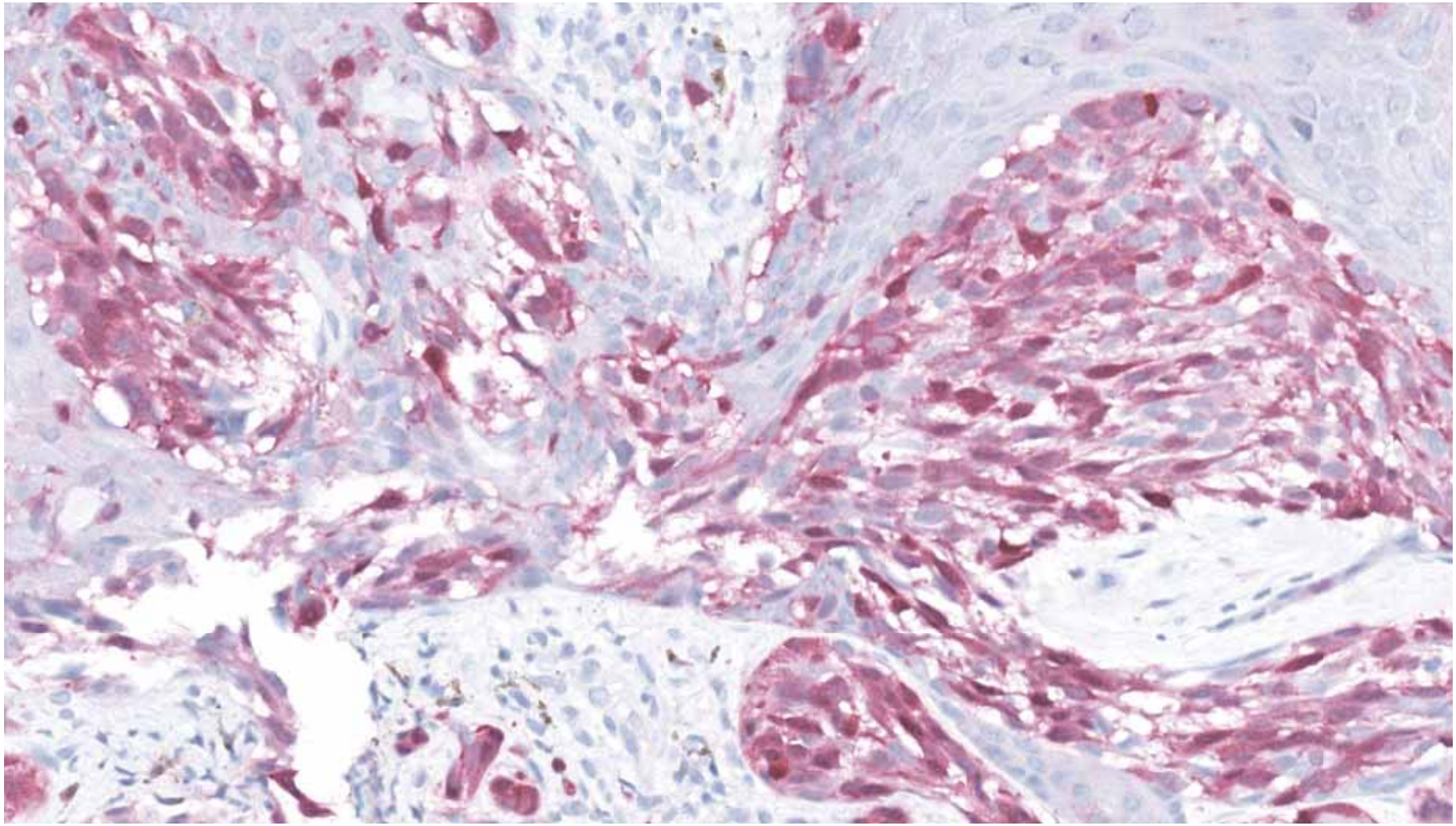
- Checkerboard stain
- Absence of staining
- Clonal loss
- Diffuse positivity
- Melanoma ex-nevus staining pattern
- Inverted gradient

Bottom heavy staining

Inverted /bottom heavy P16 expression



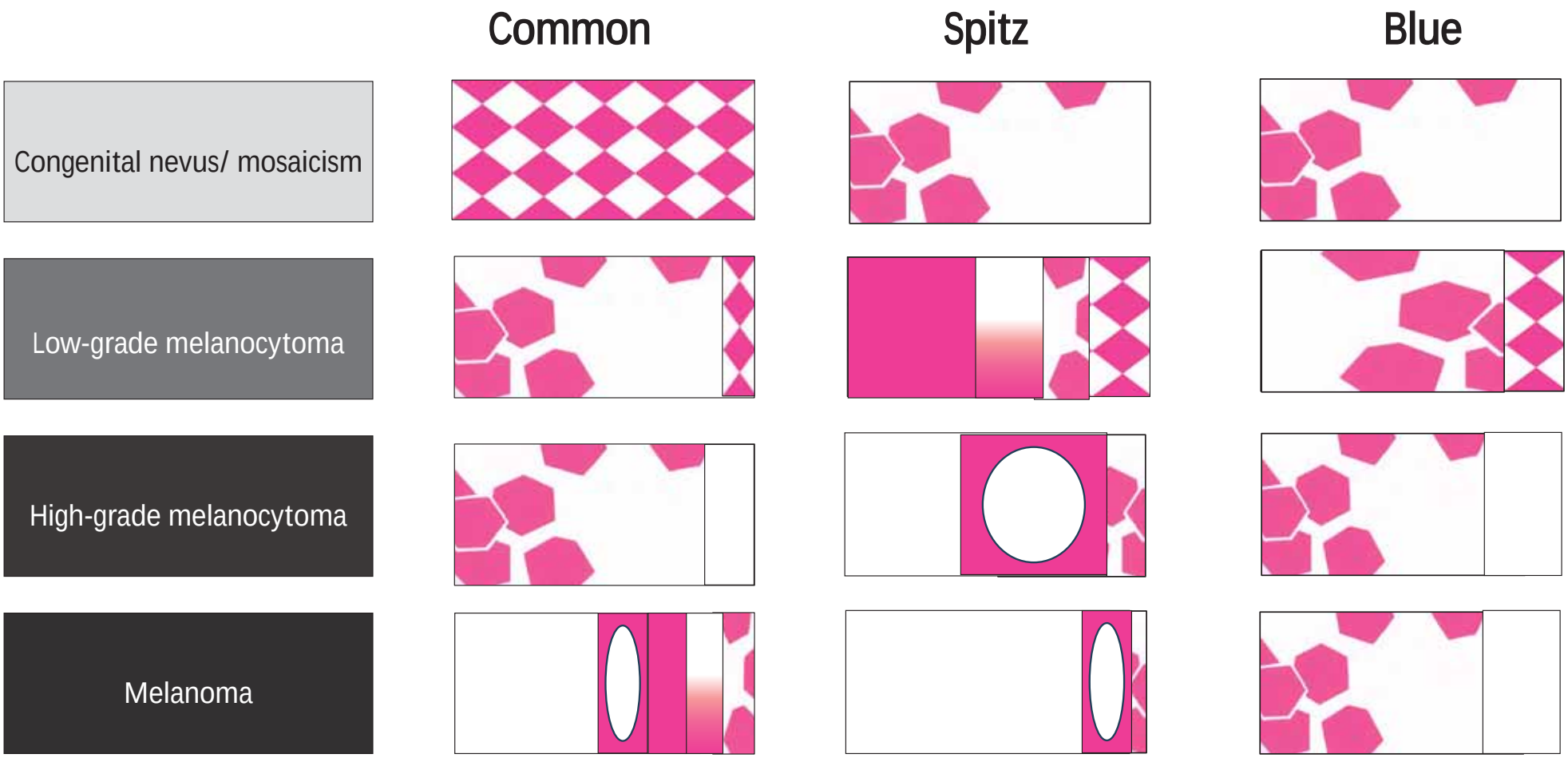
P16 Nuclear positivity



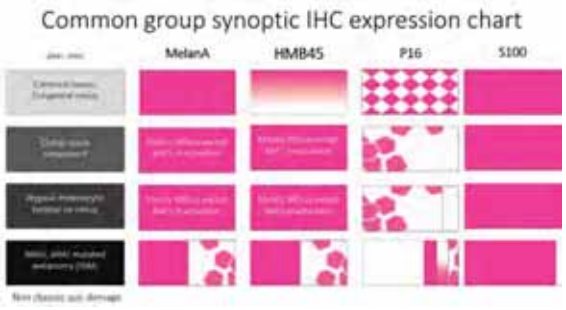
Negative trend

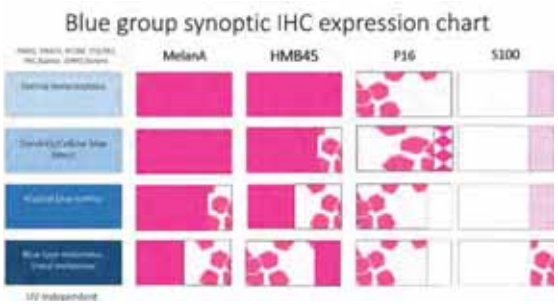
- Swapping out P16 for PRAME is a very bad idea

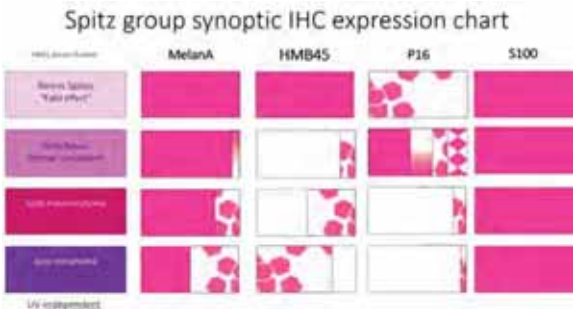
P16 synoptic IHC expression chart



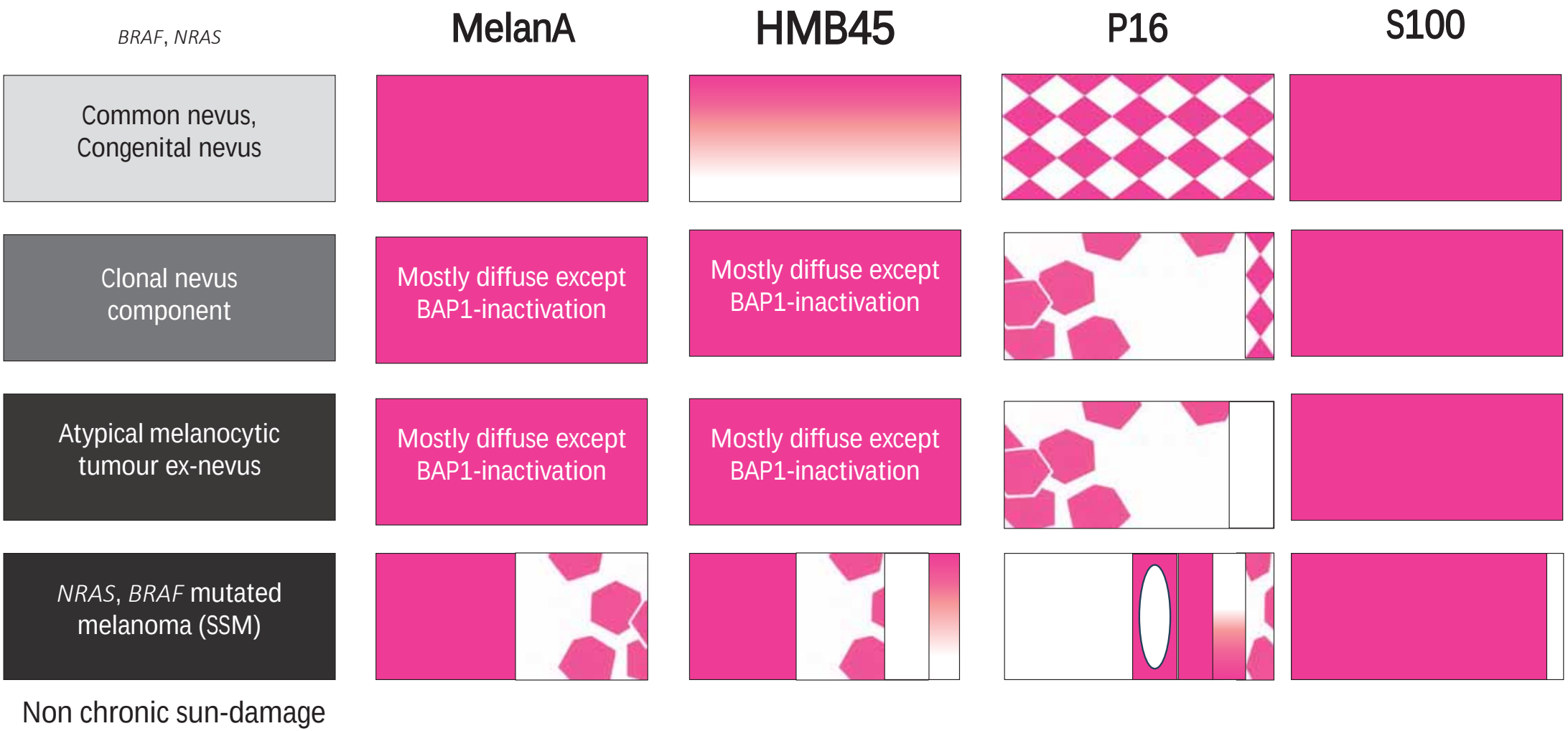
Contextual/combined IHC diagnosis approach





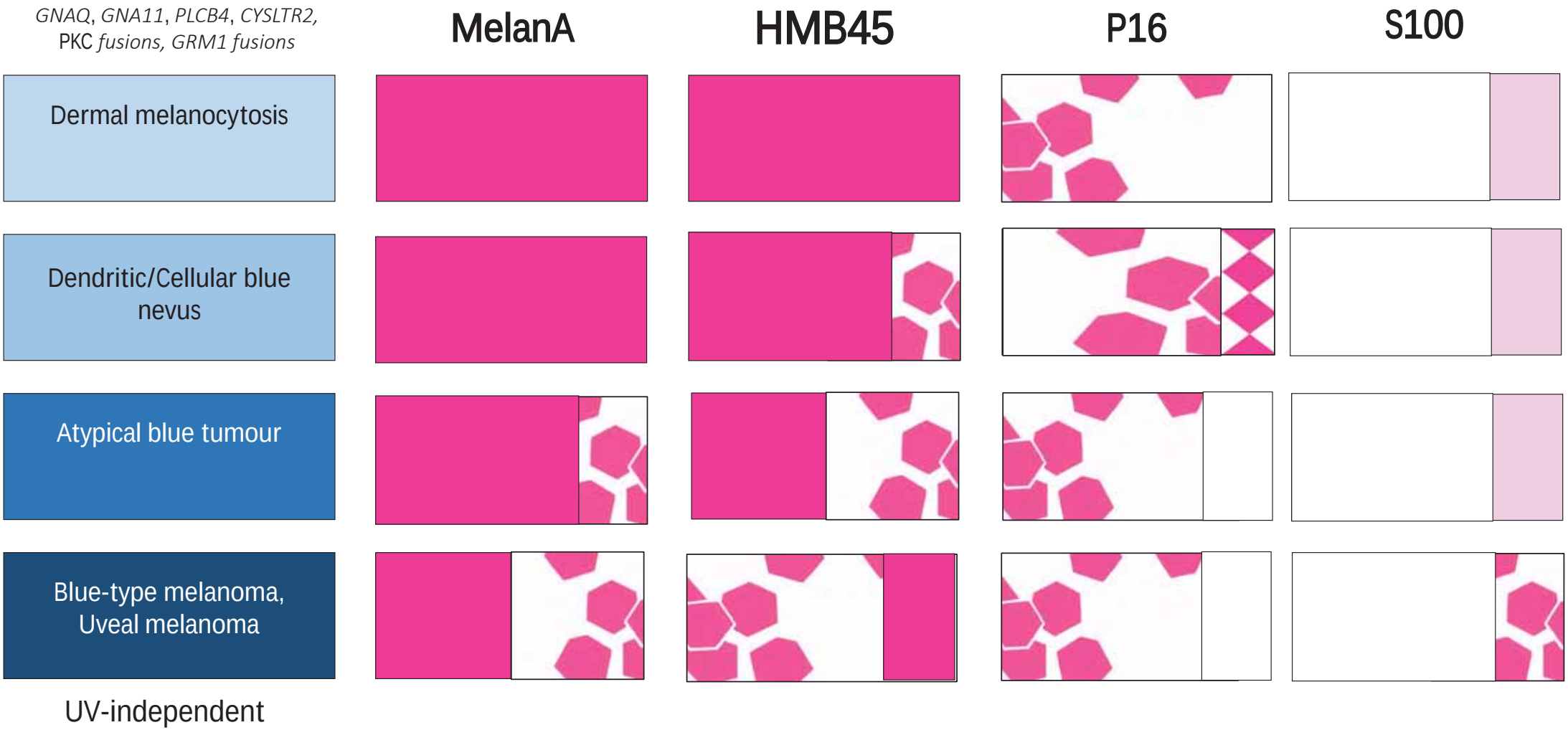


Common group synoptic IHC expression chart

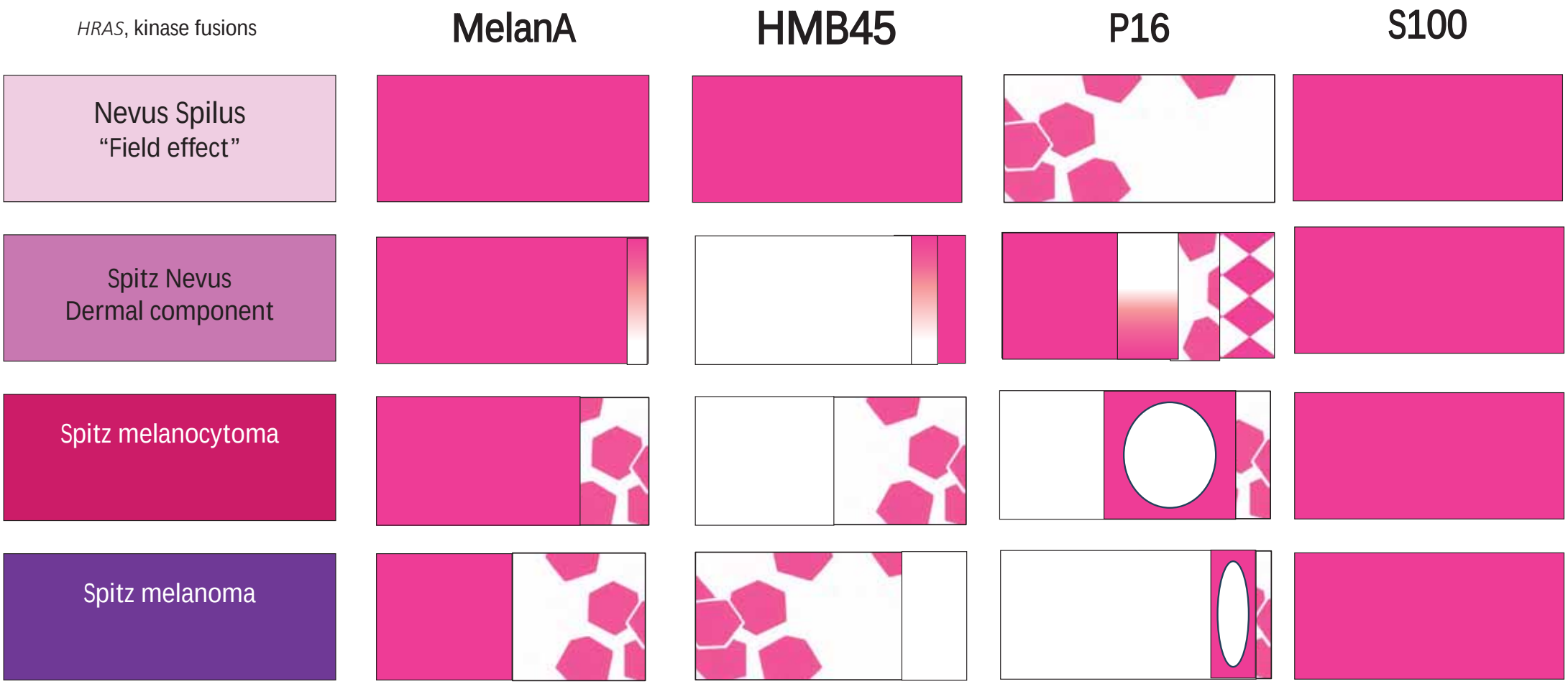


Non chronic sun-damage

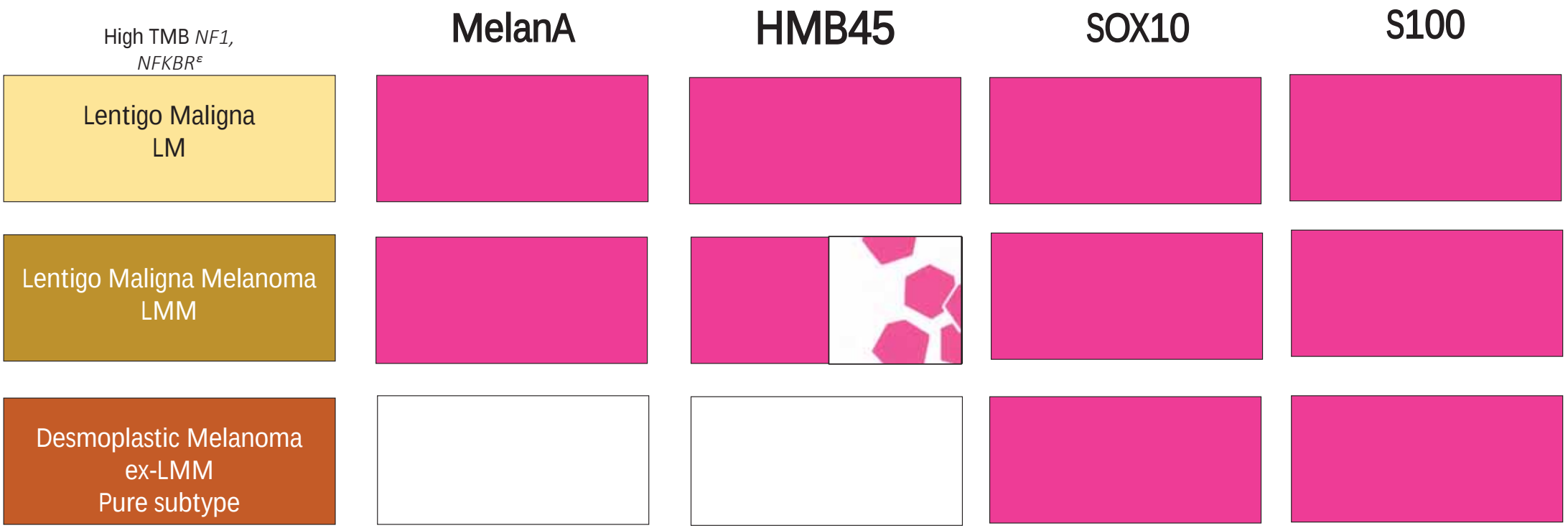
Blue group synoptic IHC expression chart



Spitz group synoptic IHC expression chart



LMM/DM synoptic IHC expression chart



Chronic sun-damage

LMM/DM synoptic IHC expression chart

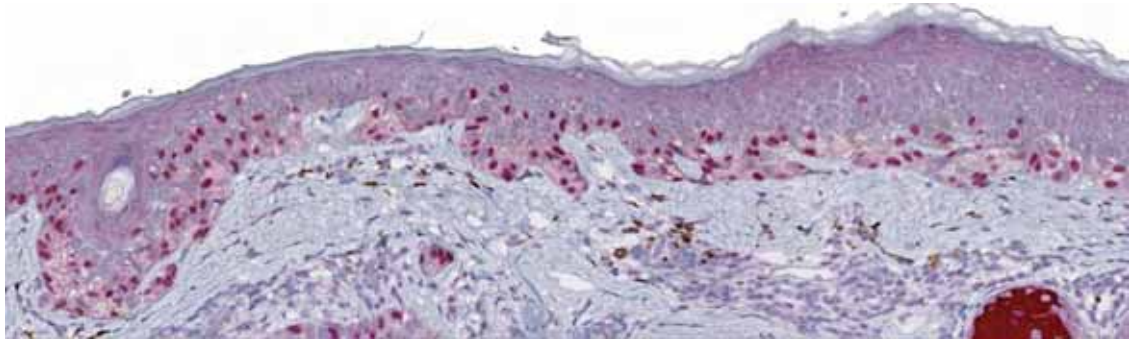
High TMB *NF1, NFKBR*[ε]

PRAME

Lentigo Maligna
LM

Lentigo Maligna Melanoma
LMM

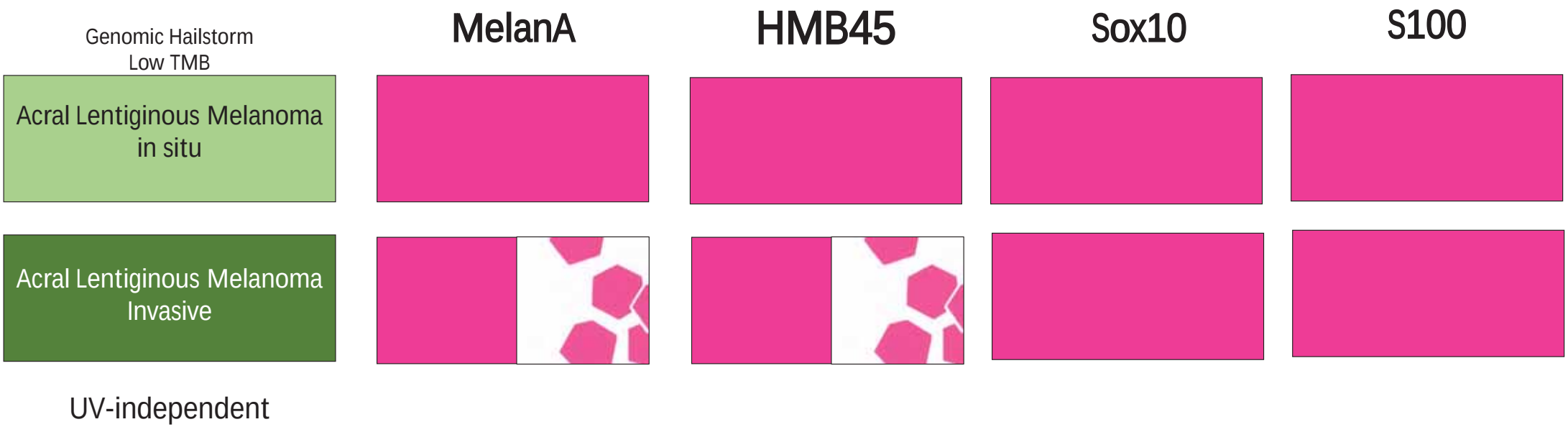
Desmoplastic Melanoma
ex-LMM
Pure subtype



Chronic sun-damage

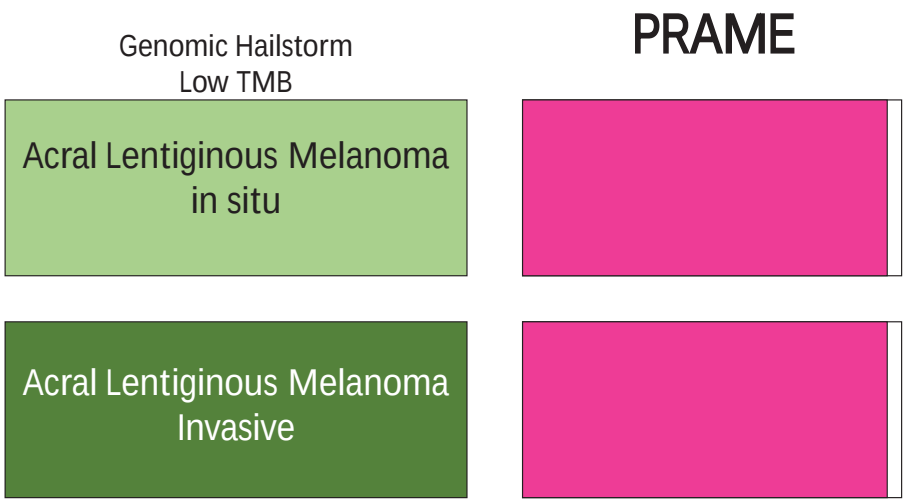
ALM synoptic IHC expression chart

Genomic Hailstorm
Low TMB

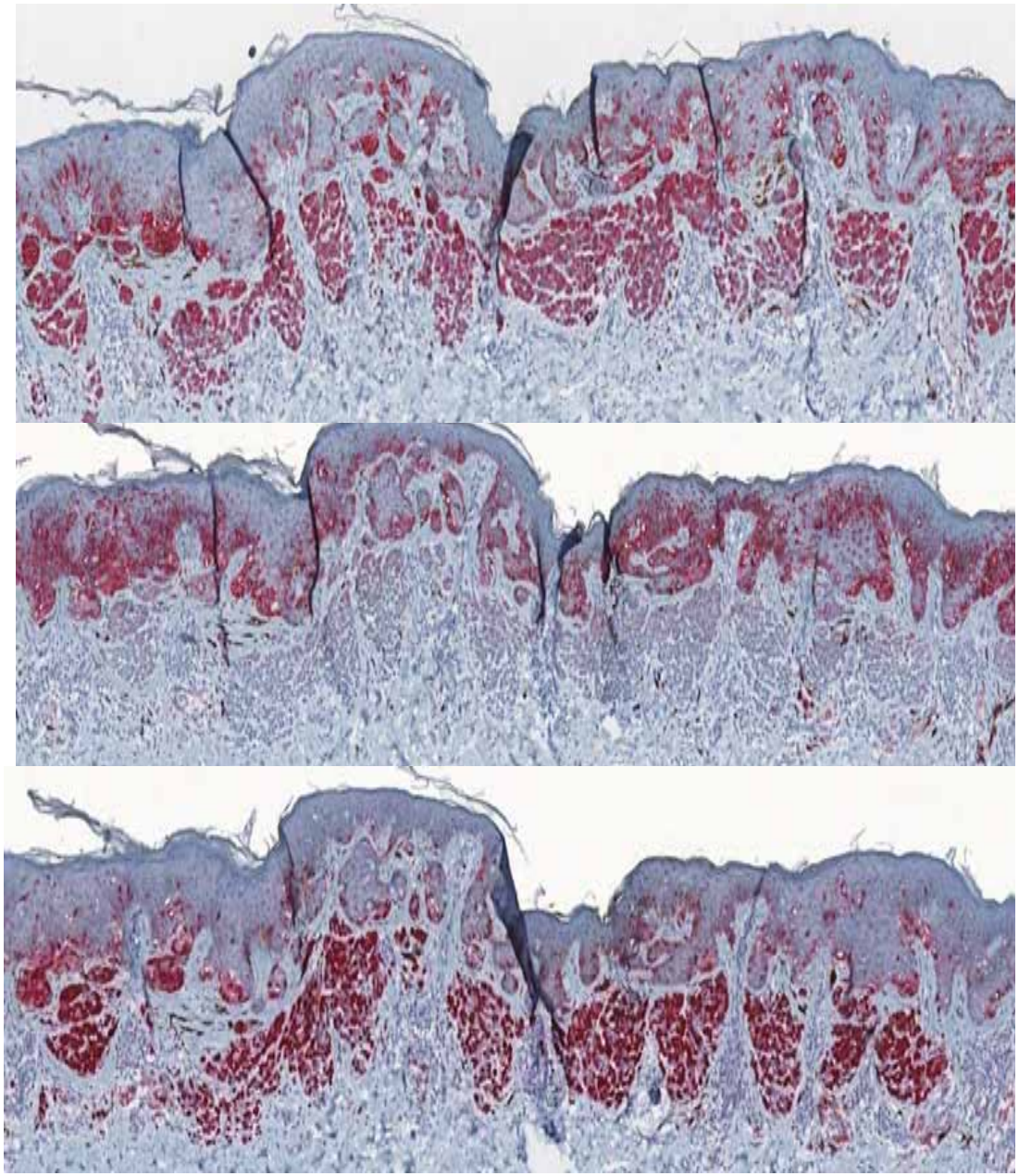


UV-independent

ALM synoptic IHC expression chart



Having a standard immunophenotype in a common compound nevus is almost a failsafe system



MelanA

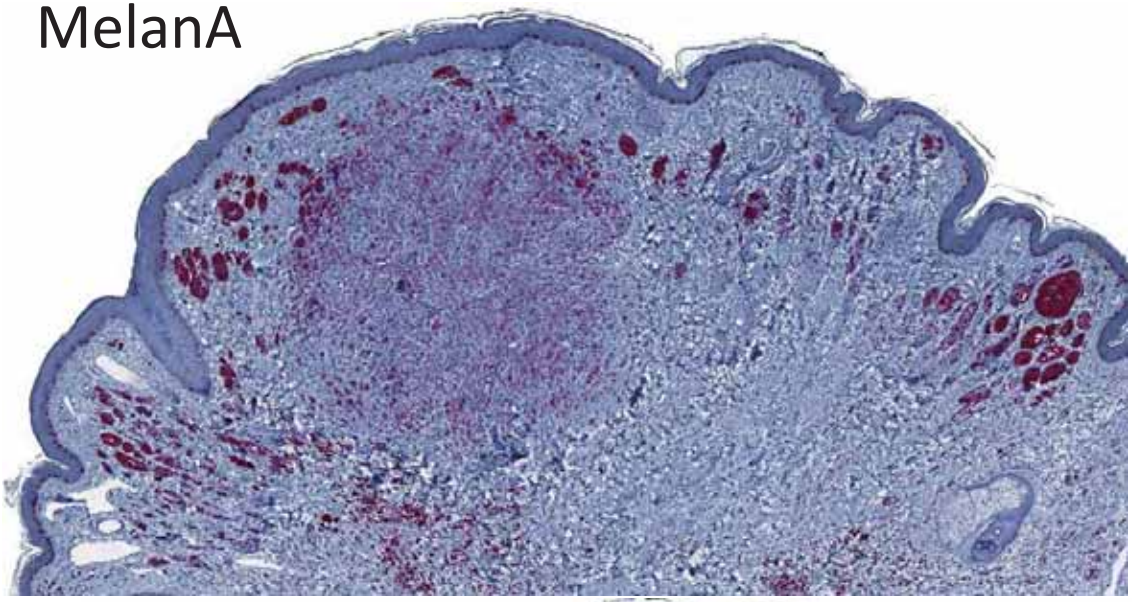
HMB45

p16

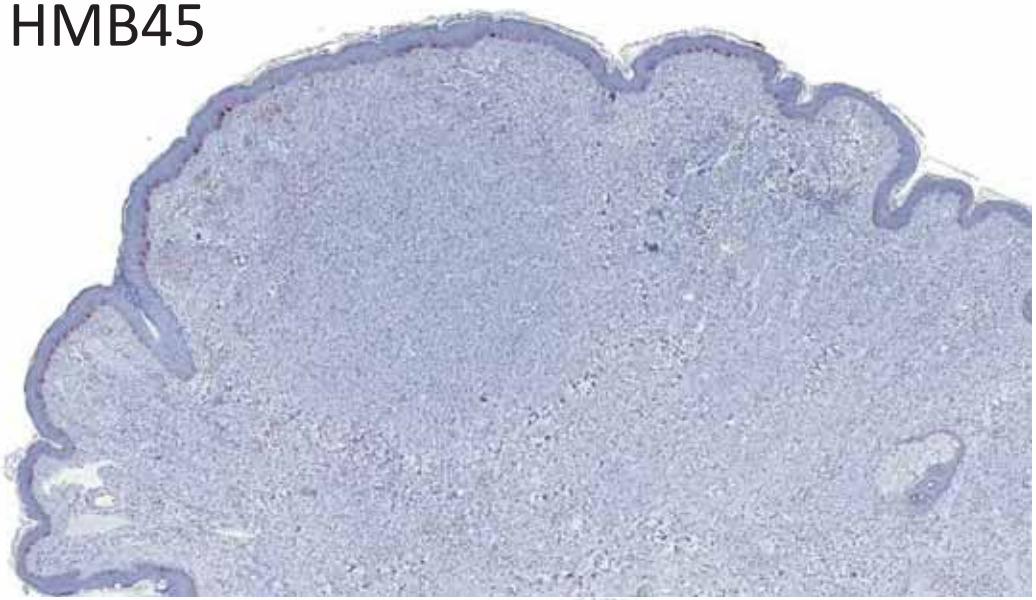
BAP1 inactivated melanocytoma profile

- Helps decide if BAP1 immunohistochemistry is useful
- Unpublished data from >250 cases

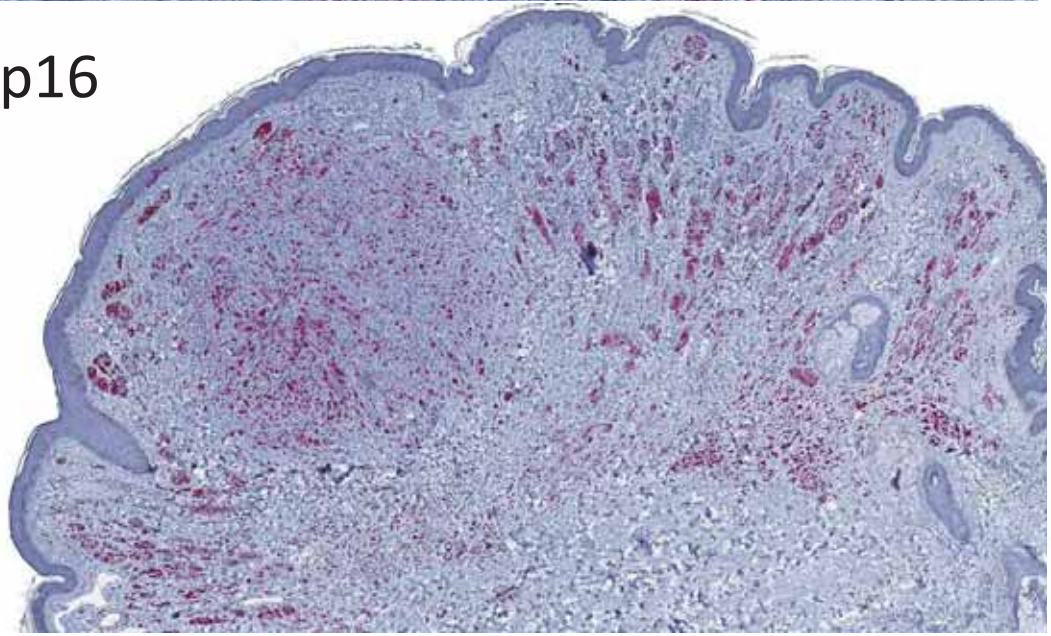
MelanA



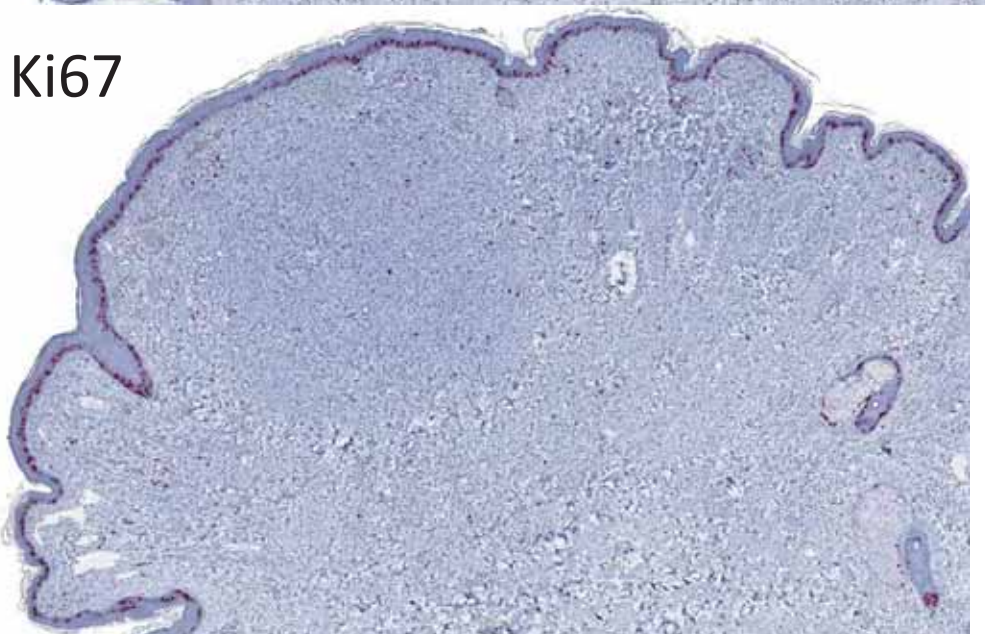
HMB45

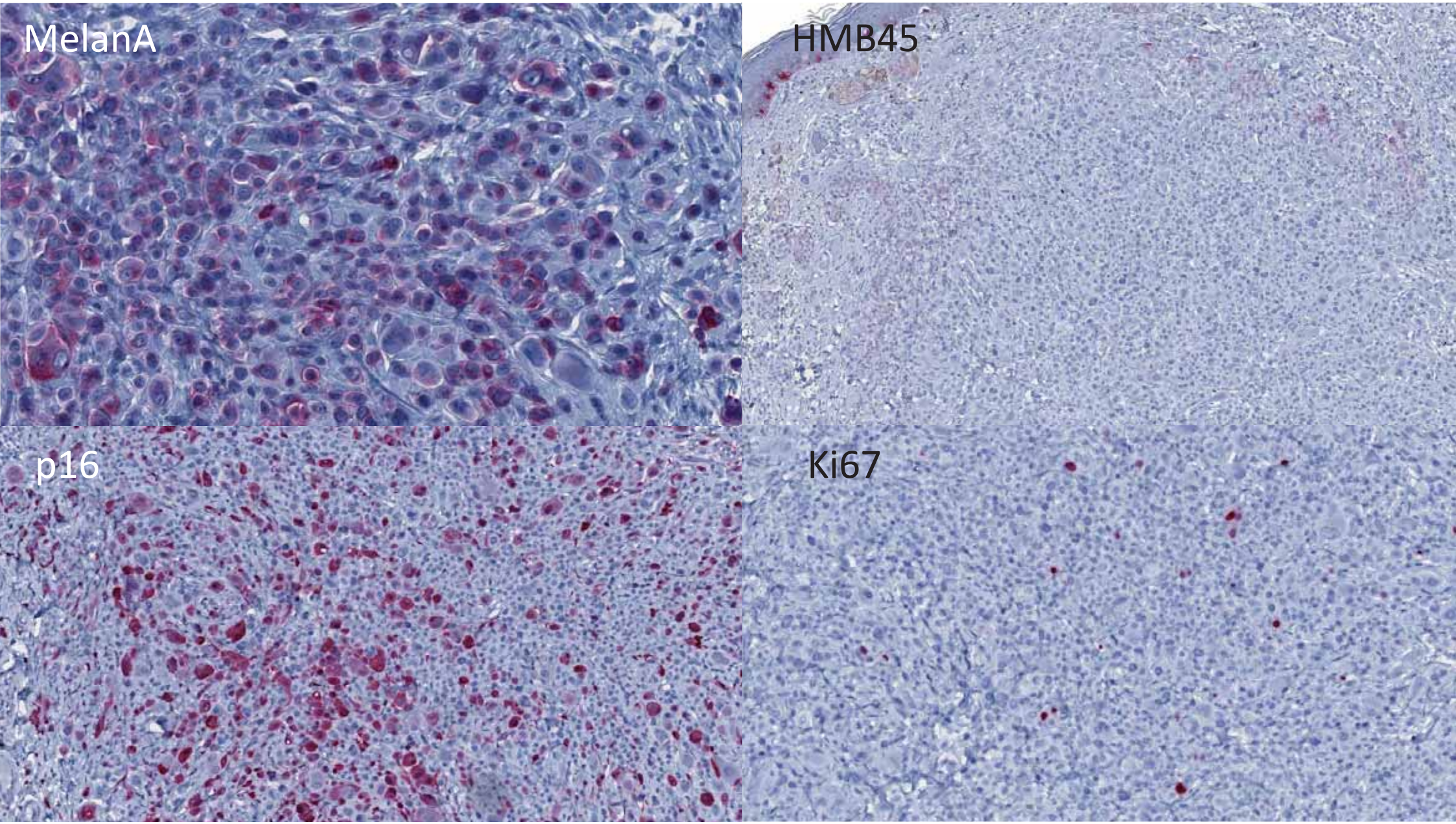


p16



Ki67




MelanA
HMB45
p16
Ki67

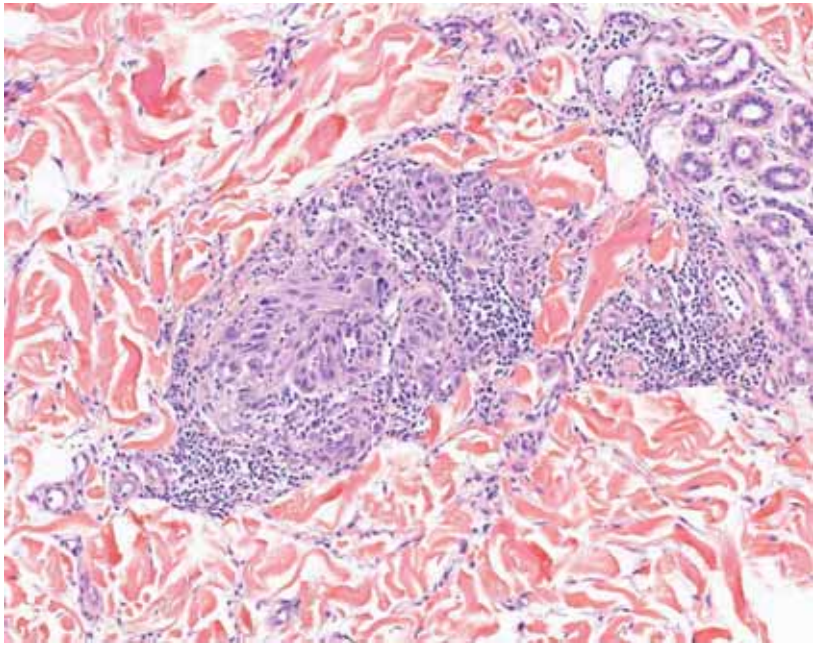
Case analysis approach

- Time-check with Lorenzo

Sox10 vs MelanA

- Sometimes one is a better choice than the other, not always same…
- Sometimes both are needed and show different patterns

Sox10 vs MelanA or HMB45
Melanoma re-excision: in-transit metastasis?



Sox10 vs MelanA or HMB45
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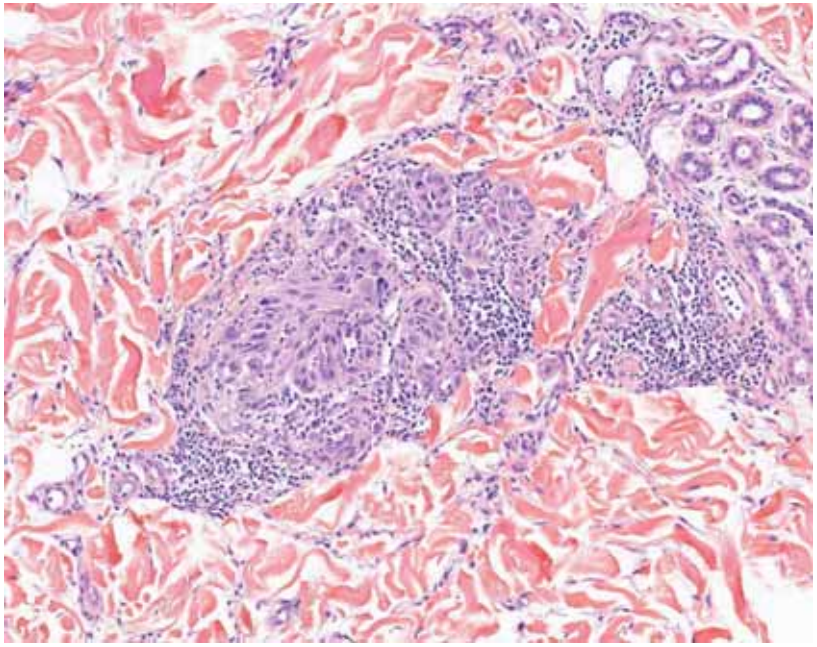

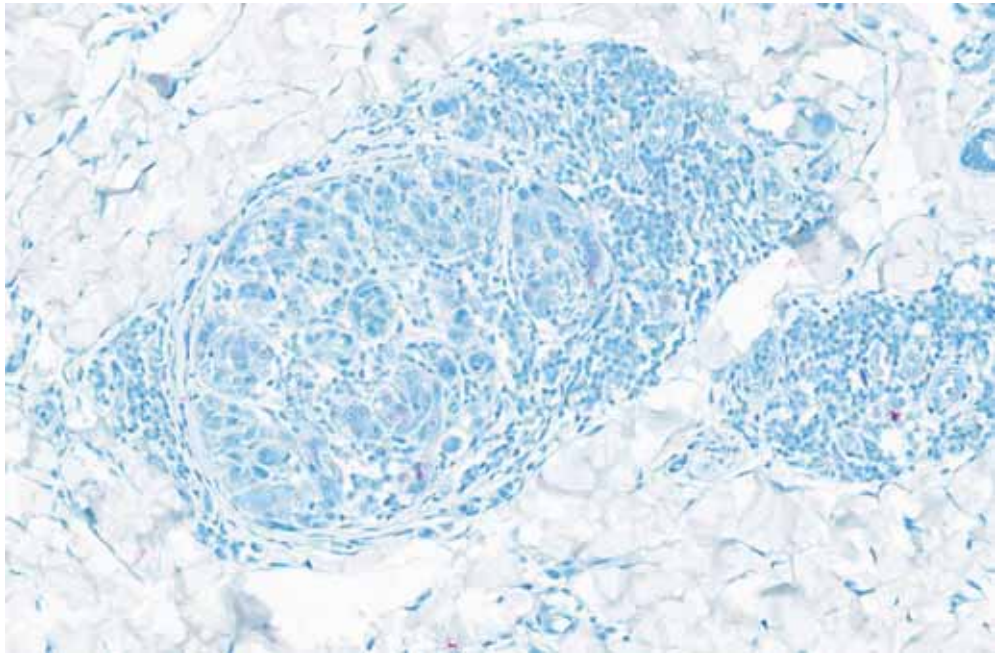

Sox10 vs MelanA or HMB45
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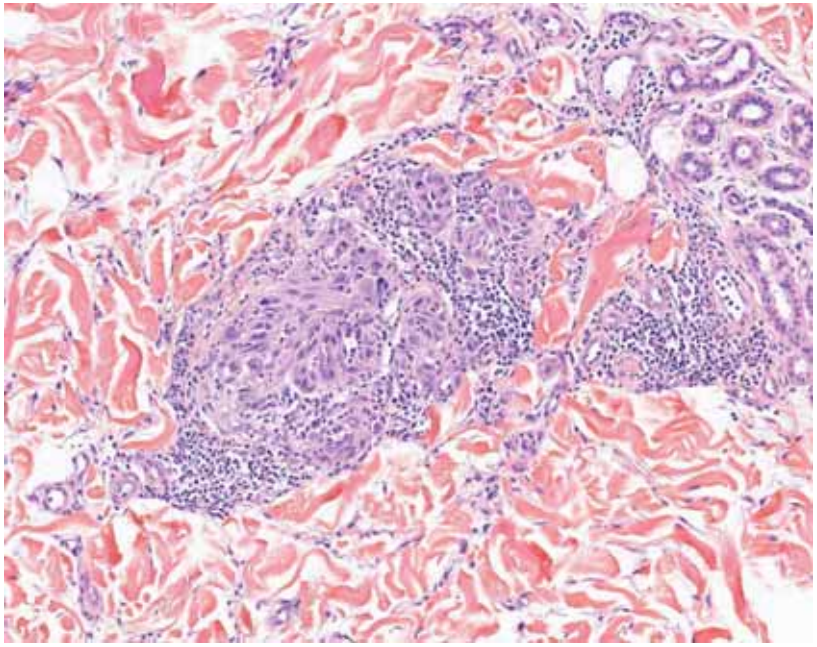

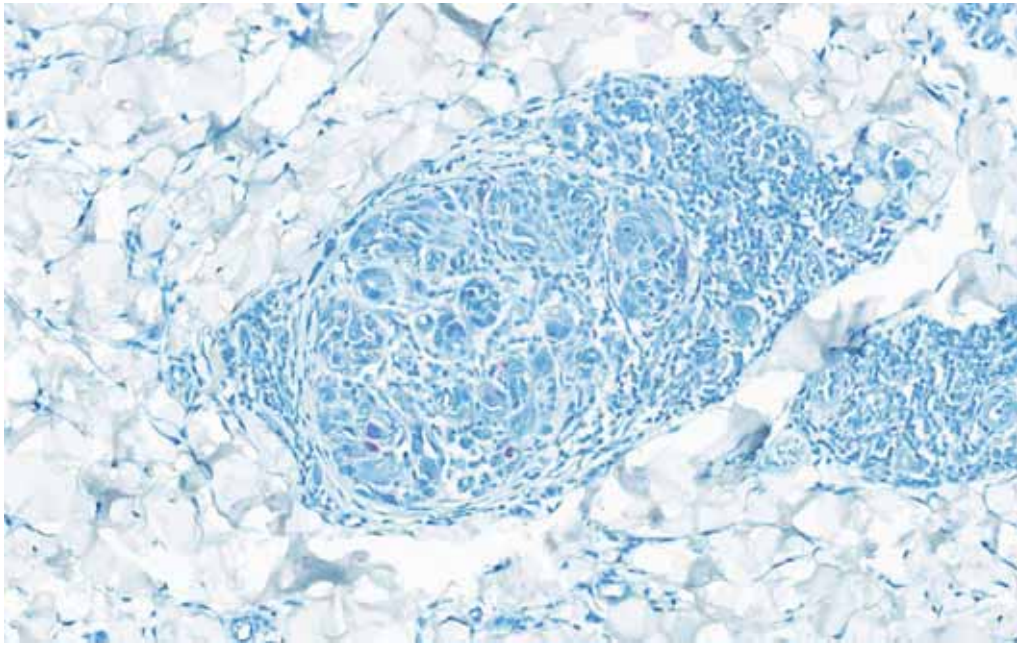

Sox10 vs MelanA or HMB45
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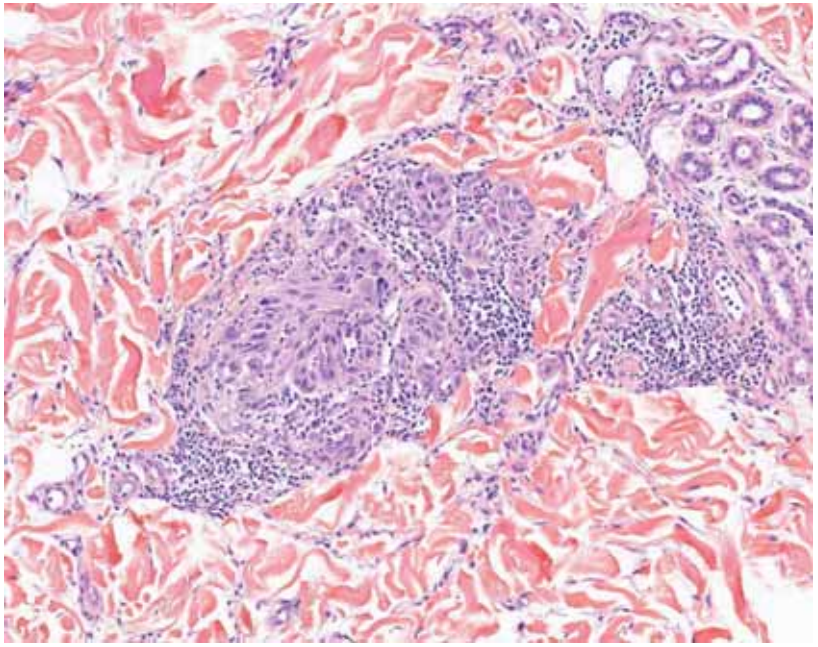
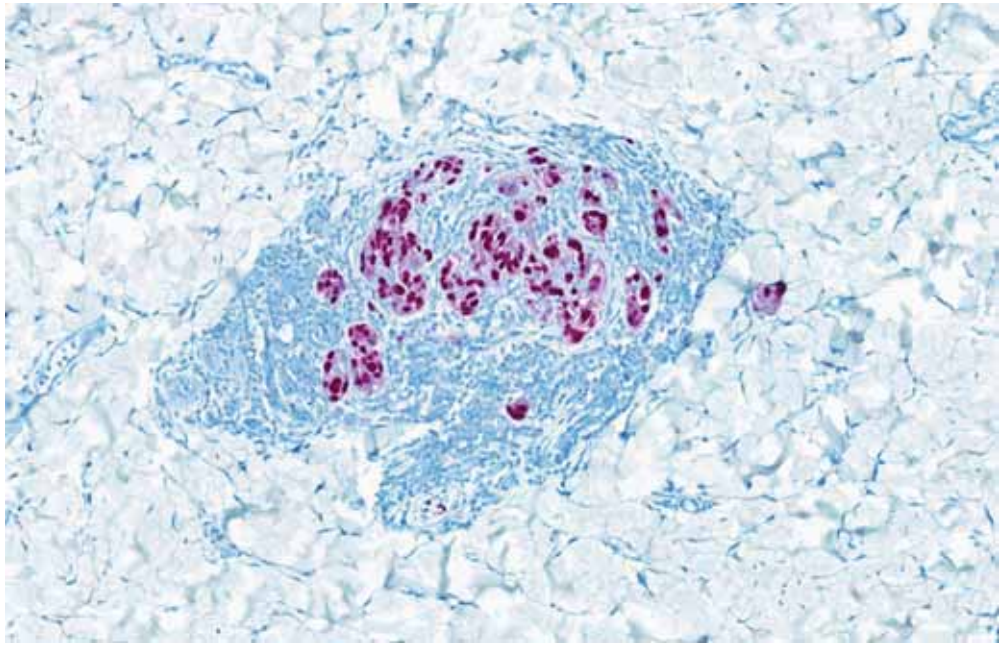
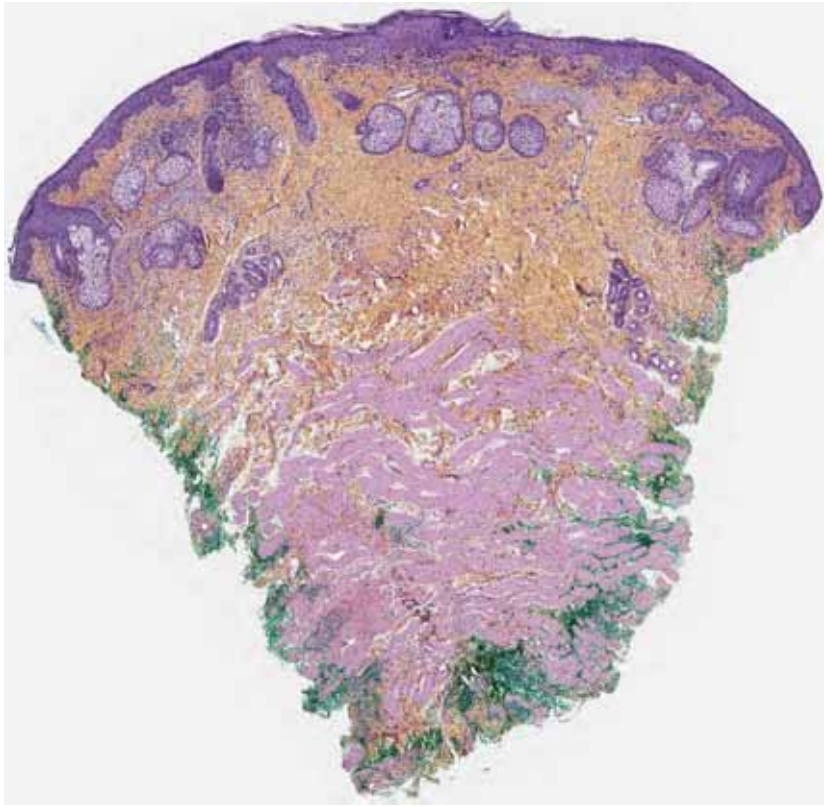




Unexpected findings MelanA
Small incidental nevi in LM resections



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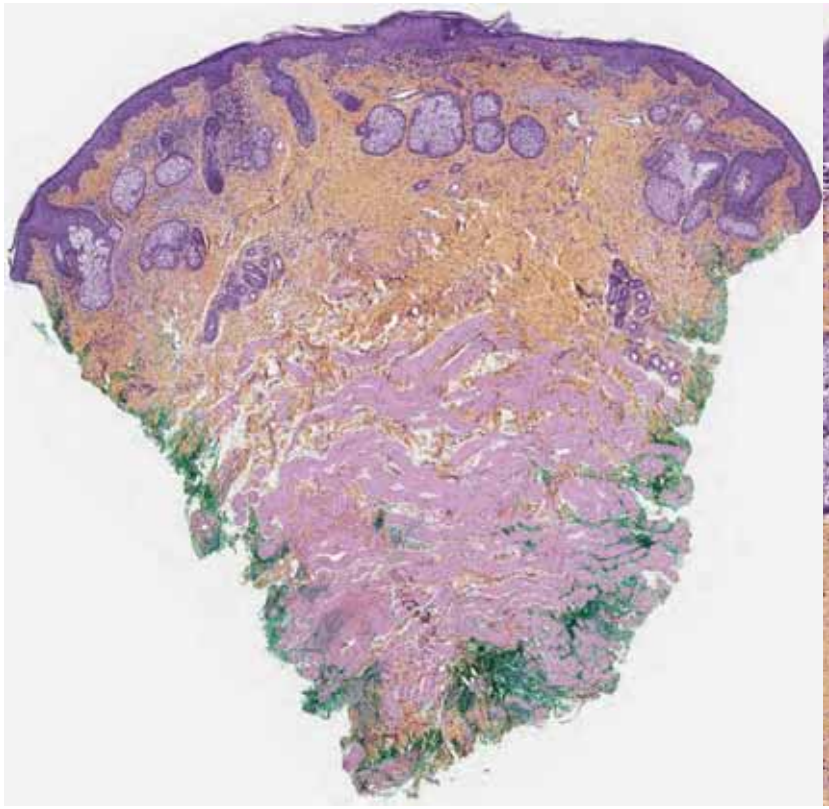

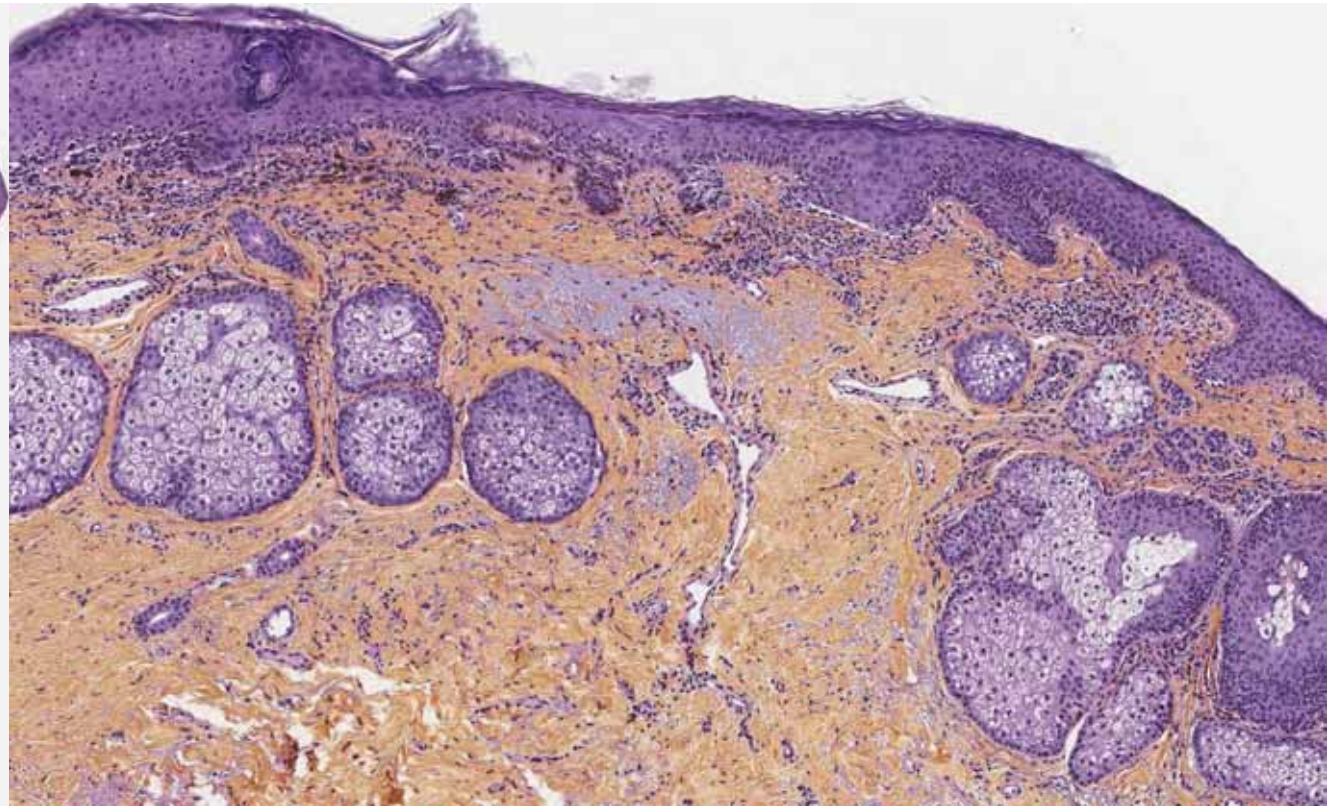

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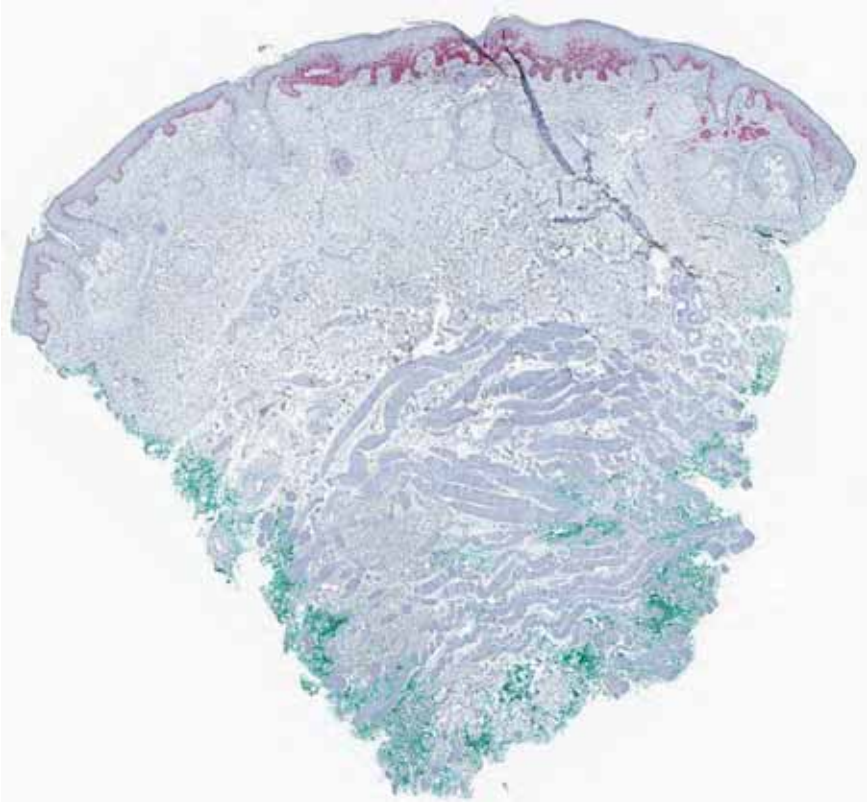
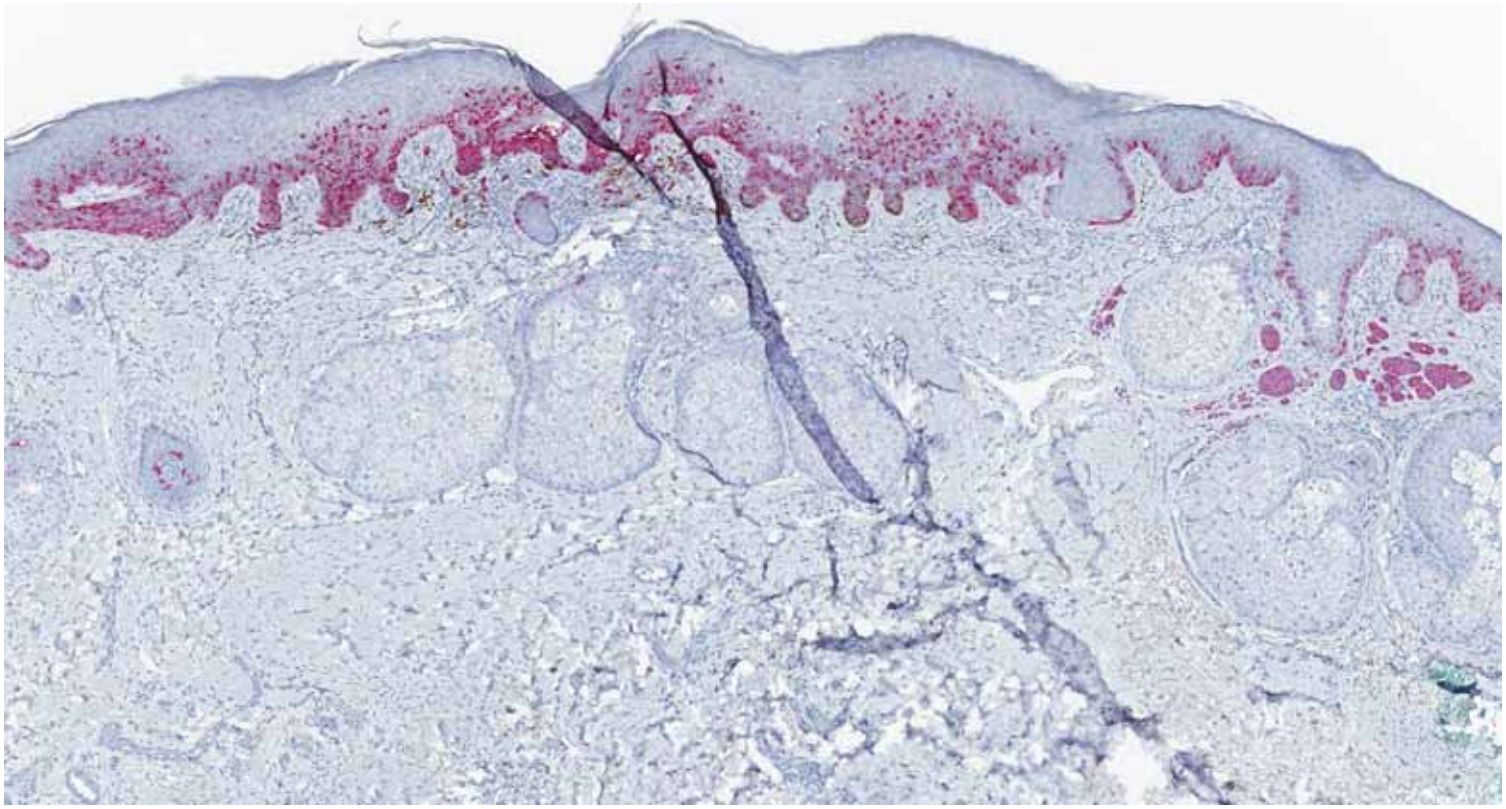
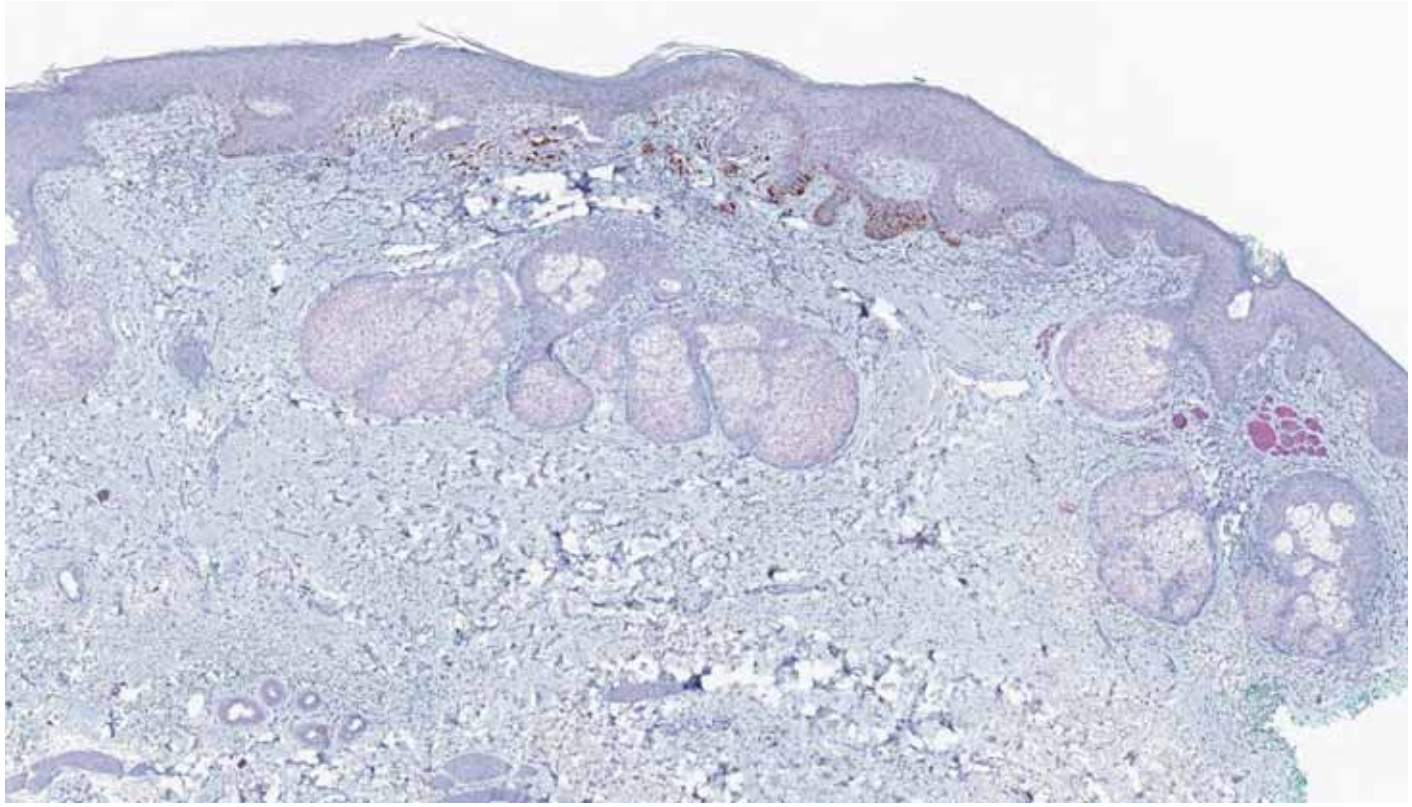


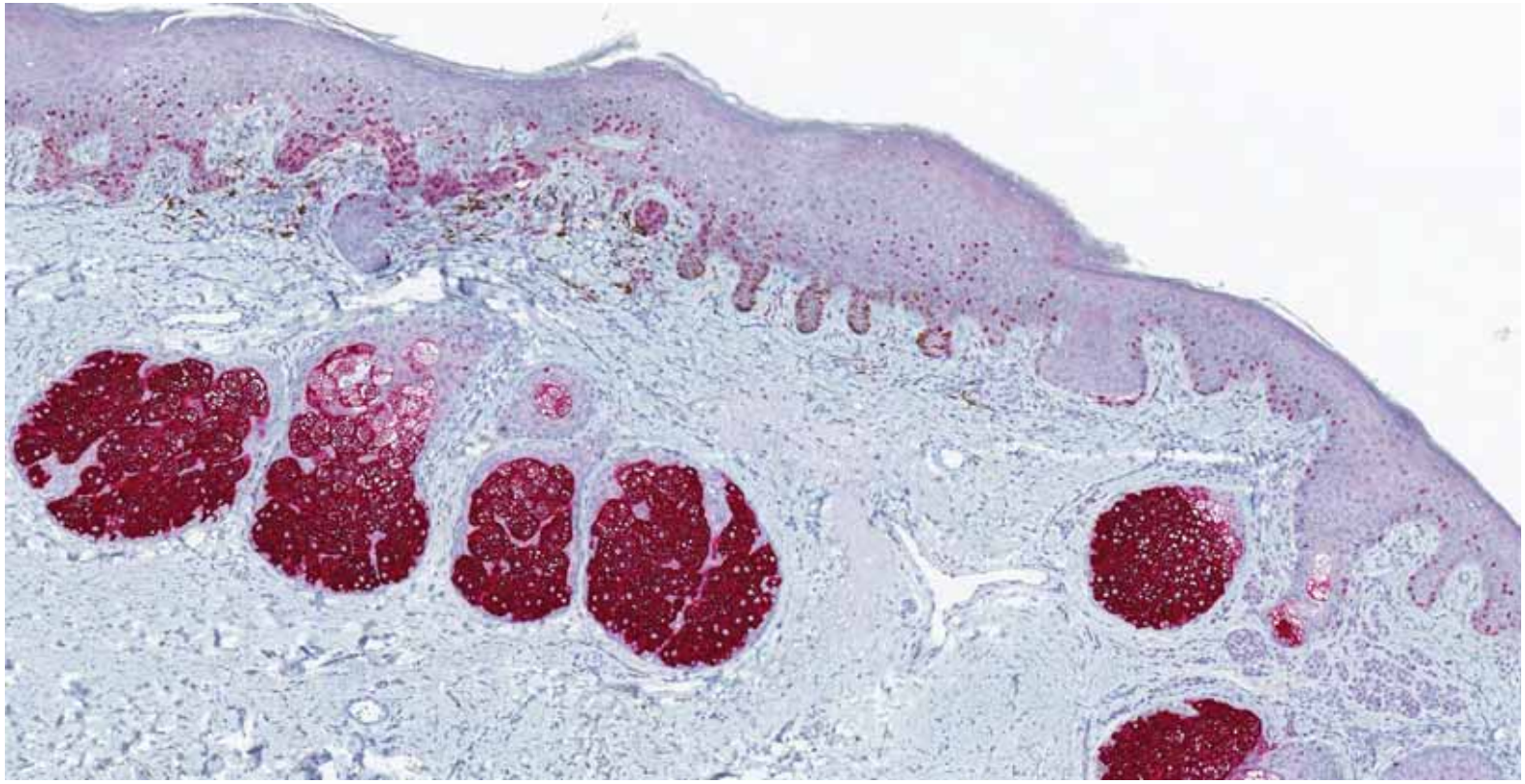
Unexpected findings MelanA
Small incidental nevi in LM resections



Unexpected findings
Small incidental BRAF V600E nevi in LM resections



Unexpected findings
Small incidental nevi in PRAME + LM



M6, modified lesion of the cheek

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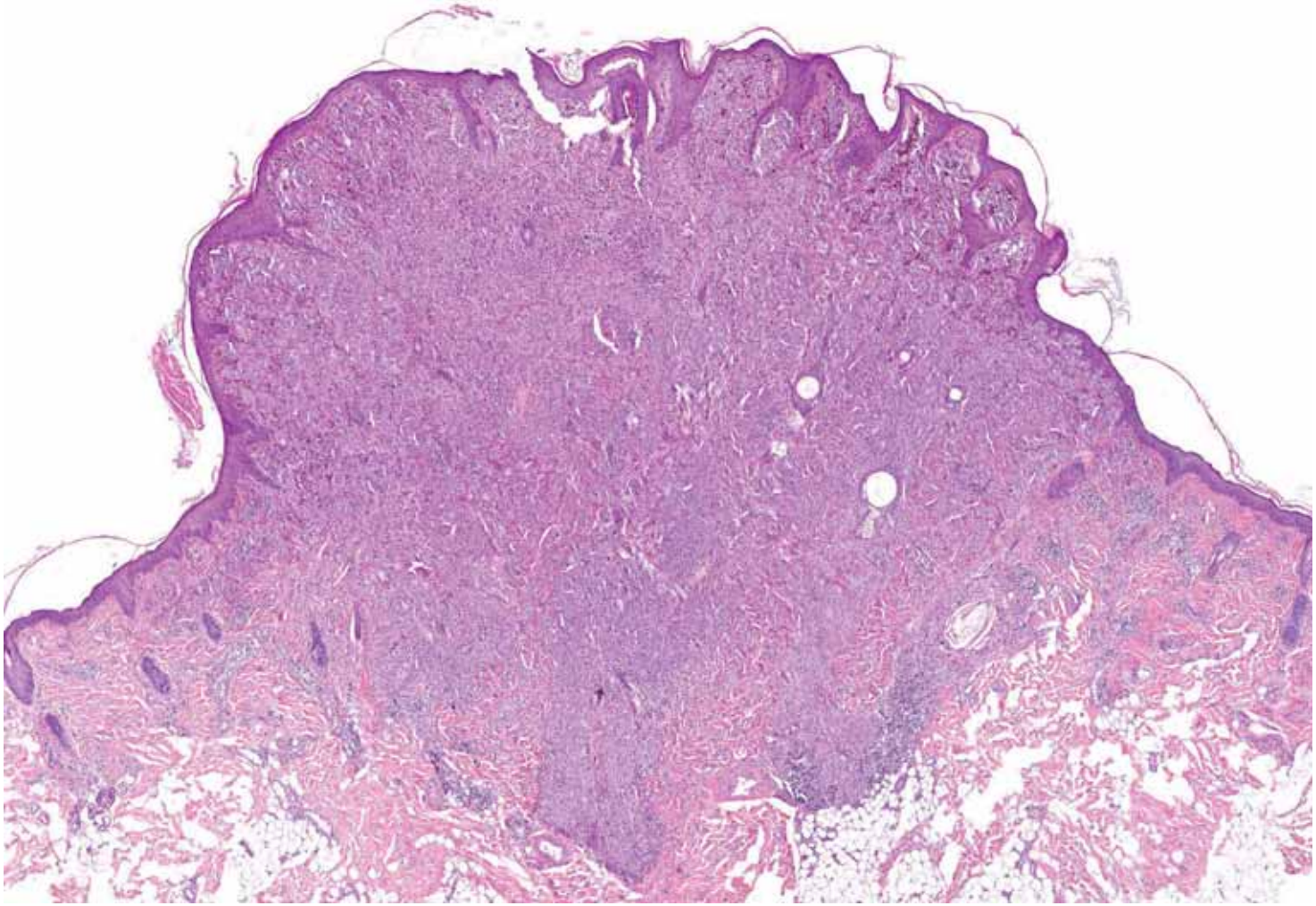
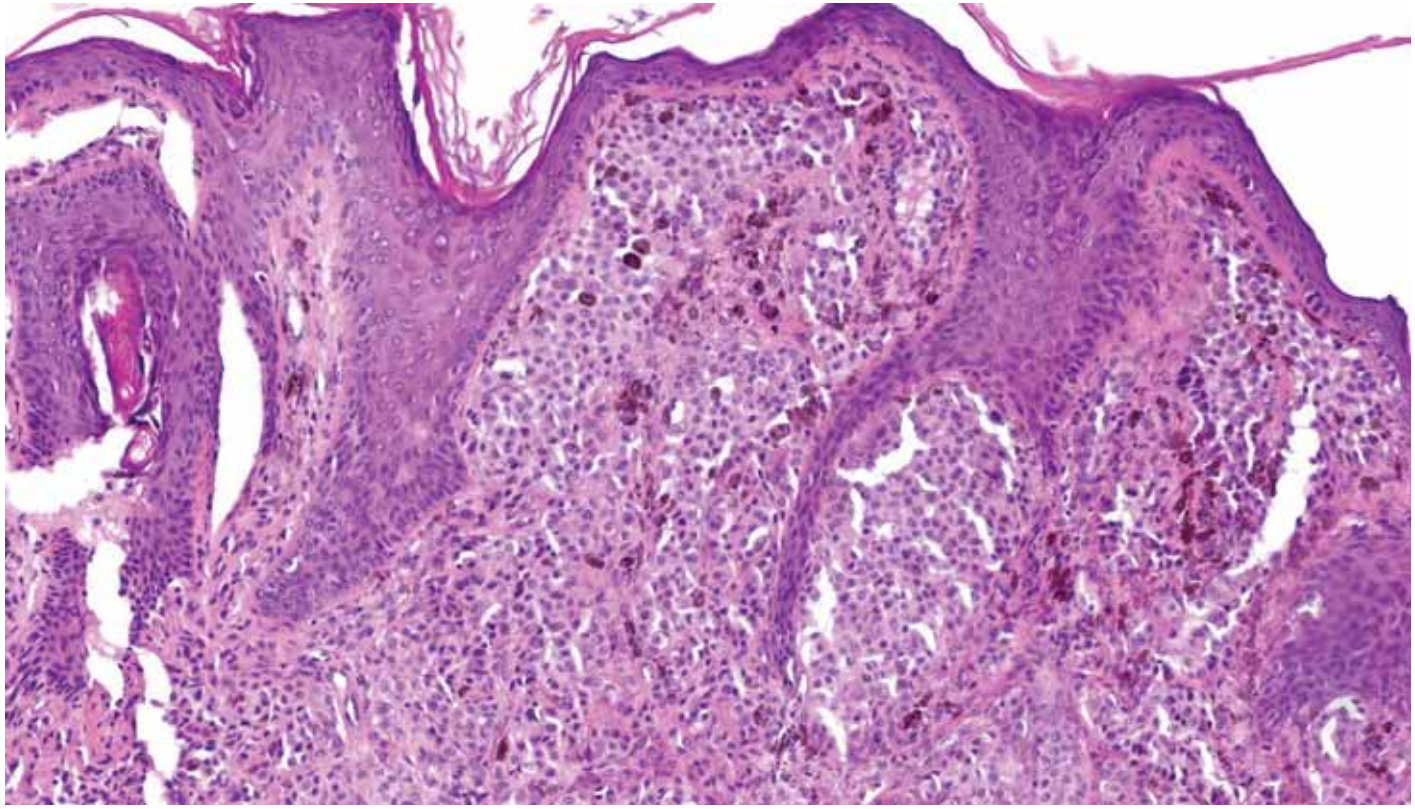
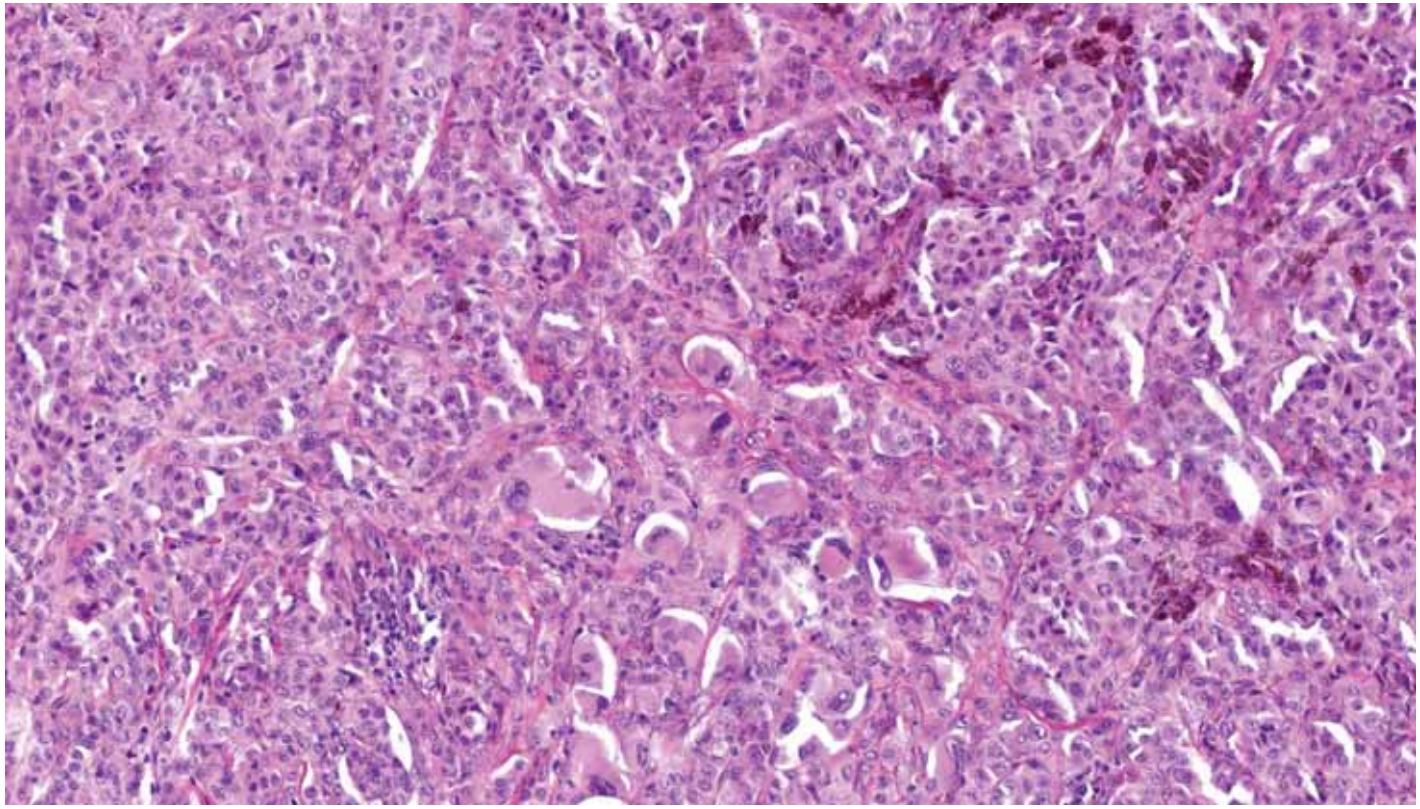


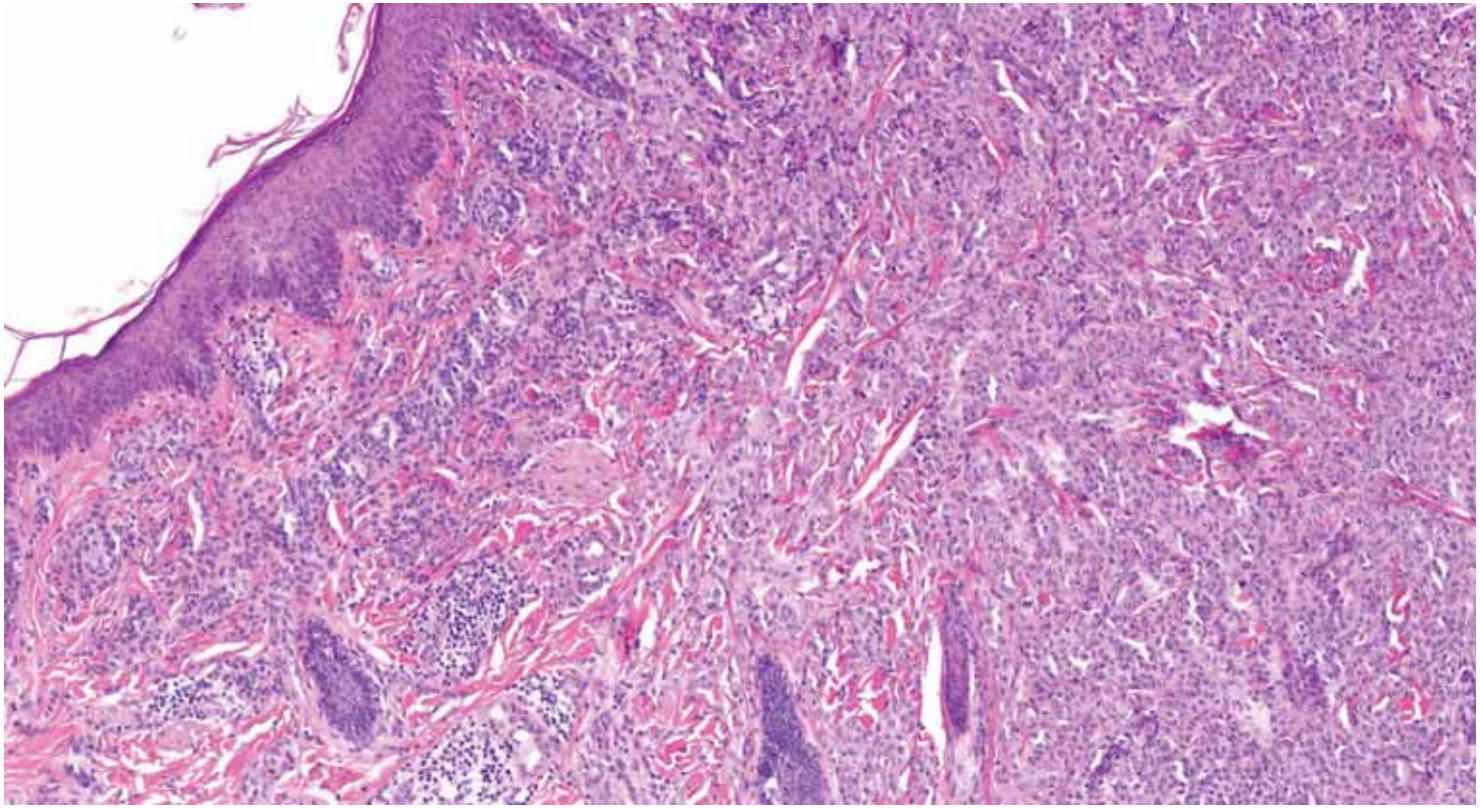
M6, modified lesion of the cheek



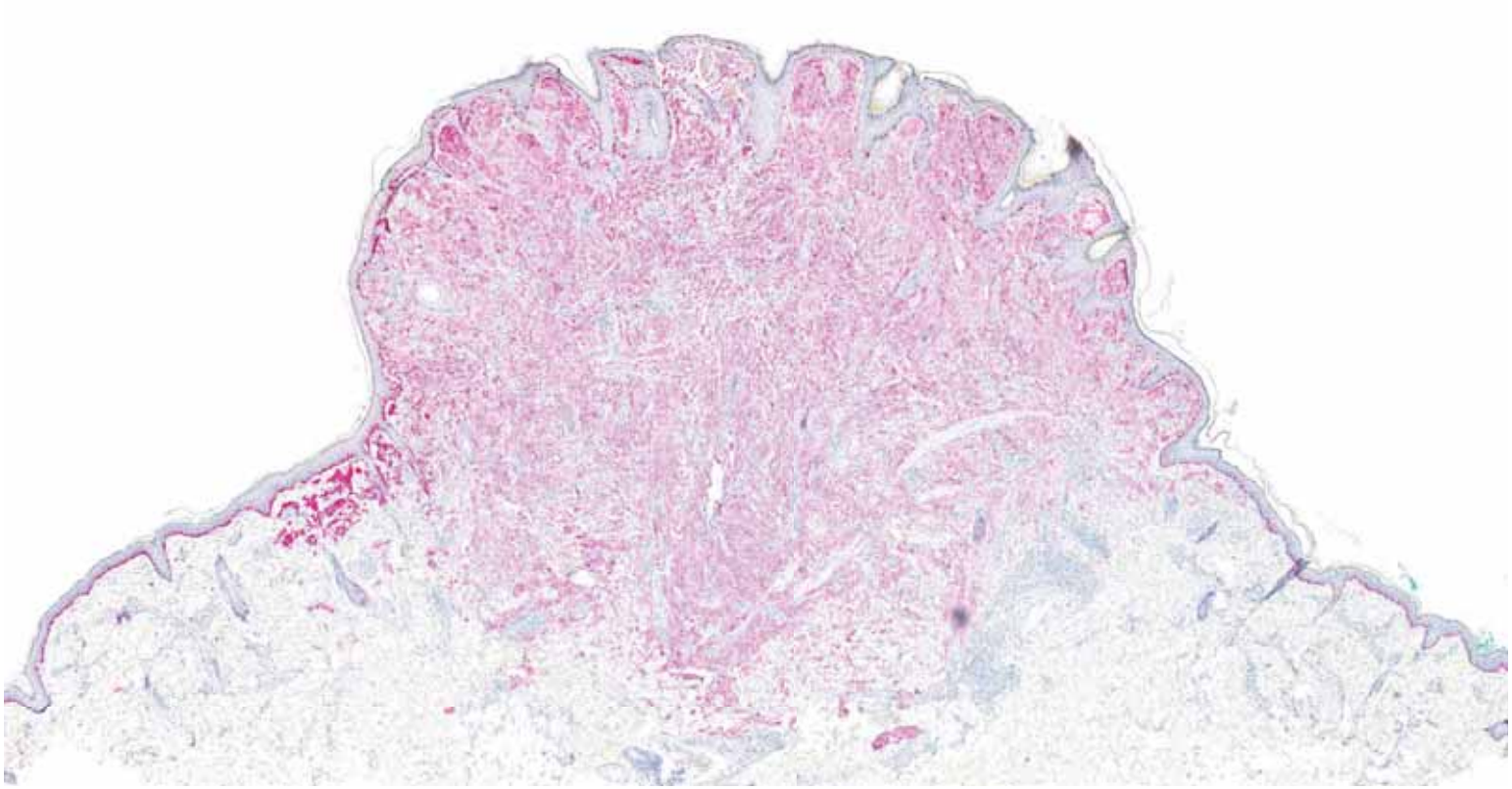
M6, modified lesion of the cheek



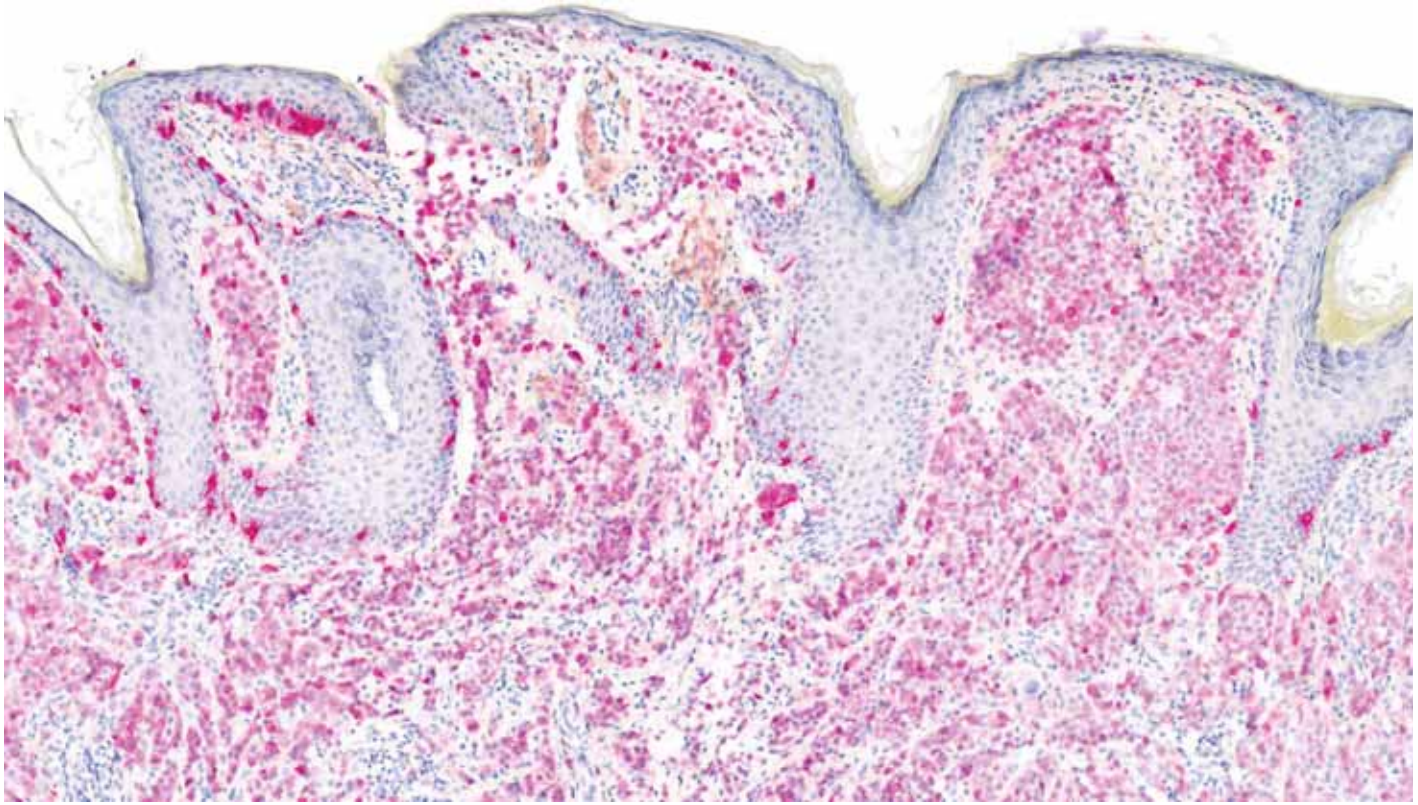
M6, modified lesion of the cheek



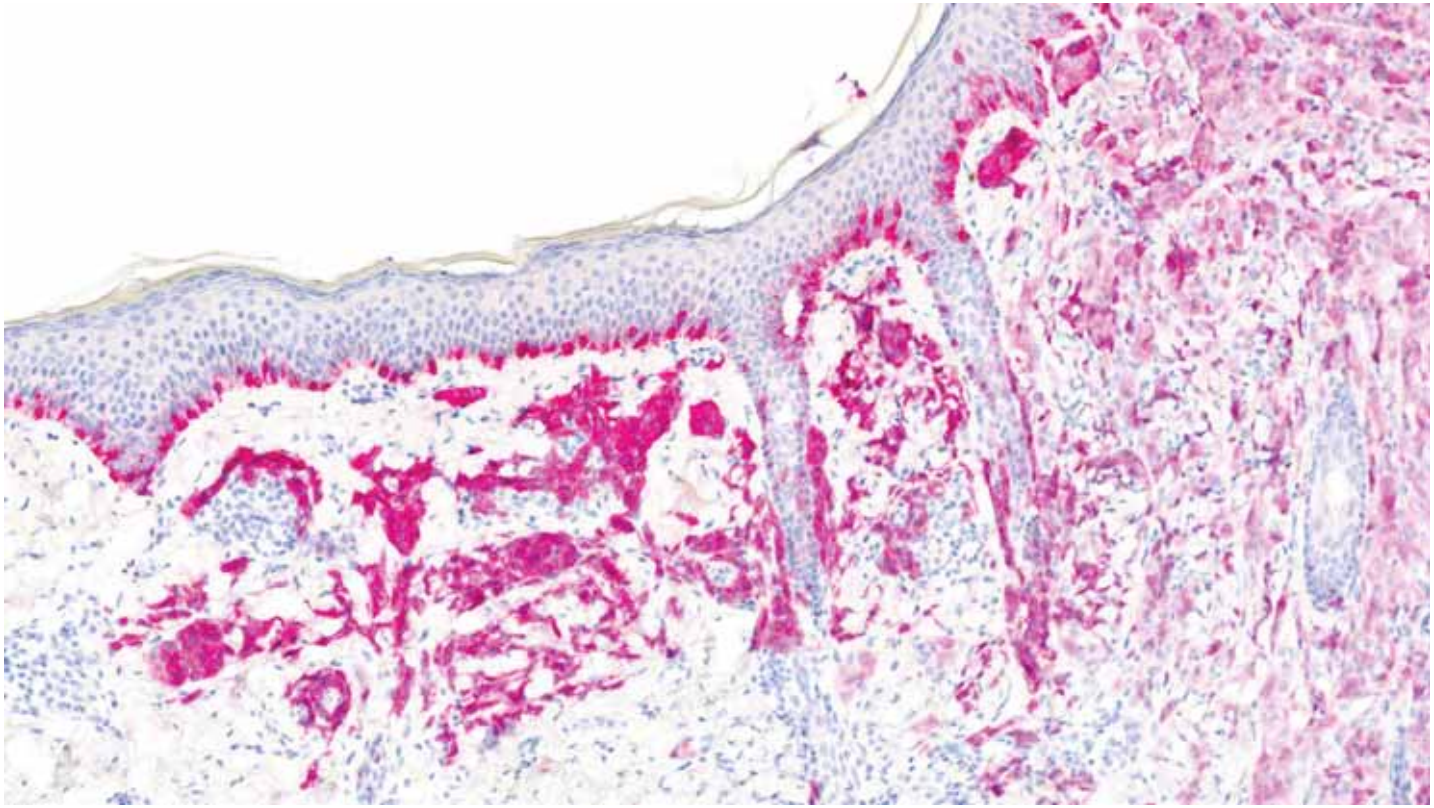
MelanA



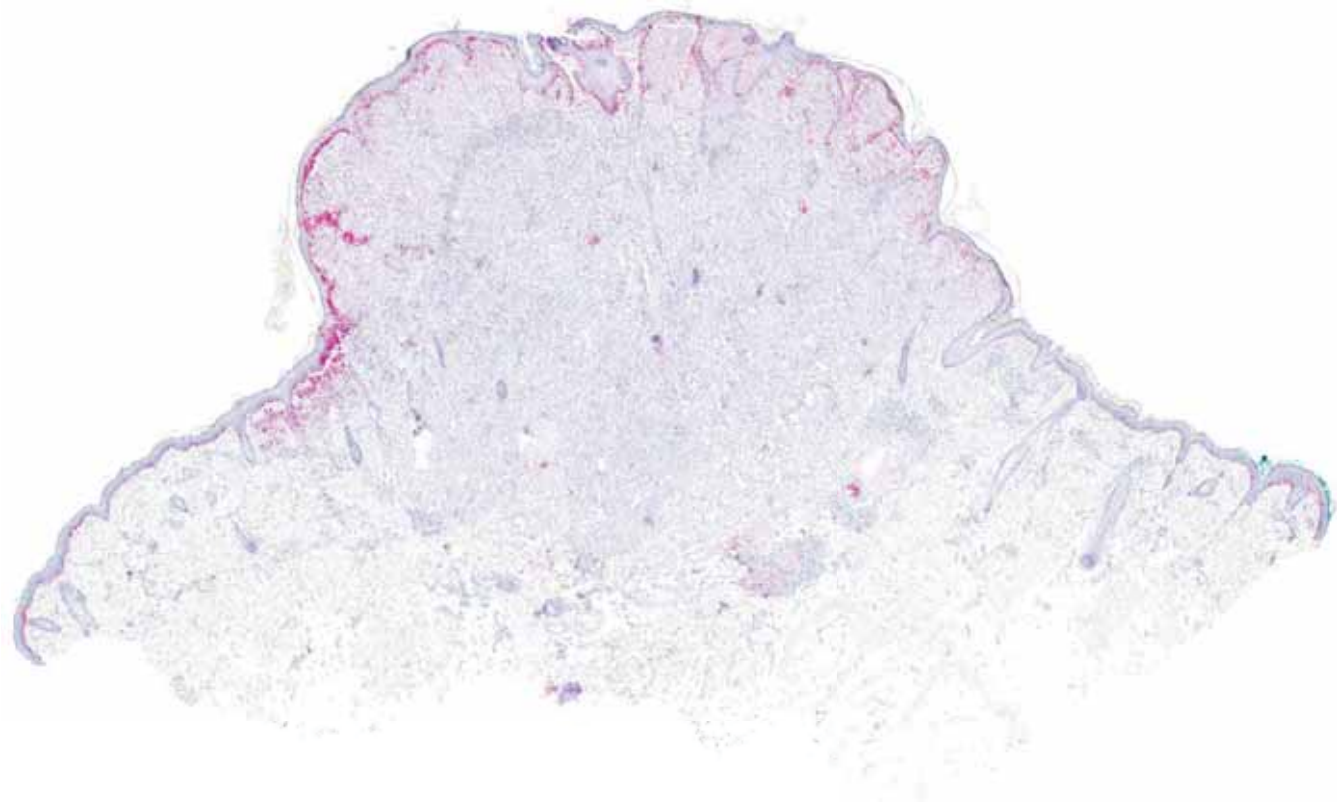
MelanA



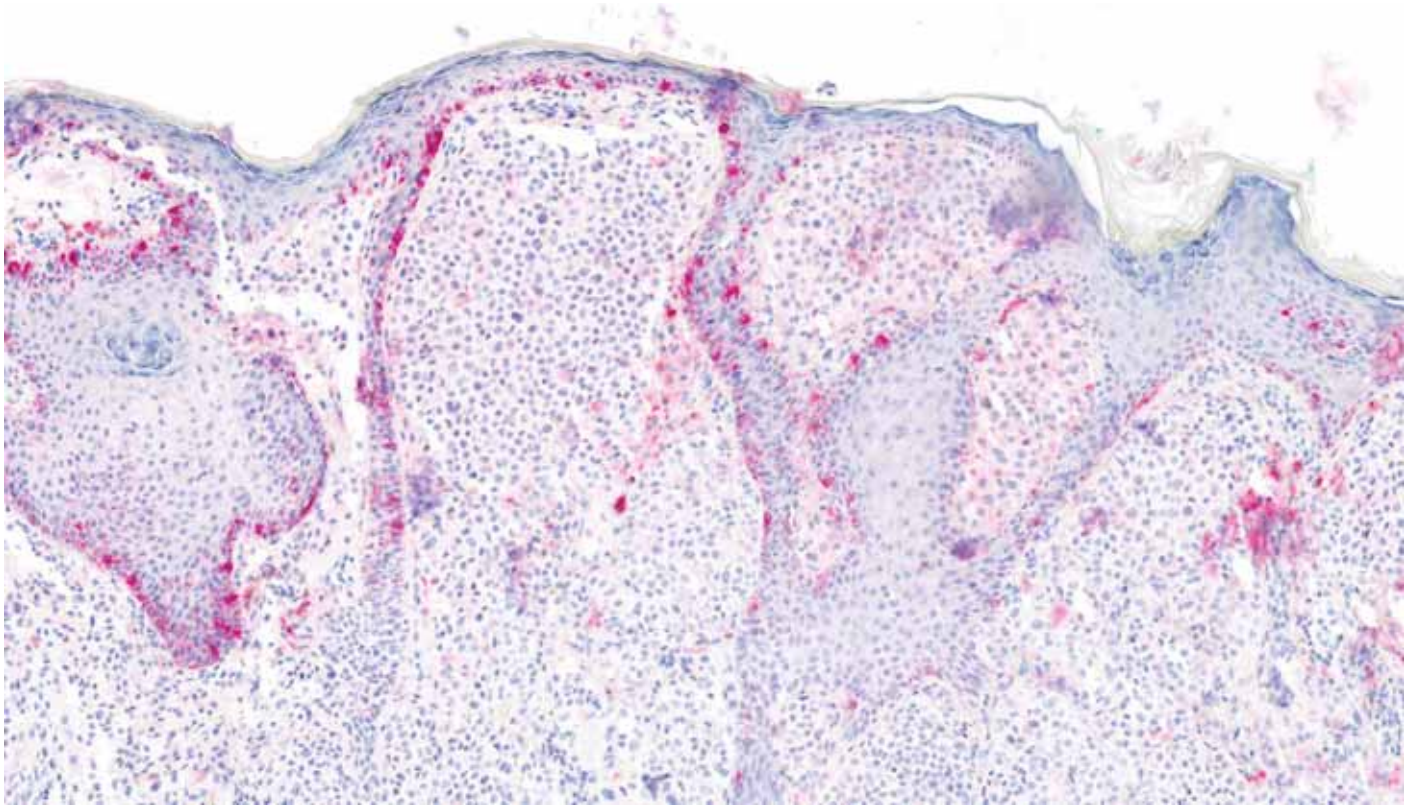
MelanA



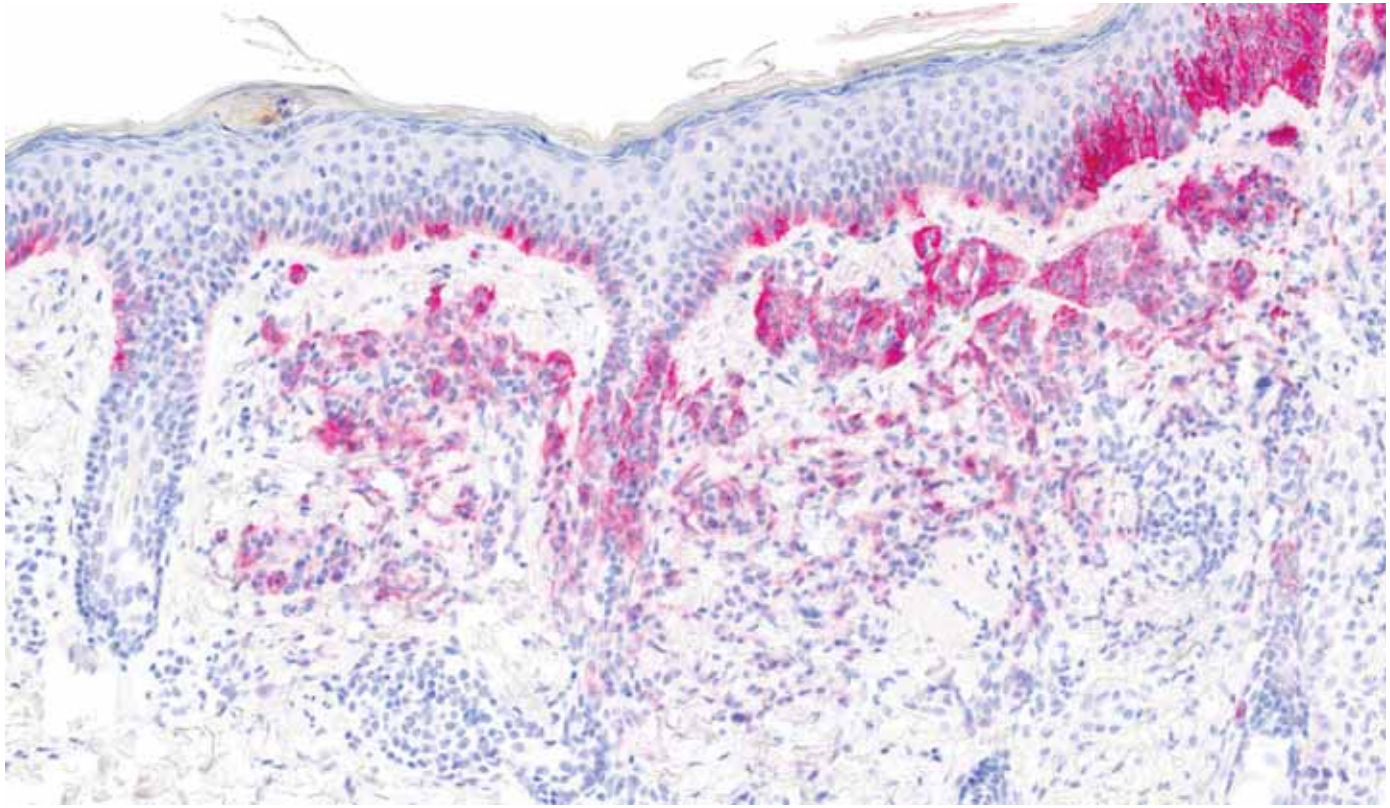
HMB45



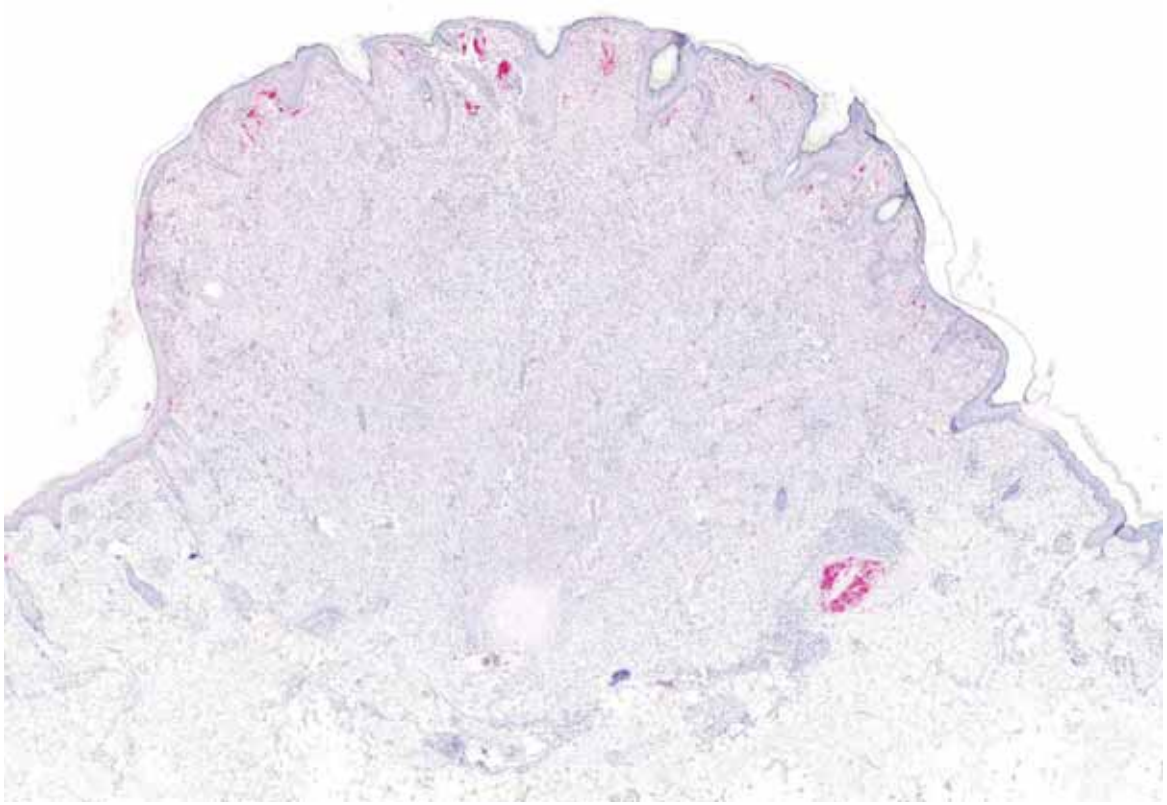
HMB45



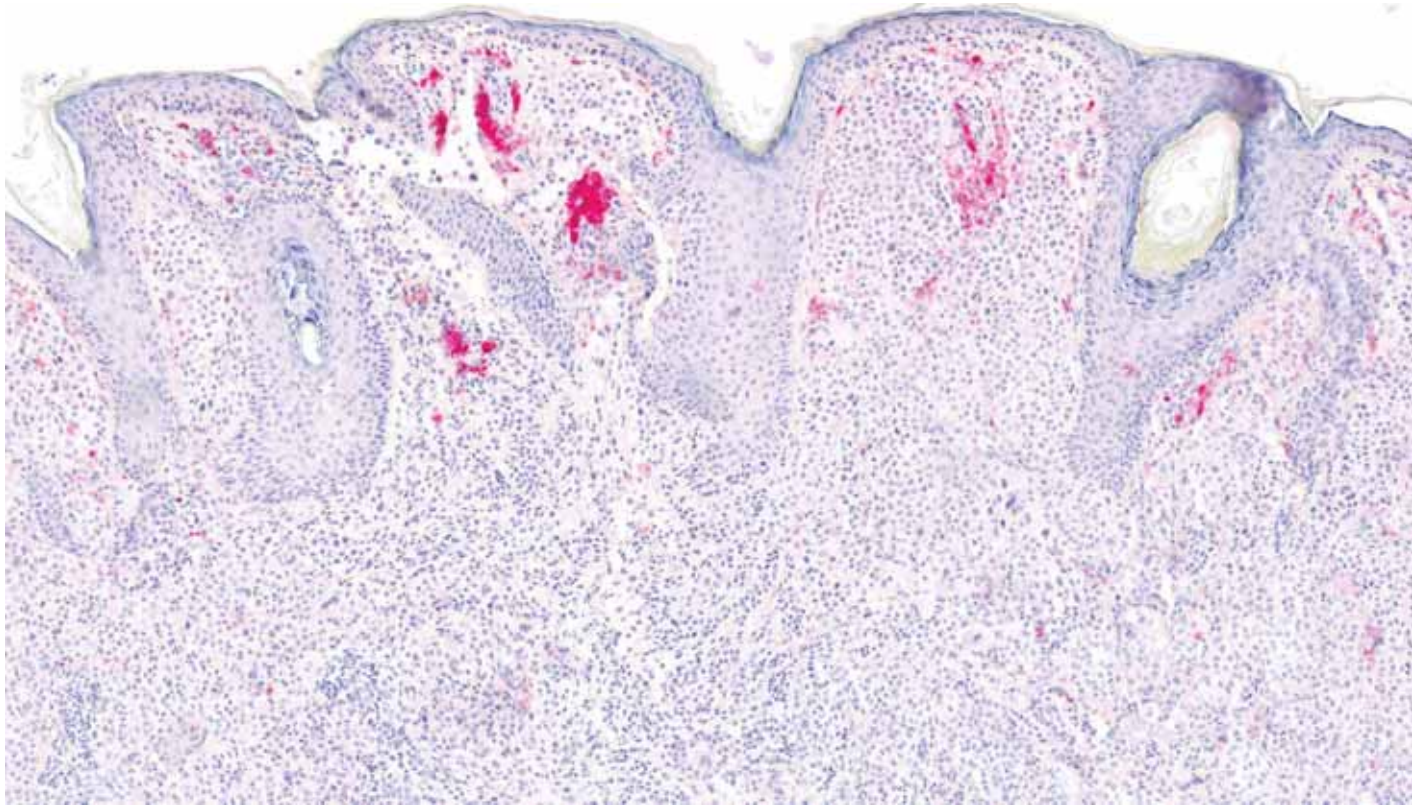
HMB45



P16



P16



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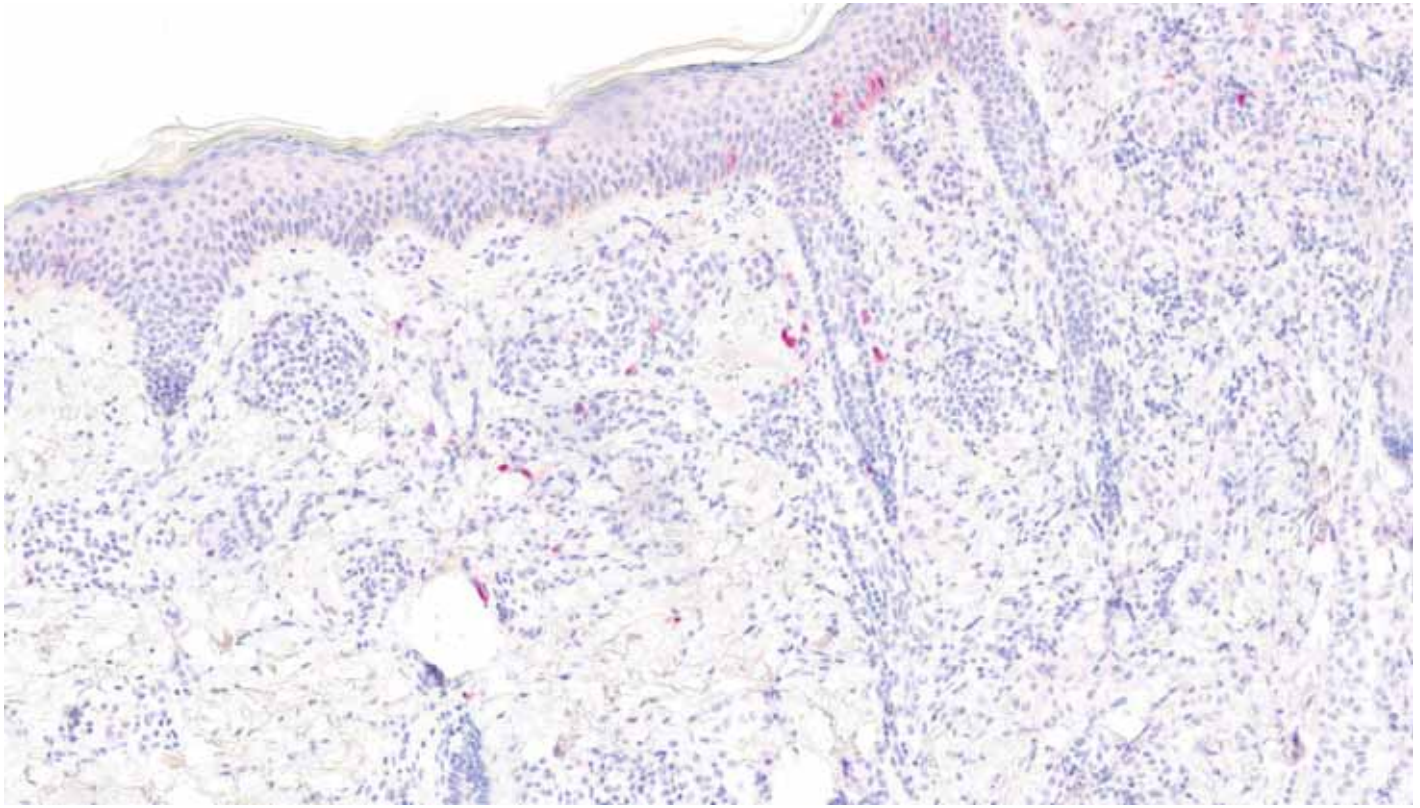
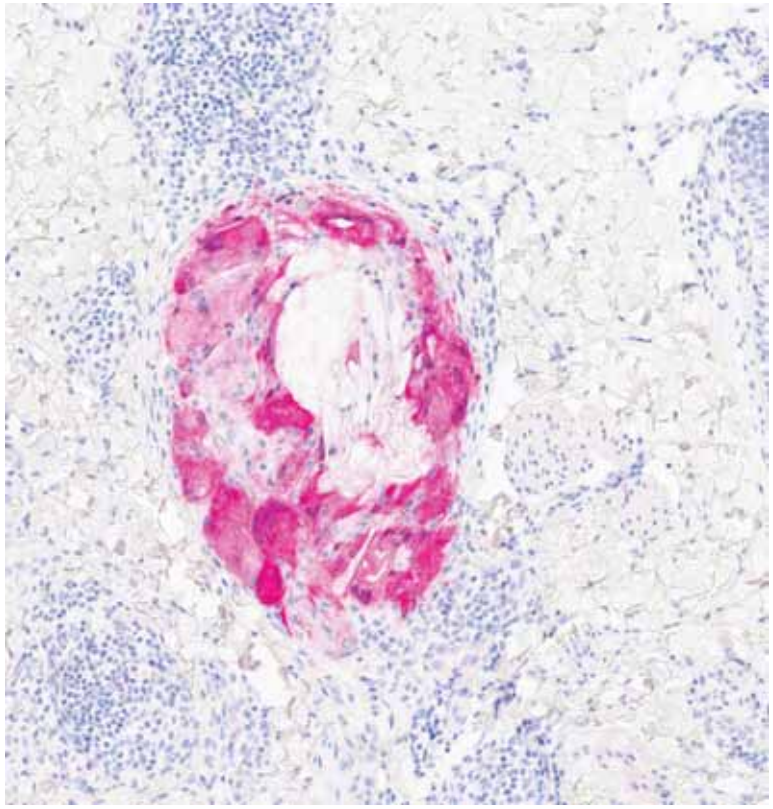
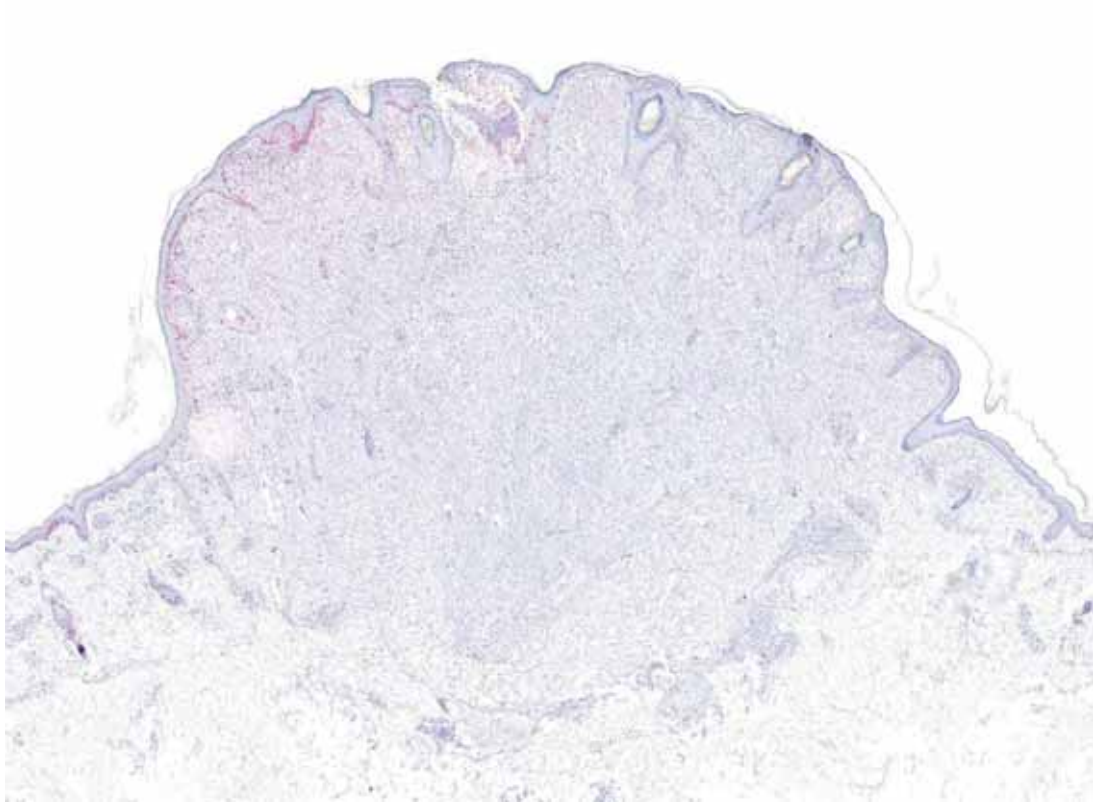


P16



Ki67



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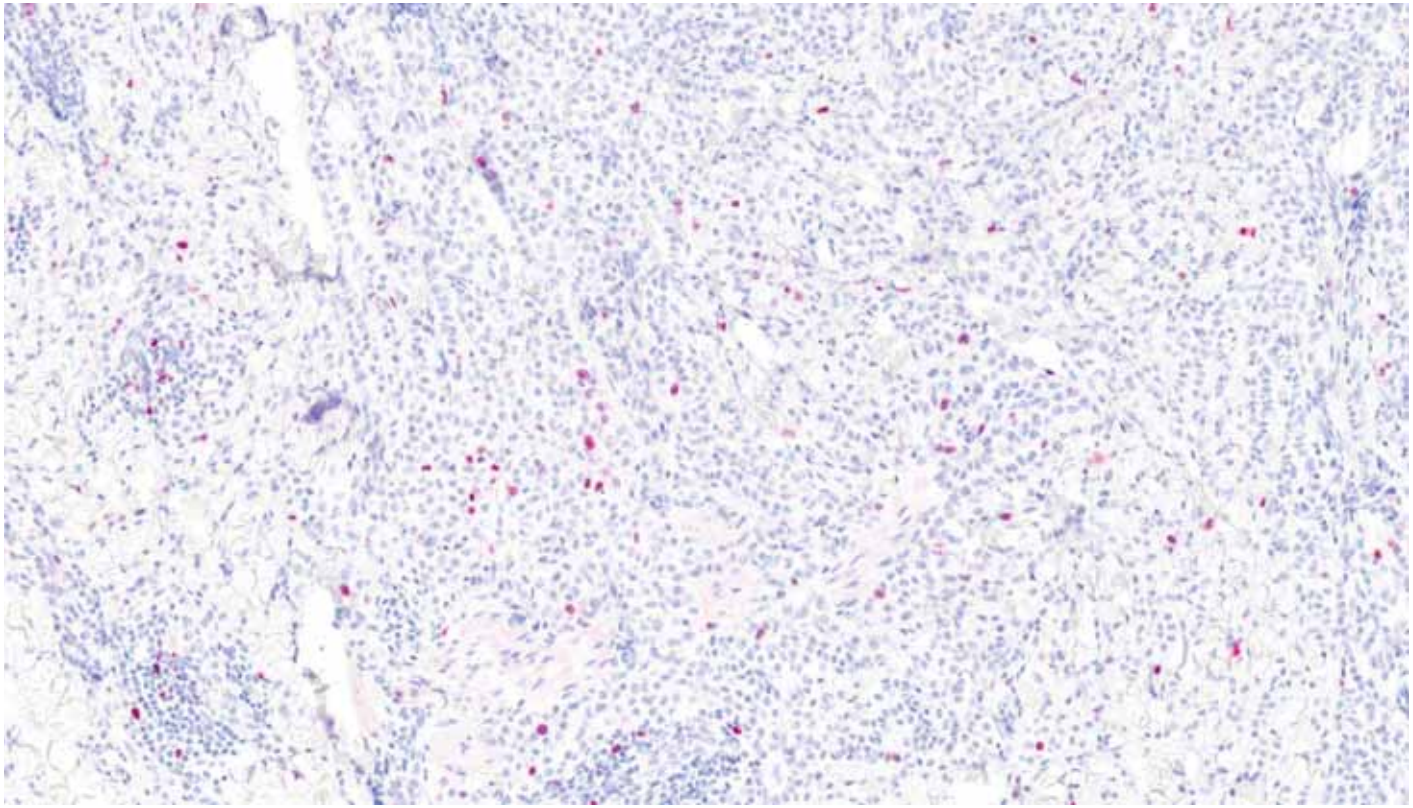
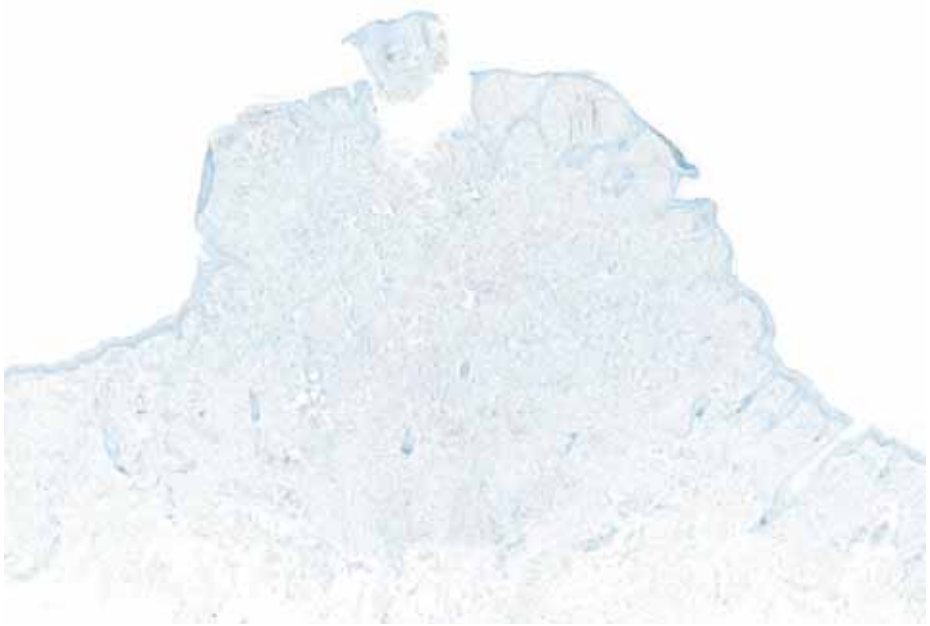
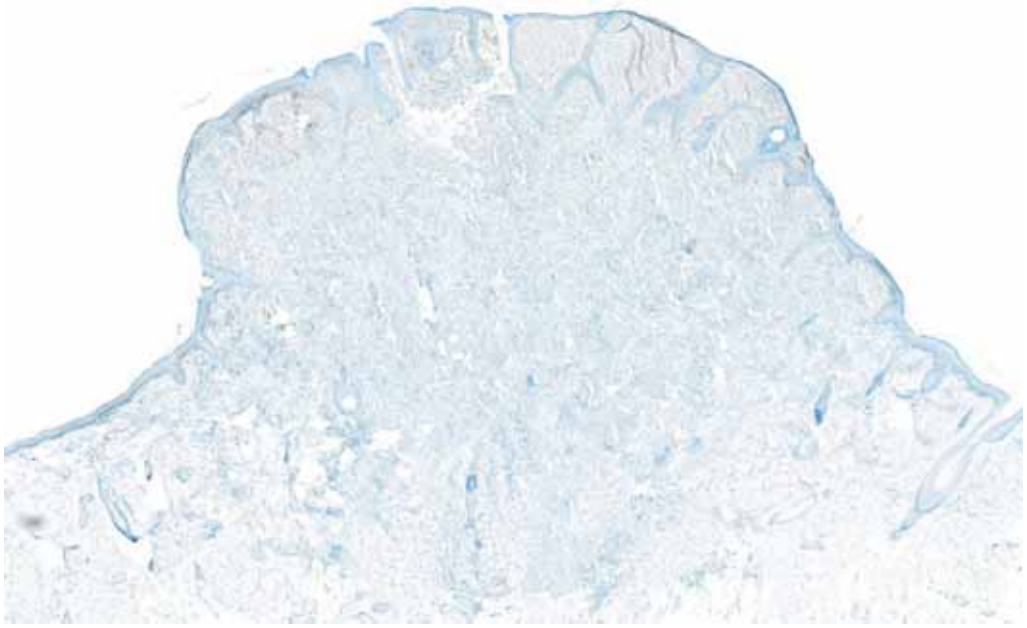


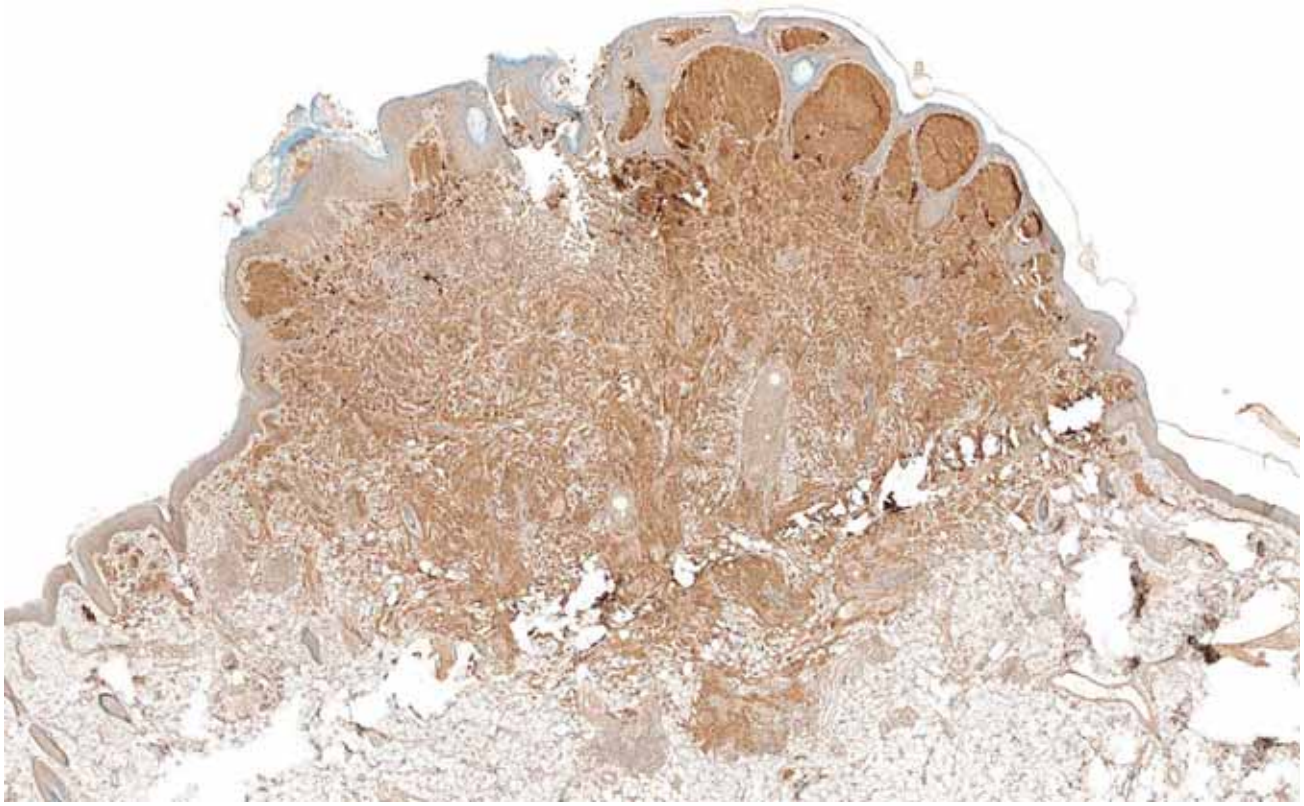
ALK



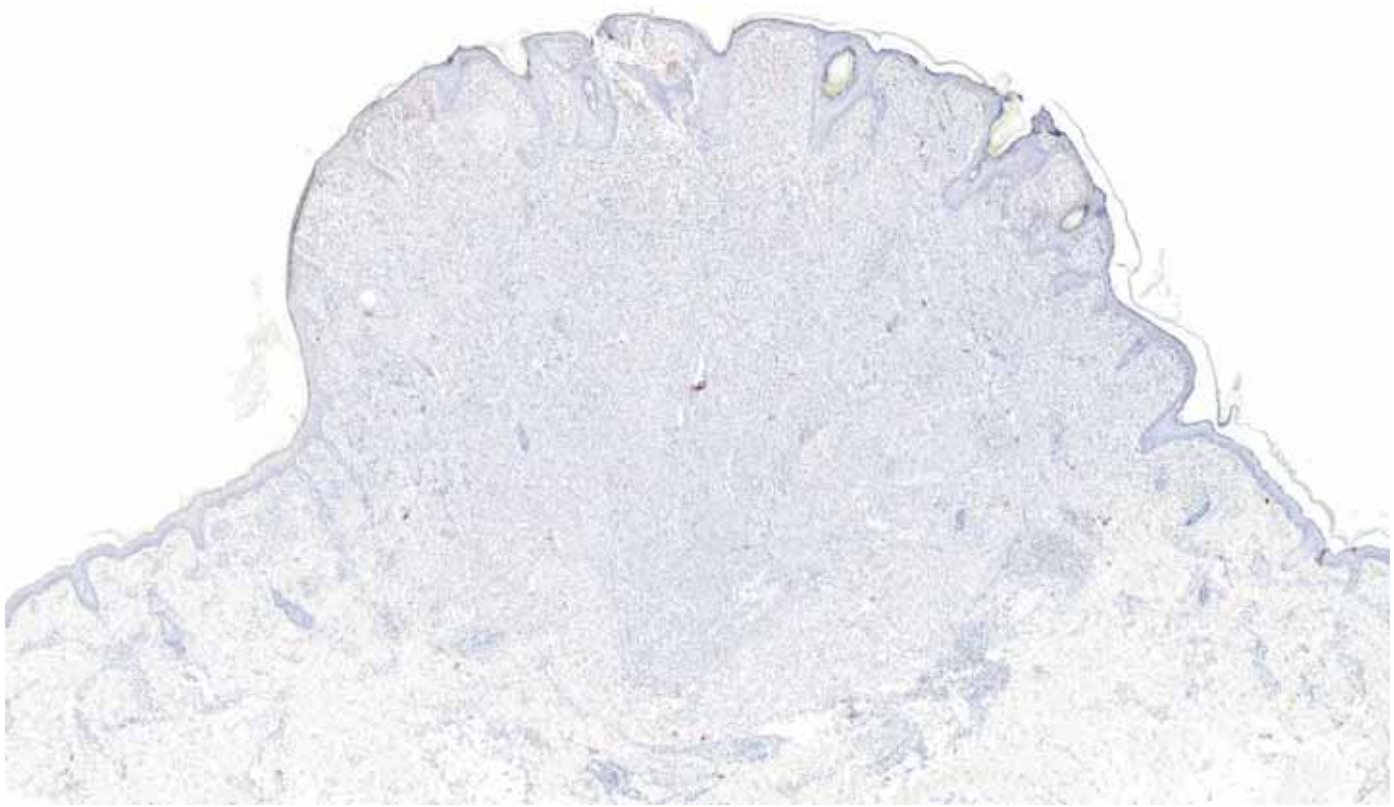
ROS1



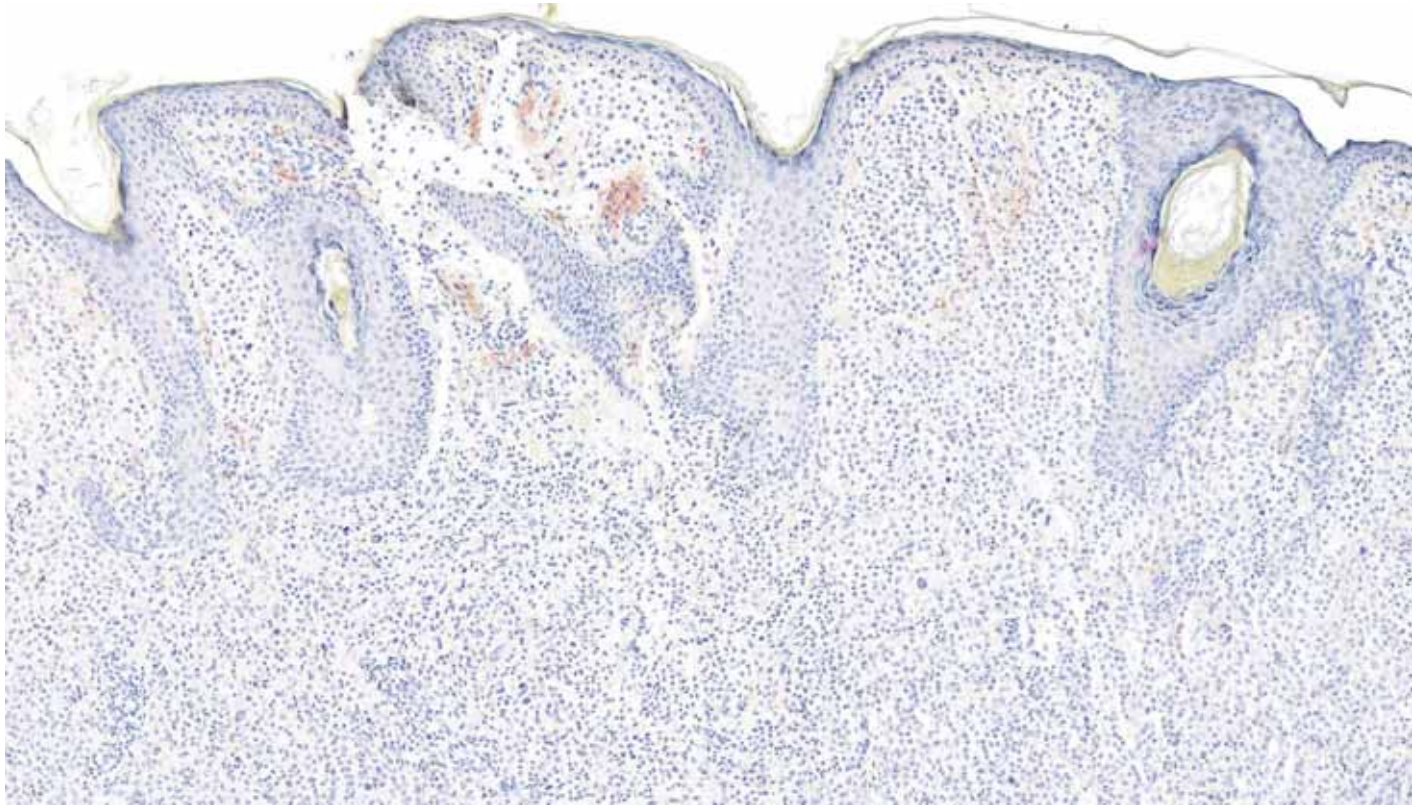
BRAF V600E



PRAME



PRAME



Final diagnosis

- Nevoid Melanoma ex-nevus
- Abnormal immunophenotype
- BRAF V600E

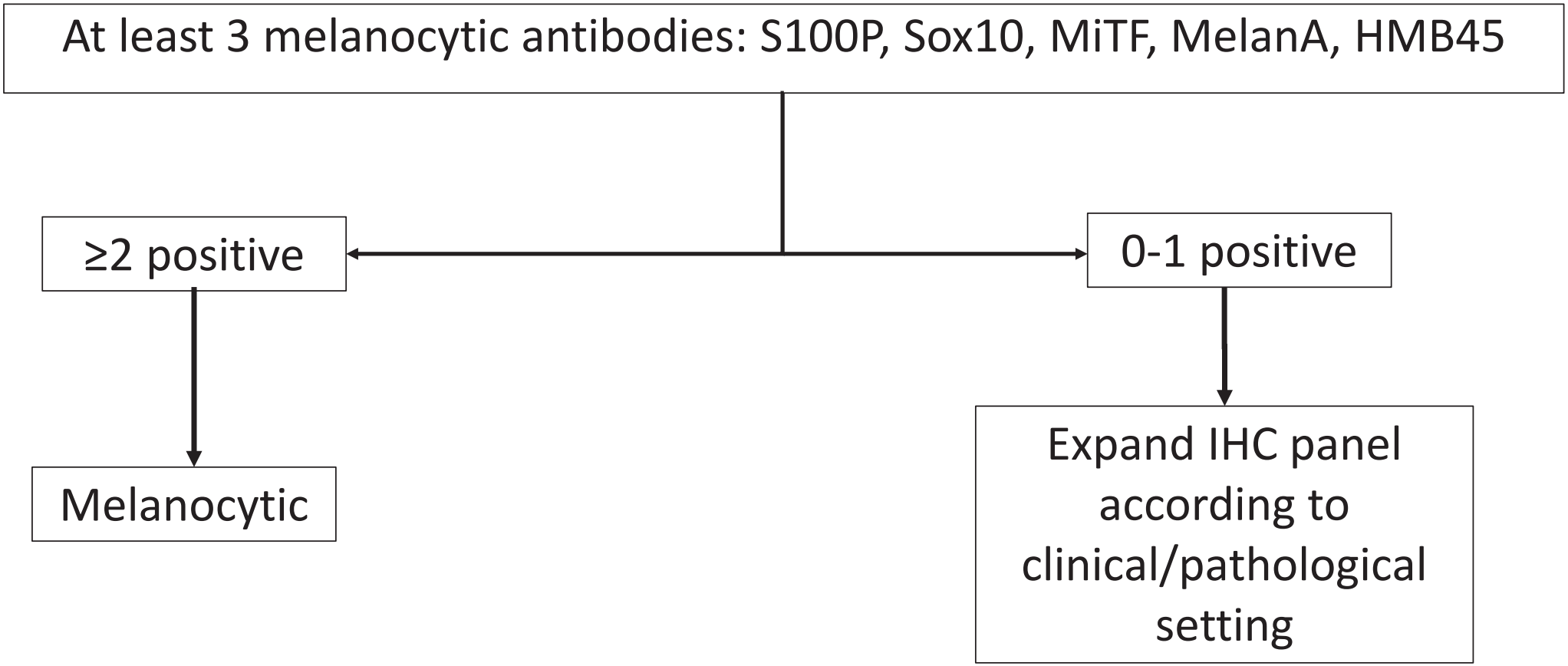
Take home messages

- IHC is a powerful tool that relies on the careful choice of antibodies adapted to a specific situation

Take home messages

- IHC is a powerful tool that relies on the careful choice of antibodies adapted to a specific situation
- To confirm melanocytic lineage always perform a panel of antibodies (S100 Protein mandatory)

Typical algorythms for IHC use Melanocytic?



Take home messages

- IHC is a powerful tool that relies on the careful choice of antibodies adapted to a specific situation
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Typical algorythms for IHC use Benign Vs Malignant?

+:
WAM, PEM or Blue morphology?

Normal immunophenotype: Very low chance of malignancy

P16 alteration: further algorythm

Fully altered Immunophenotype: Rule out nevoid melanoma

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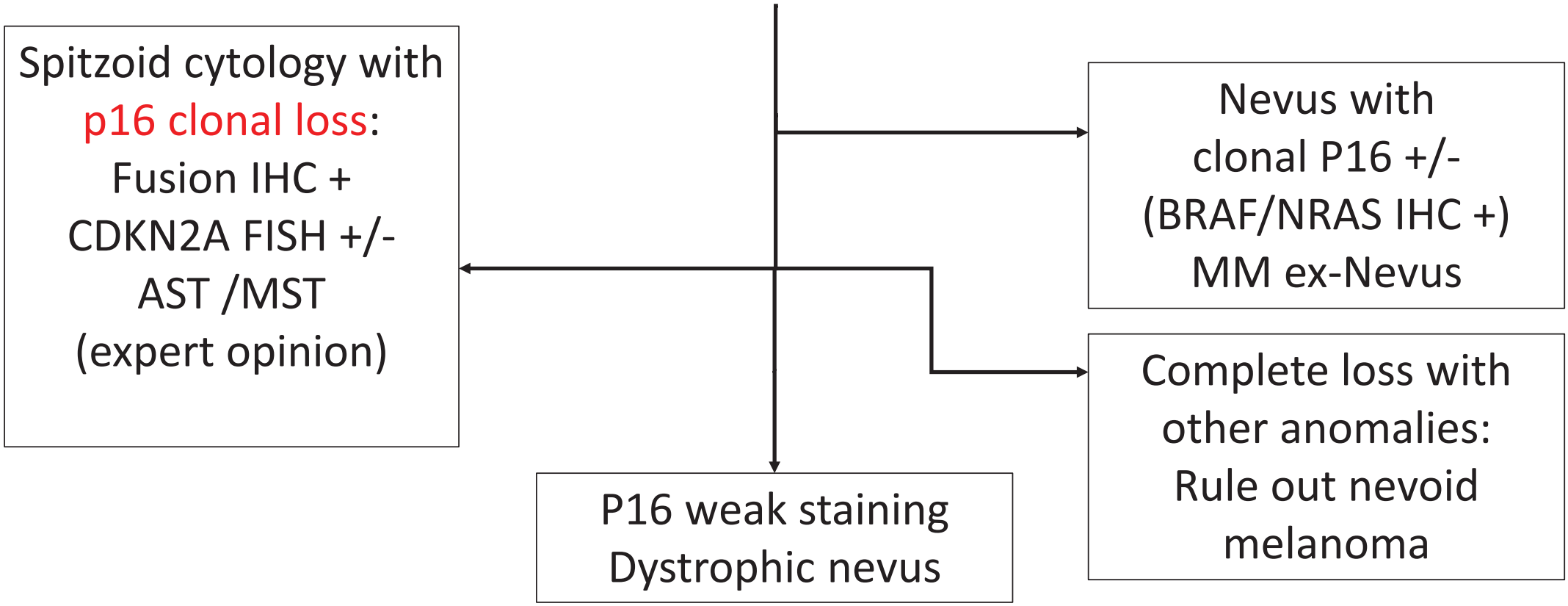
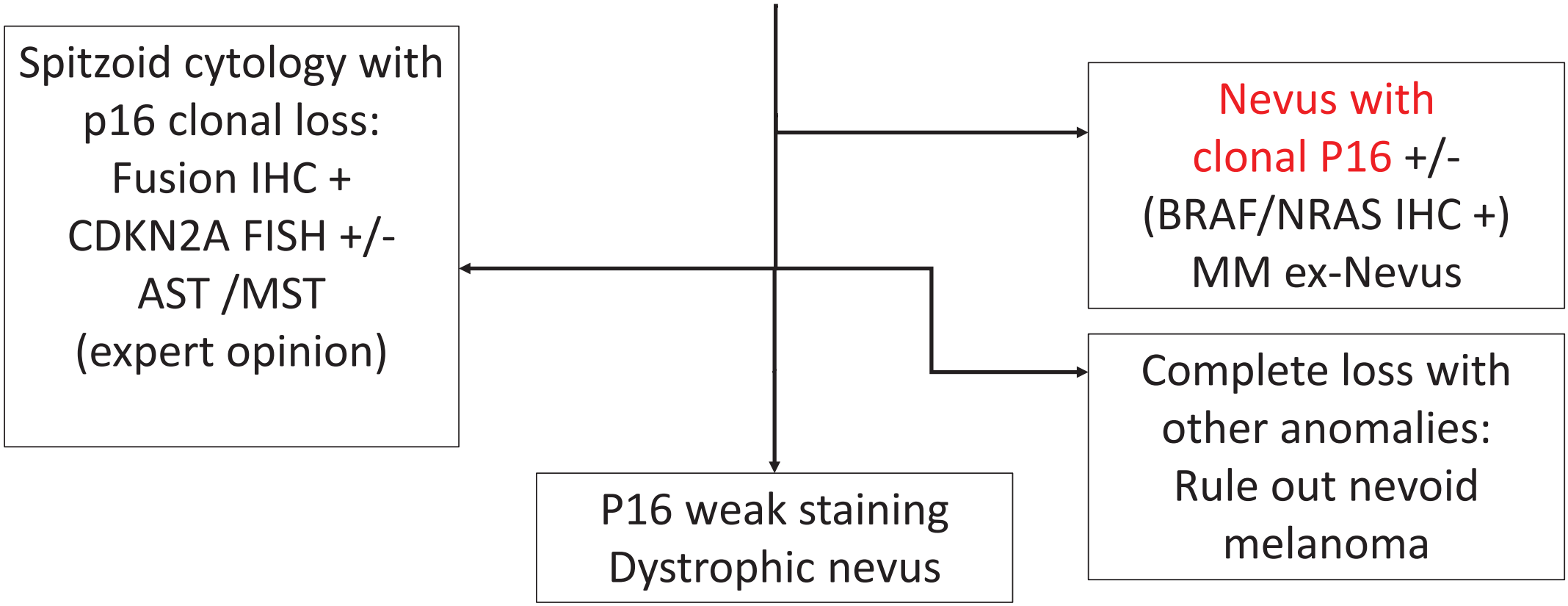
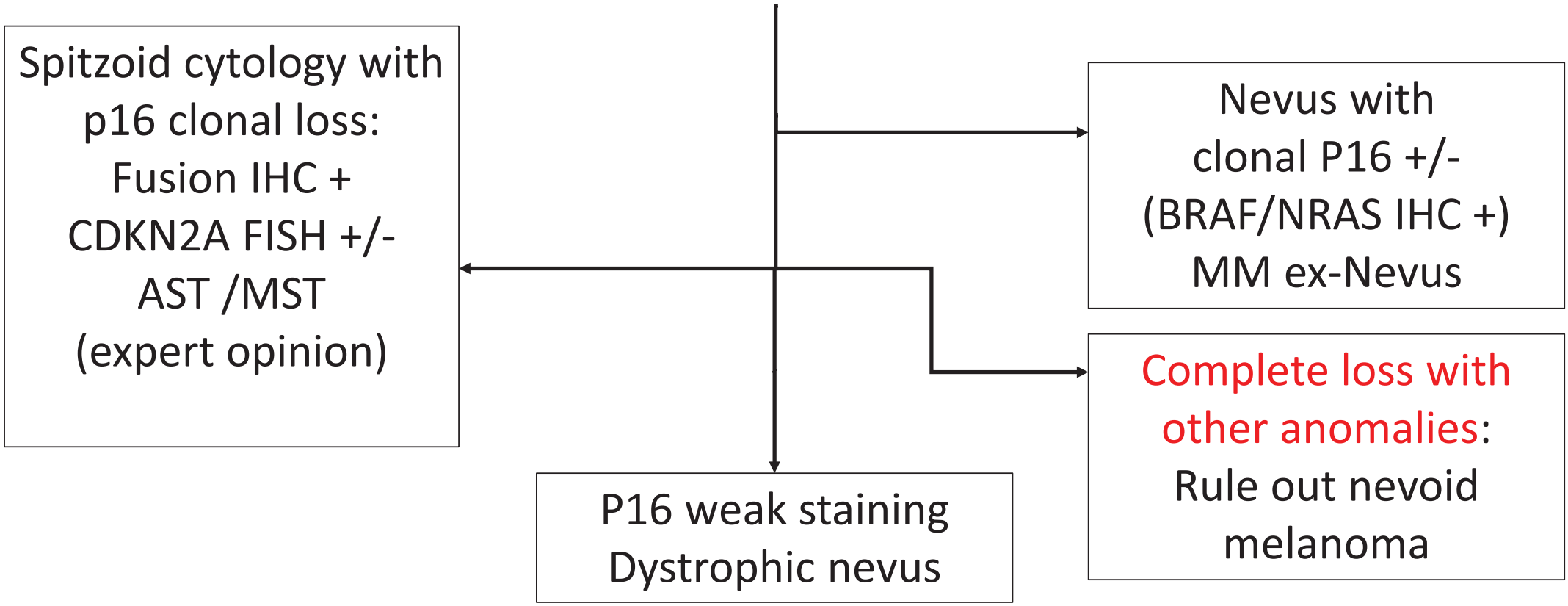
Typical algorythms for IHC use P16 alteration



Typical algorythms for IHC use P16 alteration



Typical algorythms for IHC use P16 alteration



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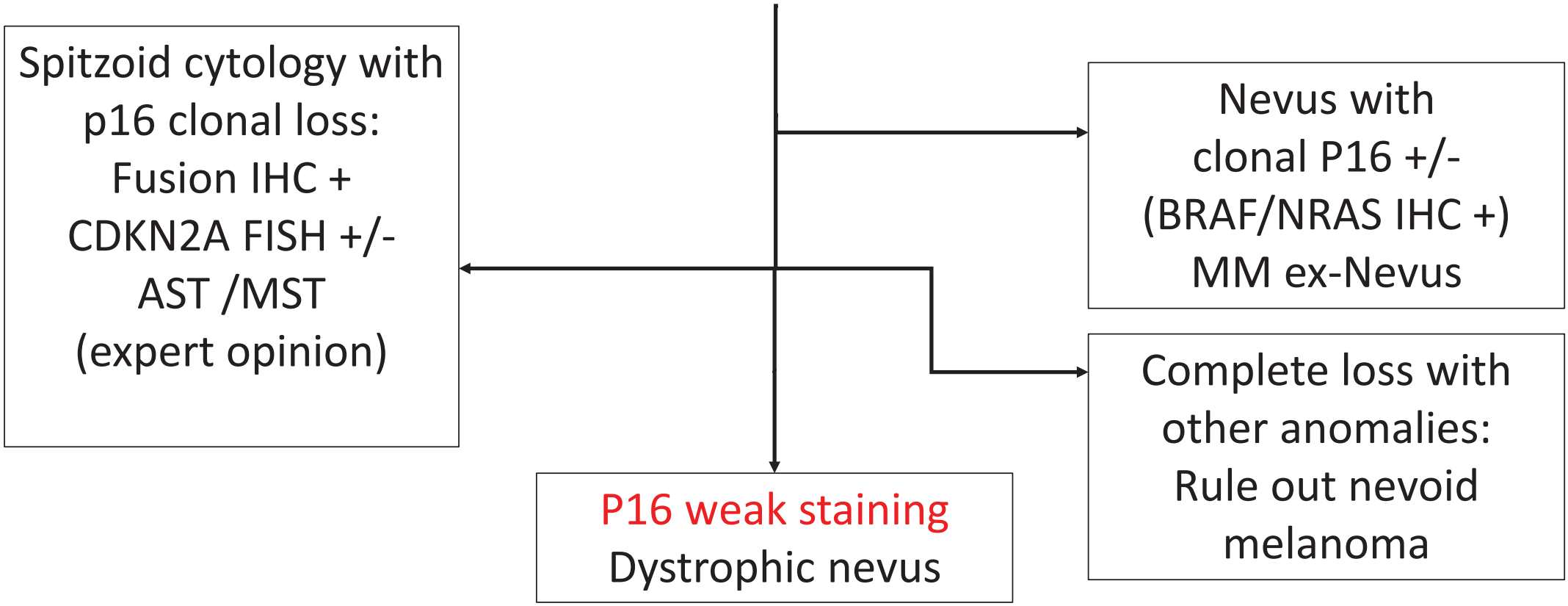

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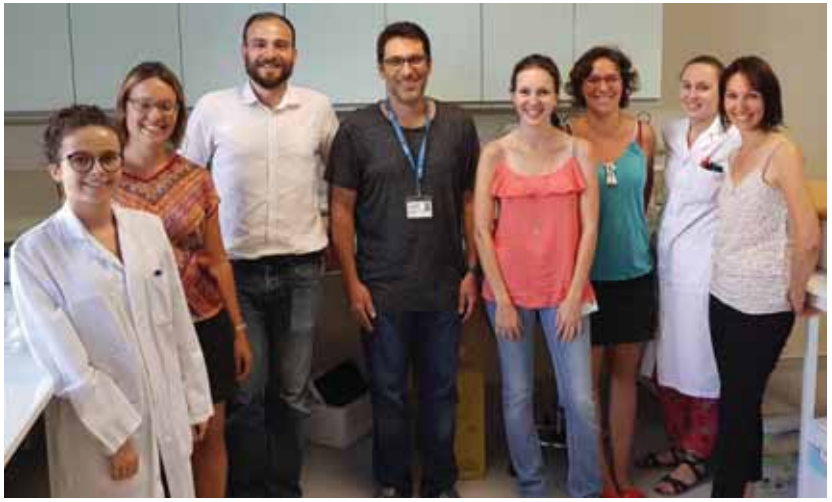
- IHC is a potential molecular screening tool

Thanks to all my staff



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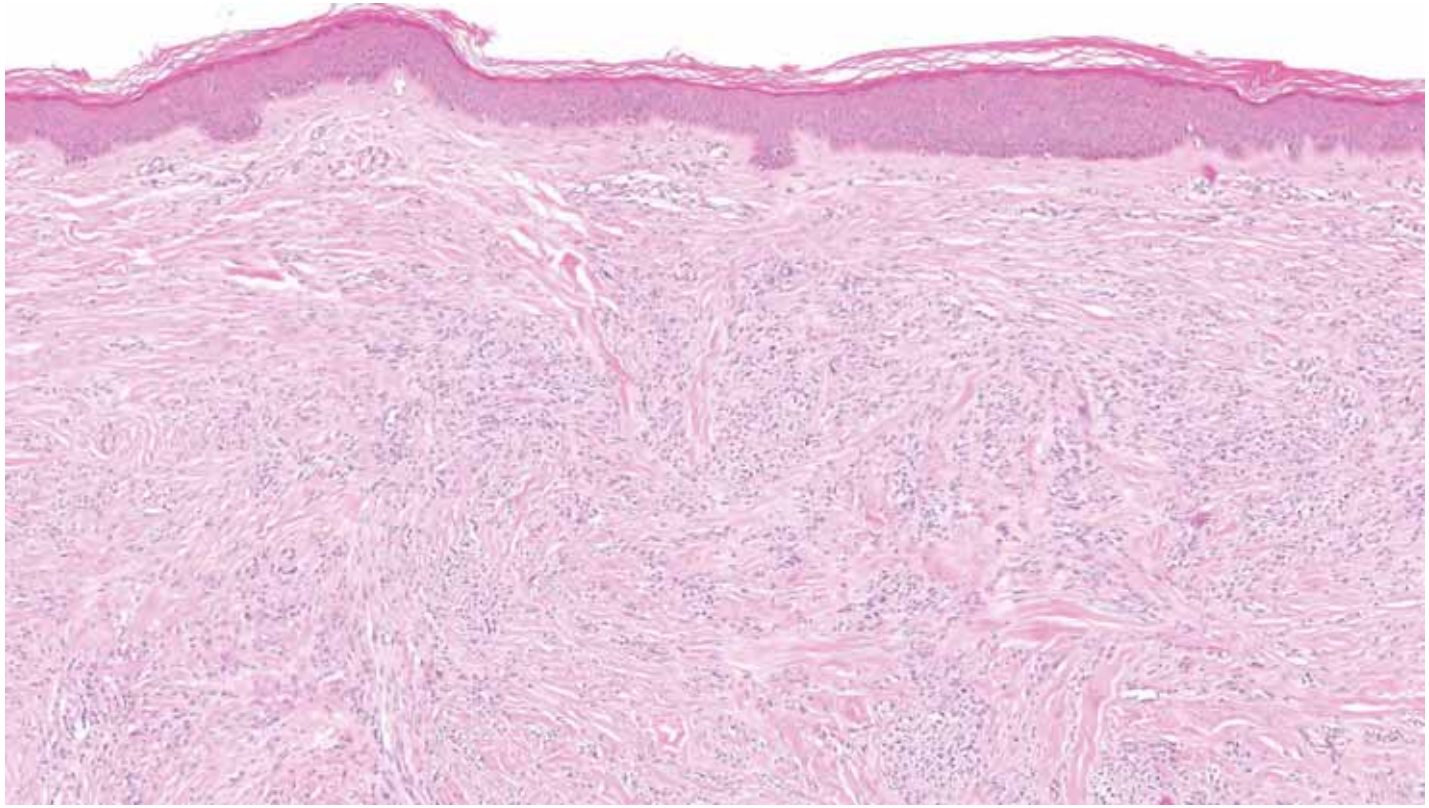
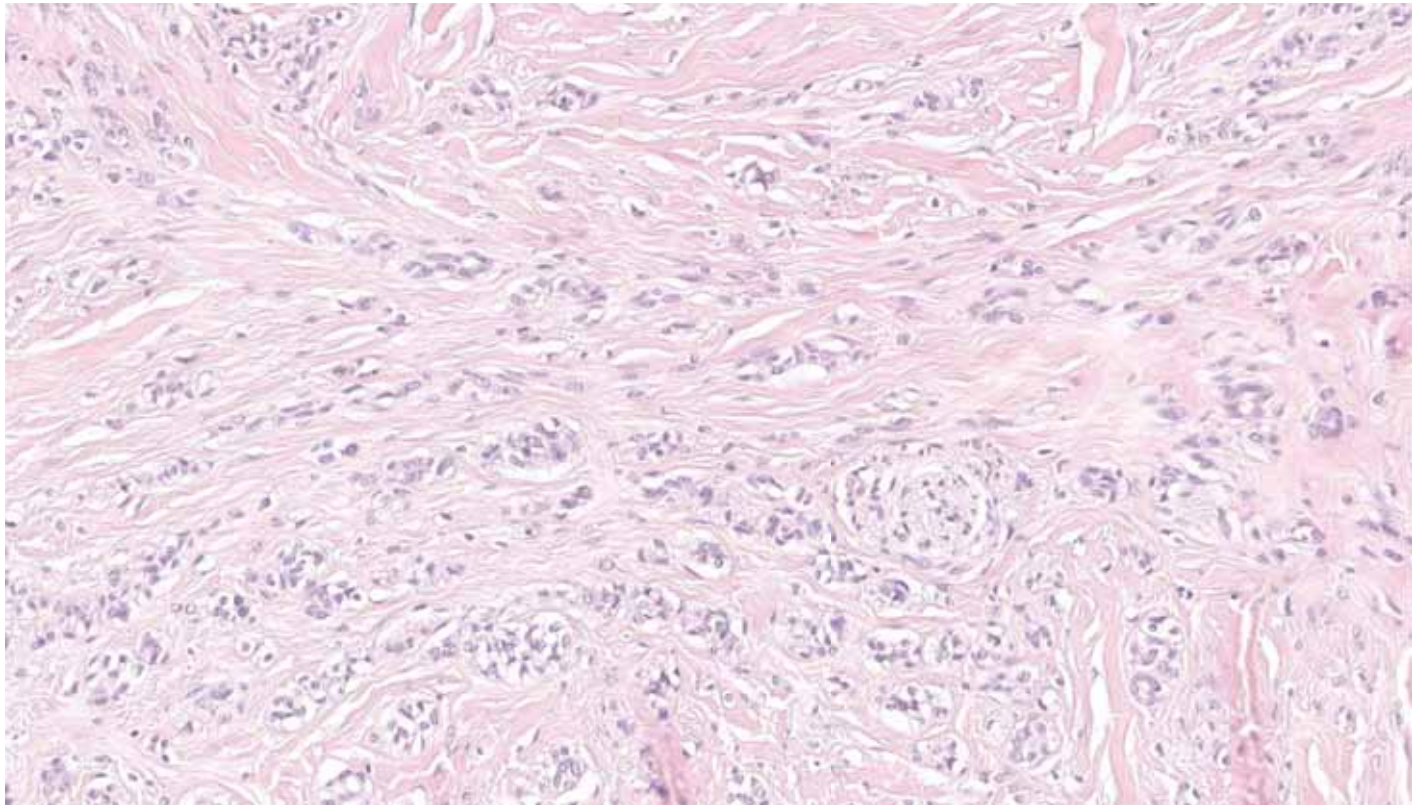
- Follow #NonAIPath hashtag for more videos

M20 Buttocks sent as ALK fused TSA

M20 Buttocks sent as ALK fused TSA

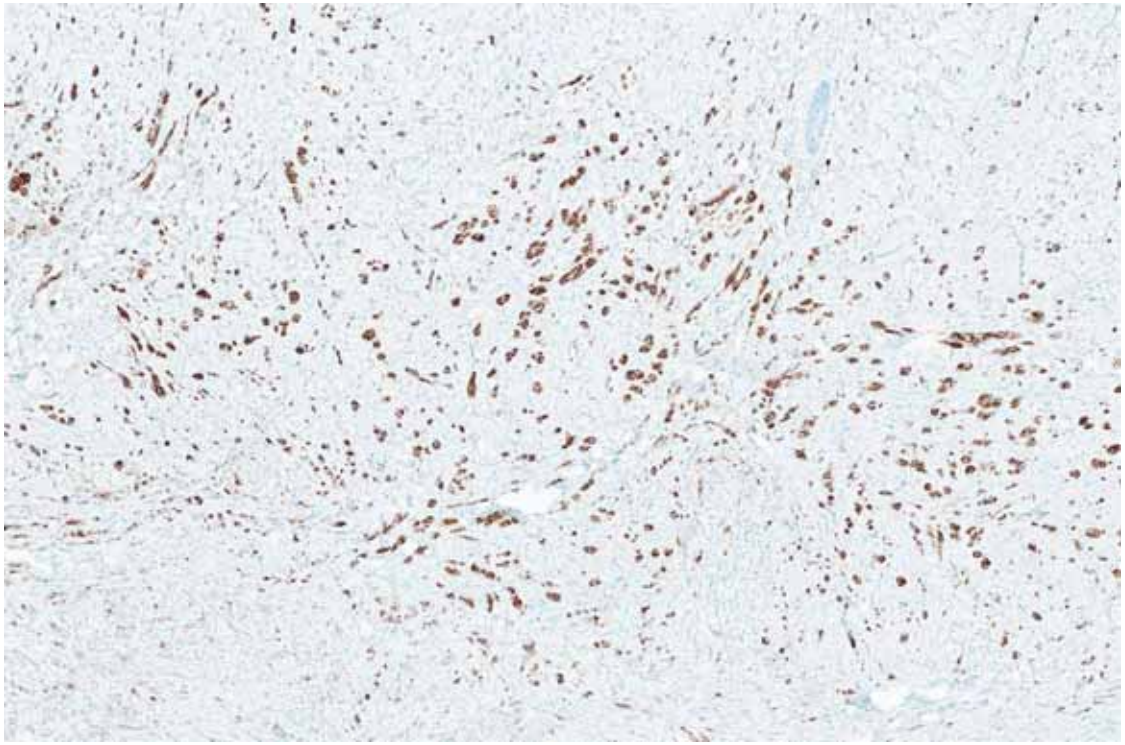


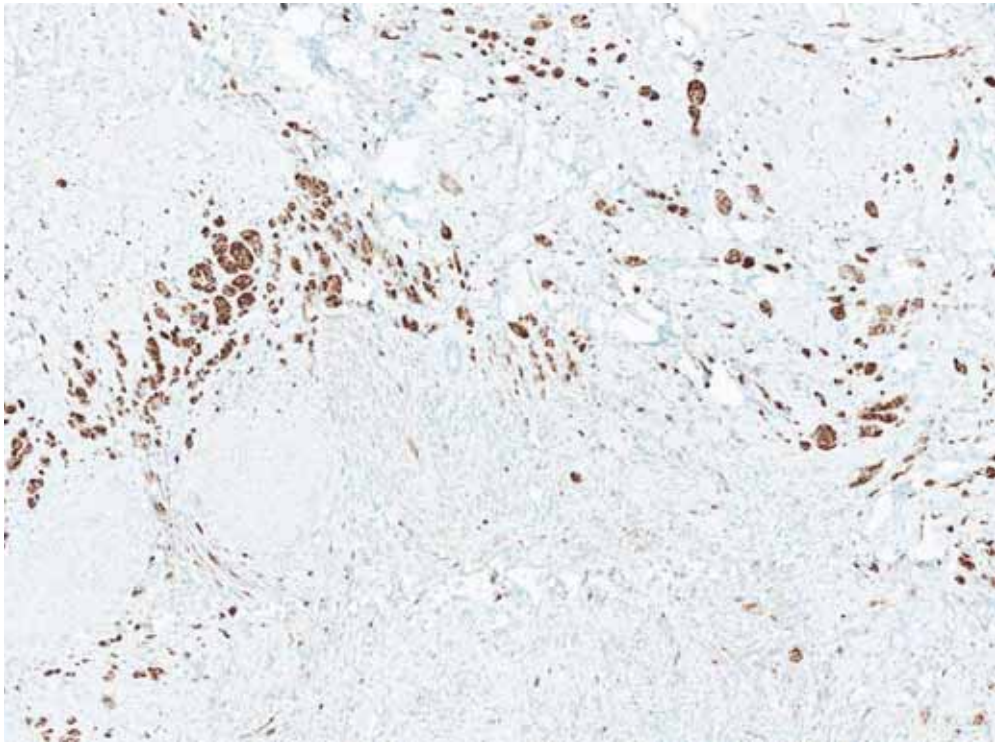
M20 Buttocks sent as ALK-fused AST



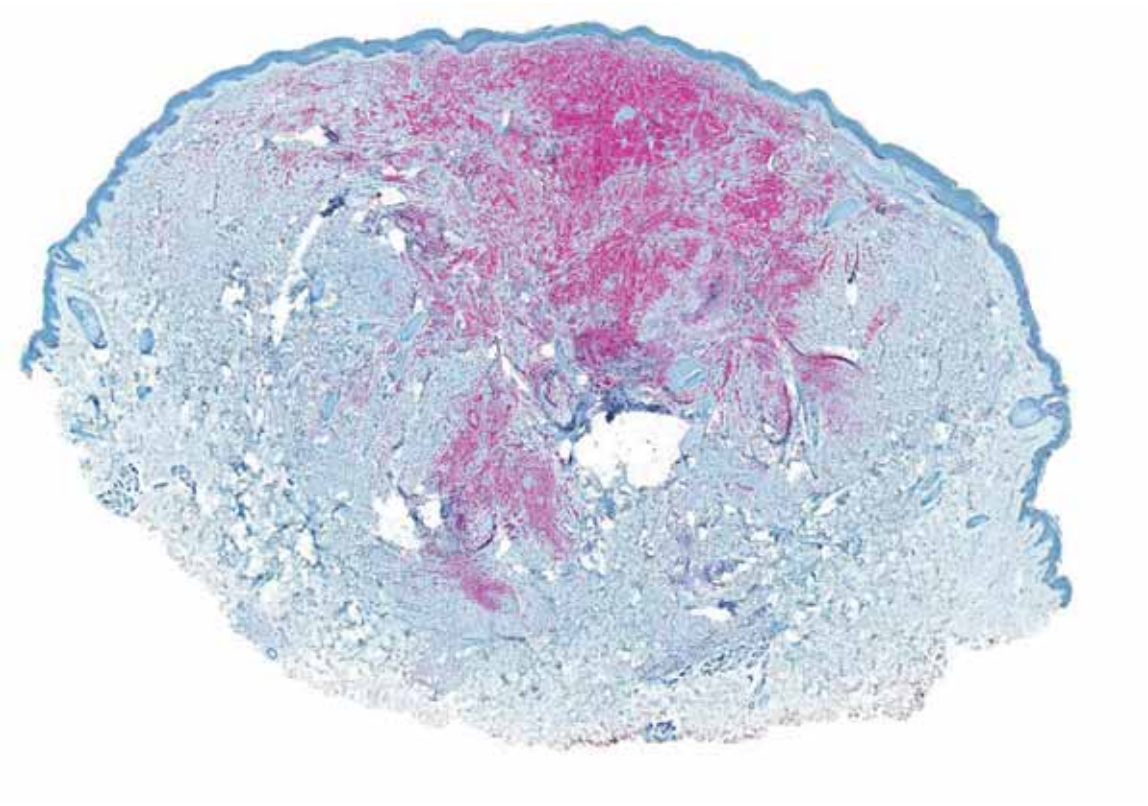
M20 Buttocks sent as ALK fused TSA

ALK IHC

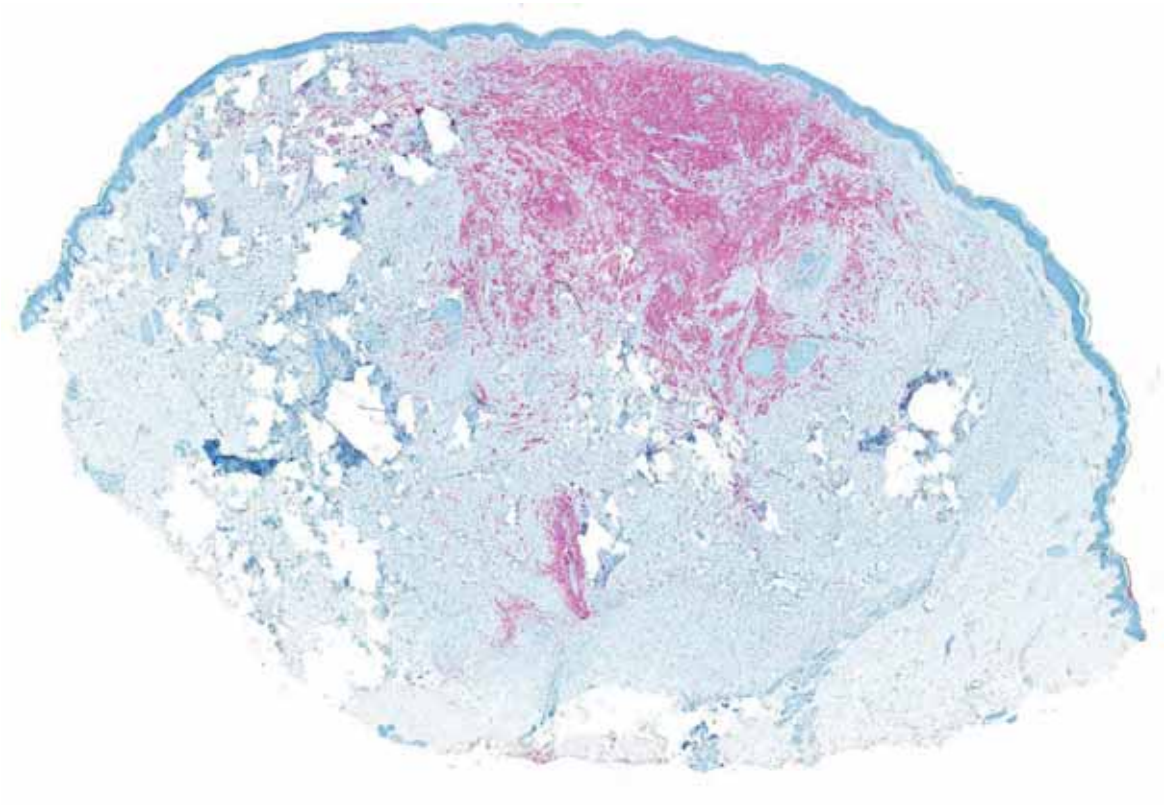




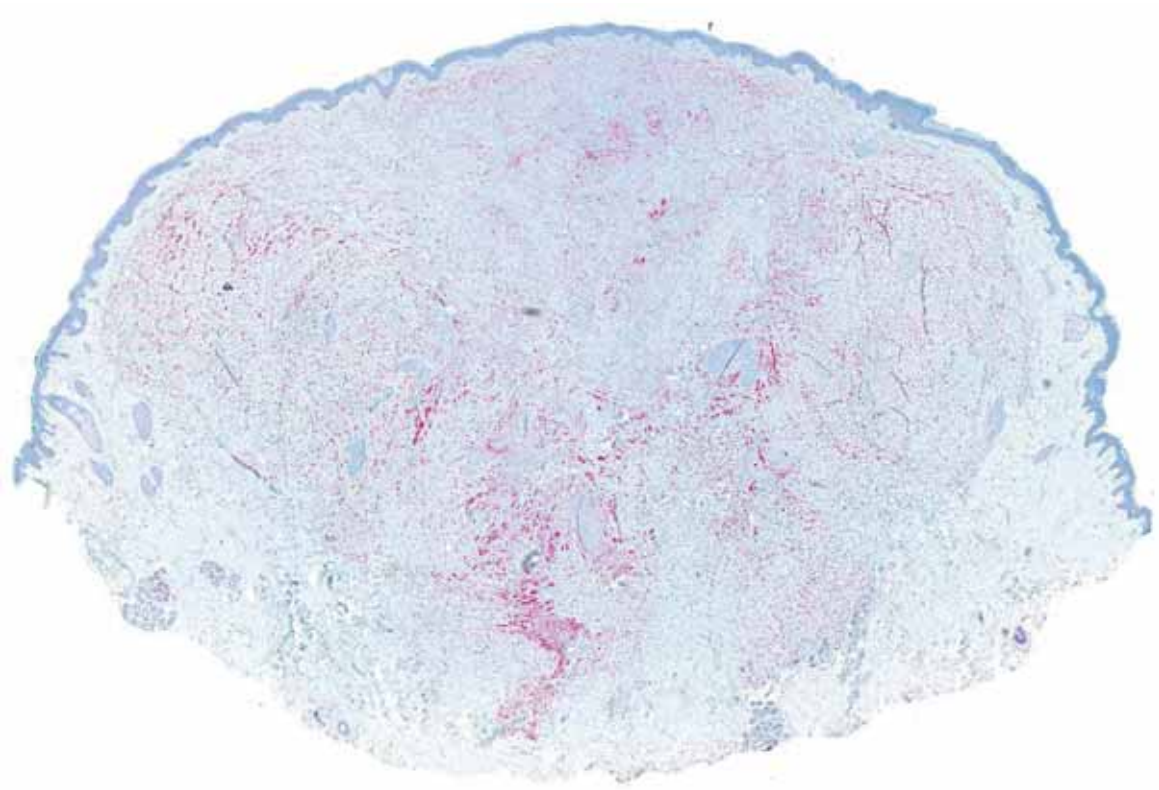
MelanA IHC



HMB45 IHC



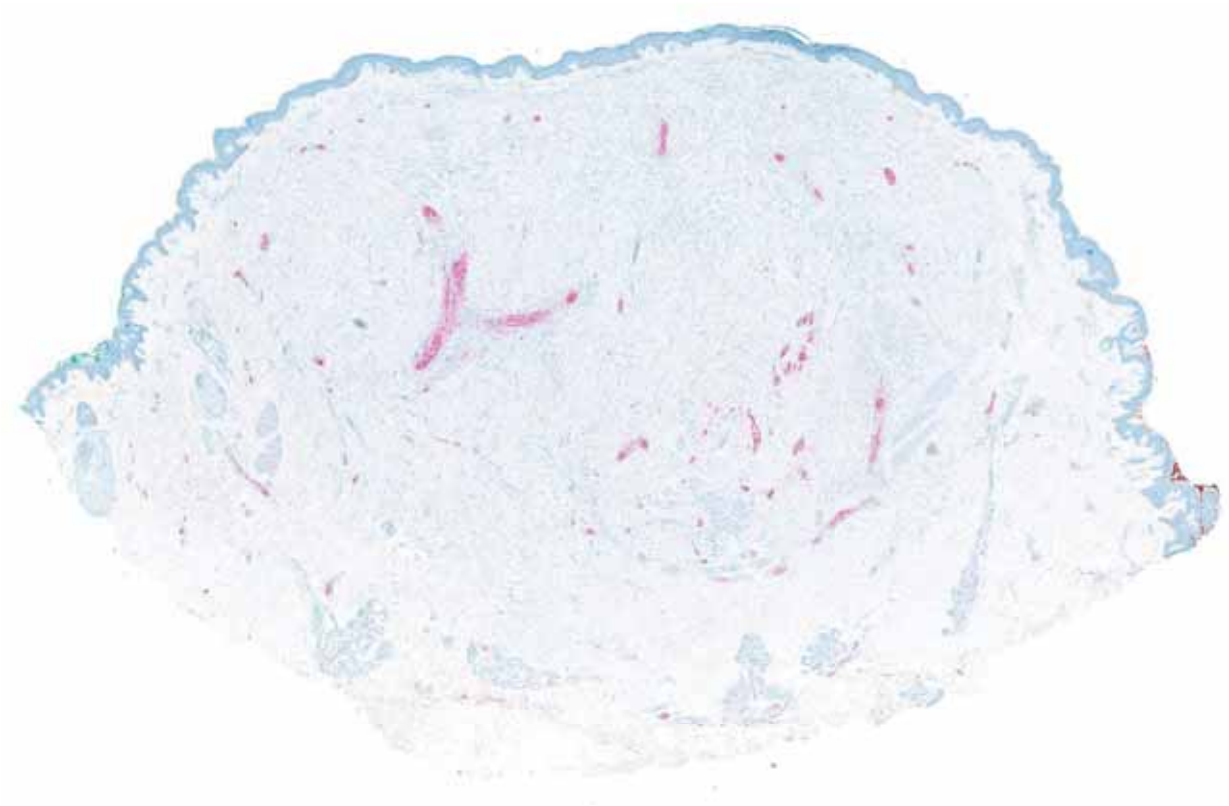
P16 IHC



Ki67 IHC

Which Ab to perform?

S100P IHC



Final diagnosis

- Unpigmented fibrosing blue naevus
- Age
- Location S100P loss
- Irregular ALK false positive expression

ALK IHC re-done