CASE REPORT

28. 6. 2019 Senec, Slovensko

MARTINA BANĚČKOVÁ
NIKOLA PTÁKOVÁ
ALENA SKÁLOVÁ

KLINICKÉ ÚDAJE

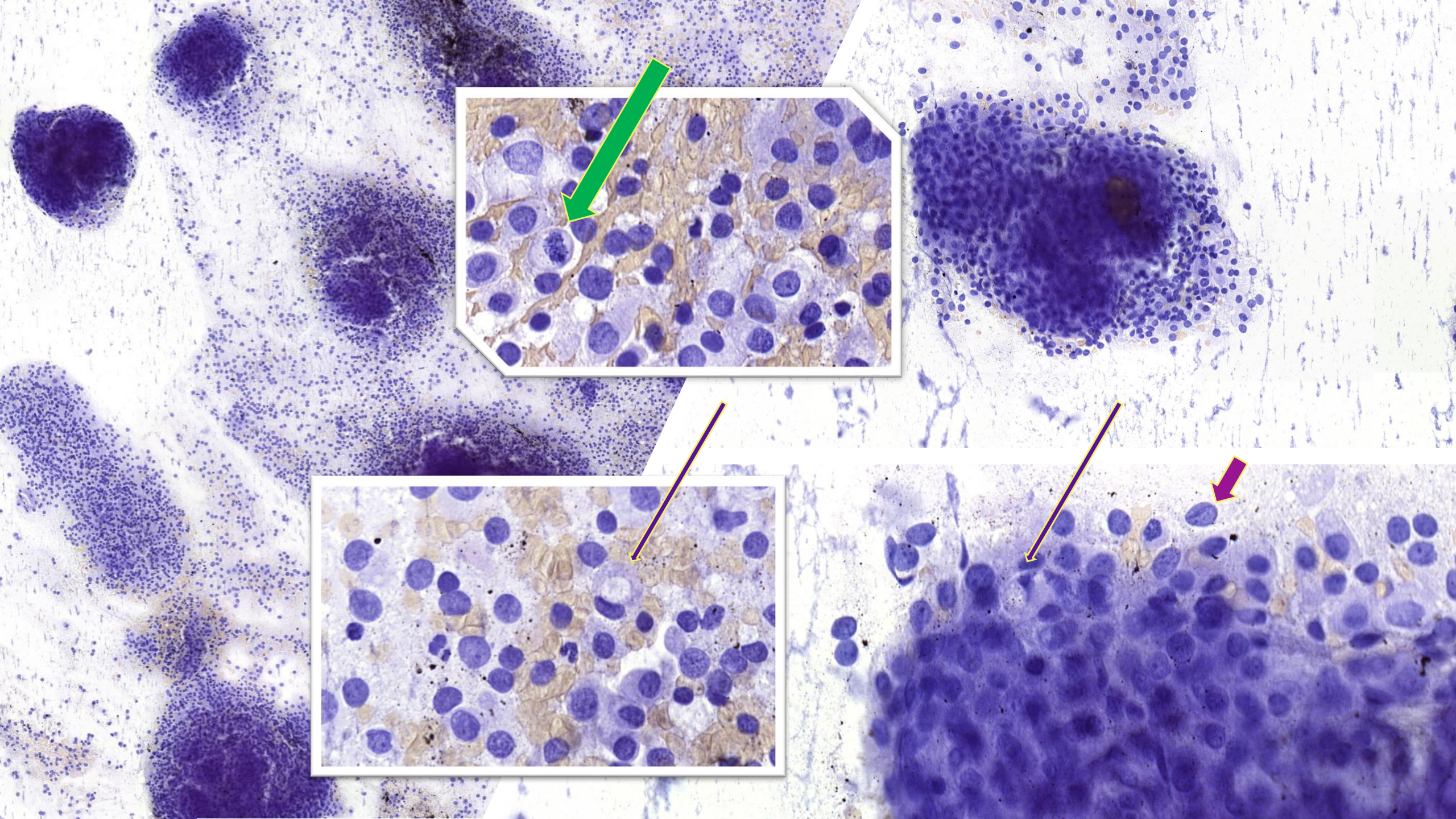
Konzultační vyšetření

1. EXCIZE v roce 2014 – muž, 56 let

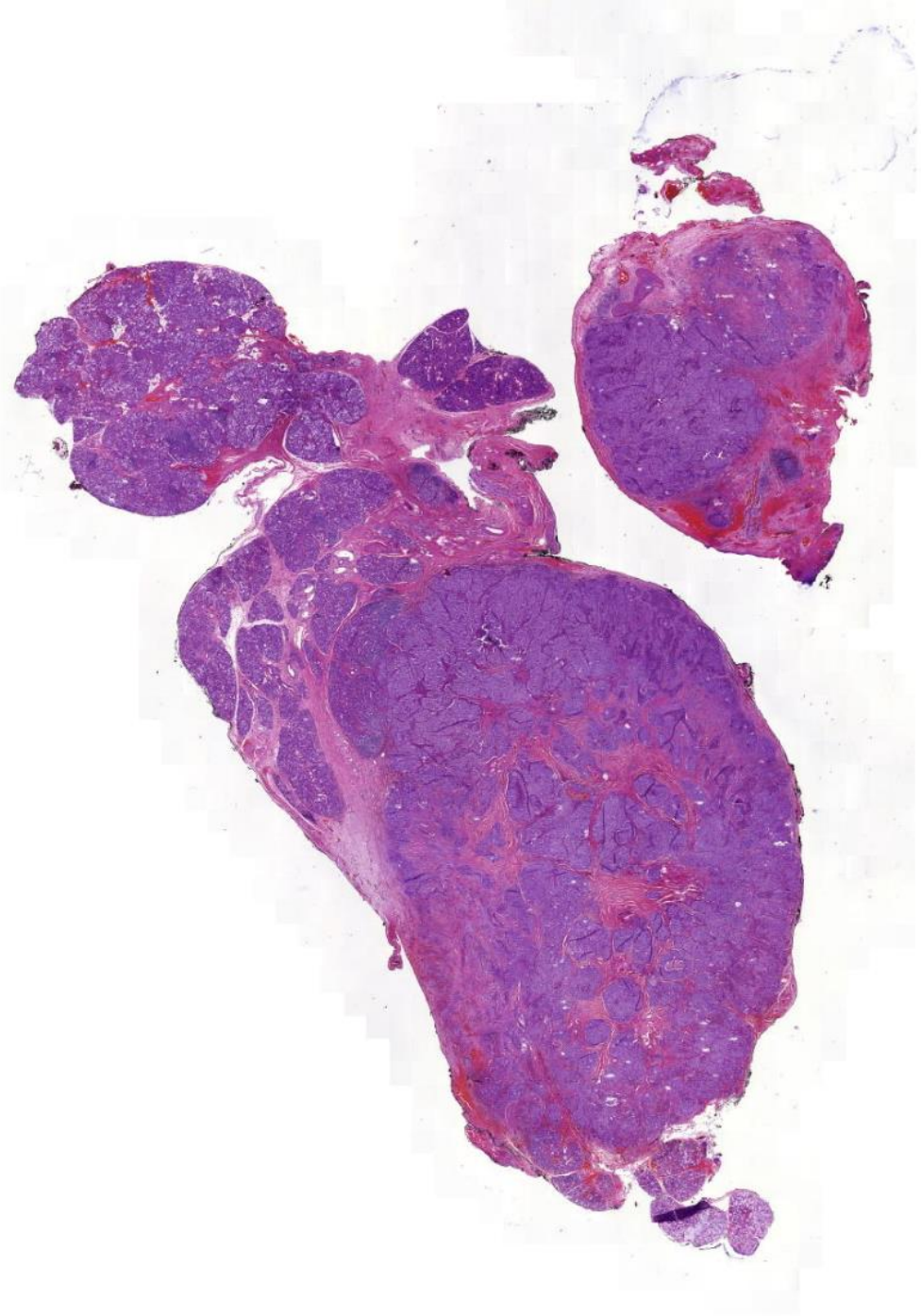
- Excize submandibulární žlázy 4 x 3 x 1,5 cm, váha – 10 g
- Na řezu: lobulární struktura, hnědočervené barvy, tumor rozměrů 1,9 x 1,5 x 1 cm
- Dg. pravděpodobně primární tumor – dif. dg. Mukoepidermoidní karcinom, Acinický karcinom či Salivární duktální karcinom

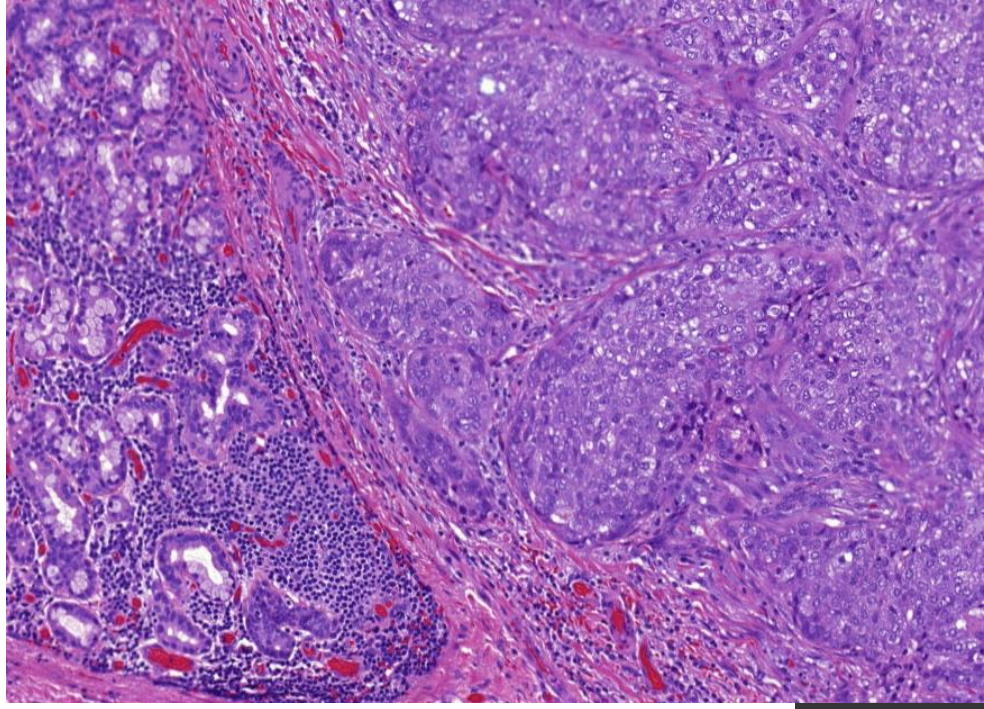
2. Recidiva po 4 letech s metastázou do jedné intraparotideální lymfatické uzliny

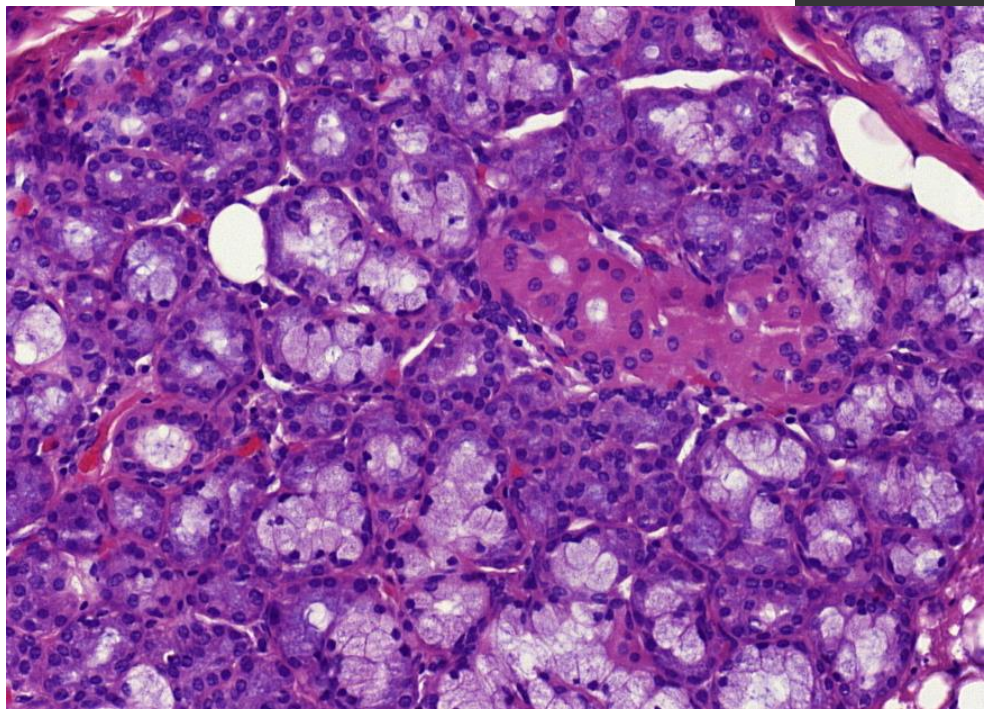
CYTOLOGICKÉ VYŠETŘENÍ

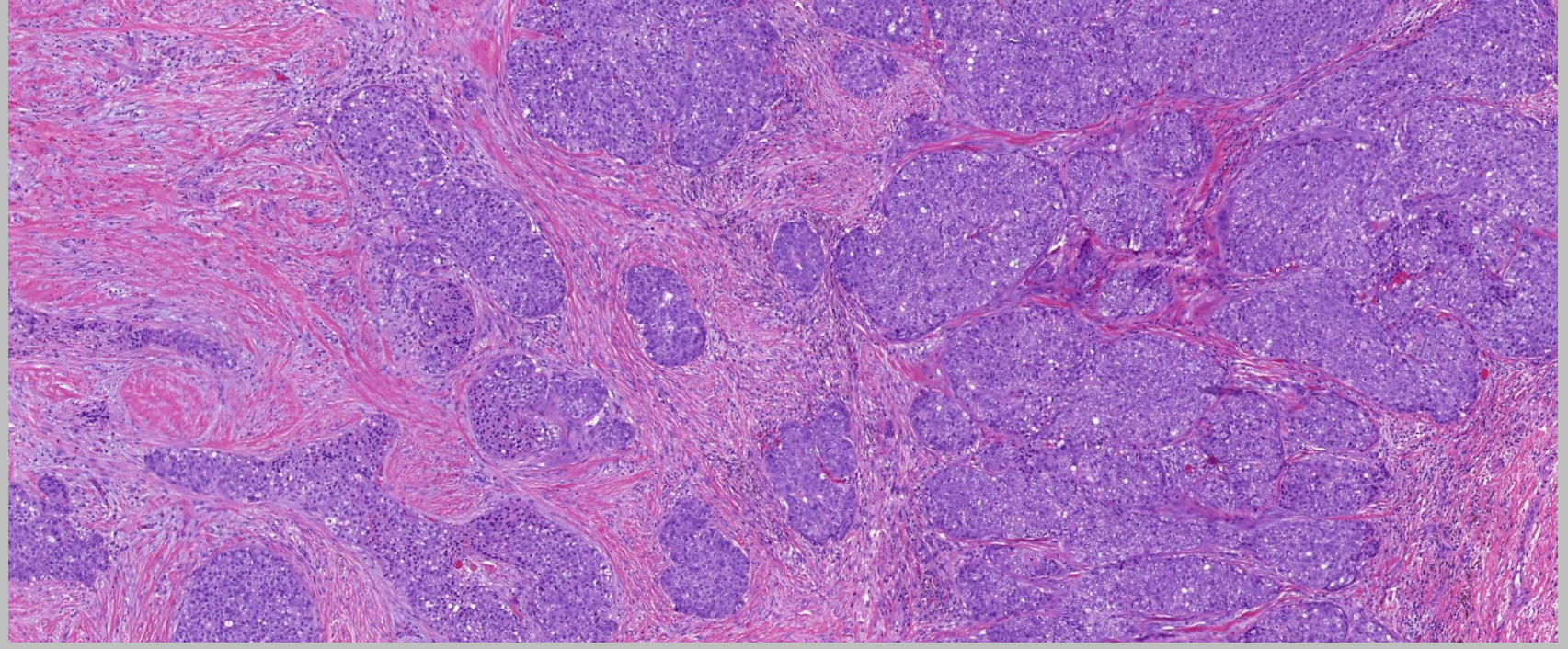


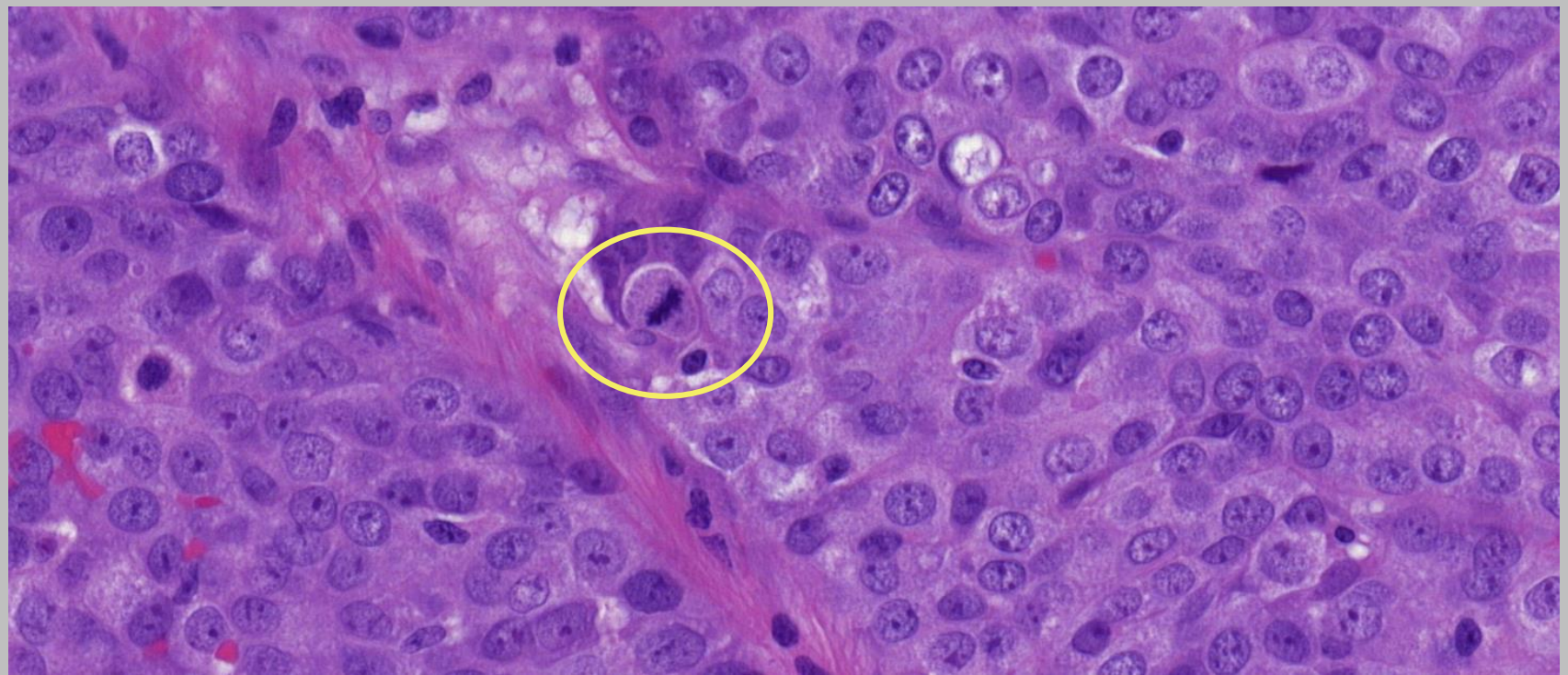
MORFOLOGIE

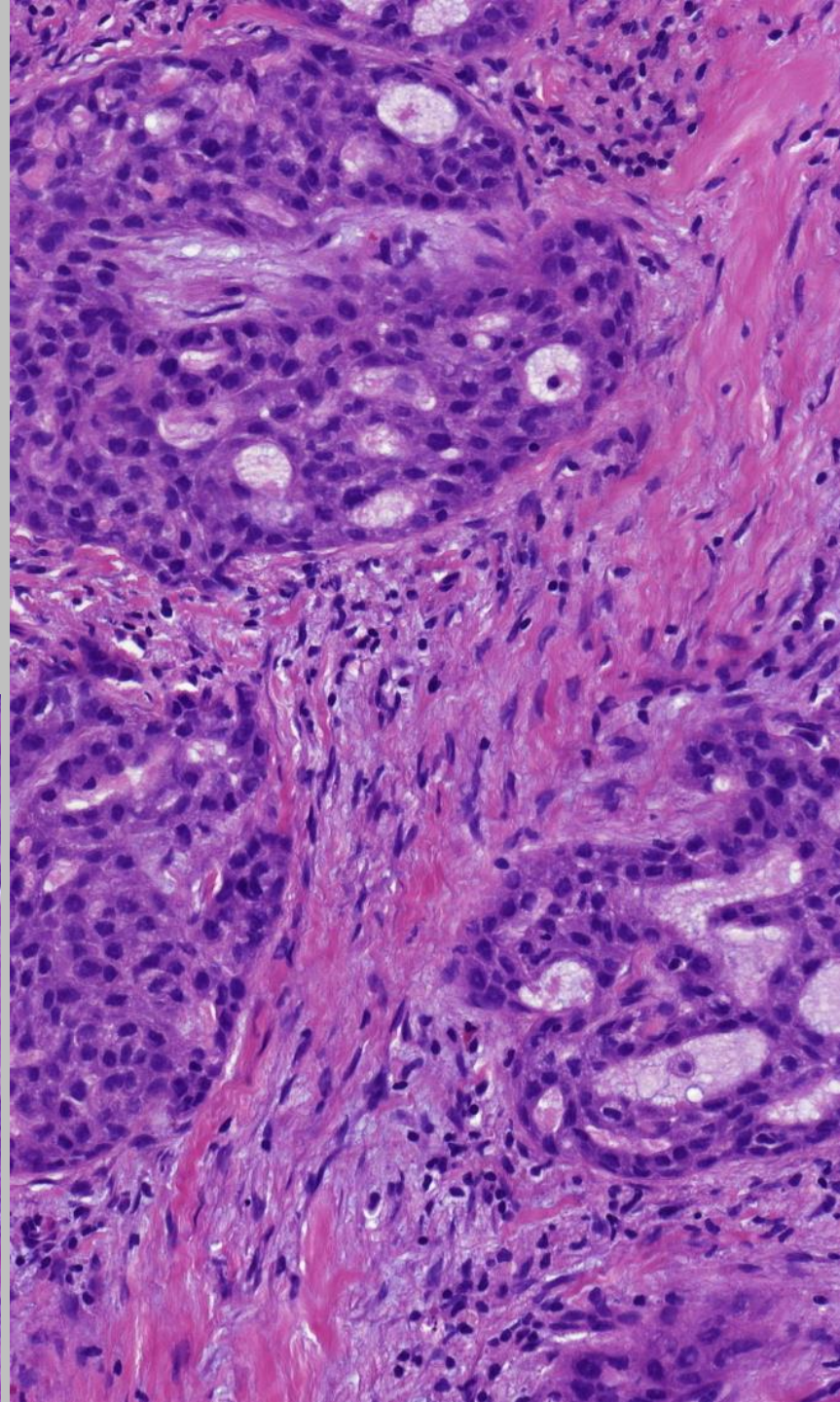


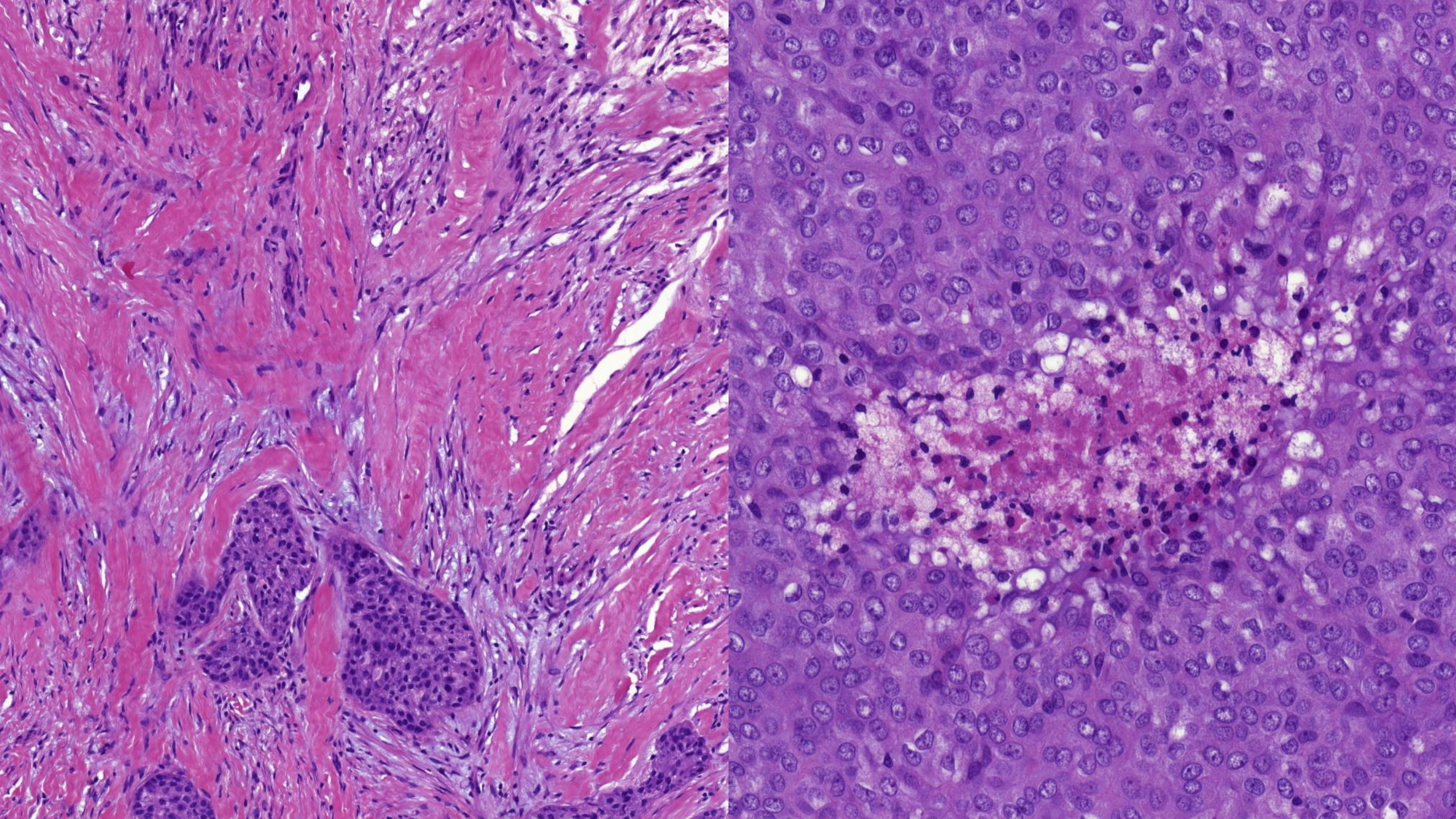




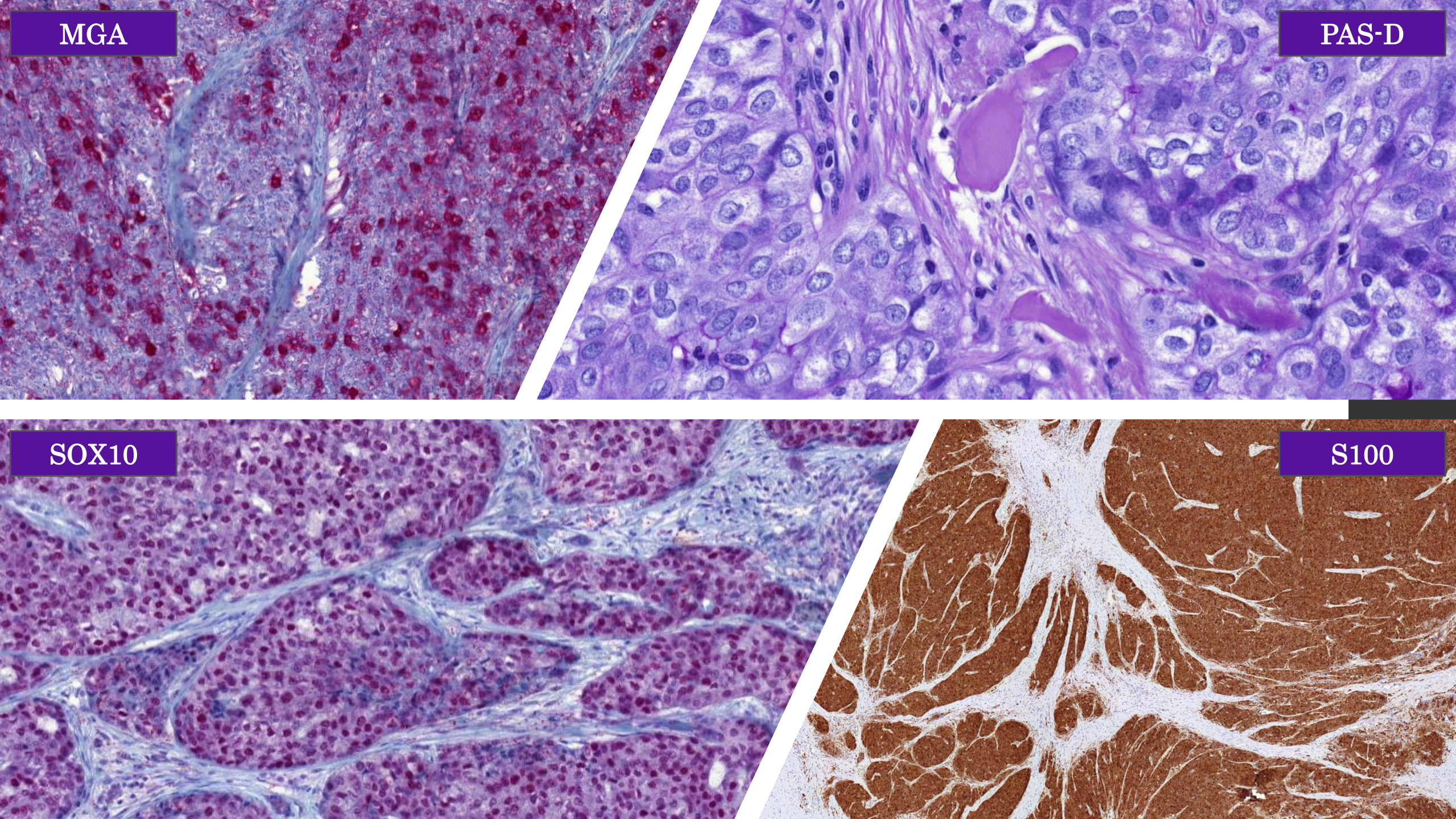


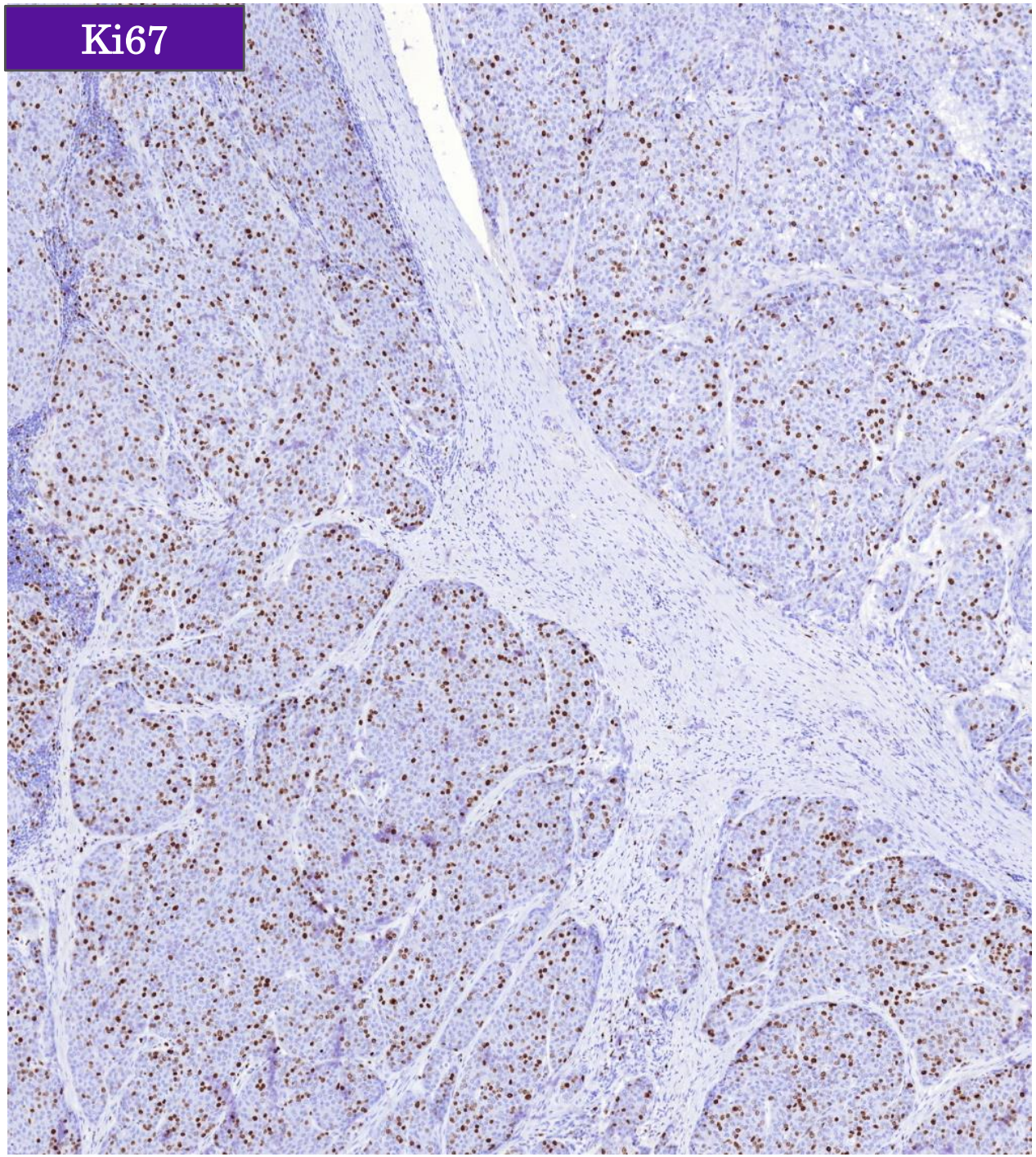


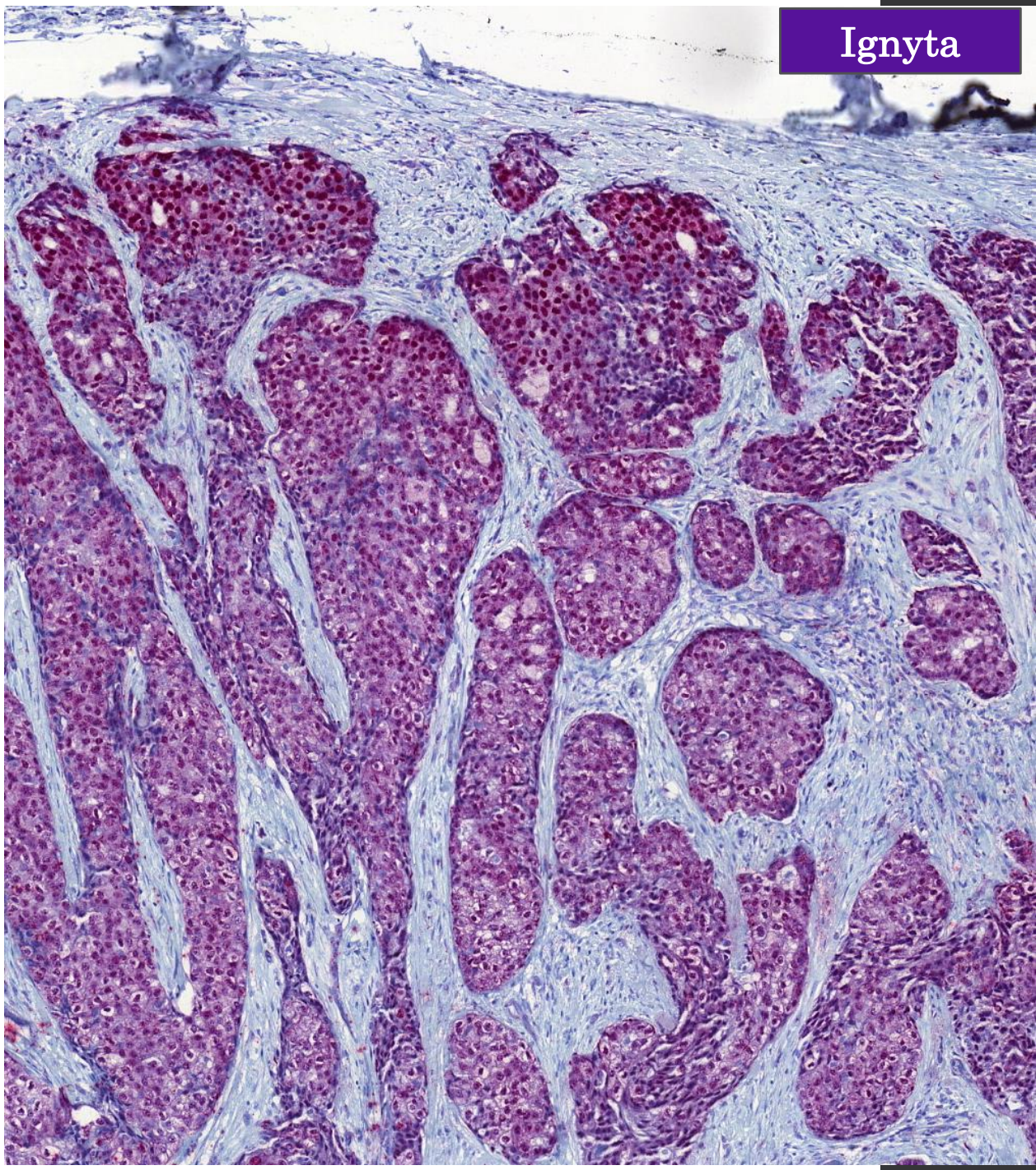




IMUNOHISTOCHEMICKÉ VYŠETŘENÍ


MGA
PAS-D
SOX10
S100


Ki67


Ignyta

GENETICKÉ VYŠETŘENÍ

Fúze t(12;15) *ETV6-NTRK3* a t(9;3)*MYB-SMR3B*

- **ETV6** – **E-T**wenty-**S**ix family – transkripční faktor – regulátor transkripce, buněčné proliferace, diferenciace a tumorigeneze
- **NTRK3 (gen)** kóduje TRKC receptor = neurotrophic tropomyosin receptor kinase, type 3
- **MYB** – (**my**elo**b**lastosis) transkripční faktor
- **SMR3B** – **s**ub**m**axillary gland androgen **r**egulated protein **3B**

HIGH-GRADE SEKREČNÍ KARCINOM

s metastázou do 1 LU

DŘÍVE MASC

Mammary Analogue Secretory Carcinoma of Salivary Glands, Containing the *ETV6-NTRK3* Fusion Gene: A Hitherto Undescribed Salivary Gland Tumor Entity

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Abstract: We present a series of 16 salivary gland tumors with histomorphologic and immunohistochemical features reminiscent of secretory carcinoma of the breast. This is a hitherto undescribed and distinctive salivary gland neoplasm, with features resembling both salivary acinic cell carcinoma (AciCC) and low-grade cystadenocarcinoma, and displaying strong similarities to breast secretory carcinoma. Microscopically, the tumors have a lobulated growth pattern and are composed of microcystic and glandular spaces with abundant eosinophilic homogenous or bubbly secretory material positive for periodic acid-Schiff, mucicarmine, MUC1, MUC4, and mammaglobin. The neoplasms also show strong vimentin, S-100 protein, and STAT5a positivity. For this tumor, we propose a designation mammary analogue secretory carcinoma of salivary glands (MASC). The 16 patients comprised 9 men and 7 women, with a mean age of 46 years (range 21 to 75). Thirteen cases occurred in the parotid gland, and one each in the minor salivary glands of the buccal mucosa, upper lip, and palate. The mean size of the tumors was 2.1 cm (range 0.7 to 5.5 cm). The duration of symptoms was recorded in 11 cases and ranged from 2 months to 30 years. Clinical follow-up was available in 13 cases, and ranged from 3 months to 10 years. Four patients suffered local recurrences. Two patients died, 1 of them owing to multiple local recurrences with extension to the temporal bone, and another owing to metastatic dissemination to cervical lymph nodes, pleura, pericardium, and lungs. We have shown a t(12;15) (p13;q25) *ETV6-NTRK3* translocation in all but one case of MASC suitable for analysis. One case was not analyzable and another was not available for testing. This translocation was not found in any conventional salivary AciCC (12 cases), nor in other tumor types including pleomorphic adenoma (1 case) and low-grade cribriform cystadenocarcinoma (1 case), whereas *ETV6-NTRK3* gene rearrangements were proven in all 3 tested cases of mammary secretory carcinoma. Thus, our results strongly support the concept that MASC and AciCC are different entities.

Key Words: salivary gland, acinic cell carcinoma, secretory carcinoma, mammary type, molecular pathology, *ETV6-NTRK3* translocation

(*Am J Surg Pathol* 2010;34:599–608)

SEKREČNÍ KARCINOM (SC)

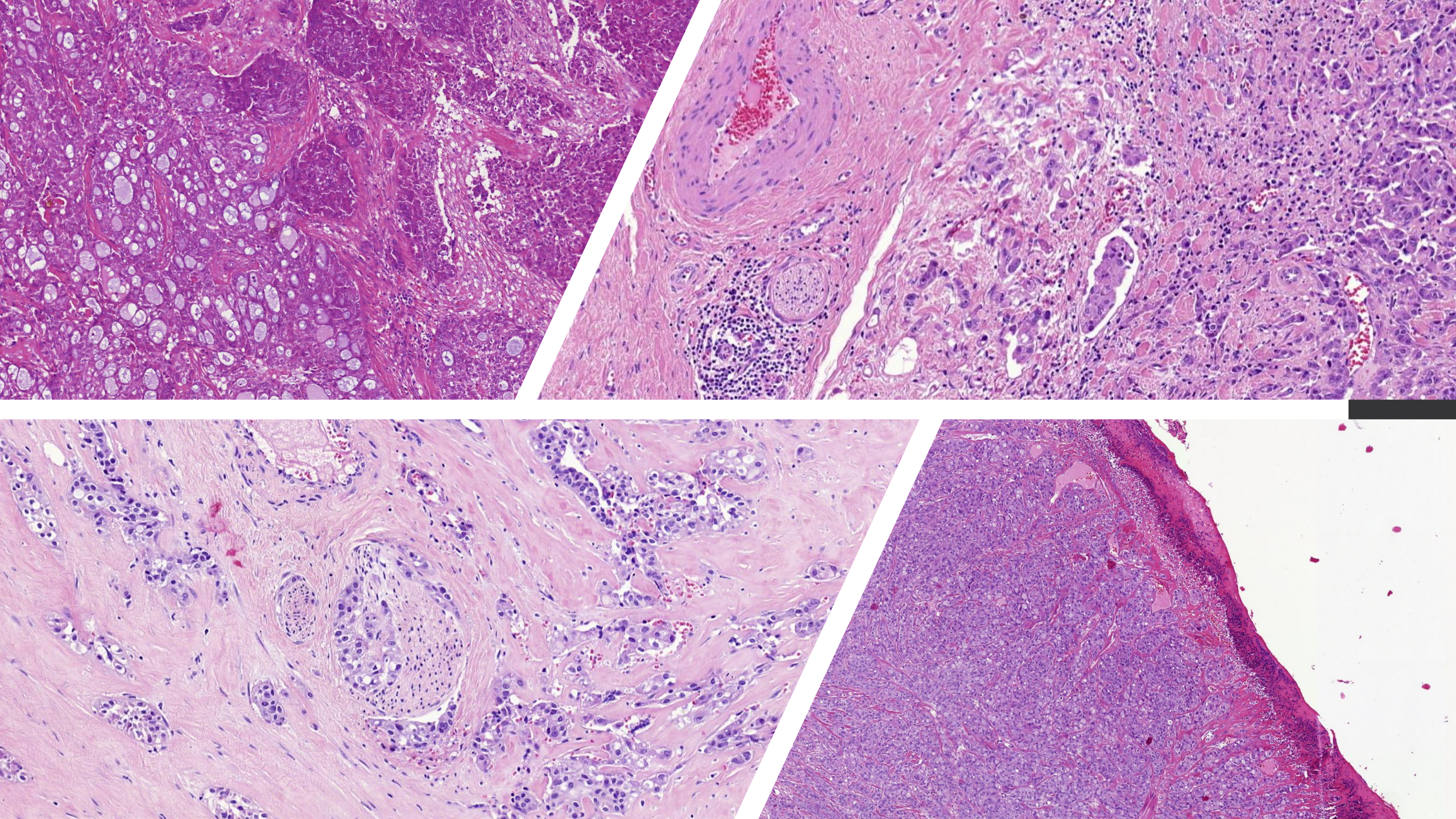
- Low grade karcinom podobný sekrečnímu karcinomu prsu – dobrá prognóza
- **Imunoprofil:**
  - CK7/S100/MMG a SOX10, GATA3 pozitivní
  - p63 a DOG1 negativní
- **Genetika:**
  - t(12;15) (p13;q25) *ETV6-NTRK3* fúze (95-98%)

Mammary Analogue Secretory Carcinoma of Salivary Glands With High-grade Transformation

Report of 3 Cases With the ETV6-NTRK3 Gene Fusion and Analysis of TP53, β-Catenin, EGFR, and CCND1 Genes

Alena Skálová, MD, PhD,† Tomas Vanecek, PhD,‡ Hanna Majewska, MD, PhD,§ Jan Laco, MD, PhD,‖ Petr Grossmann, PhD,‡ Roderick H. W. Simpson, MB, ChB, FRCPath,¶ Lukas Hauer, MD,# Pavel Andrle, MD,# Lubor Hosticka, MD,# Jindrich Branžovský, MD,* and Michal Michal, MD**

- 3 pacienti věk 36 – 73 let
- umírají od 2-6 let s diseminací primárního onemocnění
  - 2 případy *ETV6-NTRK3* v LG i HG komponentě
  - 1 případ ETV6 zlom v LG i HG komponentě
- !! Genetický profil!! – targeted terapie TRK inhibitory



t(12;15)(p13;q25) s *ETV6-NTRK3*

- NESPECIFICKÁ, 95-98%
- Sekreční karcinom prsu (juvenilní)
- Hematologické malignity – jako varianty AML
- Kongenitální fibrosarkom a kongenitální mesoblastický nefrom
- tzv. Černobylský karcinom – podtyp papilárního karcinomu štítné žlázy
- Sekreční karcinom jiných lokalit: kůže, nosní sliznice, štítná žláza, plíce

- NENACHÁZÍ SE V ŽÁDNÉM JINÉM NÁDORU SLINNÝCH ŽLÁZ

Mammary Analogue Secretory Carcinoma of Salivary Glands

A Clinicopathologic and Molecular Study Including 2 Cases Harboring ETV6-X Fusion

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Abstract: Mammary analogue secretory carcinoma (MASC) is a recently described low-grade carcinoma with morphologic and genetic similarity, including *ETV6-NTRK3* fusion, to secretory carcinoma of the breast. *ETV6* is frequently involved in other epithelial and nonepithelial tumors, and many fusion partners of *ETV6* have been reported. In the present study, 14 Japanese

novel insight into the oncogenesis, histopathology, diagnosis, treatment, and prognosis of this newly recognized carcinoma.

Key Words: mammary analogue secretory carcinoma, *ETV6-NTRK3*, gene rearrangement, linicopathologic analysis

(*Am J Surg Pathol* 2015;39:602–610)

Mammary Analogue Secretory Carcinoma of Salivary Glands

Molecular Analysis of 25 ETV6 Gene Rearranged Tumors With Lack of Detection of Classical ETV6-NTRK3 Fusion Transcript by Standard RT-PCR: Report of 4 Cases Harboring ETV6-X Gene Fusion

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Abstract: *ETV6* gene abnormalities are well described in tumor pathology. Many fusion partners of *ETV6* have been reported in a variety of epithelial and hematological malignancies. In salivary gland tumor pathology, however, the *ETV6-NTRK3* translocation is specific for mammary analogue secretory carcinoma (MASC), and has not been documented in any other salivary tumor type. The present study comprised a clinical and molecular analysis of 25 cases morphologically and immunohistochemically typical of MASC. They all also displayed the *ETV6* rearrangement as visualized by fluorescent in situ hybridization but lacked the classical *ETV6-NTRK3* fusion transcript by standard reverse-transcriptase-polymerase chain reaction. In 4 cases, the classical fusion transcript was found by more sensitive, nested reverse-transcription-polymerase chain reaction. Five other cases harbored atypical fusion transcripts as

of appropriate tissue material. Finally, in the 4 remaining cases whose profile was *NTRK3* split-negative and *ETV6* split-positive, unknown (non-*NTRK*) genes appeared to fuse with *ETV6* (*ETV6-X* fusion). In looking for possible fusion partners, analysis of rearrangement of other kinase genes known to fuse with *ETV6* was also performed, but without positive results. Although numbers were small, correlating the clinico-pathologic features of the 4 *ETV6-X* fusion tumors and 5 MASC cases with atypical fusion transcripts raises the possibility of that they may behave more aggressively.

Key Words: mammary analogue secretory carcinoma, MASC, *ETV6-NTRK3*, *ETV6-X* fusion transcript, clinicopathologic analysis

(*Am J Surg Pathol* 2016;40:3–13)

SALIVARY SECRETORY CARCINOMA WITH A NOVEL *VIM-RET* AND DUAL *ETV6-NTRK3/MYB-SMR3B* FUSIONS:

NGS based molecular profiling of 49 cases revealed an expanding molecular spectrum of a recently described entity.

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BACKGROUND

Secretory carcinoma (SC), originally described as mammary analogue secretory carcinoma, is a predominantly low-grade salivary gland neoplasm characterized by a recurrent t(12;15)(p13;q25) translocation, resulting in *ETV6-NTRK3* gene fusion (1). Recently, novel *ETV6-RET* (2) and *ETV6-MET* (3) fusions were found in a subset of SCs lacking the classical *ETV6-NTRK3* fusion transcript, but harboring *ETV6* gene rearrangements.

DESIGN

Forty nine cases of SC revealing typical morphology and immunoprofile were analyzed by NGS using the Fusion Plex Solid Tumor kit (ArcherDX). All 49 cases of SC were also tested for *ETV6* break by FISH and for common *ETV6-NTRK3* fusions using RT PCR.

RESULTS

Among the 49 studied cases, 37 (76%) occurred in the parotid gland, 7 (14%) in the submandibular gland, 2 (4%) in minor salivary glands, 1 (2%) in nasal mucosa, 1 (2%) in facial skin and 1 (2%) in the thyroid gland. Most cases were diagnosed in males (27/55%). Patient age at diagnosis varied from 15 to 80 years, with a mean age of 49.9 years. Regarding molecular analysis, 38 cases (78%) presented the classical *ETV6-NTRK3* fusion, 9 cases (18%) were negative for the *ETV6-NTRK3* fusion and 2 cases (4%) were not analyzable. Among the 9 negative cases for *ETV6-NTRK3* fusion, 8 cases presented *ETV6-RET* fusion. Using NGS analysis, a novel *VIM-RET* fusion transcript was identified in one case of SC of parotid gland. In addition, one recurrent high grade SC of submandibular gland was simultaneously positive for *ETV6-NTRK3* and *MYB-SMR3B* fusion transcripts.

CONCLUSION

A novel finding in our study has been a discovery of a *VIM-RET* fusion in one patient with SC of parotid gland who could possibly benefit from *RET*-targeted therapy. In addition, in one recurrent high-grade SC two different fusions *ETV6-NTRK3* and *MYB-SMR3B* were found. The expanded molecular spectrum of SC provides a novel insight into oncogenesis and molecular diagnosis of this recently described entity.

REFERENCES

1. Skalova A, Vanecek T, Sima R, et al. Mammary analogue secretory carcinoma of salivary glands, containing the ETV6-NTRK3 fusion gene: a hitherto undescribed salivary gland tumor entity. Am J Surg Pathol 2010;34:599-608.
2. Skalova A, Vanecek T, Martinek P, Weinreb I, Stevens TM, Simpson RHW, et al. Molecular profiling of mammary analog secretory carcinoma revealed a subset of tumors harboring a novel ETV6-RET translocation: Report of 10 cases. Am J Surg Pathol 2018; 42:234-26.
3. Rooper LM, Kennecke T, Ning Y, Bishop JA, Gordon SW, Kang H. Salivary Secretory Carcinoma With a Novel ETV6-MET Fusion: Expanding the Molecular Spectrum of a Recently Described Entity. Am J Surg Pathol. 2018 Aug;42(8):1121-1126. doi: 10.1097/PAS.0000000000001065.

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Case 1. Salivary secretory carcinoma with a novel *VIM-RET* fusion.
58-year old woman presented with a slowly growing mass in the parotid gland. Superficial parotidectomy was performed. The patient refused any additional treatment. Eight months after the surgery is patient well, without signs of clinical or radiological disease recurrence.

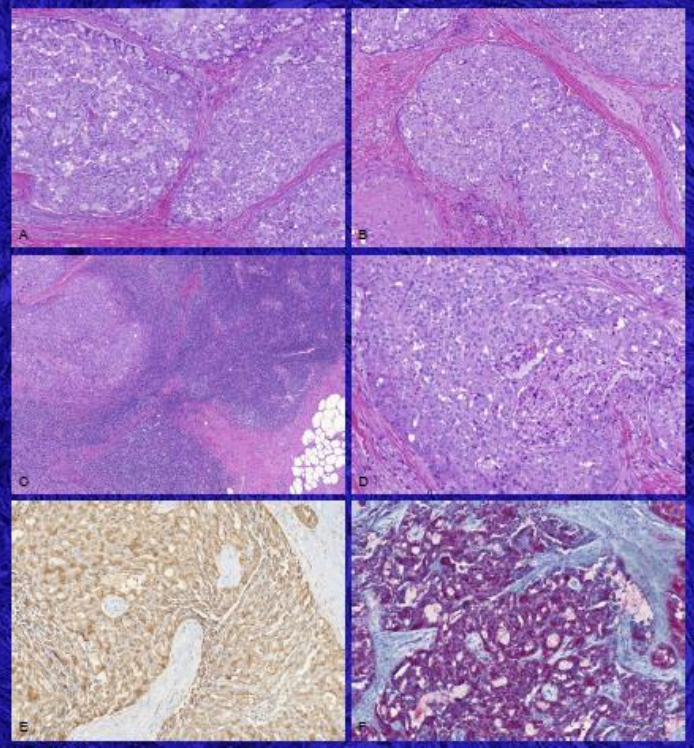

Fig. 1. Case 1.
SC is characterized by solid and microcystic growth with a multilobular structure divided by thin fibrous septa (Fig. 1A). Invasive growth of SC close to nerve is seen ((Fig. 1B). Neoplastic cells infiltrating periglandular tissue within an abundant lymphoid infiltrate (Fig. 1C) and focal comedo-like necrosis are present (Fig. 1D). Using immunohistochemical stains, the tumor cells are positive for mammaglobin (Fig. 1E) and S100 protein (Fig. 1F).

Fig. 3. Case 1.
Schematic representation of the fusion transcript identified by ArcherDX assay, involving the VIM exon 7 with RET exon 12.

Case 2. Salivary secretory carcinoma with dual *ETV6-NTRK3* and novel *MYB-SMR3B* fusions
61-year old man presented with a recurrent tumor of the submandibular gland, that measured 1,9x1,5x1,0 cm. The affected salivary gland was resected and a single lymh node metastasis was found. After the excision of the neck metastasis, the patient had a PET and lung CT scans performed and he is clear of disease 12 months after surgery.

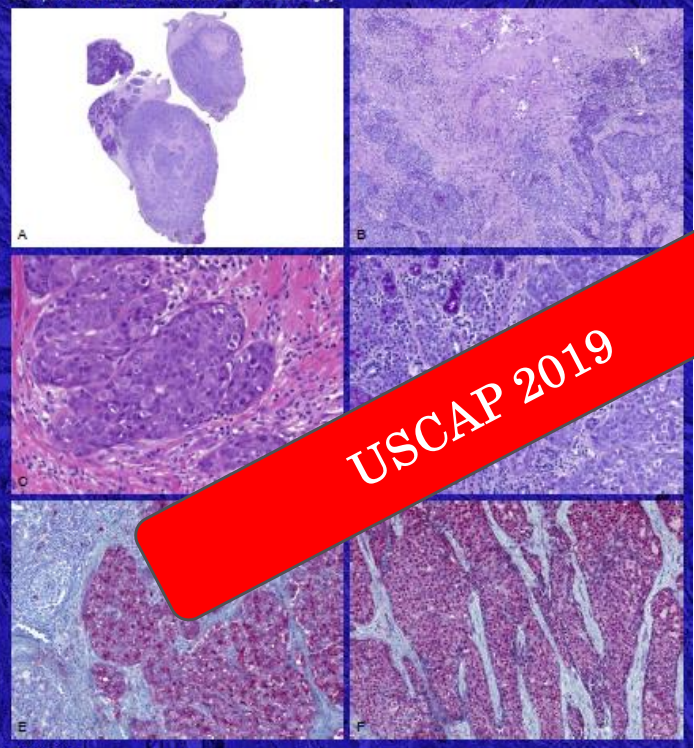

Fig. 2. Case 2.
SC of submandibular gland shows multifocal (Fig. 2A) and widely invasive growth pattern (Fig. 2B). High power view shows nuclear polymorphism and predominantly solid growth with minimal secretory activity (Fig. 2C). Invasion into the glandular tissue is apparent in PAS stain (Fig. 2D). S100 protein (Fig. 2E) and pan-TRK immunohistochemical stains are diffusely positive in all neoplastic cells (Fig.2F).

Fig. 4. Case 2.
Schematic representation of the fusion transcript identified by ArcherDX assay, involving the MYB exon 9 with SMR3B exon 3.

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Original contribution

Novel gene fusions in secretory carcinoma of the salivary glands: enlarging the *ETV6* family☆,☆☆,★,★★

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Keywords:
Secretory carcinoma;
Salivary gland tumor;
ETV6-NTRK3;
ETV6-RET;
MAML3

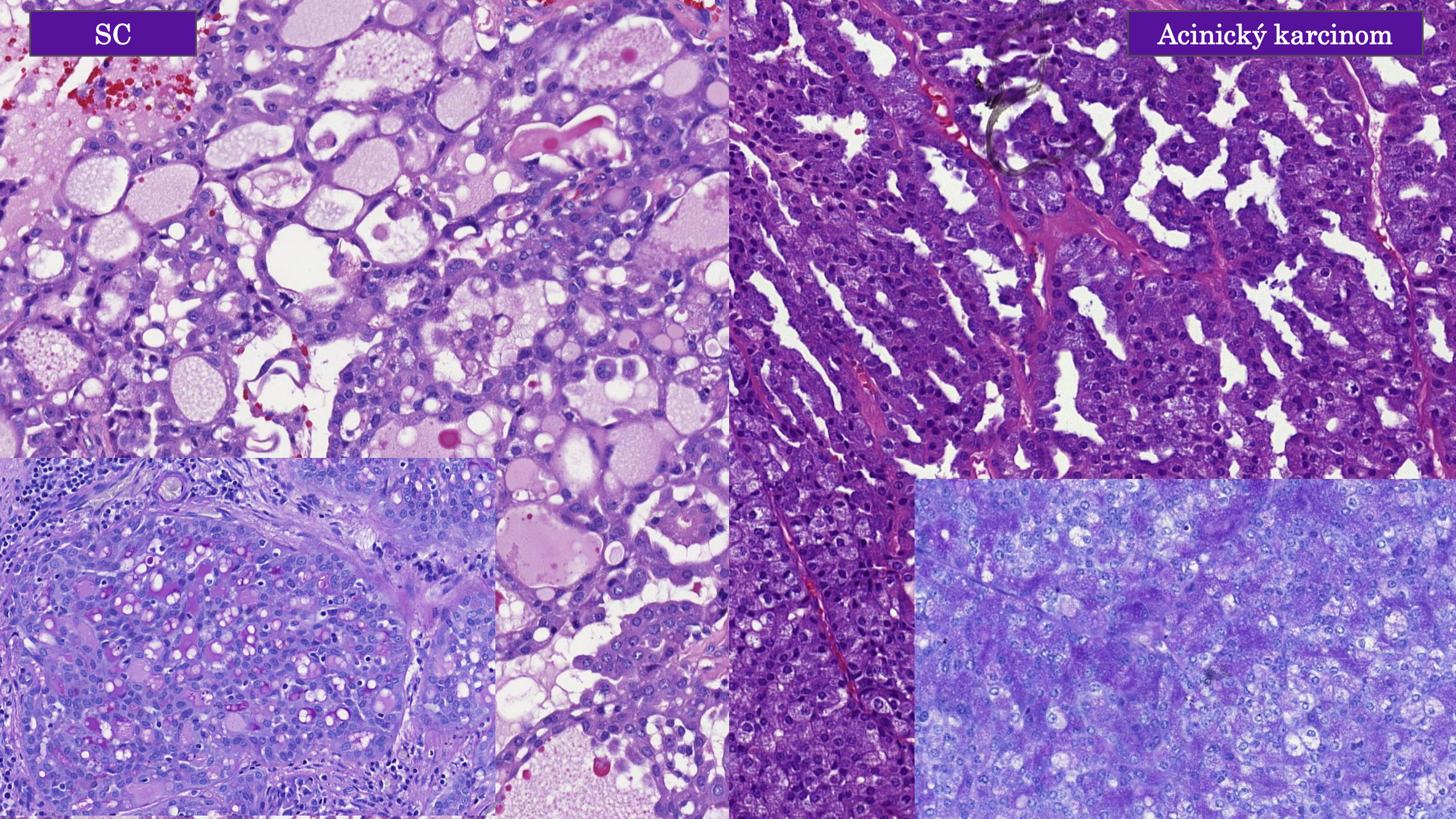
Summary Secretory carcinoma (SC) of the salivary gland is a low-grade malignancy associated with a well-defined clinical, histologic, immunohistochemical, and cytogenetic signature. Although the t(12;15) (p13; q25) translocation resulting in an *ETV6-NTRK3* gene fusion is well documented, advances in molecular profiling in salivary gland tumors have led to the discovery of *RET* as another *ETV6* gene fusion partner in SC. Here, we applied an RNA-based next-generation sequencing (NGS) approach for fusion detection on 14 presumed SC. The cases included 7 SC with classic *ETV6-NTRK3* gene fusion and 3 SC harboring

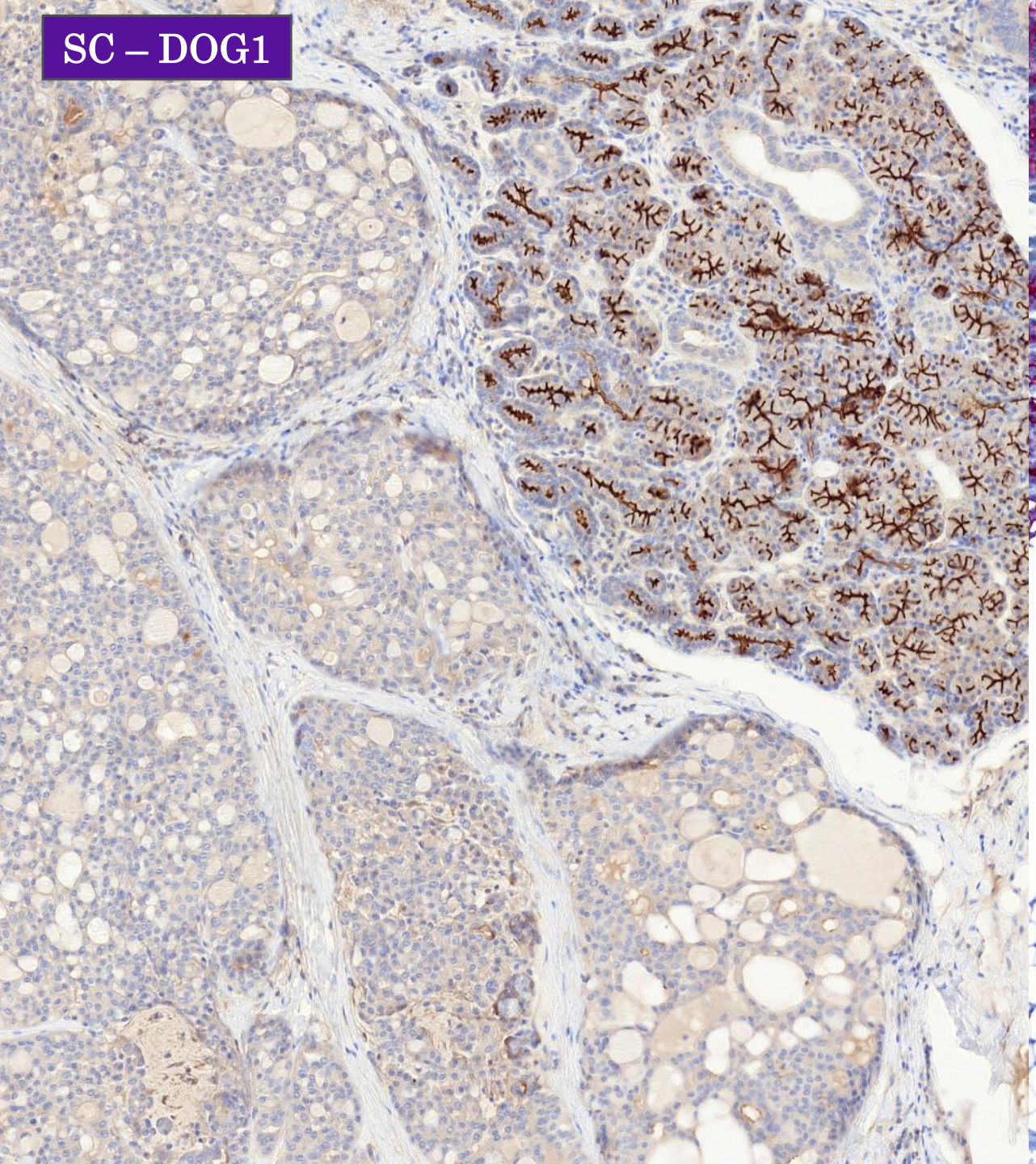
ETV6 exon 5 a *NTRK3* exon 15; *ETV6* exon 2 a *MAML3* exon 2

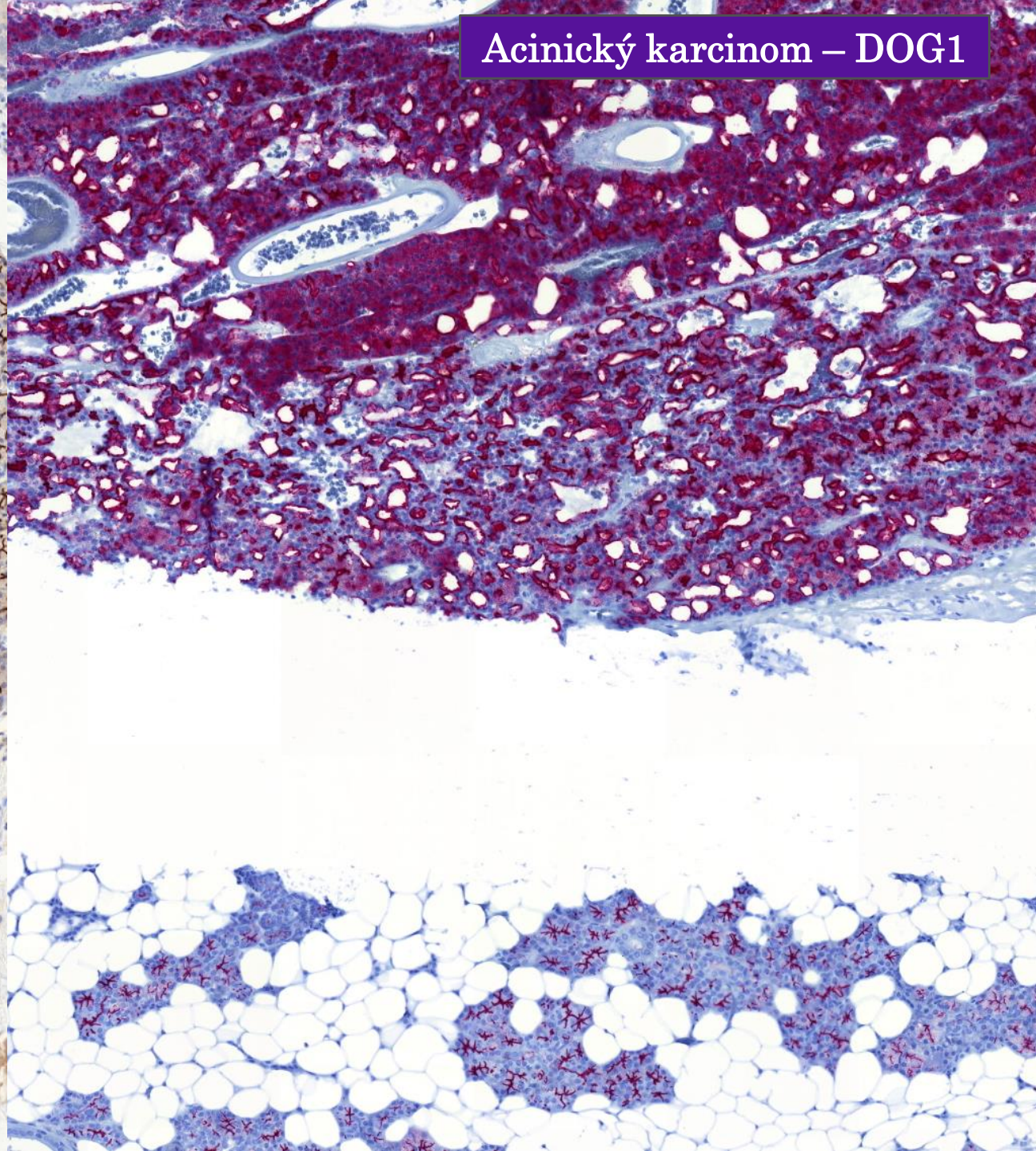
DIFERENCIÁLNÍ DIAGNÓZA

SC a Acinický karcinom

Sekreční karcinom	Acinický karcinom
bez PAS+ zymogenních granul	PAS+ zymogenních granula v cytoplazmě
morfologie: homogenní, mikrocysty, žlazové prostory se sekrečním PAS+ materiálem	morfologie: strukturální různorodost
imunoprofil: S100+/MGA+/CK7+/DOG1-	DOG1+
genetika: *ETV6-NTRK3* translokace (95-98%)	ETV6 intaktní, *HTN3-MSANTD3*


SC
Acinický karcinom


SC – DOG1


Acinický karcinom – DOG1

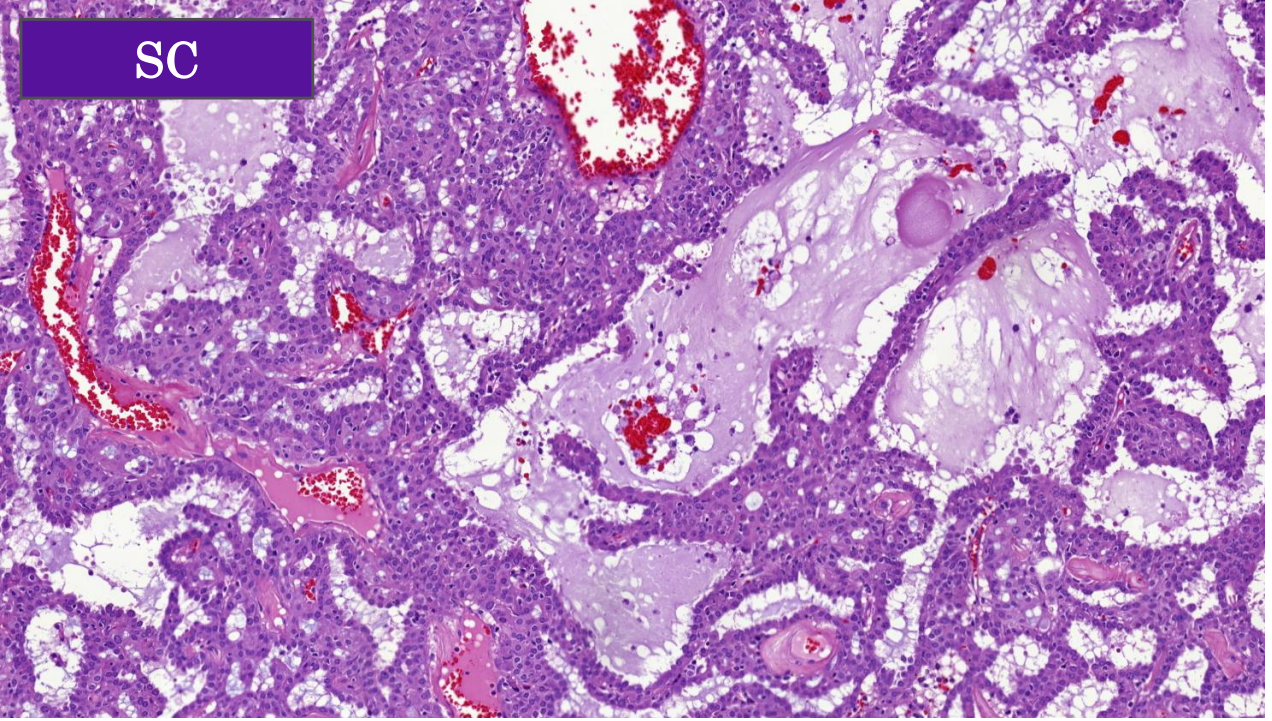
SC a LG mukoepidermoidní karcinom

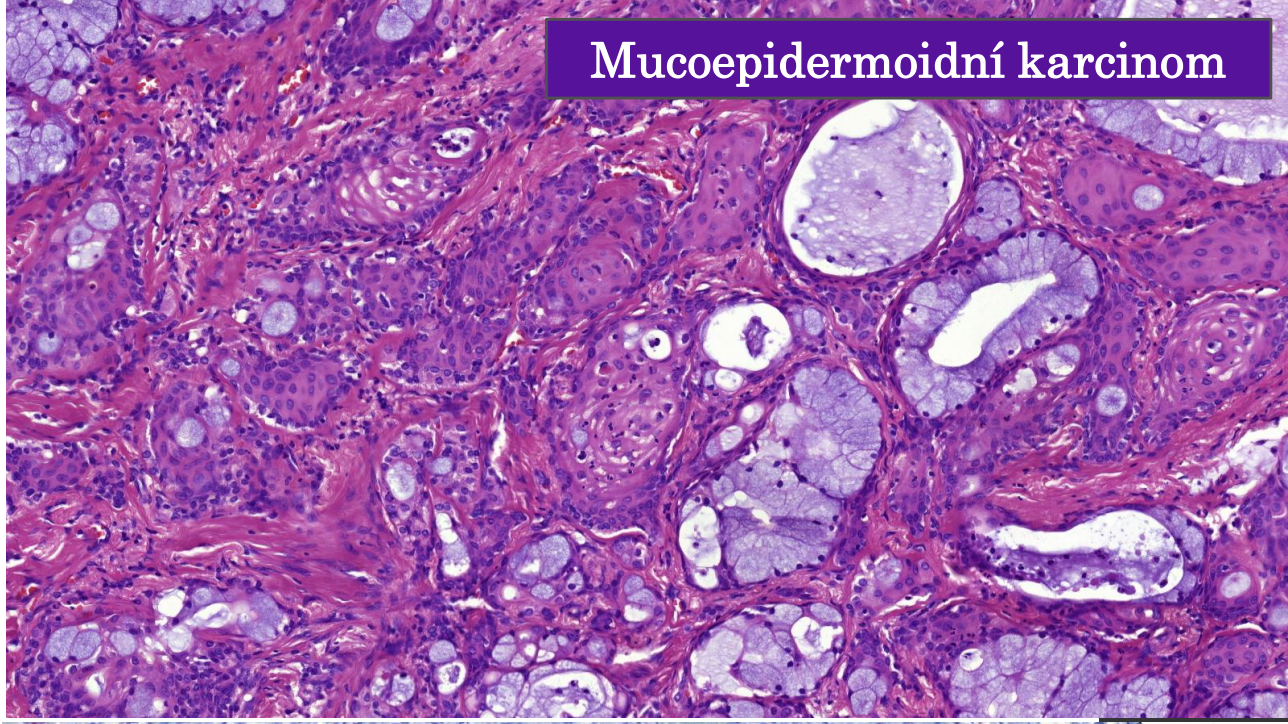
Sekreční karcinom

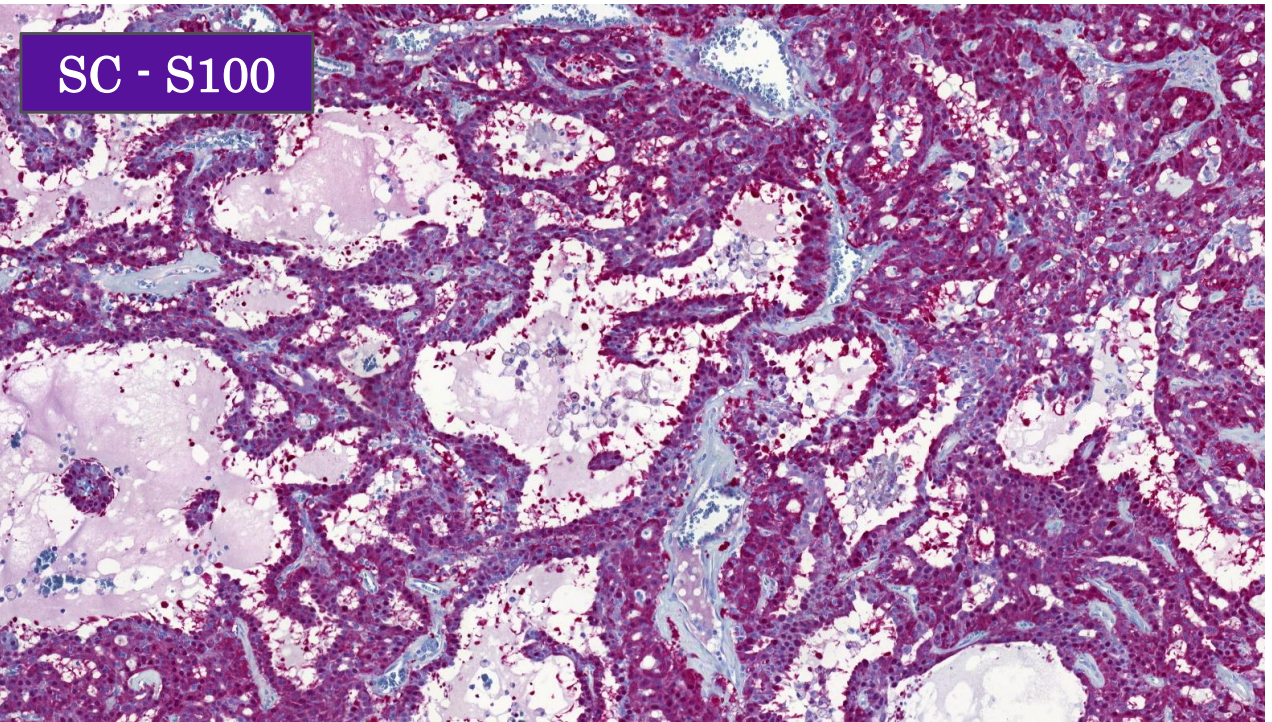
- fokálně HMWCK a mucinózní diferenciace
- obvykle p63 negativní, S100++
- t (12;15) *ETV6-NTRK3*

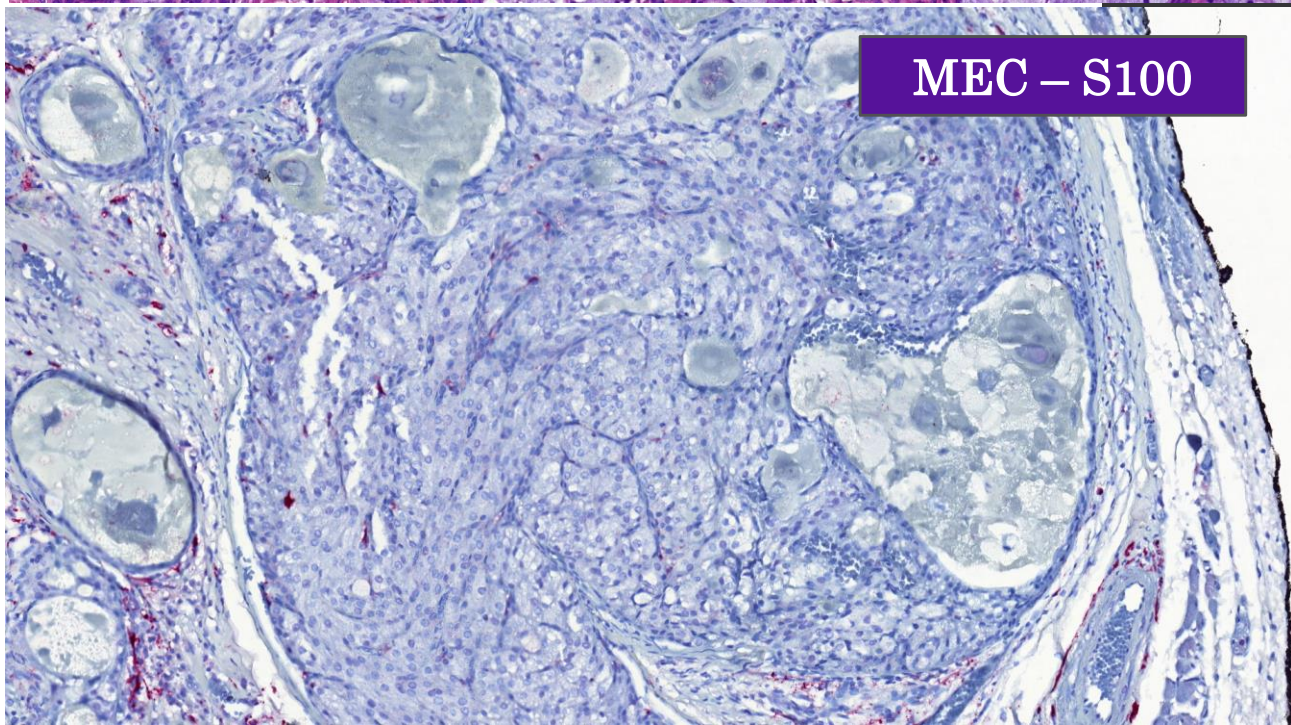
Mukoepidermoidní karcinom

- chybí vzhled dlažebních kostek, přítomny mezibuněčné můstky, dlaždicobuněčná metaplázie a intermediární buňky
- obvykle S100 negativní, HMWCK+
- t (11;19) *CTRC1/3-MAML2*


SC


Mucoepidermoidní karcinom


SC - S100


MEC – S100

SC a „Intraduktální karcinom dle WHO 2017“

Sekreční karcinom

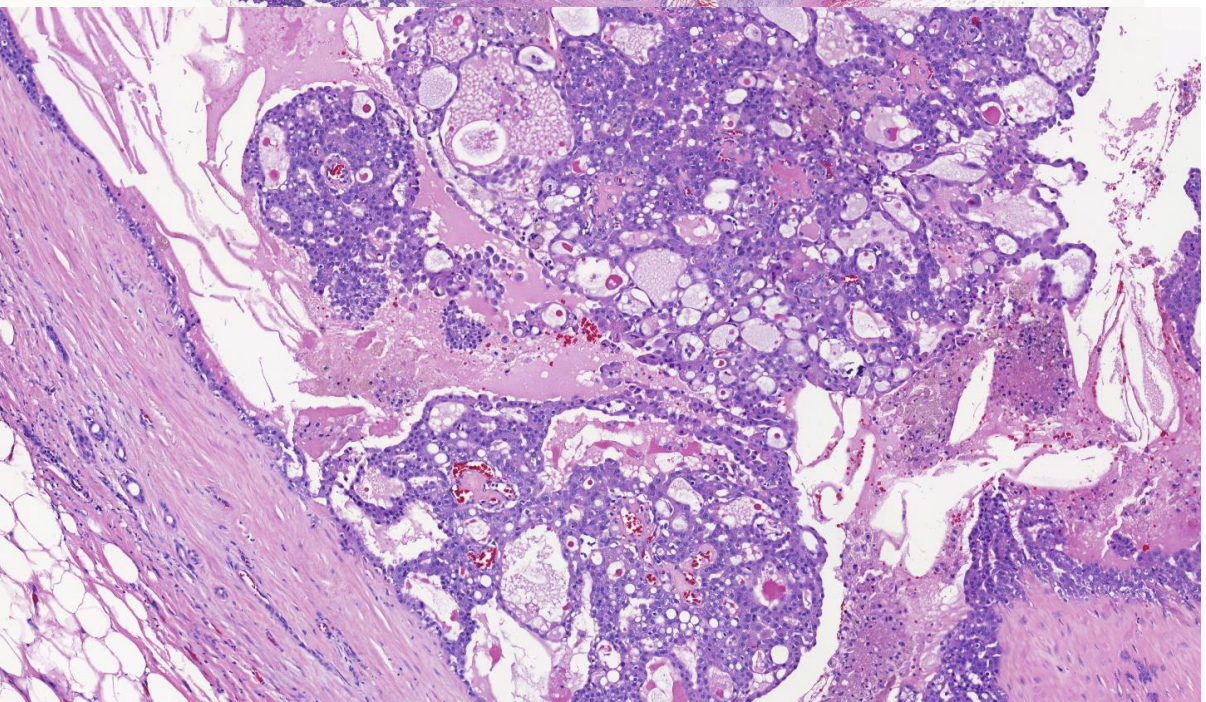
- S100 +
- p63 negativní
- *ETV6* zlom, RET zlom ve zlomku SC

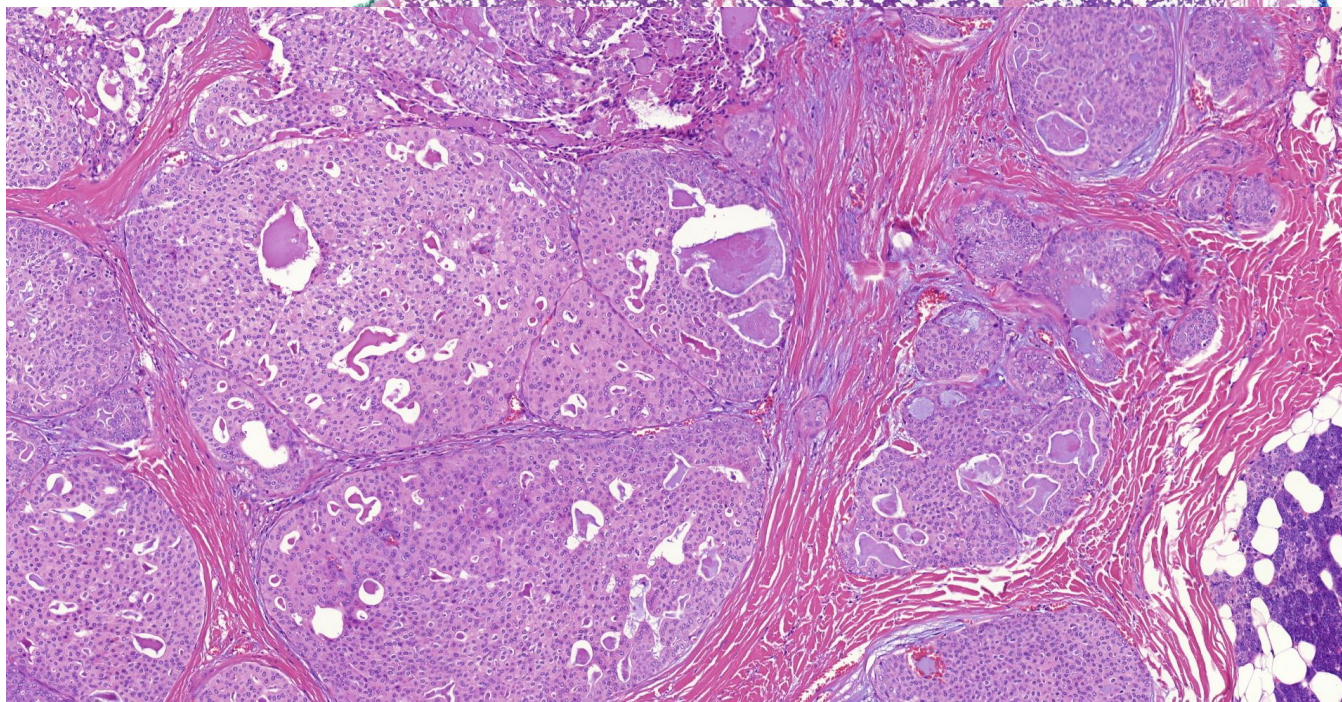
IDC

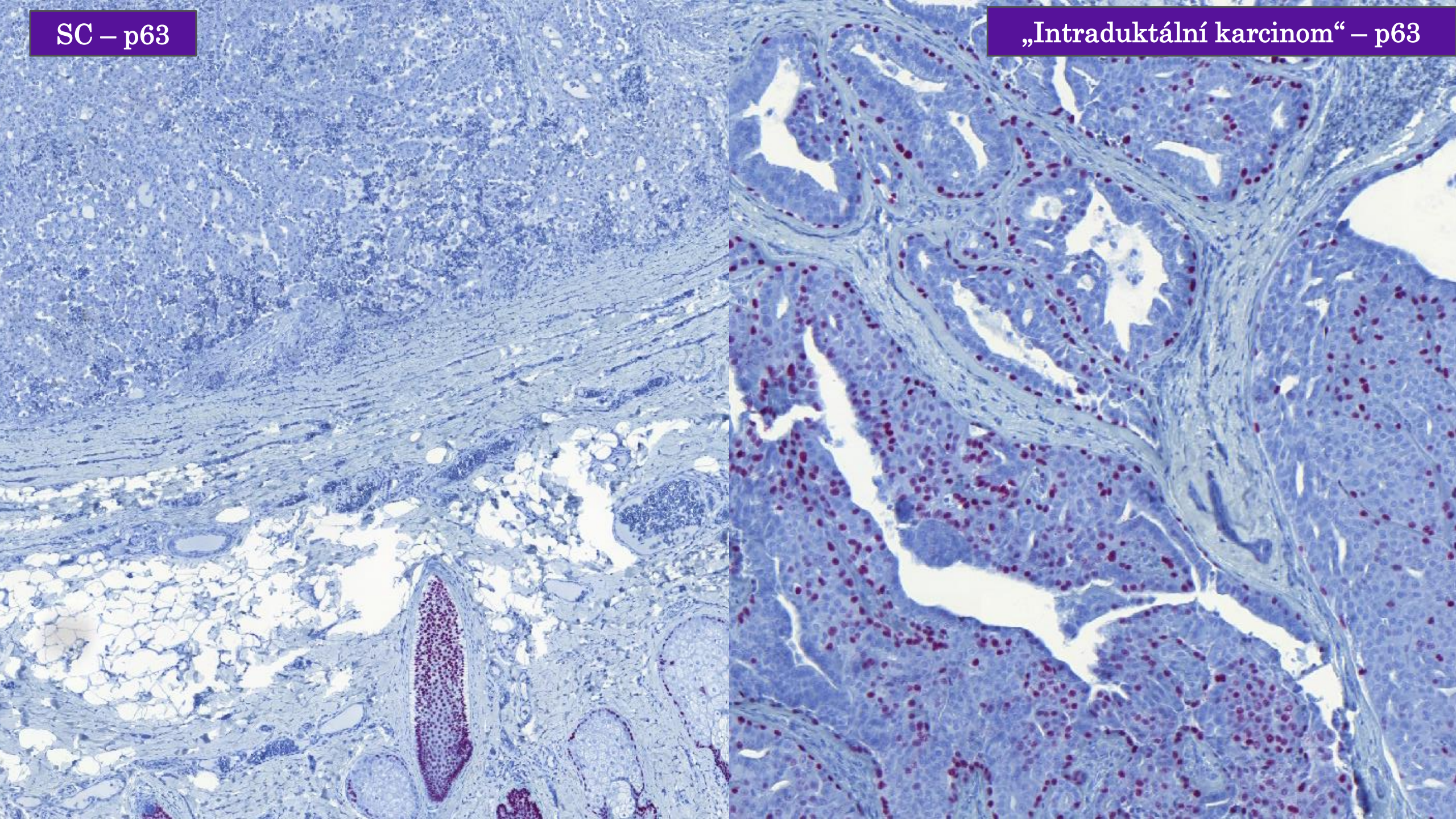
- *LG salivární duktální karcinom (Delgado 1996) / LG kribriformní cystadenokarcinom (dle WHO 2005)*
- S100 +
- p63 pozitivita kolem hnízd tu bb
- ETV6 negativní, ale *NCOA4-RET a TRIM27-RET*

SC

„Intraduktální karcinom“






SC – p63
„Intraduktální karcinom“ – p63

SHRNUTÍ

- převážně LG
- 10% umírá s dg. SC
- Vysoký klinický stage a HG varianty = špatné prognostické faktory
- Terapie – prostá excize u LG
  - pokročilý nález a metastatická diseminace – paliativní terapie
  - !! molekulární testování!!
- Biologická léčba:
  - **Vitrakvi (larotrectinib) či Entrectinib** – TRKA,B,C
  - **RET-inhibitory** –Blue-667 a LOXO_292

Drilon et al. What hides behind MASC:…Ann Oncol 2016;27:920-6

Jackson Pollock

DĚKUJI ZA POZORNOST

28. 6. 2019, Senec, Slovensko

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