Assessment Run 75 2025

Transcription factor SOX-10 (SOX10)

Updated 13.01.2026

Purpose
Evaluation of the technical performance, level of analytical sensitivity and specificity of IHC tests among the NordiQC participants for SOX10, identifying malignant melanomas and triple negative breast carcinoma in the characterization of tumours of unknown origin. Relevant clinical tissues, both normal and neoplastic, were selected to represent a broad spectrum of antigen densities for SOX10 (see below).

Material
The slide to be stained for SOX10 comprised:

1. Skin, 2. Colon adenocarcinoma, 3. Appendix, 4. Triple negative breast carcinoma, 5-6. Malignant melanoma

All tissues were fixed in 10% neutral buffered formalin.

Criteria for assessing a SOX10 staining as optimal included:

- A strong, distinct nuclear staining reaction of virtually all melanocytes in the skin and Schwann cells in the appendix.
- An at least moderate, distinct nuclear staining reaction of the majority of myoepithelial cells and scattered luminal cells lining sweat glands in the skin.
- An at least moderate, distinct nuclear staining reaction of virtually all neoplastic cells in the triple negative breast carcinoma.
- An at least moderate, distinct nuclear staining reaction of the majority of neoplastic cells in the malignant melanoma, tissue core no. 5.
- A strong, distinct nuclear staining reaction of the majority of neoplastic cells in the malignant melanoma, tissue core no. 6.
- No staining reaction in other cellular structures including the neoplastic cells of the colon adenocarcinoma.

A weak cytoplasmic staining reaction in cells with a strong nuclear staining reaction was accepted. For certain primary antibodies e.g. mAb clone EP268, a weak cytoplasmic staining reaction of ganglion/neuronic cells in the appendix was accepted, provided that interpretation of the specific nuclear staining reaction was not compromised.

KEY POINTS FOR SOX10 IMMUNOASSAYS
- The mAb clone **BC34, BS7** and rmAb clone **EP268** are all recommendable as concentrated Abs.
- The rmAb clone **SP267** and **EP268** are recommendable as RTUs.
- Efficient HIER in an alkaline buffer is important for an optimal performance.

Participation

Number of laboratories registered for SOX10, run 75	454
Number of laboratories returning slides	416 (92%)

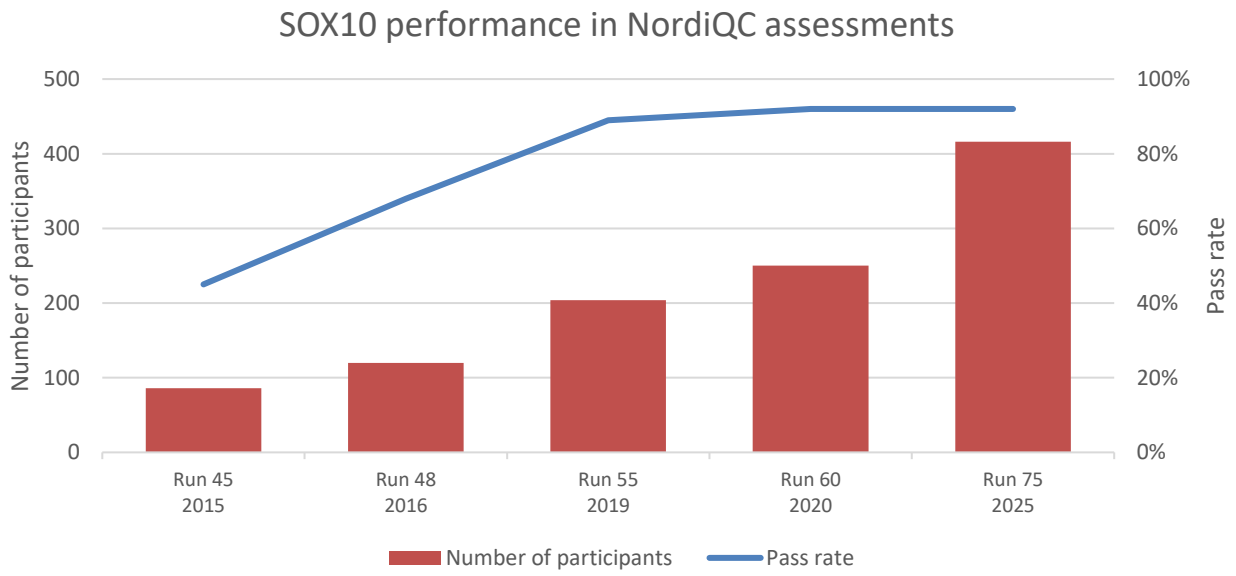
At the date of assessment, 92% of the participants had returned the circulated NordiQC slides. All slides returned after the assessment were assessed and laboratories received advice if the result was insufficient, but the data were not included in this report.

Results
416 laboratories participated in this assessment. 384 (92%) achieved a sufficient mark (optimal or good) – see Table 1a (page 3). Tables 1b and 1c summarizes the antibodies (Abs) used and assessment marks (see page 3 and 4).

The most frequent causes of insufficient staining reactions were:
- Less successful primary antibody
- Insufficient HIER (too short heating time or HIER in acidic buffer).
- Less sensitive detection systems

Performance history
This was the fifth NordiQC assessment of SOX10. The overall pass rate of 92% is the same as obtained in run 60, 2020 despite a significantly increased number of participants (see Graph 1).

Graph 1. **Proportion of sufficient results for SOX10 in the five NordiQC runs performed**



Controls
Skin and colon/appendix are recommended as positive and negative tissue controls for SOX10. In skin, strong nuclear staining reaction in virtually all melanocytes must be seen. The vast majority of myoepithelial cells and scattered luminal cells lining sweat glands must show an at least moderate nuclear staining reaction. In colon/appendix, virtually all Schwann cells must display an as strong as possible nuclear staining reaction without any staining reaction of epithelial cells and e.g. smooth muscle cells. At present, and as specified in previous assessments, no reliable tissue component with consistent low-level expression of SOX10 has been identified, monitoring the reproducibility and overall analytical sensitivity of the assay. Thus, both skin and colon/appendix are needed as tissue controls for SOX10.

Conclusion
The mAb clones **BC34, BS7** and the rmAb clones **EP268** and **SP267** were the most widely used antibodies and could all be used to obtain an optimal result for SOX10. Irrespective of the clone applied, efficient HIER (preferable in an alkaline buffer), a precise calibration of the primary Ab and the use of a 3-step multimer/polymer based detection system, were the main prerequisite for an optimal result. The RTU system 760-4968 (Ventana) based on the rmAb clone SP267, showed superior performance and following vendor recommended protocol settings, 100% (117/117) were assessed as sufficient of which 96% (112/117) were optimal.

Table 1a. **Overall results for SOX10, run 75**

	n	Optimal	Good	Borderline	Poor	Suff.[1]	OR[2]
Concentrated antibodies	134	73	53	5	3	94%	54%
Ready-To-Use antibodies	282	215	43	17	7	91%	76%
Total	416	288	96	22	10		
Proportion		69%	23%	5%	3%	92%	

1) Proportion of sufficient results (optimal or good).
2) Proportion of optimal results.

Table 1b. **Concentrated antibodies and assessment marks for SOX10, run 75**

Concentrated antibodies	n	Vendor	Optimal	Good	Borderline	Poor	Suff.[1]	OR[2]
mAb clone **BC34**	3	Abcam	0	2	0	1	-	-
	43	Biocare Medical	16	25	2	0	95%	37%
	2	Zytomed Systems	0	2	0	0	-	-
mAb clone **BS7**	9	Nordic Biosite	6	3	0	0	100%	67%
mAb clone **BSB-62**	1	BioSB	0	1	0	0	-	-
mAb clone **ZM10**	1	Zeta Corporation	0	1	0	0	-	-
mAb clone **A-2**	1	Santa Cruz	0	1	0	0	-	-
mAb clone **IHC010**	2	GenomeMe	1	1	0	0	-	-
Ab clone **GM005**	1	Gene Tech	1	0	0	0	-	-
rmAb clone **EP268**	58	Cell Marque	44	12	1	1	97%	76%
	2	Epitomics	1	0	1	0	-	-
	4	BioSB	0	4	0	0	-	-
	1	Pathnsitu	0	1	0	0	-	-
rmAb clone **GR24**	1	GeneBioSolution	0	0	1	0	-	-
rmAb clone **QR006**	2	Quartett	2	0	0	0	-	-
rmAb clone **ZR275**	1	Zeta Corporation	1	0	0	0	-	-
rmAb clone **E6B6I**	1	Cell Signaling	1	0	0	0	-	-
pAb **AB5727**	1	Millipore	0	0	0	1	-	-
Total	134		73	53	5	3		
Proportion			54%	40%	4%	2%	94%	

1) Proportion of sufficient results (optimal or good). (≥5 assessed protocols).
2) Proportion of optimal results (OR).

Table 1c. **Ready-to-use antibodies and assessment marks for SOX10, run 75**

Ready-To-Use antibodies	n	Vendor	Optimal	Good	Borderline	Poor	Suff.[1]	OR[2]
mAb clone **BC34** **API 3099**[3]	1	Biocare Medical	0	1	0	0	-	-
mAb clone **BC34** **API 3099**[4]	13	Biocare Medical	5	4	4	0	38%	69%
mAb clone **BC34** **MSG117**	1	Zytomed Systems	1	0	0	0	-	-
mAb clone **SOX10/991** **PA0010-U**	2	Leica Biosystems	0	1	1	0	-	-
mAb clone **ZM10** **8308-C010**	3	Sakura Finetek	2	1	0	0	-	-
Ab clone **BP6024** **I10152E-05**	1	Biolynx Biotechnology	1	0	0	0	-	-
Ab clone **DY49970** **4912102**	1	Dakewe	0	1	0	0	-	-
Ab clone **GM005** **GT221002**	1	Gene Tech	1	0	0	0	-	-
rmAb clone **SP267** **760-4968 (VRPS)**[3]	117	Ventana/Roche	112	5	0	0	100%	96%
rmAb clone **SP267** **760-4968 (LMPS)**[4]	95	Ventana/Roche	69	21	5	0	95%	73%
rmAb clone **EP268** **383R**	29	Cell Marque	22	4	2	1	90%	76%
rmAb clone **EP268** **MAD-000656QD**	7	Master Diagnostica	0	1	2	4	14%	0%
rmAb clone **EP268** **RMPD077**	2	Diagnostics BioSystems	0	0	1	1	-	-
rmAb clone **EP268** **BSB 258-0/1/2**	5	BioSB	0	3	2	0	60%	0%
mAb clone **C611** **CSM-0181**	2	Celnovte	1	0	0	1	-	-
rmAb clone **MXR026** **RMA1058**	1	Fuzhou Maixin	1	0	0	0	-	-
rmAb clone **QR006** **P-S001-30/70/150**	1	Quartett	0	1	0	0	-	-
Total	282		215	43	17	7		
Proportion			76%	15%	6%	3%	91%	

1) Proportion of sufficient results (optimal or good). (≥5 assessed protocols).
2) Proportion of optimal results (OR).
3) Vendor Recommended Protocol Settings (VRPS) to a specific RTU product applied on the vendor recommended platform(s) (≥5 asessed protocols).
4) Laboratory Modified Protocol Settings (LMPS) to a specific RTU product applied either on the vendor recommended platform(s), non-validated semi/fully automatic systems or used manually (≥5 assessed protocols).

Detailed analysis of SOX10, Run 75

The following protocol parameters were central to obtain optimal staining:

Concentrated antibodies

mAb clone **BC34**: Protocols with optimal results were all based on Heat Induced Epitope Retrieval (HIER) using an alkaline buffer as Cell Conditioning 1 (CC1, Ventana/Roche) (2/13)*, Target Retrieval Solution (TRS) pH 9 (Dako/Agilent) (9/22) or Bond Epitope Retrieval Solution 2 (BERS2, Leica Biosystems) (5/9) as retrieval buffer. The mAb was typically diluted in the range of 1:25-1:150 and a 3-step detection system was applied. Using these protocol settings, 44 of 46 (96%) laboratories produced a sufficient staining (optimal or good).
* (number of optimal results/number of laboratories using this buffer)

mAb clone **BS7**: Protocols with optimal results were all based on HIER using an alkaline buffer as TRS pH 9 (Dako/Agilent) (5/7) or CC1 (Ventana/Roche) (1/2). The mAb was diluted in the range of 1:100-1:200 depending on the total sensitivity of the protocol employed. Using these protocol settings, 9 of 9 (100%) laboratories produced a sufficient staining

rmAb clone **EP268**: Protocols with optimal results were all based on HIER using either CC1 (Ventana/Roche) (11/18), TRS pH 9 (Dako/Agilent) (24/27), BERS2 (Leica Biosystems) (8/15), Bond Epitope Retrieval Solution 1 (BERS1, Leica Biosystems) (1/2) or Tris-EDTA buffer (1/1) as retrieval buffer. The rmAb was typically diluted in the range of 1:50-1:500 depending on the total sensitivity of the protocol employed. Using these protocol settings, 61 of 63 (97 %) laboratories produced a sufficient staining.

Table 2. **Proportion of optimal results for SOX10 for the most commonly used antibodies as concentrate on the four main IHC systems***

Concentrated antibodies	**Dako/Agilent Autostainer[1]**		**Dako/Agilent Omnis**		**Ventana/Roche BenchMark[2]**		**Leica Biosystems Bond[3]**	
	TRS pH 9.0	TRS pH 6.1	TRS pH 9.0	TRS pH 6.1	CC1 pH 8.5	CC2 pH 6.0	BERS2 pH 9.0	BERS1 pH 6.0
mAb clone **BC34**	3/3**	-	6/19 (32%)	-	2/13 (15%)	-	5/9 (56%)	-
mAb clone **BS7**	-	-	5/7 (71%)	-	1/2	-	-	-
rmAb clone **EP268**	3/5 (60%)	-	21/21 (100%)	-	11/18 (61%)	-	8/15 (53%)	1/2

* Antibody concentration applied as listed above, HIER buffers and detection kits used as provided by the vendors of the respective systems.
** (number of optimal results/number of laboratories using this buffer).
1) Autostainer Link 48.
2) BenchMark GX, Ultra, Ultra plus
3) Bond III, Prime

Ready-To-Use antibodies and corresponding systems
rmAb clone **SP267,** product no. **760-4968,** Ventana/Roche Benchmark XT/GX/Ultra/Ultra plus:
Protocols with optimal results were typically based on HIER using CC1 (efficient heating time 32-64 min. at 95-100°C), 16-40 min. incubation time of primary Ab, UltraView with or without amplification (760-500 + 760-080) or OptiView with or without amplification (760-700 + 760-099) as the detection system. Using these protocol settings, 204 of 208 (98%) laboratories produced a sufficient result.
The product was used by four laboratories on a non-intended platform. These data are not included here.

Table 3 summarizes the proportion of sufficient and optimal marks for the most commonly used RTU systems (≥10 assessed protocols). The performance was evaluated both as "true" plug-and-play systems performed strictly accordingly to the vendor recommendations and by laboratory modified systems changing basal protocol settings. Only protocols performed on the intended IHC stainer device are included.

Table 3. **Proportion of sufficient and optimal results for SOX10 for the most commonly used RTU IHC system**

RTU system	**Recommended protocol settings***		**Laboratory modified protocol settings****	
	Sufficient	Optimal	Sufficient	Optimal
VMS XT/GX/Ultra/Ultra Plus rmAb SP267 **760-4968 UltraView**	100% (71/71)	94% (67/71)	93% (50/54)	57% (31/54)
VMS XT/GX/Ultra/Ultra Plus rmAb SP267 **760-4968 OptiView**	100% (46/46)	98% (45/46)	100% (37/37)	97% (36/37)

* Protocol settings recommended by vendor – Retrieval method and duration, Ab incubation times, detection kit, IHC stainer/equipment.
** Significant modifications: retrieval method, retrieval duration and Ab incubation time altered, detection kit – only protocols performed on the specified vendor IHC stainer were included.

Comments
In this assessment and in concordance with the observations in previous NordiQC assessments of SOX10, the prevalent feature of an insufficient staining reaction was a too weak or false negative staining reaction of cells expected to be demonstrated and was seen in 59% (19/32) of the insufficient results. The majority of the laboratories were able to stain for SOX10 in Schwann cells of the appendix, the neoplastic cells of the melanoma tissue core no. 6, whereas demonstration of SOX10 in the neoplastic cells of the melanoma tissue core no. 5, the neoplastic cells of the triple negative breast carcinoma and myoepithelial cells of the skin was more challenging and required a carefully calibrated protocol. In 41% (13/32) of the insufficient results, a general poor signal-to-noise ratio and/or false positive staining reaction was seen.

In this assessment, the pass rate was high and grouped together, both for laboratory developed (LD) and Ready-to-Use (RTU) assays, 92% (384/416) of the participants produced a sufficient result.

32% (134/416) of the participants used a concentrated Ab within a LD assay, with a pass rate of 94%, 54% optimal, compared to a slightly lower pass rate of 91% if using a RTU system, used by 68% (282/416), however, with an increased proportion of optimal results – 76% (see Table 1a).

The mAb clone **BC34** and the rmAb clone **EP268** were the most widely used antibodies within a LD assay. The **mAb clone BC34** gave an overall pass rate of 94% (45/48). However, only 33% (16/48) was optimal (see Table 1b). As shown in Table 2, optimal results could be obtained on all automated and semi-automated IHC platforms from Dako/Agilent, Leica Biosystems and Ventana/Roche. All optimal results were based on HIER in an alkaline buffer and a 3-step detection system.

The LD assays based on **rmAb clone EP268** provided a pass rate of 95% (62/65) of which 69% (45/65) were assessed as optimal. When using a 3-step detection system, a 100% pass rate (37/37) was obtained, 81% optimal (n=30). When using a 2-step detection system, a lower pass rate of 89% (25/28) was seen, only 54% optimal (n=15). As shown in Table 2, the rmAb clone EP268 could provide optimal results on all main IHC platforms from the three major vendors.

Nine laboratories used the **mAb clone BS7** within a LD assay and all (9/9) were assessed as sufficient. 67% (6/9) of the protocols were assessed as optimal. The results marked as good were typically characterized by reduced signal-to-noise ratio and/or a reduced intensity/proportion of cells demonstrated. Data from internal NordiQC reference laboratories have previously revealed that the clone is highly recommendable but can be challenging to calibrate accurately to find right level of analytical sensitivity versus unwanted background reaction.

In this assessment, the RTU system **760-4968 (Ventana/Roche)** based on the rmAb clone SP267 was the most widely used assay for the demonstration of SOX10. The number of participants using this system, has increased significantly during the last runs; from 5 laboratories in run 48 (2016) to 208 in this run 75. The RTU system has shown to be very robust, and in the previous three runs, the accumulated pass rate has been 98% (163/166) being the same obtained in this run 75 (204/208). As shown in Table 3, the highest proportion of sufficient (100%) and optimal results (96%) were obtained using the RTU system according to the protocol recommendations provided by the vendor using either UltraView or OptiView as detection system. Laboratory modified protocol settings (typically adjusting HIER and incubation time of the primary Ab) could also provide optimal results, but proportion of optimal results decreased from 96% to 73% compared to vendor recommended protocol settings. Four laboratories applied the RTU system on another platform and 3 of 4 with a sufficient result.

The RTU **MAD-000656QD (Master Diagnostica)** product, based on rmAb clone EP268, was used by 7 participants. The protocols were typically based on HIER in an alkaline buffer and 2- or 3-step detection system. 86% (6/7) were insufficient due to an extensive, aberrant cytoplasmic staining reaction, which might be related to a contamination with another Ab or other lot issues.

One laboratory obtaining a good result, used the RTU system **API 3099 (Biocare Medical)** based on the mAb clone BC34 developed for the IHC stainer IntelliPATH. However, 14 laboratories used the RTU format on platforms other than the IntelliPATH (Biocare). Off-label use of an RTU format, validated for a given IHC system e.g. platform including immuno-reagents, is not recommended despite obtaining a relative high pass rate (see Table 1c). This "inappropriate/incorrect" use of an RTU product was also seen with other RTU formats e.g. **383R (Cell Marque)** based on the rmAb clone EP268. Overall, the off-label use of the RTU formats API 3099 and 383R on non-compliant platforms gave a relatively high pass rate but reduced proportion of optimal results compared to e.g. the Ventana RTU system (see Table 2c).

This was the fifth assessment of SOX10 in NordiQC (see Graph 1). The pass rate is the same as in the latest run 60, 2020. Several parameters contributed to the high proportion of sufficient results: 1) The extended use of robust primary Abs (e.g. rmAb clone EP268) on the expense of less successful primary Abs as pAbs, 2) The superior performance of the RTU system 760-4968 (Ventana/Roche) based on the rmAb clone SP267 and applied by 50% (208/416) of the participants, 3) Laboratories following advice giving by the NordiQC organization in past runs, typical providing specific recommendations to use HIER in an alkaline buffer, careful calibration of the primary Ab, use monoclonal Abs and the use of an 3-step multimer/polymer detection system.
Importantly, the protocols must give staining results accordingly to the expected pattern and antigen level within the recommended tissue control material (see below). This seems highly beneficial and central for both the initial validation process and verification of IHC test reproducibility for SOX10.

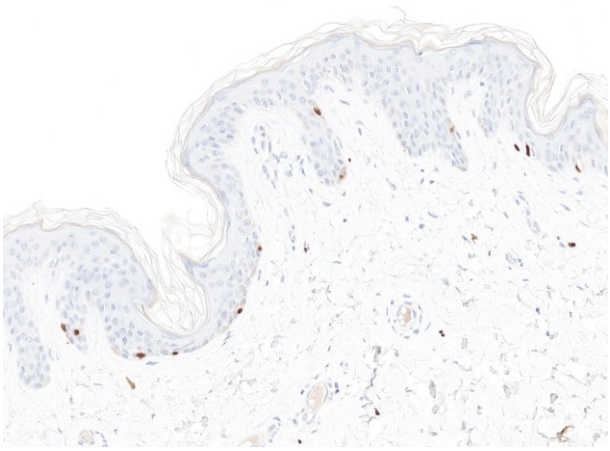


Fig. 1a
Optimal SOX10 staining result of the skin using the rmAb clone SP267 (RTU system 760-4968, Ventana/Roche) following recommendations given by the vendor - HIER in CC1 (32 min.), 32 min. incubation time in primary Ab and OptiView as detection system. All melanocytes show a strong, distinct nuclear staining reaction. No background is seen.

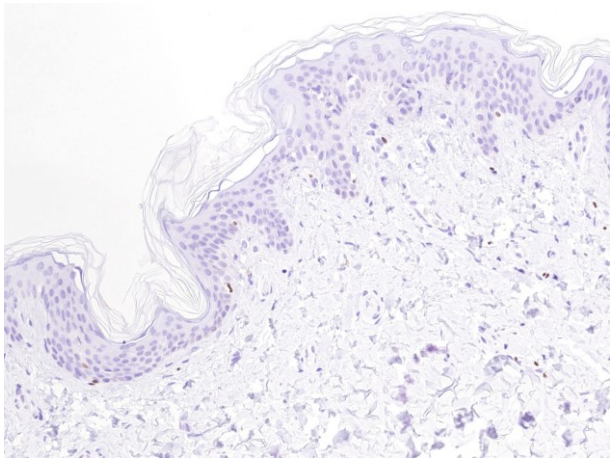


Fig. 1b
Insufficient SOX10 staining result of the skin, using the mAb clone BC34 within an LD assay with HIER in TRS low ph buffer. Melanocytes are only faintly positive.

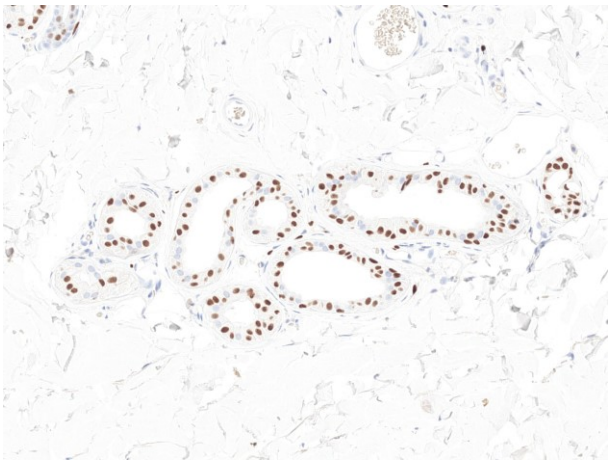


Fig. 2a
Optimal SOX10 staining result of the skin using same protocol as in Fig. 1a. The majority of myoepithelial cells and scattered luminal cells show an at least moderate, distinct nuclear staining reaction.

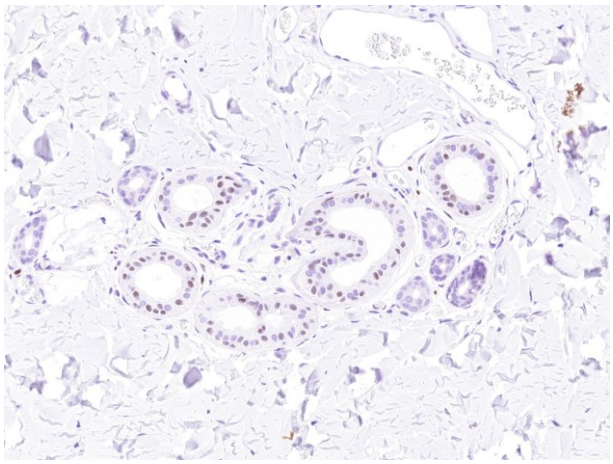


Fig. 2b
Insufficient SOX10 staining result of the skin, using same protocol as in Fig. 1b. A too weak staining reaction of the myoepithelial and luminal cells is seen – compare with Fig. 2a for optimal result.

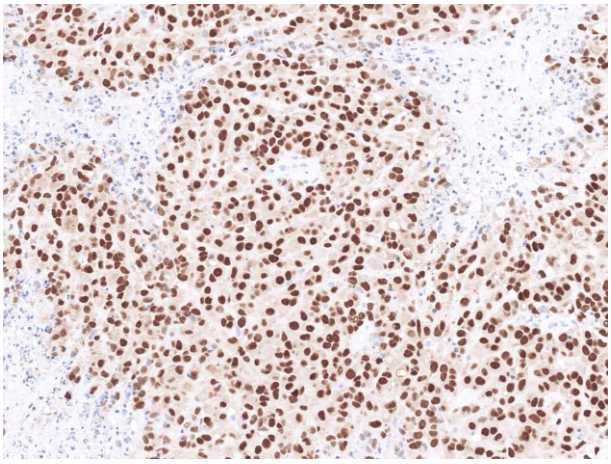


Fig. 3a
Optimal SOX10 staining result of the melanoma, tissue core no. 6, using same protocol as in Figs. 1a – 2a. Virtually all the neoplastic cells show an at least moderate but distinct nuclear staining reaction. A weak cytoplasmic staining was accepted in the neoplastic cells.

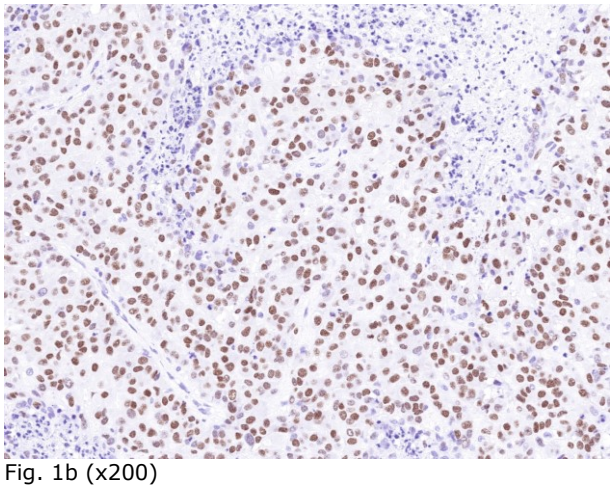


Fig. 1b (x200)
SOX10 staining result of the melanoma, tissue core no. 6, using same protocol as in Figs. 1b – 2b. The neoplastic cells show a weak nuclear staining reaction. The result overall assessed as insufficient.

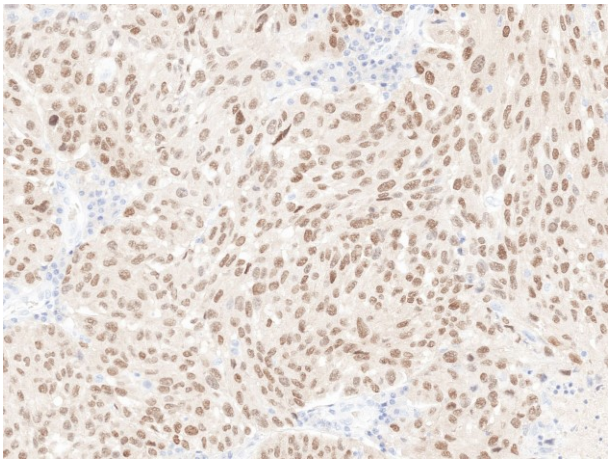


Fig. 4a
Optimal SOX10 staining result of the melanoma, tissue core no. 5, using same protocol as in Figs. 1a – 3a. Virtually all the neoplastic cells show an at least weak nuclear staining reaction.

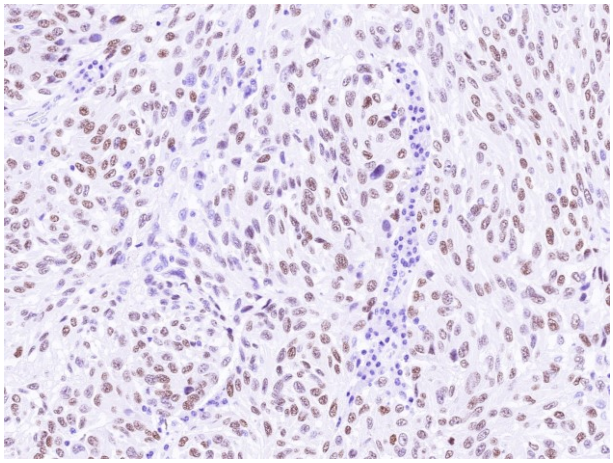


Fig. 4b
SOX10 staining result of the melanoma, tissue core no. 5, using same protocol as in Figs. 1b – 3b. The staining intensity of the neoplastic cells is weaker, compared to optimal result in Fig. 4a. The result overall assessed as insufficient.

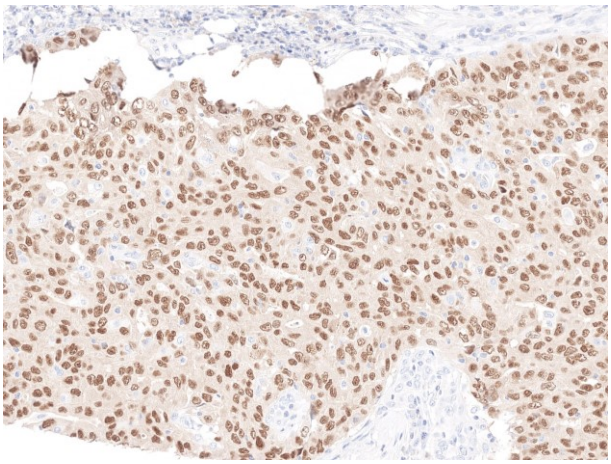


Fig. 5a
Optimal SOX10 staining result of the triple negative breast carcinoma using same protocol as in Figs. 1a – 4a. Virtually all neoplastic cells show a moderate nuclear staining reaction.

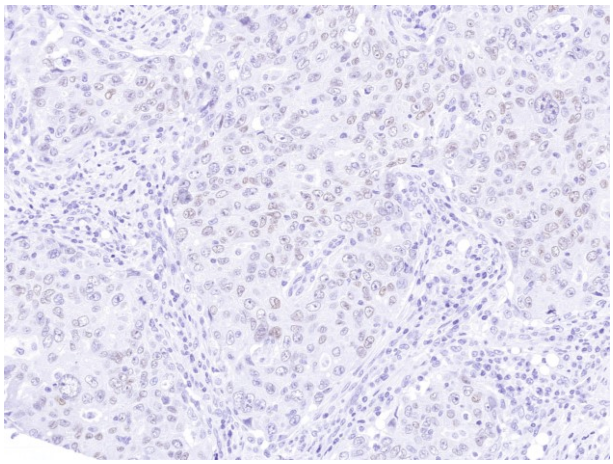


Fig. 5b
Insufficient SOX10 staining result of the triple negative breast carcinoma using same protocol as in Figs. 1b – 4b. Virtually all neoplastic cells are negative.

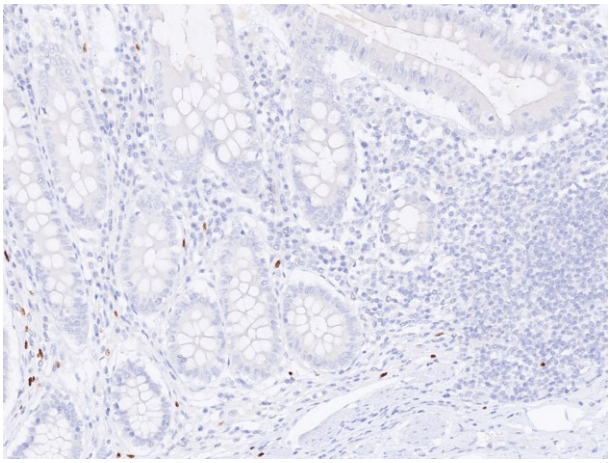


Fig. 6a
Optimal SOX10 staining result of the appendix using same protocol as in Figs. 1a – 5a. Virtually all Schwann cells in lamina propria mucosa show a strong nuclear staining reaction. The epithelial cells and lymphocytes are negative.

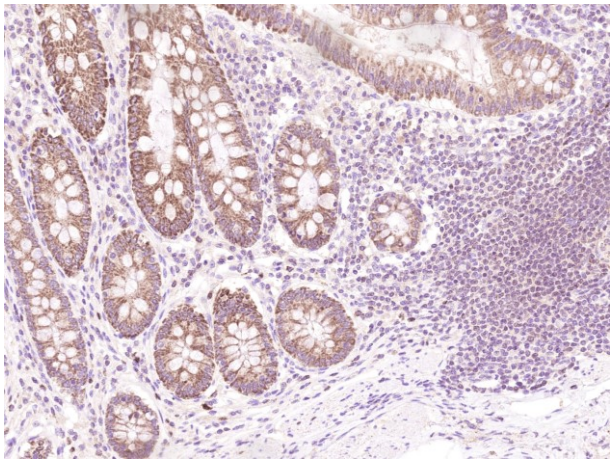


Fig. 6b
Insufficient SOX10 staining result using the rmAb clone EP268 with HIER in an alkaline buffer and a 3-step detection system. An aberrant cytoplasmic granular staining reaction is seen in both epithelial cells and lymphocytes. This aberrant and extensive cytoplasmic reaction was seen in 6/7 protocols based on this RTU format of EP268 from Master Diagnostica and might be related to a contamination with another Ab or other lot issues.

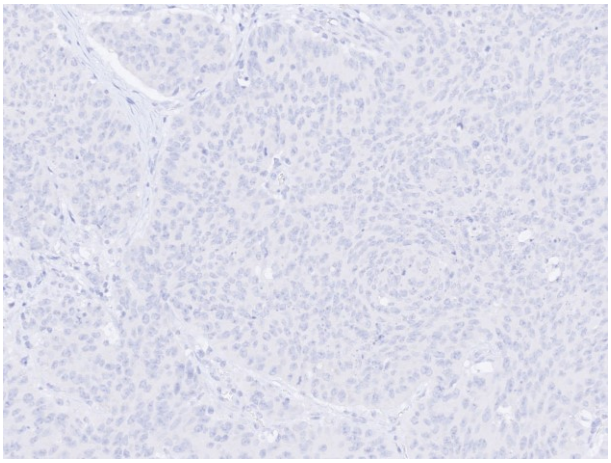


Fig. 7a
Optimal SOX10 staining result of the colon adenocarcinoma using same protocol as in Figs. 1a – 6a. The neoplastic cells are negative as expected.

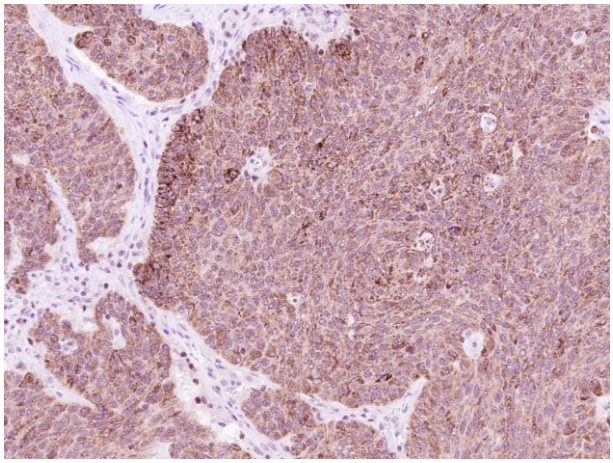


Fig. 7b
Insufficient SOX10 staining result of the colon adenocarcinoma using same protocol as in Fig. 6b. The neoplastic cells show an aberrant cytoplasmic granular staining reaction, complicating the interpretation.

HLK/LE/RR/SN 15.12.2025

Version	Description of change and reason	Date	Authorized by
2	Table 1c updated as error found in data entry	13.01.2026	HLK/TJU