HCB1/22: Histological and Cytological Staining

This EQA round was accomplished according to the document ***EQA Plan 2022***.

Typing conventions: *We are using comma as a decimal separator and dates in day.month.year format.*

Samples

The samples for this round were prepared by the subcontractor. Each participant received:

- 2 histological slides (labelled A and B).
  Sample A: Gastric resection, female, born 1962.
  Sample B: Gastric resection, female, born 1962.
- 2 cytological slides (labelled C and D).
  Sample C: Ascites, female, born 1974.
  Sample D: Fluidothorax, female, born 1974.

The staining to be performed by each participant was prescribed for each slide

Assessment of the participants' results

The tasks of the participants were:

1. Perform staining using a standard procedure that is routinely used in the laboratory (or perform an alternative staining) and mark the staining really used in the result form.
2. Send both stained slides (EQA samples) and filled in result form back to SEKK.

Assessment of participant's staining is performed by a team of 3 experts. This team evaluates the staining quality for each slide separately. The experts evaluate **the quality of staining** on the scale from **0 to 2 points** for each individual slide as follows:

Score (points)	***Description***	***Criteria***
2	Excellent staining	Staining without comments from the experts.
1	Acceptable staining	For HE staining (sample A) and MGG/HE (samples C and D) weak staining of the cores, which still allows to assess the details of the nuclear architecture. For methods detecting helicobacteria (sample B) any staining intensity that allows to identify the bacteria and determine their morphology.
0	Unacceptable staining	For HE staining (sample A) very weak staining of the cytoplasm of cells with hematoxylin, practically not allowing tissue evaluation, very weak staining of cell nuclei with eosin, not allowing to assess in detail the architecture of the nuclei. For methods detecting helicobacteria (sample B) the intensity of staining which no longer made it possible to identify bacteria and determine their morphology. For MGG and HE staining (samples C and D) very weak staining of the cytoplasm of cells, practically not allowing tissue evaluation and very weak staining of cell nuclei, not allowing to assess in detail the architecture of the nuclei.

Virtually every routinely used staining has many variants that are used according to the local customs and traditions of workplaces. Whether or not individual experts like a particular staining is usually the subject of discussion during the evaluation, but it does not affect the scoring of individual preparations - a key parameter of the assessment is the applicability of the staining in routine operation.

The staining quality of a particular slide is not evaluated if an expert has marked the slide as not assessable, or if the participant used other than the prescribed or alternative staining, or has not done the staining at all.

We do not process the slides and the results sent by the participants after the expert group meeting.

Experts assess all samples anonymously, i.e. without knowledge of the participant that sent the sample.

Team of the experts	Pavel Fabian, MD, PhD assoc. prof. Tomáš Jirásek, MD, PhD Iva Staniczková Zambo, MD, PhD

Using several anonymous model cases, the experts verified their assessment criteria and discussed possible points of dispute in order to ensure the maximum possible objectivity in the interpretation among all experts.

The scores for individual samples from individual experts are summated, so the sums could range from **0 to 6 points** for each slide. The scores achieved were then evaluated as follows:

Score	***Evaluation***	***Recommendation***
6 or 5	Excellent result	Without comments.
4 or 3	Acceptable result	It is advisable to improve the staining (the staining is not optimal).
2 and less	Unacceptable result	It is a warning signal and an impulse for an immediate action

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If a participant's result is evaluated as “excellent result” or “acceptable result” on the basis of the scoring, then the result is evaluated as **successful** in the EQA.

Supervisor’s comment

There were 84 participants in this round.

Sample A (histology)

HE staining (success rate 100 %): HE staining is the absolute basis of the methodology of any histomorphological laboratory and the participants manage this staining in a quality suitable for diagnostics.

Sample B (histology)

Detection of the helicobacteria staining (success rate 100 %): The participants are capable to detect helicobacteria histochemically. The assessors did not distinguish between the two dominantly used histochemical methods for the detection of helicobacteria, i.e. modifications of Giemsa staining and Warthin-Starry silvering (the representation of both procedures was approximately proportional in set of the results). Both methods yielded satisfactory results and allowed helicobacteria to be identified in the samples. In individual cases with a lower score, we recommend to revise the work procedure of the staining. The possible discoloration of the sample area during silver staining according to Warthin-Starry ("mirror") was not a reason for a lower score.

A total of three B samples provided by the participants with detection of helicobacteria by immunohistochemical methods were not assessed at all because the HCB programme is designed to check the quality of histological staining procedures, including histochemistry, while other EQA programmes are designed to evaluate immunohistochemical staining, such as *General Immunohistochemistry* - Staining (VIB).

Sample C (cytology)

MGG staining (success rate 99 %): In rare cases the slides were assessed as acceptable - this was due to the low intensity of nucleus staining. We recommend that laboratories whose results have been evaluated as acceptable to check the staining procedure for this method. Completely unacceptable results did not allow distinguishing diagnostically significant cell details.

HE staining (success rate 100 %): The results were fine here.

Sample D (cytology)

The same applies to the sample D as mentioned above for the sample C.

Summary

In the opinion of the experts the vast majority of the submitted preparations demonstrated good quality staining and routine practice usability; section thickness is a matter of local habit, as is the intensity of a tissue staining with hematoxylin and eosin. Samples that some laboratories consider excellent may be evaluated by another workplace as thick and unsatisfactory, or discolored (and vice versa). We reiterate that the measure of evaluation is usability in routine practice, not the "artistic impression" of the sample before the "jury".

Long term success rate

You can find in the following table the overview of the total success of the participants of this round over last 2 years. Individual ranges of success are defined in the column headers (0 % … no success; 50 % … success from 1 to 50 %; 75 % … success from 51 to 75 % etc.). Next 2 lines contain both absolute and relative number of participants that reached the success rate specified in the header.

	Success	***0 %***	***50 %***	***75 %***	***80 %***	***85 %***	***90 %***	***95 %***	***99 %***	***100 %***
Count	absolute	0	0	0	0	0	2	10	0	72
	relative	-	-	-	-	-	2,4 %	12 %	-	86 %
Note: You can find your individual success over last 2 years in your result sheet.										

The table shows that the most participants in this round show a long-term success rate of over 90 %.
A success rate of 90 % or less should be considered an impulse for the improvement.

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Supplements

As a supplement to this report individual participants receive:

Name of the supplement	***Remark***
Confirmation of attendance	Issued only to those participants who have met the conditions for its issuance.
Result sheet (qualitative results)	Issued only to those participants who sent us the results. In the result sheet you can find the scoring of the staining which was performed by a team of experts for individual glasses (the symbols are explained in the legend). Here you can compare your results with the anonymised results (points) of other participants.

The supplements are labelled by its name, the code of the EQA round, and the code of the participant and are intended for the participant's private purposes only.

Also we return all the slides that we received from the participants.

Additional information

The final report, with the exception of the supplements, is public. Further information is freely available to both participants and other professionals at www.sekk.cz, in particular:

- The summary of the results of this round, including this final report.
- The document ***EQA Plan*** (contains information that applies both to this round and also the EQA in general).
- Explanation of the content of the particular supplements mentioned above.
- Contact to the EQA provider and the EQA coordinator and the list of all supervisors, including contacts.