




DSCH

Best Laboratory Practise and Standardization of Immunohistochemistry Testing

Seminar, Copenhagen, May 18th 2011

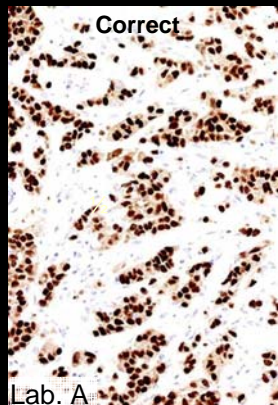
Mogens Vyberg
 Assoc professor
 Scheme director, NordiQC
 Inst. of Pathology, Aalborg Hospital
 Aarhus University Hospital
 Denmark

Ductal breast carcinoma



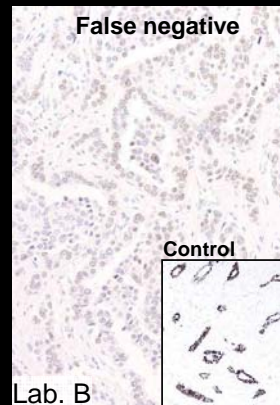
- Classifying
- Subtyping
- Staging
- Grading
- Identifying
 - predictive markers
 - therapeutic markers

External Quality Assurance: Estrogen Receptor



Correct

Lab. A

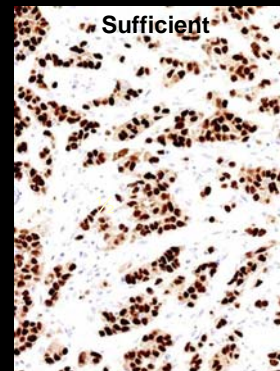


False negative

Lab. B

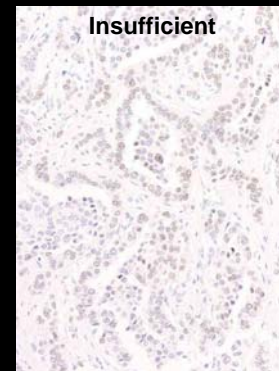
Control

NordiQC EQA: Estrogen Receptor 2003



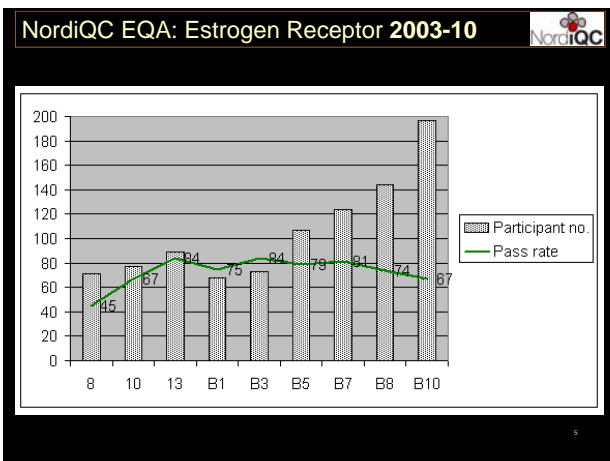
Sufficient

32 / 71 labs = 45%



Insufficient

39 / 71 labs = 55%



External Quality Assurance

- Staining quality varies greatly between different laboratories depending on the individual selection of methods and the technical expertise
- Internal quality control will often not identify a poorly calibrated IHC system giving insufficient staining results
- Standardization of staining methods is not possible but standardization of staining results is mandatory
- External quality assurance of staining results through laboratory proficiency testing is mandatory
- External quality assurance of antibodies, reagents and platforms through producer proficiency testing is mandatory

Establishment of a Nordic EQA program for IHC

Central assessment with consensus between experienced pathologists

- Correlate stains with central protocol parameters
 - Identify less successful Abs
 - Identify inappropriate protocol settings
- Publish general results and recommended protocols on an open website
- E-mail individual results to the participants
 - Give specific explanations for insufficient results
 - Give tailored recommendations for improvement

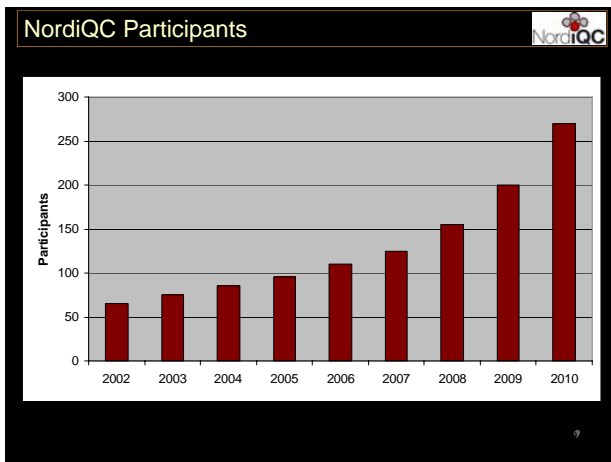
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Establishment of a Nordic EQA program for IHC

Nordic Immunohistochemical Quality Control
NordiQC founded 2003 by Nordic pathologists

- Independent, scientific, not-for-profit organisation
- Institute of pathology Aalborg Hospital
- General module: 3 annual runs
 - ~16 different markers/tests
- Breast cancer module: 2 annual runs

HER-2 IHC & BRISH, ER/PR

NordiQC Website: www.nordiqc.org

Home • Participation • Assessments • Episodes • Protocols • Techniques • Links

NordiQC is an independent scientific organization, promoting the quality of immunohistochemistry by arranging schemes for pathology laboratories, assessing tissue stains, giving recommendations for improvement and providing good protocols.

Last update: 15-04-2011

General results from **Run 31** (General module) and **G1** (Gastric cancer pilot module) are available by 7th April, see [Newsletters](#). Individual results are e-mailed.

Run 32 and B11 are closed for protocol submission (Deadline see 11th April). See the tests on [Participation](#). Unstained slides are circulated about 10th April.

NordiQC Seminar in collaboration with Danish Society for Color- and histochemistry - 18th May in Copenhagen! [Send Laboratories Details and Standardisation of Immunohistochemical Testing](#)

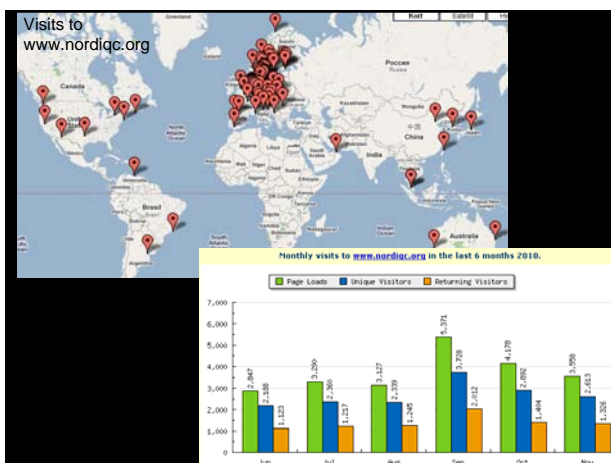
There are still seats available at the NordiQC Workshop September 2012, click on [Workshop 2012](#)



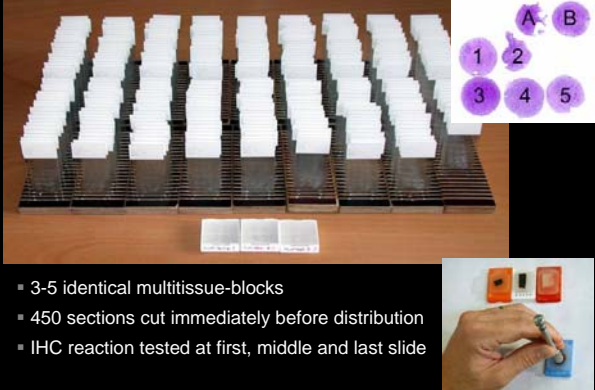
Fig. CD36 staining of nerves in appendix (A,B) and a pancreatic neuroendocrine carcinoma (C,D). A. Optimal staining of appendix. B. Staining of appendix using clone 123C3 on a BenchMark platform, the staining is only slightly weaker than in (A). C. Optimal staining of neuroendocrine carcinoma. D. Staining of endocrine carcinoma using clone 123C3 on a BenchMark platform, the staining is false negative. Clone 123C3 is platform dependent.



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NordiQC serial sections



- 3-5 identical multitissue-blocks
- 450 sections cut immediately before distribution
- IHC reaction tested at first, middle and last slide

NordiQC assessment

40 runs =
 > 25,000 slides =
 > 125,000 tissue sections
 assessed

Assessment and recommendations

A pdf-file e-mailed with individual marks, comments and suggestions

Marker	CD23	CR	CyD1	Ki67	Podop	TTF1
Assessment:	Poor	Optimal	Optimal	Good	Good	Borderline
Comments to the protocol:	False negative	-	-	Excessive counterstain	Weak	Weak*
Suggestions for improvement:	Consider change of primary Ab and recalibrate	-	-	-	-	Increase primary Ab conc. and/or prolong HIER

Marker	CR	Ep-CAM	IgK	IgM	TTF-1
Assessment:	Good	Inappropriate Ab	Poor	Poor	Borderline
Comments to the protocol:	Excessive background	Looks like CAM 5.2	False negative	False negative	Weak
Suggestions for improvement:	-	-	Use HIER as pre-treatment e.g. CC1 mild and consider change of the primary Ab and recalibrate	Consider change of primary Ab and recalibrate	Please read the epitope description and assessment summary carefully, as the choice of the Ab clone will influence the sensitivity and specificity

Over-all assessment results

NordiQC consensus marks (average 2003-08)

- Optimal: **36 %**
- Good: **33 %**
- Borderline: } **31 %** { too weak / false neg.: ~ 90 %
- Poor: } { over-stained / false pos.: ~ 10 %

NordiQC over-all assessment results

Major causes of insufficient stains: "Leading"

- Less successful / problematic antibodies **18 %**
- Inappropriate antibody dilution **39 %**
- Inappropriate epitope retrieval **31 %**
- Other inappropriate lab. performance **12 %**
 - Platform problems
 - Inappropriate calibration / home brews
 - Endogenous biotin reaction (EBR)
 - Section drying-out after HIER
 - Technical stainer errors
 - ...
 - Unexplained


Results of NordiQC tailored recommendations

Lab response to 419 advices (11 markers)

	No.	Improved	%
Positive	268	195	73
Negative	151	21	14

External IHC Quality Assurance

- Almost 1/3 of all IHC stains produced by NordiQC participants are still insufficient !
 - New labs
 - New antibodies, techniques, platforms
 - New challenges
- How many IHC stains produced by labs not participating in an EQA scheme are insufficient ?
- How many scientific publications are based on insufficient IHC stains ?
- What are the consequences for the patients ?

External Quality Assurance – ER 

JNCI Journal of the National Cancer Institute Advance Access published June 10, 2008

NEWS

Breast Cancer Testing Scandal Shines Spotlight on Black Box of Clinical Laboratory Testing

By Karyn Hede

“Through the inquiry, the public learned that between 1997 and 2005 nearly 400 of about 1,000 breast cancer patients received incorrect test results of the ER status of their breast tumors.”

“There are no good data on the quality of ER testing in the United States. The scary thing about the debacle in Canada is that we would never have known about this if results hadn't been checked in a central lab.”

Craig Allred

13.15 – 13.45	Best laboratory practice for examination of prognostic and predictive markers Prof. Emina Emilia Torlakovic, MD, PhD, FCAP UHN/Toronto General Hospital, Canada
13.45 – 13.50	Discussion
13.50 – 14.20	Labeling of reagents (IVD, ASR, RUO, CE, FDA) and what are the differences for you as a laboratory and for the patients? Frank Petersen, DS CERT A/S - Sundhed og fødevarer, Copenhagen
14.20 – 14.45	Discussion and break
14.45 – 15.15	Requirements for internal and external quality controls Søren Nielsen, HT, Scheme organizer, NordiQC
15.15 – 15.45	Pitfalls in immunohistochemistry Assoc. prof. Mogens Vyberg, MD, NordiQC, Aalborg Hospital, Aarhus University Hospital
15.45 – 16.30	Qualitative and quantitative interpretation of immunohistochemistry staining Prof. Emina Emilia Torlakovic, MD, PhD, FCAP UHN/Toronto General Hospital, Canada
16.30 – 16.50	Discussion
16.50 – 17.15	Networking. Wine and sandwich will be served.