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NORDIC COMMITTEE ON METHODS FOR FOODS

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The next NMKL Secretary General,

Nina Skall Nielsen, Technical University of Denmark, DTU

After the closure of the NMKL Annual Meeting 24 August 2016, the office of NMKL Secretary General will move from Norway to Denmark and Nina Skall Nielsen becomes the NMKL Secretary General.

Nina Skall Nielsen has a Master's Degree from the University of Southern Denmark in biotechnology, specialising in molecular biology. Further, she has a Ph.D. in biochemistry and nutrition from the Technical University of Denmark (DTU).

She has worked as a researcher / senior researcher, project manager and university teacher at the Danish Fisheries Institute, DTU Aqua and at the DTU Food Institute. Her work has mainly been related to edible fats in vegetables, fish and milk. The focus has been on the chemical and sensory quality, and protection of fats against rancidity by amongst others using different processing methods, and by using compounds with potential anti-oxidative effects, including natural anti-oxidants.

For the last five years, Nina has concurrently been the safety coordinator for the National Food Institute with about 400 employees.

Nina is relatively new in NMKL, as she joined the committee in January 2016.

Nina Skall Nielsen, DTU
the next NMKL Secretary General

The URL of NMKL will remain the same: **www.nmkl.org**

There will be no changes in the subscription of NMKL Publications.

The secretariat of NordVal International, which is a part of NMKL, will also move to DTU. There will be no changes in the certification process due to the change of office.

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NMKL methods vs ISO Standards according to Microbiological Criteria

COMMISSION REGULATION (EC) No 2073/2005 of 15 November 2005

From time to time, NMKL gets questions from private and public laboratories regarding NMKL methods versus ISO methods. They wonder if they can continue using NMKL methods, or if they need to incorporate ISO methods at the laboratory. Many laboratories, especially in the Nordic countries, prefer NMKL methods, as these are available in their own language, written in simple form, and have validation data included. The reason for the questions is that the methods of analysis referred to in the European Commission Regulation EC 2073/2005 are ISO methods only. However, the Regulation does allow the use of other analytical methods. Article 5, section 5, third and forth paragraph states:

(III) The use of alternative analytical methods is acceptable when the methods are validated against the reference method in Annex I [i.e. ISO standards] and if a proprietary method, certified by a third party in accordance with the protocol set out in EN/ ISO standard 16140 or other internationally accepted similar protocols, is used.

(IV) If the food business operator wishes to use analytical methods other than those validated and certified as described in paragraph 3 the methods shall be validated according to internationally accepted protocols and their use authorised by the competent authority.

According to *(III)*, methods certified by NordVal International or other certification bodies can be used.

According to *(IV)*, methods validated by standard developing organisations such as NMKL and AOAC International, can be used, as the methods are validated according to internationally accepted protocols, - if authorised by the competent authority. Being linked to the Nordic Council of Ministers, NMKL therefore asked to the Nordic countries to authorise the use of NMKL methods.

The following responses are received:

From Finland: Evira maintains a list of recommended practices on its website. The above NMKL methods can be used within the limits specified in the table of studies under the Regulation 2073/2005. See the list here.

From Iceland: With reference to Article 5, section 5, fourth paragraph of Commission Regulation (EC) 2073/2005, the Food Safety Authority in Iceland (Matvælastofnun / Icelandic Food and Veterinary Authority) allows that NMKL methods are used in line with ISO methods in microbiological analysis of foodstuffs.

From Norway: The Norwegian Food Safety Authority allows that NMKL methods are used as an alternative to the ISO methods in accordance with EU Regulation 2073/2005. A comparison of EN/ISO methods and NMKL methods has been presented to ESA on several occasions, and ESA has accepted the comparison. Generally, NMKL methods are at least as well tested as EN/ISO methods before being accepted.

National Food Agency, Sweden allows NMKL methods used as an alternative to ISO methods for analysis and sampling pursuant to Regulation 2073/2005.

Conclusion: It is OK to continue using NMKL methods!

The comparisons between NMKL and ISO methods are available here

NEW NMKL PROCEDURE: VERIFICATION OF MICROBIOLOGICAL METHODS

(NMKL PROCEDURE NO. 32, 2016)

Coming soon

Before an analytical method is taken into use, e.g. for routine testing, a laboratory must verify that it is capable of performing the analytical method. This includes evaluation of relevant performance characteristics to ensure that the method is fit for purpose within the laboratory.

This procedure provides guidelines on how

- a laboratory may verify that it is capable of performing an analytical method which has previously been externally validated,
- a laboratory can evaluate the possibility of analysing additional matrices not covered by the primary validation, or include new matrices in the scope of an already verified method, and
- a laboratory should plan a verification and elaborate a verification report.

The procedure is applicable to the verification of methods already validated properly in an interlaboratory study, and also to well recognised standardised methods without validation and specified performance characteristics.

The procedure also describes how measurement uncertainty could be established while verifying the method, as the data obtained in the verification can be used in the evaluation thereof.

The table below lists relevant method performance characteristics that should be evaluated in method validation and verification.

Method performance characteristics	Method Validation		Method Verification	
	Qualitative methods	Quantitative methods	Qualitative methods	Quantitative methods
Robustness	X	X		
Selectivity	X	X		
Sensitivity	X			
Specificity	X			
Level of Detec-	X		X	
Precision (repeatability / reproducibility)		X*		X
Trueness		X		X

*In the method validation, the precision may be given as an accuracy profile, which is a graphical illustration of the median of the results including the standard deviation.

How to carry out the verification of the different parameters is described in the procedure elaborated by:

- Charlotta Engdahl Axelsson, Eurofins, Sweden (Project leader)
- Majbritt Moos, Akzo Nobel Salt A/S, Denmark
- Sven Qvist, Denmark
- Saija Hallanvuo, Evira, Finnish Food Safety Authority, Finland
- Margrét Geirsdóttir, Matis, Iceland
- Hege Johnsrud, Eurofins, Norway
- Hilde Skår Norli, Norwegian Veterinary Institute / NMKL, Norway

Charlotta Engdahl Axelsson, Eurofins

SEMINAR ON MOLECULAR METHODS

PFGE PCR Whole- genome sequencing

DATE: Thursday 25 August 2016
PLACE: Stratos Culture and Event Youngstorget 2a, Oslo
LANGUAGE: Mainly Scandinavian languages
TARGET GROUP: Food authorities and food laboratories (private and public)

BACKGROUND: At the seminar, we will look at the use of molecular methods from a practical point of view, focusing on foodborne bacteria, in both control and research context. The seminar will examine the basis for the methodology (technology), practical applications and challenges, and discuss how we are going to use this technology in the future.

FOR PROGRAM AND REGISTRATION CLICK HERE
Are you interested in exhibiting, please contact NMKL General Secretariat on nmkl@vetinst.no.

INTERNATIONAL SYMPOSIUM - LABS IN A CRYSTAL BALL
NEW TRENDS FOR MULTI-COMPONENTS ANALYSIS

DATE: Tuesday 18 and Wednesday 19 October 2016
PLACE: Parque Científico de Barcelona, Torre D C/ Baldiri Reixac 4, Barcelona
LANGUAGE: English

Marine biotoxins
Veterinary drugs residues
Environmental contaminants
Pesticides and mycotoxins
Food packaging contact material
General quality assessment

This symposium is arranged by a scientific committee representing AOACI Europe Section, ACE Analytical Community Europe and Inkemia-IUCT Group

The symposium will include presentations from the foremost experts in different areas.

There will be exhibitions and poster presentations.

Abstracts for poster presentations are welcomed before 22 July 2016.

Students are strongly recommended to attend the Symposium for a reduced fee.

Program, information about registration etc. are available on **www.aoaceurope.com**.

AOAC INTERNATIONAL EUROPE SECTION OF AOAC INTERNATIONAL

Workshop on National Reference Laboratories (NRL) and coming Control Regulation 882 regarding NRL

Date: 8 November 2016
Place: Norwegian Veterinary Institute/ Norwegian Food Safety Authority
Language: English
Target group: Nordic contact persons from food authorities and Nordic representatives of NRL.
The workshop is sponsored by the Nordic Council of Ministers.

Aim of the workshop: To discuss the organisation of NRL, and the duties and requirements imposed by the current and the Control Regulation 882 on NRL and food authorities.

Experiences from NRL - How to fulfil the requirements and perform the tasks in an effective way? As the number of laboratories has decreased, and the scopes of the NRL are about to be widened, cooperation will be increasingly important. How can we facilitate Nordic cooperation and networking in these areas?

For program and registration click HERE or contact us at nmkl@vetinst.no

70th NMKL Annual Meeting

Date: 21—24 August 2016
Place: Røros, Norway
Target group: NMKL members
NMKL members are appointed experts from Denmark, Finland, Iceland, Norway and Sweden. During the Annual Meeting, the sub committees will discuss all the subjects on the working program. There will also be time for networking.

NMKL will celebrate its 70 year anniversary in a historic setting. Røros is sometimes called *Bergstaden* which means "the mining town" due to long history of copper mining. The inhabitants of Røros still work and live in the characteristic 17th and 18th century buildings which led to its designation as a UNESCO World Heritage Site in 1980. Røros has about 80 wooden houses, most of them standing around courtyards. Many retain their dark log facades, giving the town a medieval appearance.

New NordVal International protocol for the validation of microbiological alternative (proprietary) methods against a reference method

In line with the revision of ISO 16140 (validation protocol), in which NordVal International has participated, NordVal International has revised its Protocol. Both the ISO 16140-2 and the new NordVal Protocol were published this month, June 2016.

The ISO standard and the NordVal Protocol describe comprehensive validations where an alternative method is compared against a reference method. The major work is carried out at an expert laboratory. The laboratory also organises an inter-laboratory study. The main difference from the previous Protocol applies to quantitative determination and the statistical evaluation of results. One of the advantages with the new ISO standard, is that it describes acceptance level, i.e. gives guidance in when the alternative method should be considered satisfactory in performance.

New certifications of methods shall be performed in accordance with the new protocol. Methods that are already certified will be gradually revalidated and evaluated against the new protocol. Existing validation data may be used where appropriate, and evaluated statistically according to the new protocol. Any additional validations will be performed in expert laboratories, and should not be performed by each laboratory. Description of measures that must be re-evaluated for existing certified methods are available here.

The new Protocol and lists of matrices suggested tested for different organisms are available at www.nmkl.org under “NordVal”.

Number of samples needed in accordance with the new Protocol

Qualitative methods

Step 1: Comparison study, carried out at expert laboratory

For application to a broad range of foods, test at least 5 food categories. The following number of samples is required for the determination of:

- Sensitivity, specificity, relative accuracy and agreement between the methods: 5 food categories and 60 samples per category (whereof about 50% positive), i.e. 5 x 60 = 300 analyses with both the alternative method and reference method.
- Selectivity: 50 target organisms (100 for *Salmonella*) as a measure for the inclusivity + 30 non-target organisms for the exclusivity.
- Relative detection level: 1 matrix per food category; 3 levels for each matrix; blind, low level (25% -75% fractional recovery) and one with somewhat higher level. The number of replicates for the low level should be at least 20, and 5 for the other two levels.

Step 2: Inter-laboratory study arranged by the expert laboratory

It is required to obtain results from at least 10 collaborators, originating from at least 5 different laboratories. A maximum of 3 data sets can come from the same laboratory. The collaborators analyse one matrix with 4 levels and 8 replicates with both the alternative method and the reference method.

Quantitative methods

Step 1: Comparison study, carried out at one lab

For applicability to a broad range of foods, test at least 5 food categories. The following number of samples is required for the determination of:

- Relative trueness: 5 samples of 3 food types of the 5 categories, i.e. 75 samples analysed with both methods.
- Accuracy profile: 3 contamination levels per category with 2 different samples and 5 replicates, i.e. 150 samples per method.
- Selectivity: 50 target organisms for the inclusivity, and 30 non-target organisms for the exclusivity.

Step 2: Inter-laboratory study arranged by the expert lab

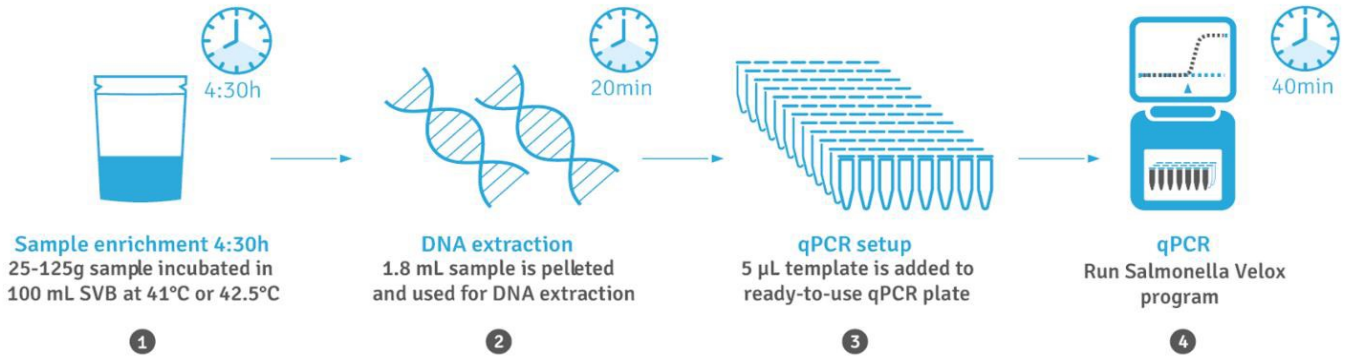
It is required to obtain results from at least 8 collaborators, originating from at least 4 different laboratories. Maximum 3 data sets can come from the same laboratory. The collaborators analyse one matrix with 3 levels and 2 replicates with both the alternative method and the reference method.

SALMONELLA VELOX - CERTIFIED BY NORDVAL INTERNATIONAL (NORDVAL 046)

Salmonella Velox is a rapid test for detection of the *Salmonella* spp. within only 5.5 hours!

NordVal International has evaluated and concluded that it has been adequately demonstrated that Salmonella Velox performs satisfactorily for meat and meat products (including poultry), fish and seafood. AnalyTech Miljølaboratorium AS, Denmark, has carried out extensive validations of Salmonella Velox from DNA Diagnostic A/S, Denmark.

The method includes an enrichment step, a DNA extraction step and a qPCR step. The enrichment is carried out with Salmonella Velox Broth for 4.5 h at 41°C or 42.5°C ± 0.5°C. The DNA extraction step uses the Salmonella Velox DNA Extraction Kit for DNA extraction from 1.5 -1.8 mL of the enrichment. The qPCR step uses the Salmonella Velox qPCR Kit and 5 µL DNA extract. The DNA sample is added to a 96 well qPCR plate containing qPCR reaction mix. The qPCR reaction runs for approximately 40 minutes.



In February 2016, the comparison study was carried out according to the NordVal Protocol of June 2016, i.e. ISO 16140-2:2016. Salmonella Velox was compared against ISO 6579:2002.

For testing the selectivity, i.e. the method's ability to detect the target microorganism from a wide range of strains (the inclusivity), and the lack of interference from a relevant range of non-target microorganisms (the exclusivity), 110 *Salmonella* strains and 30 non-*Salmonella* bacteria were tested with Salmonella Velox and ISO 6579. Two samples with *Salmonella* strains were excluded due to failure of spiking. The remaining 108 *Salmonella* strains, representing 101 different *Salmonella* serovars, all tested positive with both Salmonella Velox and the reference method. All non-*Salmonella* strains were negative with both methods. The selectivity was therefore 100% for both methods.

For the sensitivity study, i.e. the ability to detect the analyte, a total of 186 samples were tested with both methods. The matrix categories tested were raw meat (including poultry), ready-to-cook meat products (including poultry), and raw and ready-to-cook fish and seafood. Overall, the results were satisfactory. The only deviation in the results between the methods, was that five of the results were positive, and confirmed positive, with the alternative method, but were negative with the reference method. Hence, the sensitivity is somewhat better with the alternative method than for the reference method.

The level of detection (LOD) for the Salmonella Velox method was compared against the LOD of the ISO 6579, by analysing 92 samples at low levels. The results were satisfactory with both methods, thus the relative level of detection (RLOD) for Salmonella Velox is satisfactory.

In April /May 2016, AnalyTech Miljølaboratorium AS organised an interlaboratory study (ILS) of the Salmonella Velox method. Eight laboratories analysed pre-enriched samples in duplicates. The matrix was raw unprocessed pork samples, inoculated with cold stressed *Salmonella typhimurium* at three different contamination levels.

The results document no statistical difference in the performances between the Salmonella Velox method and the reference method ISO 6579:2002. For the certificate click here.

Renewed NordVal Certificates

The following NordVal Certificates were renewed in June 2016:

- **TRANSIA® PLATE Listeria (NordVal 002)**

The TRANSIA® PLATE Listeria from Biocontrol is an ELISA method, tested for detection of Listeria in foods and environmental samples. See the certificate here.

- **RAPID'Salmonella method short protocol & RAPID'Salmonella method double enrichment protocol (24h) (NordVal 032)**

RAPID'Salmonella from Bio-Rad is a chromogenic agar medium, and the principle of the method relies on demonstration of two enzymatic activities.
The RAPID'Salmonella method with short protocol and RAPID'Salmonella method with double enrichment protocol are applicable to foods, animal feeds and environmental samples. The sensitivity and the detection level were satisfactory. The agreement between the RAPID'Salmonella method with double enrichment protocol and the reference method were not satisfactory due to a high number of false positives with the alternative method. Thus, according to this study, the double enrichment protocol will cause false positives in 20% of the cases, and hence confirmation is necessary. See the certificate here.

Visit NordVal at www.nmkl.org for certificates and the new NordVal Validation Protocol

THANK YOU!

I am about to finalise my very last NMKL Newsletter, my 57th issue in 19 years. At the coming NMKL Annual Meeting, I am resigning as NMKL Secretary General, as the Norwegian Veterinary Institute (NVI) will no longer be hosting the office.


Nina Bakkelund, Secretary

Since October 1997, NVI has held the secretariat, and together with Nina Bakkelund (secretary) I have had the honour and pleasure of working for a thriving and vibrant organisation. I have been privileged in working with wonderful, dedicated and knowledgeable experts, both within the Nordic cooperation and internationally.

Thank you so very much for good cooperation and for some fantastic and interesting years!


Hilde S.Norli, Secretary General

Both Nina Bakkelund and I will miss working in NMKL. However, we will be dealt new and interesting tasks at the NVI. Nina will continue to work as the secretary for the Norwegian National Committee. I feel reassured that the secretariat will be in good hands at the Technical University of Denmark, and I am convinced that NMKL will continue as the unique network as it is, keeping up the good work in elaborating analytical methods and guidelines, and arranging workshops and symposiums. All the best for the years to come!

Available NMKL Procedures

- 1, 2. Ed. 2005 Calibration and performance checking of laboratory balances
- 3, 2. Ed., 2016 Control charts and control materials in internal quality control in food chemical laboratories
- 4, 3. Ed., 2009 Validation of chemical analytical methods
- 5, 2. Ed. 2003 Estimation and expression of measurement uncertainty in chemical analysis
- 6, 1998, (Adm 2002, Adm 2006) Generelle retningslinier for kvalitetssikring af sensoriske laboratorier. (only available in Danish and Finnish)
- 7, 1998 Checking of UV/VIS spectrophotometers
- 8, 4. Ed. 2008 Measurement of uncertainty in quantitative microbiological examination of foods
- 9, 2. Ed., 2007 Evaluation of method bias using certified reference materials.
- 10, 2001 Control of microbiological media
- 11, 2. Ed. 2010 Procedure for sensory analysis of drinking water
- 12, 2. Ed., 2014 Guide on sampling for analysis of foods
- 13, 2003 Volumetric control
- 14, 2004 SENSVAL: Guidelines for internal control in sensory analysis laboratories
- 16, 2005 (2007) Sensory quality control
- 17, 2006 Guidelines for requirement specifications for food analyses
- 18, 2006 The use of reference materials, reference strains and control charts in a food microbiological laboratory
- 19, 2007 Guidelines for sensorial analysis of food containers/packages
- 20, 2007 Evaluation of results from qualitative methods
- 21, 2008 Guide for sensory analysis of fish and shellfish
- 22, 2008 Considerations regarding evaluation of immunochemical test kits for food analysis
- 23, 2008 Guide on quality assurance in microbiological laboratories
- 24, 2010 Guidelines for quality assurance for food chemical laboratories
- 25, 2014 Recovery information in analytical measurement
- 26, 2012 Control and internal calibration of thermometers and temperature control on microbiological laboratories
- 27, 2013 Measurement uncertainty in sensory analysis
- 28, 2014 Guidelines for reporting sensory data
- 29, 2014 Guidelines for sensory analysis of meat and meat products
- 30, 2014 Statistical evaluation of results from quantitative microbiological methods
- 31, 2015 Guidelines for sensory evaluation of bread