5. newsletter, June 2012

EAVLD Newsletter

This newsletter...

Foreword

Dear colleague

This fifth EAVLD Newsletter comprises updated information about the Congress 1-4 July 2012 in Poland and different issues relevant for the progress of the association. Special attention should be focused on the need for EAVLD country representatives and new board members to be elected at the general assembly on 3 July at the Congress. Further, you find description of new diagnostic tools on page 4-7.

EAVLD Congress 2012

Updated information from June 1st, 2012
provided by The Organizing Committee

- congress website at www.eavld2012.org has had >11.000 hits.
- scientific program is available on the congress website as well as the instructions for preparation of oral presentations and posters.
- 5 invited speakers will give keynote lectures on 4 main topics of the congress.
- 2 young scientists have been awarded the Young Scientist Award which is free registration for the congress.
- 39 oral presentations and 77 posters were selected. Abstracts for posters can still be submitted.
- approx. 170 people have registered so far from 26 countries including Australia, Canada, Europe, Iran, Sudan and USA. We expect 200-220 participants in total.
- 88 participants have registered for shuttle buses from Warsaw airport directly to Kazimierz Dolny and back; we strongly advise to register for this mode of transfer: it's cheap, fast and convenient.
- the congress hotel is fully booked – if problems with accommodation in other hotels are encountered, please contact the Organizing Committee for assistance.
- 19 sponsors will be present in the field of veterinary diagnostics (all available slots).

EAVLD Newsletter

Election of EAVLD board members

During the upcoming EAVLD congress, there will also be elections for new or additional board members. We are currently in a possition that present board members can remain in the board for another 2 years, but most will have to step down in 2014. If most of the board would step down in 2014, all at the same time, this would be a serious continuity problem. To allow for future continuity in the board, the number of boardmembers will be increased for the next two years. This will allow for a few board members to become part of the team now, and continue their work after 2014. This will be a one-time solution to the current problem. After that, it is expected that we end up with a schedule in which more or less half the board will step down each 2 years.

If you are willing to become a board member, please contact the secretary of the EAVLD (Willie Loeffen: secretary@eavld.org) for more information. The association will not run itself, but is in need of volunteers who want to spend some time in managing some activities of the board. Besides regular board members, we are also looking for a secretary and vice president. The vice president will automatically become president in another 2 years, and past-president 2 years after that. We are especially looking for board members from countries that are not yet represented in the current board. Also women and young diagnosticians are especialy requested to respond, as these are currently underrepresented in the board. For young diagnosticians, this is a good way to become more involved with an international association, with the ability to expand your horizon and network.

The present board members are from Denmark, Germany, Poland, Spain, Sweden, The Netherlands and UK. Look for details at http://www.eavld.org/joomla/index.php/governing-board-of-eavld

Progress of EAVLD

The establishment of EAVLD was announced during the WAVLD Congress in Madrid in June 2009, to create an inclusive forum for veterinary laboratory scientists working in all disciplines across the full range of animal species, and include everyone with an interest in the veterinary laboratory diagnostics in Europe. Since then the growth of the EAVLD has been quite significant, and now we are more than 200 members in most European countries. Our first Congress was held in Lelystad (Netherlands) in 2010, and it was a great and successful event with participations of many colleagues from all over Europe. It was very helpful to strengthen the already existing collaborations, and to establish new ones. Now we are preparing the second Congress next 1-4 July in Kazimierz Dolny (Poland), which we expect to be at least as successful as the first.

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Members are encouraged to contribute to the Newsletter

During these years we have also set up a newsletter that is published twice every year. We would like it to become a communication tool for all members of EAVLD, and it will include scientific short communications as well as news and events that may be of relevance for colleagues working in the Veterinary Laboratory Diagnostics field. We would like to have members' opinions, thoughts and views about the laboratory diagnosis of animal diseases and everything related to it. We encourage you to send contributions to the editor Sven Erik Jorsal: selj@vet.dtu.dk

Join as country representatives

The EAVLD cannot grow without the contribution of its members, and unfortunately, Board members cannot reach everywhere. For that reason the Association needs to have country representatives in each country. They can make the EAVLD known to their colleagues, increase the number of members, become a first level of communication between members in their countries and the Association, help Board in any matter related to their country, and find and suggest possible contributions and relevant news for our Newsletter. But they should preferably take an active role and have an ongoing communication with the Board, sending opinions, suggestions and anything they may find relevant for the association.

Please, if you want to collaborate in making the EAVLD bigger and stronger, or you know somebody that could be of help, send a message to: Jose A. Garcia, Dpto. Email: gcabrera@vet.ucm.es. or contact any Board member.

The present country representatives with their email addresses are listed below:

Country	Representative	E-mail
Belgium	Ann-Brigitte Caij	ann.brigitte.caij@var.fgov.be
Denmark	Sven Erik Jorsal*	selj@vet.dtu.dk
Finland	Varpu Hirvelä-Koski	varpu.hirvela-koski@evira.fi
Germany	Martin Beer*	Martin.Beer@fli.bund.de
Greece	Katerina Marinou	katmarinou@gmail.com
Ireland	Rónan O'Neill	ronan.oneill@agriculture.gov.ie
Italy	Gian Luca Autorino	gianluca.autorino@izslt.it
Netherlands	Willie Loeffen* Gerard J. Wellenberg*	willie.loeffen@wur.nl g.wellenberg@gddeventer.com
Poland	Tadeusz M. Wijaszka*	t.wijaszka@ur.krakow.pl
Portugal	Gertrude Thompson	gat1@mail.icav.up.pt
Romania	Mihail Turcitu	Mihai.Turcitu@idah.ro
Slovakia	Zuzana Dirbakova	dirbakova@svuzv.sk, molbiol@svuzv.sk
Slovenia	Brane Krt	Brane.Krt@vf.uni-lj.si
Spain	Jose A. Garcia*	gcabrera@vet.ucm.es
Sweden	Frederik Widén*	Frederik.Widen@sva.se
United Kingdom	Andrew Soldan*	Andrew.Soldan@ahvla.gsi.gov.uk

*: Present board members

Links for EAVLD website

EAVLD is currently working on expanding the link pages on our website:

http://www.eavld.org/

If you have any interesting web-links you want to share with your colleagues, please contact Frederik Widén, our treasurer and webmaster (treasurer@eavld.org).

Links to relevant websites on veterinary diagnostics are welcome. Also links to your own institute or company could be put on-line. This way we hope to create a good overview of relevant diagnostic institutes and companies in Europe.

Development of new tools for multiplex analysis

To increase the efficacy and speed in the test of a vast number of serum samples via high throughput screening, it is crucial to develop methods that facilitate simultaneous analysis of several parameters in a serum sample. Multiplex analysis denotes analytical methods where several factors such as antigens, antibodies, cytokines etc. can be measured simultaneously in a sample. Multiplex analysis may be based on mixtures of microparticles. Each microparticle has a color code and e.g. a particular bacterial component/antigen bound to its surface. When this particular color code shows a positive response in the analysis read out this shows the sample contains antibodies which corresponds to a particular microorganism/infection.

Bead based multiplex flow cytometry applies fluorescent microparticles that can be separated by fluorescence intensity or different fluorescence color. The fluorescence code identifies the different beads modified with different bacterial components. The method with which the antigen is coupled to the micro bead surface is crucial to achieve high recognition between the bead bound antigen and antibodies in the sample, and thereby enhancing the sensitivity of the multiplex assay method.

We have developed a new, efficient and generic chemical method to couple bacterial polysaccharide antigens to microparticles or other types of surfaces. Existing methods for the binding or coupling of polysaccharide antigens to surfaces, generally results in lower recognition of antibodies in the sample due to the formation of unstable binding, or as a result of binding methods that compromise the antigenic molecular structure under the coupling process.

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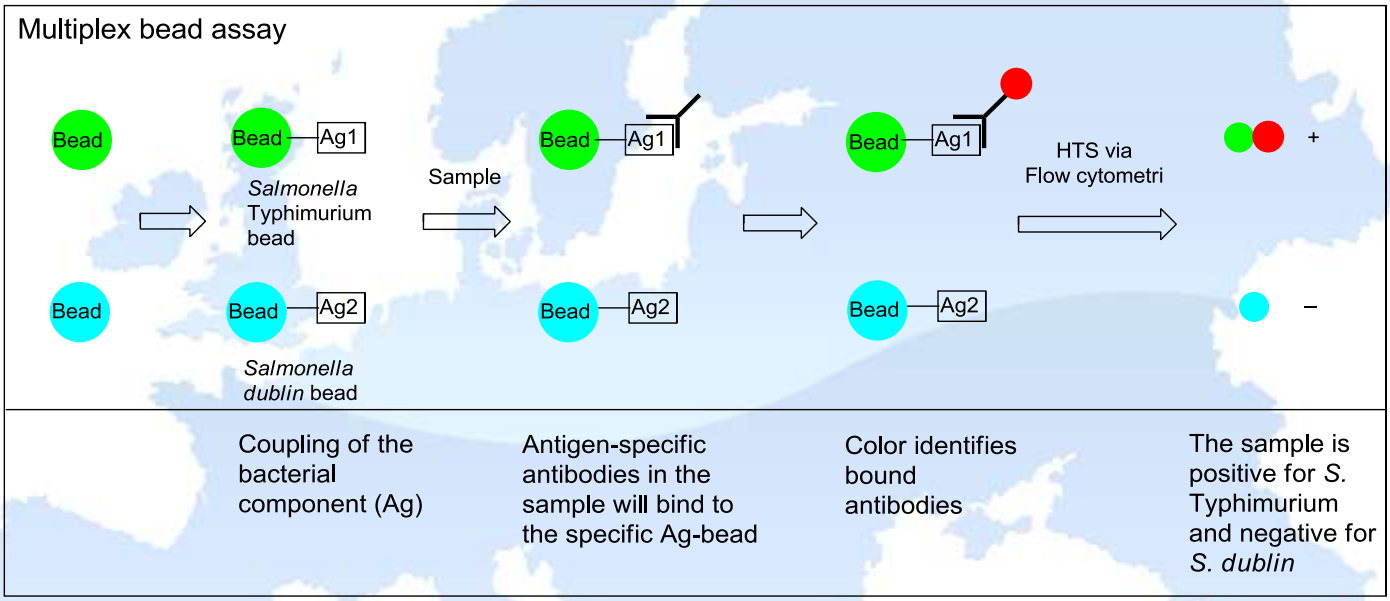
The new chemical method involves coupling to a region of the LPS antigen which is not involved in the recognition of antibodies. This coupling ensures a high recognition of antibodies in the sample. Although the method is generic the project has investigated the coupling method in combination with flow cytometry using two LPS antigens from *Salmonella* Typhimurium and *Salmonella dublin* as a model for the development of sensitive flow cytometry multiplex assays.

By Ulrik Boas and Ulla Riber, National Veterinary Institute, Technical University of Denmark.



Metagenomic pathogen detection

Metagenomics is "the analysis of genomic DNA from a whole community" (2) or, more specifically, "the application of modern genomics techniques to the study of communities of microbial organisms directly in their natural environments, bypassing the need for isolation and lab cultivation of individual species" (1). The latter definition highlights the most important features of metagenome analysis for the detection of novel or unexpected pathogens especially for diagnostic purposes.

Due to the fact that all pathogens comprise a genome made up of nucleic acids, it is possible to detect and identify these by sequencing the complete nucleic acid content of a given sample. The use of metagenome analysis for pathogen detection was fuelled by the availability of the different Next Generation Sequencing (NGS) technologies.

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These various technologies enable unbiased sequencing at reasonable prices per base with an unprecedented throughput. The unbiased sequencing is a key feature because it enables analyzing the complete community with equal probability for detection of all its components. The high throughput is at the same time a possibility and a challenge for metagenomics. Because the resulting datasets can contain up to millions of reads (short 50-500 bp stretches of sequences each representing an individual molecule of the input nucleic acids), the sensitivity of the approach is easily scalable, i.e. the chance to detect the pathogen only depends on the total number of reads generated. On the other hand, this poses a problem for the efficient analysis of the resulting datasets because there may only be single sequencing reads which represent the pathogen.

The general workflow of metagenomics pathogen detection is depicted in figure 1. First of all, an appropriate sample is drawn from the diseased animal (Fig 1 A). This may be body fluids or tissue samples. From this sample, all nucleic acids are extracted and are converted into a sequencing library (Fig 1 B) in a multistep process which includes reverse transcription of RNA into DNA in order to enable detection of pathogens with an RNA genome. This library is then prepared for sequencing and finally sequenced to yield thousands or even millions of individual sequencing reads (Fig 1 C). After sequencing, the complete set of sequences has to be analyzed in order to group every single read according to the taxon it is closest related to. For this purpose, all reads have to be compared to all known sequences available (Fig 1 D). This is a costly process requiring powerful computing resources and elaborate algorithms for efficient and reliable classification. The final output of this computational analysis of a metagenomics dataset is a listing of all taxa identified in the read sequences (Fig 1 E). Within this information, a diagnostician has then to identify the potential pathogen.

The pathogen detection procedure outlined above has been established at the Friedrich-Loeffler-Institut, the German Federal Research Institute for Animal Health, in the last years. This included especially optimization of the sample preparation and sequencing procedures as well as programming of a computational pipeline for data analysis. For data analysis, i.e. classification of all reads into the various taxa, a powerful software pipeline was set up. This analysis pipeline fully automatically generates a clear tabular report which can be used by the diagnostician to get an overview of all detected potential pathogens. Making use of the established routines, we were able to identify the novel orthobunyavirus Schmallenberg virus in serum samples from cattle suffering from an unknown disease causing fever, diarrhea, and a decrease in milk production.

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By Dirk Hoeper, Matthias Scheuch and Martin Beer

1. Chen, K., and L. Pachter. 2005. Bioinformatics for whole-genome shotgun sequencing of microbial communities. PLoS Comput Biol 1:106-112.
2. Gilbert, J. A., and C. L. Dupont. 2011. Microbial metagenomics: beyond the genome. Ann Rev Mar Sci 3:347-371.