In the spotlight

News from the CONffIDENCE project

Dear stakeholder,

The CONffIDENCE project team is proud to present the 7th edition of the CONffIDENCE e-newsletter. In this newsletter you will find recent developments in the CONffIDENCE project and related information in the area of contaminants in food and feed.

If you want to subscribe to CONffIDENCE News, please fill in the registration form on http://www.conffidence.eu/Stakeholders/registration.php

Best regards,

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In the spotlight

CONffIDENCE: 2nd Open Day in Brazil

During the International Triennial Meeting of World Aquaculture Society (WAS) and FENACAM, held from 6-10th June 2011 in Natal Brazil, the CONffIDENCE project held its 2nd Open Day together with a booth presentation. The Open Day with the title: Rapid tests for contaminants was organized on Thursday the 9th of June during the afternoon session, as part of the session with the title International Cooperation in S&T and Aquaculture research under the European Union’s Seventh Framework Programme (FP7), organized by the European Commission (delegation of the EU in Brazil & Research & Innovation DG). The Open Day included 4 oral presentations, giving an overview of CONffIDENCE, the industrial needs regarding rapid tests for contaminants, and key results obtained thus far for persistent organic pollutants (POP’s) and heavy metals. During the break a poster session was held where various project members presented CONffIDENCE results. The combination of posters and oral presentations proved to be effective in generating interest and stimulating discussions with the
A full report and overview of the posters presented during this open day can be found on the CONffIDENCE internet site.

Read more about World Aquaculture 2011…

CONFfIDENCE at APIMONDIA 2011

APIMONDIA was held at Buenos Aires on 21 and 25 September 2011. It is the most complete event because of the great variety in the audience, from Argentina, Latin America and all over Europe, but also because of the number and diversity of offers in terms of information. International Scientific Presentations, Round tables with specific topics, Conferences and symposiums designed and presented by worldwide recognized specialists, that covered many areas so the beekeeper can increase his production and make the most out of his resources.

Stefan Weigel (RIKILT) and Vincent Chabottaux (UNISENSOR) gave a lecture to explain the general CONffIDENCE activities and the development of fast multiplex dipstick assays for screening of antibiotics in honey. During this meeting, a CONffIDENCE exhibitor booth was crewed by UNISENSOR and RIKILT partners. Two scientific posters were shown: one about the development of a new multiplex dipstick for the simultaneous detection of sulfonamides, fluoroquinolones, tylosin and chloramphenicol in honey and the second about the effect of acidic hydrolysis on sulfamides detection in honey with new generic antibody-based dipstick assay.

The prototype multiplex kit was shown and principles of the test explained to the booth visitors (more than 500) such as beekeepers, honey quality responsible, honey packers, honey exporters and responsible of cooperatives from different countries including Argentina, Belgium, The Netherlands, Germany, Chili, Africa, USA, Portugal, Spain, Russia, China, New Zealand, Greek, Turkey, France… A total of 1500 leaflets (related to Conffidence project or honey dipstick) were distributed. A call was also launched to the parties that are interested to participate in a field survey using the new dipsticks.

Read more about the field evaluation on honey dipstick…

Read more about APIMONDIA 2011…
CONffIDENCE at FIAPP-VICTAM International 2011

FIAPP - VICTAM International 2011, held in Köln on 3-5 May is the most important event for the feed industry. The exhibition is the only trade show (600 exposants) and series of conferences and seminars organised specifically for producers and suppliers of specialist ingredients and additives for the animal, petfood and aqua feed industries.

The use restrictions of animal proteins in feed led the sector to turn towards other protein sources. In the framework of the scientific conference FIAPP organized in association with the European Feed Technology Center (EUFETEC), several presentations were dedicated to sunflower meal and distiller’s dried grains with solubles (DDGS) as alternative protein sources.

Another issue developed in the framework of this event was the concern from feed companies to improve the industrial processes in order to reduce the cross-contaminations and the energetic costs. Jacob de Jong from RIKILT presented on behalf of the NUTRECO Company, the industrial needs for the feed control and on behalf of the partners in the CONffIDENCE project, the first results achieved on rapid methods in development for chemical contaminants in feed.

Read more on the presentation…

Read more on FIAPP - VICTAM International 2011…

CONffIDENCE: Next Open Day

Since the start of this project on May 1, 2008, much effort has been put into the development of rapid methods for food and feed safety. The project team would like to share its results so far with all interested stakeholders. For this, the third Open day will be organized on 3rd November 2011, in the framework of the RAFA 2011 Conference. By this way, stakeholders can be informed regarding the project results. More information on this event can be found in the upcoming events section of this newsletter.

Read more about the 3rd CONffIDENCE OpenDay…

Field evaluation on honey multiplex dipstick for antibiotics

A simple and cheap test has been developed within the EU project CONffIDENCE that allows collectors, intermediaries and staff at cooperatives to test individual lots of honey for antibiotics before purchase. The multiplex dipstick detects sulfonamides, tylosin, fluoroquinolones and chloramphenicol. The test can be carried out under field conditions. The entire procedure does not take more than 45 min/8 samples from sampling to result. We intend to test the dipstick under real field conditions to prove its applicability and robustness. Therefore, we look for parties that are interested to participate in a field survey using the new dipsticks.

Read more about this test…
News from the CONffIDENCE project

Developing detection methods for organic pollutants; POPs, PFCs, Pesticides

Regarding the POPs (WP1a), the results concerning the newly developed sample preparation method and alternative possibilities of final instrumental analysis were presented by K. Kalachova during the 5th international meeting on Chemistry & Life 2011 conference held in Brno (Czech Republic) on 14-16th September 2011. The presentation was focused on the possibility to employ triple quadrupole ion analyzer (MS/MS) for detection of target analytes following their gas chromatographic (GC) separation. GC-MS/MS was shown to be a good alternative to comprehensive gas chromatography (GCxGC) employing time of flight (TOF) MS in terms of highly specific detection of even trace levels of brominated flame retardants (BFRs), polychlorinated biphenyls (PCBs) including dioxine-like and polycyclic aromatic hydrocarbons (PAHs).

Read more about the conference....

Persistent organic pollutants (POPs) are food contaminants of a global public health concern and known to be carcinogenic and endocrine disruptors. Their monitoring is essential and an easy-to-use, rapid and affordable multi-analyte screening method with a simplified sample preparation can be a valuable tool prior to instrumental analysis. For this purpose, a flow cytometric immunoassay (FCIA), based on a spectrally-encoded microbeads technology, was developed for the multiplex detection of polycyclic aromatic hydrocarbons (PAHs), polychlorinated biphenyls (PCBs) and the emerging polybrominated diphenyl ethers (BDEs) in buffer and fish extracts. A paper from Meimaridou et al. entitled “Multiplex Screening of Persistent Organic Pollutants in Fish using Spectrally-Encoded Microspheres” was recently published in Analytical Chemistry.

Read more about this publication....

The development of a new sample preparation method for the simultaneous determination of PCBs, PBDEs and PAHs in fish and shrimps and results of validation study were described in the paper “Simplified and rapid determination of polychlorinated biphenyls, polybrominated diphenyl ethers, and polycyclic aromatic hydrocarbons in fish and shrimps integrated into a single method” by Kalachova et al. The manuscript was accepted in September 2011 for the publication in Analytica Chimica Acta. Briefly, the ethyl acetate-based extraction followed by silica minicolumn clean-up was used for the sample preparation and GC-TOFMS instrumental technique was applied for the simultaneous determination of all target analytes as well as for potential non-target screening. DART–TOFMS was shown to be an effective tool for the fat control in sample extracts.

Read more about this publication....
During recent years, mass spectrometry (MS) and hyphenated chromatographic instrumentation and techniques have been a subject of dramatic developments, resulting in the introduction of various useful tools for the analysis of halogenated persistent organic pollutants (POPs) and polycyclic aromatic hydrocarbons (PAHs) in food and environmental matrices. A chapter entitled “Halogenated persistent organic pollutants and polycyclic aromatic hydrocarbons in food” published in “Mass Spectrometry in food Safety – Methods and protocols” and written partially in the framework of the CONffIDENCE project, describes state-of-the-art in the field of MS as a primary detection tool for the halogenated POPs and PAHs previously separated using either gas chromatography (GC) or liquid chromatography (LC). Since sample preparation practice plays a crucial role for obtaining optimal performance characteristics of a particular analytical method, a brief overview of sample extraction and clean-up procedures in the POPs/PAHs analysis is also briefly outlined. 

Read more about this publication...

In the 1st Interlaboratory exercise, 8 participants were invited to take part in this study pertaining to expert laboratories in PFCs analysis from 6 European countries. However, just 7 participants finalized the interlaboratory exercise. Laboratories participated in order to assess the performance characteristics of a simplified analytical method for 3 selected PFCs (L-PFOS, PFOA and FOSA) in food matrices (fish tissue and milk). This study was performed in order to establish the applicability of a developed and validated method by ICT-Praha and IDAEA-CSIC under the frame of WP 1b CONffIDENCE project. Read more on PFCs study...

The occurrence of organohalogenated compounds including major persistent chlorinated pollutants, such as polychlorinated biphenyls (PCBs) and DDT and its metabolites, brominated flame retardants (BFRs), represented by polybrominated diphenylethers (PBDEs) and hexabromocyclododecane (HBCD), together with currently widely discussed perfluorinated compounds (PFCs), mainly perfluorooctanesulfonic acid was monitored in several fish species collected from Czech rivers. Eleven sampling locations in highly industrialized areas were chosen. In addition, wild species of 14 farmed fish (grown in dedicated ponds) were also analysed. Detailed results are presented by Hradkova et al., in the publication entitled “Occurrence of Halogenated Contaminants in Fish from Selected River Localities and Ponds in the Czech Republic” as published in “Archives of Environmental Contamination and Toxicology”. Read more about this publication...

The main development focus within WP1c over the last six months were directed to pre-validation of a coulombimetric immunosensor for detection of paraquat in potato extracts and further development of this sensor to allow multiplex detection of other contaminants. The pre-validation showed that paraquat could be reliably detected down to 0.01 mg/kg (i.e. half the MRL) while the signal obtained was still easily distinguished from the blanks. By using metal nanoprobes the feasibility of multiplex detection was demonstrated. In this extended version of the assay, paraquat was detected through cadmium nanoparticles and another contaminant, DON, through lead nanoparticles, enabling simultaneous detection of multiple classes of contaminants in one
combined assay (see opposite Figure). The applicability of the assay has been demonstrated for cereals.

Earlier work on a fast detection of dithiocarbamates by ambient mass spectrometry has now been published in Food Additives and Contaminants: Tomas Cajka, Katerina Riddellova, Paul Zomer, Hans Mol & Jana Hajslova, “Direct analysis of dithiocarbamate fungicides in fruit by ambient mass spectrometry”.

Read more about this publication...

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**Developing detection methods for Veterinary Pharmaceuticals: Coccidiostats, Antibiotics**

Regarding coccidiostats (WP2a), the developed five-plex flow cytometry-based immunoassay was further optimised in order to cope with newly set maximum coccidiostats' levels for some of the targeted coccidiostats on one hand and with the known high variability of feeds. In general and except for one coccidiostat in feed, namely diclazuril, the immunoassay confirmed to be suitable for the detection of the target coccidiostats in feed and eggs. The first major outcome of the single-laboratory validation in eggs is that the method is mature enough to undergo complete validation through collaborative trial in this matrix.

As a preparation step of the upcoming collaborative trial, all WP2a partners received the first training on both the instrumentation and on the protocol. The picture on the left illustrates a three-days training event that was organised at RIKILT, Wageningen, The Netherlands under the supervision of the partner responsible for the method development. The next step will consist of performing the ring trial in the following months for both matrices and the six coccidiostats. Current output of WP2a as well as a demonstration of the protocol will be shown at the next CONFIDENCE open day to be held at RAFA 2011, Praha, Czech Republic in November 2011 [www.rafa2011.eu](http://www.rafa2011.eu). The next stakeholder event for WP2 will be organised in combination with the next EuroResidues VII upcoming conference in May 2012 (http://www.euroresidue.nl) where latest results will also be presented and discussed.

Read more on coccidiostats...

In the last 6 months, the multiplex dipstick protocol and the Readsensor method for detecting antibiotics in honey were transferred from Unisensor to FERA for the single-laboratory validation. The multiplex dipstick assay was further optimized at FERA and more than 1000 tests were produced at Unisensor for this external evaluation. Ruggedness, specificity and some stability parameters of the kit were performed by FERA. Further work is underway to establish the sensitivity of the test to an increased number of target analytes. Logistical aspects regarding the preparation/organization of the inter-laboratory study of the multiplex assay (planned in January/February 2012) were addressed.
Developing detection methods for Heavy Metals

Regarding heavy metals (WP3), the method development of a HPLC-ICPMS method for methylmercury in marine samples has been finalized and the method was in-house validated at DTU Food. The method approach includes acidic extraction in an ultrasonic bath, allowing a large number of samples to be extracted simultaneously. Detection is performed by HPLC for separation of methylmercury from inorganic mercury on-line coupled to ICPMS for mercury-specific determination. In-house validation of the method included e.g. the determination of the linearity range, precision, accuracy, the limit of detection (LOD) and limit of quantification (LOQ). The method is planned to be put into collaboratively trial later this year.

The focus in CONffIDENCE WP3 is the development of methods for the determination of metal species of highest toxicological interest, exemplified by inorganic arsenic and methylmercury. An overview of the importance of determining toxicological important metal species for improved risk assessment has been highlighted in two recent invited lectures by workpackage leader Dr. Jens J. Sloth at the 4th International IUPAC Symposium for Trace Elements in Food (TEF-4), Aberdeen, UK, 19-22 June 2011 (title: Novel developments and future trends in methods of analysis for trace element species in food and feed control) and at the 125th Annual AOAC meeting & exposition, New Orleans, Louisiana, 19-21 September 2011 (title: Speciation analysis of trace elements in food and feed - status and future developments). In both lectures emphasis was put on the need for development of methods for the specific determination of trace elements species in food and feed, e.g. inorganic arsenic and methylmercury.

The recent article by the CONffIDENCE scientists Rantala et al., entitled “Luminescent bacteria-based sensing method for methylmercury specific determination” published in Analytical and Bioanalytical chemistry describes a bacterial biosensor method for the selective determination of the bioavailable fraction of the organomercurial compound, methylmercury.

Read more about this publication...

Developing detection methods for Biotoxins: Alkaloids, Marine Biotoxins, Mycotoxins

As part of the IUPAC International Congress on Analytical Sciences 2011 (ICAS-2011), Kyoto, Japan, 22-26 May 2011, a Food Science Symposium was organized under the title "New Developments of Methods and Reliability of Analytical Results". Hans van Egmond (RIKILT) was invited to deliver a keynote presentation at this symposium, with the title “Natural Toxins; occurrence, intoxications, analysis and regulations”. Among other topics he gave an outline of the CONffIDENCE project and provided - with assistance of Noan Nivarlet, Unisensor - in particular some insight into the progress and prospects with the development of multiplex dipsticks for pyrrolizidine, tropane and ergot alkaloids. Other presentations at the Food Science Symposium dealt with analytical aspects of mycotoxins, contaminants, amino acids, cellulosic fiber biopolymer and ergosterol.

Read more about the Symposium’s programme...
After finalizing the protocol to detect ergot in cereals by imaging method using a line scan NIR imaging system combined with a conveyor belt, the last 6 months were dedicated to the transfer and validation of the methodology developed by CRA-W to an industrial laboratory, NUTRECO. For this purpose, a full commercial package, available on the market for routine analysis, including a similar NIR imaging instrument using trays as sample support and a chemometrics and image processing software (UmBio Evince) was installed in the company facilities in July. New discrimination models were built and tested on site on wheat and rye samples contaminated with ergots. Laboratory assistants were trained to the technique and first analyses were performed by themselves. The next step will consist to validate the methodology with the 2011 cereal harvest.

A publication “Online detection and quantification of ergot bodies in cereals using near infrared hyperspectral imaging” has been accepted to be published in the journal “Food Additives and Contaminants” and a poster on the subject was presented at the 15th ICNIRS international conference in May 2011.

Read more about this poster…

The 8th World Congress on Alternatives and Animal Use in the Life Sciences was held in Montreal, Canada in August 2011. This meeting provides a forum that supports both the ethical use of animals and quality science. Dr Katrina Campbell from Queen’s University, Belfast gave an oral presentation on the work performed in CONffIDENCE to develop a multiplex shellfish toxin assay which will provide a scientifically better and more ethically acceptable method to detect a wide range of important biotoxins.

Read more about this conference…

Further information of the progress in the Work Package will be presented at the RAFA meeting held in Prague, November 2011.

In the first two years of the CONffIDENCE project, multiplex dipstick immunoassays for the simultaneous determination of major *Fusarium* toxins, namely zearalenone, T-2 and HT-2 toxins, deoxynivalenol and fumonisins, in cereals, cereal foods and feed have been developed. The optimized immunoassays are able to detect the presence/absence of 6 mycotoxins at levels close to EU regulatory levels within 30 min. The kit has been recently launched on the market. During last six months a “single-laboratory validation design” has been elaborated and applied to evaluate performances of multiplex dipstick immunoassays for the determination of *Fusarium* toxins in wheat and maize. Statistical evaluation of the results (ANOVA) showed the ruggedness of the assays, since neither the matrix (i.e. wheat/maize origin) nor the day effect inflated the variation of the results. Rate of false positive of blank samples was generally lower than 10% for all toxins. When differentiating bank samples from samples at or above the EU limits the test resulted to be absolutely fit-for-purpose. Future activity will be the evaluation of assay performances through interlaboratory study involving 10 to 12 participants. On farm application of multiplex dipsticks is planned for incoming months, with the aim of demonstrating the applicability of developed methods in HACCP monitoring programmes.
Noan Nivarlet (Unisensor, Belgium) presented a poster in: International Symposium. Mycotoxins: Challenges and Perspectives, Ghent, Belgium, 24th May 2011 on Multiplex lateral flow immunoassay for Fusarium toxins in cereals. The poster described main results obtained within the CONffIDENCE project in development of multiplex dipsticks and simplified sample preparation protocols. [Read more about this poster...](#)

[Read more about this Food Mycotoxin Symposium...](#)

ISPA (National Research Council of Italy) team presented the multiplex dipstick immunoassays during the event « Made in Italy Agroalimentare » held in Bari (Italy) on September 21st-25th, 2011. An exhibit dedicated to dipstick kits developed in the CONffIDENCE project was set up within the whole exhibition focused on recent innovations on quality and safety of mediterranean agro-food productions. [Read more about Made in Italy Agroalimentare...](#)

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**News from other projects**

**How to become a QSAFFE stakeholder?**

The QSAFFE team would like to invite you to become a stakeholder for an EU Collaborative project addressing the work programme topics KBBE-2010-2-4-03: Quality and safety aspects of feed.

The project, Quality and Safety of Feeds and Food for Europe (QSAFFE) is coordinated by Professor Chris Elliott, Queen’s University, Belfast and started in March 2011.

In a nutshell, the concept of QSAFFE is to deliver better, faster and more economically viable means of ensuring the quality and safety of animal feeds in Europe.

The QSAFFE consortium is composed of academics and government scientists with substantial experience in animal feed quality & safety research along with industrial companies who are dedicated to supplying and producing safer and higher quality animal feed products. Together, their vision is an integrated approach to the reduction and management of chemical and microbiological contamination in animal feeds in Europe.

The Ultimate Goal: to provide Europe with a framework for improving the quality and safety of animal feeds entering at ports from outside the EU as well as products produced within Europe.

To get more information and if you would be interested in becoming a stakeholder in this project, please visit [http://www.qsaffe.eu/](http://www.qsaffe.eu/) and/or register via the website.
Upcoming Events

5th INTERNATIONAL SYMPOSIUM ON RECENT ADVANCES IN FOOD ANALYSIS

01-04 Nov 2011

On behalf of the Institute of Chemical Technology, Prague (ICT Prague, Czech Republic) and RIKILT - Institute of Food Safety (Wageningen, the Netherlands), we would like to invite all food researchers and representatives of national and international agencies, control bodies, governmental and commercial laboratories and industry to attend the 5th International Symposium on Recent Advances in Food Analysis (RAFA 2011) at Prague. In line with previous events, the RAFA 2011 will focus on recent advanced analytical & bioanalytical technologies and emerging food related applications. The programme includes oral presentations by young scientists, satellite workshops, interactive and vendor seminars and nice social events. A CONffIDENCE open day will be organized including lectures, posters, demonstration activities.

Read more about this symposium...

Read more about the CONffIDENCE OpenDay...

MYCORED INTERNATIONAL CONFERENCE SOUTH & CENTRAL AMERICA 2011

15-18 Nov 2011

The MycoRed South & Central America Conference 2011 will be held in Mendoza in November 2011. preliminary actions are on going to organize the Conference on "Reducing mycotoxins in the food and feed chains in Latin America in a global context". Topics: • Pre-harvest mycotoxins reduction: Plant resistance, Cultural Practices, Biocontrol approaches, Predictive models; • Mycotoxin post-harvest • Reduction of mycotoxins during processing food and feeds • Biodiversity • Novel methodology to study toxigenic fungi and mycotoxins • Bioavailability and detoxification of mycotoxins, Fungal Genome and mycotoxins, • Exposure to mycotoxins and risk analysis, Education in Mycotoxicology. Organising Committee is chaired by prof. Sophia Chulze of UNRC (email: schulze@exa.unrc.edu.ar).

Read more about this conference...
CONTAMINANTS AND RESIDUES IN FEED AND FOOD OF ANIMAL ORIGIN

21 - 22 Nov 2011

This event held in Cologne/Germany is of interest to anyone involved in process and environmental contaminants, agricultural contaminants, residues of feed additives, veterinary drugs and pesticides in feed and food of animal origin. The highlights will be: * European Community policy on contaminants in feed and foodstuffs * New analytical developments in veterinary drugs * Unavoidable carry-over of coccidiostats into non target feed and resulting residues in food * Experience with dioxins * Fusarium toxins in feed and food * Update on marine biotoxins * Experience with perfluorinated compounds * Heavy metals in feed * Update on PBDEs in food * Progress on the review of existing EU Maximum Residue Levels (MRLs)

Read more about this conference …

EURORESIDUE VII

14 - 16 May 2012

The EuroResidue Conferences are organized to cover all aspects concerning residues of veterinary drugs such as analytical techniques, pharmaco-logical and toxicological studies, registration and regulation and others. The EuroResidue VII will be held at Egmond aan Zee in The Netherlands on 14-16 May 2012. Special emphasis will be laid upon recent developments with respect to the detection and determination of drug residues in any analytical matrix. During this conference a separate session will be devoted to CONffIDENCE accomplishments in the areas of coccidiostats and antibiotics.

Read more about the conference…

6th NORDIC CONFERENCE ON PLASMA SPECTROCHEMISTRY

09 - 12 June 2012

The Analytical Section of the Norwegian Chemical Society invites scientists and users of science to this sixth Nordic Conference on Plasma Spectrochemistry. The aim of the conference is to provide a forum to encourage the exchange of ideas and knowledge about recent developments and state-of-the-art knowledge in main areas of analytical plasma spectrochemistry (ICP-OES, ICPMS and related techniques). This is a fine opportunity to share and discuss recent experiences and to be scientifically updated through a number of short courses given by international leading specialists in typical Norwegian informal surroundings.

Read more about this conference… or contact Jens J. Sloth [jjsl@food.dtu.dk] (member of the scientic committee).
The 7th Conference of The World Mycotoxin Forum® and the XIIIth IUPAC International Symposium on Mycotoxins and Phycotoxins will be jointly organized. This unique combined event, WMF meets IUPAC, will build on the success of the previous conferences which were held separately all over the world. WMF meets IUPAC will take place in Rotterdam, the Netherlands, on 5-9 November 2012. During this conference a separate session will be devoted to CONffIDENCE accomplishments in the areas of mycotoxins, marine biotoxins and alkaloids.

Read more about this conference ...

CONffIDENCE FP7 project: www.conffidence.eu
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