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Report of the 10th Biopesticides Expert Group Seminar on Bioinformatics and Regulation of Microbial Pesticides

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Report of the 10th Biopesticides Expert Group Seminar on Bioinformatics and Regulation of Microbial Pesticides

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Foreword

This report summarises the discussion and outcomes of an OECD Expert Group on BioPesticides (EGBP) seminar on bioinformatics and regulation of microbial pesticides. This one-day seminar was held on 24 June, 2019 at OECD headquarters in Paris, France, one day before the annual meeting of the EGBP, a sub-group of the OECD Working Group on Pesticides (WGP). The seminar was the tenth in a series of EGBP (formerly the BioPesticides Steering Group, BPSG) seminars that focus on bio-pesticide related issues of interest to OECD governments and other stakeholders.

The seminar was chaired by Jeroen Meeussen (European Union Minor Uses Coordination Facility), chair of the EGBP. Fifty six experts from ten OECD member countries, the European Commission, the Business and Industry Advisory Committee to the OECD (BIAC), the International Biocontrol Manufacturers Association (IBMA) and research institutes/universities participated in the Seminar. The list of participants can be found at Annex 2.

The seminar was organised to present and discuss the potential use of genome sequencing, bioinformatic tools and databases in a regulatory decision process for microbials, as well as to initiate a dialogue how this new technology can enrich the toolbox of evaluators for microbials.

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Introduction

This report presents the results and recommendations of an OECD Seminar on "Bioinformatics and regulation of microbial pesticides". Its aim is to provide an overview of the issues associated with this topic from the perspective of research, industry and regulatory experts, and to provide input to the potential future development of recommendations for possible further OECD work.

The seminar focused on promoting a dialogue on "Bioinformatics and regulation of microbial pesticides" and initiating a process to make recommendations for improvements by exchanging information on governments' or organisations' experiences and challenges with the potential use of genome sequencing, bioinformatic tools and databases for the evaluation of microbials.

Participants

People attending the OECD Seminar included:

- members of the OECD Working Group on Pesticides (WGP) and Expert Group on BioPesticides (EGBP);
- invited experts from key stakeholder groups such as the pesticide industry and manufacturers of biopesticides (BIAC and IBMA);
- invited experts from research institutes (academia), and
- regulators, risk assessors and evaluators from governmental or intergovernmental bodies.

Purpose and Scope of the Seminar

The main objectives of the EGBP Seminar were:

- to review/discuss commonly used sequencing platforms, bioinformatic search tools and databases;
- to give an overview of how bioinformatic and sequencing tools are used in research and development within companies manufacturing microbials;
- to identify how this technology might inform the regulatory process;
- to identify strengths/weaknesses of their potential use in a regulatory decision process;
- to determine key issues and challenges of this technology when applied in a regulatory context;
- to exchange information on OECD countries' current activities in this area;
- to exchange information and needs between regulators, scientists and other stakeholders;
- to suggest and discuss options of further steps for OECD countries and key stakeholders in OECD and non-OECD countries to address the identified issues;

- to make recommendations on possible use of sequencing platforms, bioinformatic search tools and databases in the evaluations of microbial biopesticides.

In particular the following issues and statements were discussed during the Seminar:

- What is the added value of bioinformatic analysis within the current regulatory approach that is based on hazard characterisation and exposure assessment?
- The applicability of these tool in the registration process of microbials.
- How to interpret results from this technology?

Structure of the Seminar

The Seminar programme is provided in Annex 1. Invited speakers included:

- International experts in this field;
- Government representatives; and
- Representatives from industry and research institutes.

Presentations were grouped into the following three sections:

- Introduction
- Stakeholders' Experience and Perspectives
- Government Experience and Perspectives

There was a short discussion after each set of presentations and a more general discussion at the end of the seminar.

Introduction to the Seminar

Presentation on the OECD and the work of OECD-EGBP and general introduction to the seminar

by the EGBP and Seminar Chair, Jeroen Meeussen, European Union Minor Uses Coordination Facility [PPT 1]

At the beginning, the Chair invited the participants of the seminar to introduce themselves. The Chair continued by giving a presentation on the history of OECD and the work of the OECD Expert Group on BioPesticides (EGBP), as well as a general introduction to the seminar. He explained that the topic "Bioinformatics and regulation of microbial pesticides" was selected because nowadays bioinformatics and Whole Genome Sequencing (WGS) play an important role in the field of microbiology and it's a topic that is regularly in the spot light. Overall, this technology is extensively used and lately it started to be used for microbial pesticides. It seems that this technology can enrich the risk assessors' toolbox but it remains to be seen how useful it is for the regulatory process.

The Chair also explained that the scope of the seminar was to discuss the applicability of this technology in the evaluation process of microbials and potentially make recommendations about how it could be included in the registration process of microbials in the future.

Overview of the commonly used sequencing platforms, bioinformatic search tools and databases

by Michel-Yves Mistou, French agency for food, environmental and occupational health and safety, ANSES, France [PPT 2]

The presenter started by introducing the various sequencing platforms and indicated that short-read technologies are well suited for high-throughput applications, as they allow rapid, cheap and accurate whole genome bacterial analyses. Through the use of short-read technologies the molecules have to be chopped off and then need to be reassembled, and this is the step that requires bioinformatics. One the other hand, the long-read sequencing technologies are used to do the comparisons and confirm the reconstruct of plasmids. However, the actual costs and high-error rates do not make long-read technologies suitable for standard microbial genomics. After that, the presenter introduced the official mission of the Laboratory for Food Safety in Anses, which is to contribute to epidemiological surveillance, strain typing and to food borne poisoning investigations, as well as to the characterisation of dangers associated to pathogenic isolates and emphasised that for all these purposes the use of WGS is needed. He then introduced the steps of analysis for phylogenetic reconstruction and the two main approaches for bacteria to establish genetic relatedness either at nucleotide level (SNP based) or at gene level (cg/wg MLST). And he concluded that whole genome multi-locus sequence typing (cg/wgMLST) is potentially more adapted to global real-time epidemiological surveillance while single nucleotide polymorphisms (SNP analysis) is better suited in situations when a strong discriminatory power is required. The presenter further talked about the different WGS analysis tools that can be open source, commercial software and genomic portals. He emphasised that these pipelines require bioinformatics resources and available infrastructure. He continued by mentioning that there have not been many benchmarking of SNP calling pipelines, as only three pipelines were tested on the same dataset. The results indicated that they can be sensitive to data quality, collection of isolates, pipelines and parameterisation, and consequently universal SNP distance cut-off values to decide about a link between isolates cannot be easily determined. For this reason, he explained that it is important to build relevant collection of isolates and not relate with the WGS analysis but always confirm with epidemiological data and he listed some genomic databases for epidemiological purposes and genome annotation and characterisation. He pointed out that for reliability purposes, the bioinformatics workflow must be "fit-for-purpose," defined by the ISO17025 standard and that some laboratories have already accredited their internal WGS analysis pipelines. He closed his talk by presenting some perspectives that could shape the future of the field, including the importance of establishing common WGS databases for harmonisation of procedures and the need for more empirical and statistical analysis (likelihood ratio) on comprehensive dataset.

Bioinformatics and sequencing tools used in research and development

by Alexander Goesmann, University of Giessen, Germany [PPT 3]

In this presentation, the speaker began by describing how the German Network for Bioinformatics Infrastructure (de.NBI) is organised and its mission that is (i) to provide comprehensive bioinformatics services to users in basic and applied life science research, (ii) bioinformatics training to users through a wide range of workshops and courses and (iii) cooperation of the German bioinformatics community with international network structures. He explained that the network hosts and provides databases as well as simple tools or workflows, so resources can be accessed by different programmes and he provided examples. He showed how users can interact with the network, by explaining that once data are generated then can be imported in special services and the results are analysed and reported before a manuscript is prepared. With respect to the training, the network offers tailor-made training courses, webinars and online training courses for omics tools and workflows, which enable researchers to transform their raw data into actual results. He then provided an introduction to ELIXIR that was established when it was recognised that the bioinformatics researchers should be better connected. The goal of this organisation is to combine in a single infrastructure all the bioinformatics resources at European level. He explained that ELIXIR coordinates and develops life science resources across Europe so that researchers can more easily find, analyse and share data, exchange expertise, and implement best practices and it's doing this by making available 18 core data resources and 145 services. It's expected that ELIXIR will play a key role in linking global data resources. He further informed the participants that de.NBI had set up a cloud computer infrastructure, which allows researchers to process and analyse their own data and that this infrastructure is now ready to open service and training activities to industry users. He closed this first part of his talk by summarising the achievements and the vision of the de.NBI network. In the second part of the presentation, he introduced the BiGi center for microbial bioinformatics and its collection of software tools. He stressed that until now they have analysed more than 4000 genomes, provided an example on antibiotic resistance genes from 1,280 *Listeria monocytogenes* strains and demonstrated that it is possible to scale up the analysis of microbial genomes. Lastly, he presented the efforts of the Fraunhofer

Institute in the University of Giessen to collect and characterise the genome of German strain microbes used for biological plant protection. He closed his talk by concluding that the availability of data is no longer a limiting factor but the analysis capacity and he stressed the importance of data management and analysis based on FAIR (findable, accessible, interoperable, and reproducible) principles.

Use of Next Generation Sequencing techniques for characterisation of baculoviruses

by J.T. Wennmann and J.A. Jehle, Julius Kühn-Institut, Germany [PPT 4]

The presenter started by describing the characteristics of baculoviruses (family Baculoviridae) that are widely applied in the biological control of insect pests. He pointed out that baculoviruses have a circular dsDNA genome that is quite large compared to other viruses and that they have a narrow host range and high virulence. They are classified in four genera and are further subdivided into two morphological groups. Today more than 200 Baculovirus isolates are now sequenced by short-read WGS techniques. The identification of baculoviruses is done traditionally by the enzymatic digestion with restriction endonucleases of their DNA and three examples were presented. The presenter then talked about the limitations of this approach that includes: a) incomplete restriction/digest, b) star activity of restriction endonucleases, c) issues with mixtures of isolates and genotypes, d) laborious and follow-up analysis, e) need for data interpretation experience, and pre-existing and circumstantial knowledge. He further provided an overview and comparison of the methods used in their lab, including short-read based WGS techniques, and he explained that the data processing approaches is based on standardised bioinformatics pipelines. He presented the sequencing parameters on isolates of the Cydia pomonella granulovirus (CpGV) that came from different geographic locations, including mixtures of those in commercially applied products. After that, the presenter introduced the Single Nucleotide Polymorphism (SNP) approach that is based on identification of SNPs positions that are specific for single isolates or phylogenetic lineages by mapping of short-reads generated by WGS techniques to a common reference genome. The entity of isolate and group specific SNP positions can then be used for isolate identification. He emphasised that the quantitative SNP analysis can be applied not only to isolates with homogenous genetic structure but also to mixtures in commercial isolates, whereas isolates with heterogeneous genetic structure show highly variable SNP frequencies. He concluded that CpGV SNP analysis is a useful tool for quality control. As regards WGS on baculoviruses, the presenter stated that this is currently the most powerful tool for isolate identification, as WGS data contain qualitative and quantitative information, allows SNP pattern interpretation in the context of natural variability and provides superior resolution compared to DNA restriction endonuclease analysis. Lastly, the presenter talked about the need for further research to: 1) learn SNP pattern interpretation in the context of natural variability, 2) develop data analyses pipeline, and 3) develop analysis criteria.

Stakeholders' Experience and Perspectives

Bioinformatics: Building the cornerstones of Sequence Homology and its use for Biologics

by Scott McClain, Syngenta Crop Protection, and presented by Greg Watson, Bayer Crop Science [PPT 5]

In this presentation, the speaker began by introducing the science of bioinformatics and explaining how it can be applied to large genome level sequence sets. He pointed out that in most cases what is important is not the sequence information that is obtained but how meaningful are the biological questions that the data address and he emphasised the need to put context to these questions. He pointed out the importance to understand biological evidence of a gene's presence in microorganisms and their potential to interact in physiological processes. Then, the presenter talked about comparative bioinformatics as tool for applying scientific methods to large-scale data that should be seen as a scientific approach for asking many new and different types of biological questions. An example of a very focussed approach is when industry develops a new product and selects those organisms that provide uniqueness and uses bioinformatics for the characterisation of the strain and to address the associated safety questions. Nevertheless, the presenter stressed that a guided homology search can inform on the safety aspects if it is based on a complete process (i.e., relevant database and homology algorithm) that has criteria. The speaker suggested the creation of a database on proteins that act in humans as toxins and from which organisms they are derived based on available clinical data. Then the clinical science can be applied as evidence to validate those sequences that will help to build a focused, defined and curated database. Lastly, the presenter explained that industry contemplates the use of bioinformatics to identify sequence homology, but the correct first step is to compare at the amino acid level, rather than the nucleotide level because functionality can only be demonstrated at phenotype. In conclusion, bioinformatics can support an overall risk assessment of microbials by being employed early and often for establishing for example taxonomy and antimicrobial resistance, as long as limitations are recognised.

How bioinformatic and sequencing data might inform the regulatory process (industry perspective)

by Andre Silvanovich, Bayer Crop Science [PPT 6]

At the beginning, the presenter provided the two main take home messages of his talk that were: (1) the current risk assessment paradigm for microbials is fit for purpose and (2) the lack of information on protein functionality currently does not allow the meaningful use of WGS for regulatory risk assessment. He indicated that commoditisation of WGS has created unrealistic expectations for its use because WGS data are meaningless without further analysis that includes: (1) assembly of short nucleotide strings, (2) identification of single nucleotide variants (SNVs) and insertion or deletion of bases (indels), (3) use nucleotide sequence to identify taxonomy, (4) gene prediction and (5) use predicted amino acid sequence to assign predicted gene function. He pointed out that little is known about large part of the genes and consequently only sequence data are not enough to inform a microbial regulatory process. The presenter stated that knowledge of protein function is important to the regulatory process, but currently there is lack of data on bacterial protein sequence and function. Nevertheless, he mentioned that submission of selected microbial sequences to regulatory authorities could add value like in the cases of taxonomic identification and investigation of potential resistance to antibiotics. He closed his presentation by stating that now WGS coupled with genome assembly and annotation does not improve the quality of a risk assessment of microbials, but in the future as the body of experimental protein knowledge expands WGS may prove to be of value for the hazard assessment.

How can Whole Genome Sequencing information be used to address data requirements for approval of microorganisms as active ingredients in plant protection products in the EU?

by Rüdiger Hauschild[1], Agata Jakubowska[2], Gianpiero Gueli Alletti[1], Jacqueline Süß[1]

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In this presentation, the speaker began by providing some background on the use of WGS for foodborne pathogens. Then he guided the audience through the current EU data requirements for microbials and indicated the potential use or not of WGS. He started by explaining that for species assignment of strains, WGP might not be necessary as there are other tools to address this data requirement. However, it may be useful for Baculoviruses as their genome is shorter compared to bacteria. As regards pathogenicity, WGS might also not be required as phylogeny through marker genes currently provides sufficient information. For distinction between Bacillus strains (*B.thuringiensis* and *B.cereus*), WGS eventually might be used to address this phenotypic question if it takes into account plasmid. However, pathogenicity usually depends on expression levels of toxin genes, and not only on the confirmation of their presence or not. He continued by explaining that WGS for establishing production of secondary metabolites of concern has limited use, as there is need to know which genes are responsible for the biosynthesis of the metabolites. Even when the metabolic pathway is established and the genes are identified, usually it is not known if the expressed protein is functional. Only when there is proof that the gene is absent WGS can be consider helpful. For antibiotic resistance, the use of WGS data could be useful if resistance genes were sufficiently characterised but currently they are not. In addition, to assess transferability of antibiotic resistance there is a need to check plasmid sequences. However, this approach can be used at the development stage of a product and WGS information can be applied as selection or exclusion criteria of a strain. Similarly, genetic stability cannot be easily addressed, but WGS always represents a molecular population of sequencing reads and variability can derive from the technology applied. Overall, WGS for the risk assessment of microbials has limited value because risk assessment is currently relying on the phenotype and the manufacturing process rather than the genotype. In conclusion, he stated that: (1) WGS can be useful for exclusion of critical issues: e.g. toxic metabolites, antibiotic resistance located on the chromosome or on plasmids, (2) WGS can be the basis for development of strain specific markers either for PCR based approaches or detection of SNPs, (3) WGS should not become a standard data requirement as the risk assessment is based on phenotypical traits and not on genome information, and (4) the genome sequences should not be part of the dossier, only reports on analyses done based on WGS.

Governments' Experience and Perspectives

EFSA WGS activities for Food Safety: Potential value of bioinformatic analysis in regulatory process including pesticides

by Jürgen Sturma and Beatriz Guerra, European Food Safety Agency, EFSA, Parma, Italy [PPT 8]

Beatriz Guerra opened the presentation by explaining in which areas of work related to food safety and public health protection EFSA is using or is planning to use new sequencing technologies (e.g., Whole Genome Sequencing, WGS, Metagenomics). She pointed out that among all the regulated products, there is an EFSA guidance and an EFSA statement that establish the need for data generated by new sequencing technologies for the risk assessment of microorganisms used in the food chain (see below). As regards the foodborne pathogens and zoonotic bacteria, she indicated that WGS was used for all the recent cases of outbreaks. WGS is also applied in EFSA, together with the European Reference Laboratory for antimicrobial resistance and the European member States to support the EU Summary reports on antimicrobial resistance monitoring of zoonotic and indicator bacteria from humans, animals and food. EFSA, together with the European Centre for Disease Prevention and Control (ECDC) have investigated needs to collect and analyse WGS data from Salmonella, Escherichia coli and Listeria monocytogenesin a joint WGS database. A scientific opinion from the BIOHAZ panel on the application and use of next generation sequencing (including WGS) for risk assessment of foodborne microorganisms is in preparation and is expected to be published soon. This opinion should support risk managers when revising current legislation. She informed the group that EFSA recently created a cloud infrastructure to store data and all sequences reviewed from about 5987 isolates and they have a bioinformatician in house. For GMOs, the audience heard that per legislation, EFSA needs to securely host the sequences of the GMO events and all associated raw data. These sequencing data are generated by the applicants using WGS or Sanger sequencing for the characterisation of the full insert and flanking regions, for the detection of all insertion sites by junction sequence analysis, and for the demonstration of genetic stability and integrity. Regarding the characterisation of microorganisms used as such in feed additives or as production strains of feed additives/food enzymes, the applicants should sequence the whole genome for bacteria and yeasts and use the sequence to (where applicable): (1) establish the taxonomic classification, (2) evaluate the presence of antimicrobial resistance genes, (3) evaluate the potential toxigenicity and pathogenicity of the strains, and (4) describe the structure of the genetic modification for strains genetically modified analysis is also recommended for filamentous fungi but alternative methods can be used to provide the above mentioned data.

After this introduction to EFSA activities in the area of WGS, Jürgen Sturma started by introducing the regulatory framework for pesticides in Europe. He pointed out that with respect to new developments that involve use of WGS data, pesticides are behind compared to the other regulated products. However, there is ongoing work in EFSA regarding the approach to be taken for the risk assessment of the regulated products and the use of new sequencing technologies. He reminded the audience about the European data requirements for microbials which are under consideration right now. Improvements are expected if WGS data was submitted including: (1) identification of the microorganism at strain level, and (2) identification of genes related to the production of secondary metabolites/toxins and to the possible antimicrobial resistance metabolites and antimicrobial resistance. In conclusion, the speaker stated that EFSA is moving ahead and is preparing to collect and deal with WGS data and WGS-based information. EFSA is recognising that different regulated products might have different needs, but in most cases similar bioinformatics tools are needed. Technology developments make it adequate to use WGS to characterise microorganisms, including biopesticides. WGS-based information already is required/recommended for other regulatory products and they are in the process of harmonising the type of data required and the level of supporting detailed information that is needs as regards the analysis and the tools.

How bioinformatic and sequencing data might inform the regulatory process (US regulatory perspective)

by Shannon Borges, Environmental Protection Agency, EPA, Washington DC, United States [PPT 9]

At the beginning, the presenter provided an introduction to the Microbial Pesticide Product Identity Data Requirements that apply for each new isolate of microbials as they are treated like new individual strains that must be registered independently. For Product Analysis, the presenter stated that there is the Guideline OCSPP 885.1100 that details the information needed to identify the active ingredient in addition to the final product and allows the use of appropriate scientific means, including new technologies. She explained why microbial identification is important for evaluators as it allows determining: (1) the properties the microorganism that may or may not have (e.g., disease potential, toxicity), (2) the pathogenicity and potential allergens to humans, and (3) the host range by using literature or scientific rational to establish the need for potential testing of certain non-target organisms. She continued by stressing out that microbial identity is highly important as it can influence the data requirements that need to be addressed and how to address these data requirements, whereas the potential relatedness to known pathogens may dictate any additional testing or screening. The presenter also mentioned that in the past the US EPA was receiving phenotypic information, however lately they receive more sequence data, including WGS. She emphasised that as sequencing ability increases, number and types of traits analysed can increase. Lastly, the presenter explained that there are challenges related to bioinformatics use in identifying microbials that are related to depth of sequencing analysis and quality of databases, the statistical analysis and the lack of information to tie back to previous types of data used. In conclusion, the speaker suggested that to move forward there is need for adequate databases and/or reference information and sufficient depth of analysis prior of starting relying only on sequence data. As regards US EPA, there will be the need for risk assessors to learn new methods, determine how to evaluate data and build capacity for reviewing these types of data. There are no plans for new data requirements, but any new approaches should fit within the guideline recommendations and be functional for identification and risk assessment.

High-throughput sequencing data of microorganisms opens new perspectives for ecotoxicological assessment of biopesticides in soils

by Thierry Heger[1*], Sofia Pereira[1], Bertrand Fournier[1] and Chantal Planchamp[2]

[1] The University of Applied Sciences Western Switzerland, CHANGINS, [2]Swiss Federal Office for the Environment (FOEN)[2] [PPT 10]

The presenter started by explaining that the biodiversity in soil is higher than the biodiversity that exists above the ground. There are different groups of microorganisms that can be found in soil and the proportion of unknown species is very high, something that makes it difficult to evaluate the potential effect of conventional or biological pesticides. He then presented the findings of a literature survey that his research group conducted on effects of microbial biopesticides on non-target soil microbial communities. The methods used concerned data from 2004-2018 and was based on 40 peer reviewed papers. From the 40 studies, 7 showed no effect, 23 small or transient effect and 10 showed strong effect of microbials on soil microbial communities. Methods that were reported included techniques like molecular fingerprinting and culture dependant methods, whereas only few studies were based on high-throughput sequencing (HTS) techniques. Overall, most studies evaluated the impact of biopesticides on only one group of microorganisms and results from different studies are usually not comparable. Then he talked about the DNA metabarcoding technique that is a DNA identification technique based on the amplification of metabarcode sequences amplified from DNA extracts. He presented the DNA metabarcoding together with the advantages and disadvantages of this HTS approach. After that, the presenter summarised the preliminary results of a cases study that involved the evaluation of the biopesticide (fungi) and pesticide (carbamate and organometal) impact on soil bacteria, fungi and protists in mesocosms. He reported that the biopesticide affected the diversity of fungi (10 days after treatments), and by increasing the dose that affected strongly the diversity of bacteria and fungi. He pointed out that this study provides data from almost the entire soil microbiome and that after the application of biopesticide a decrease in the pH was recorded that might be due to the active ingredient and/or the formulants. However, further analyses are underway to include bigger intervals after biopesticide application (samples collected at day 50 and 150). Also, it might be worth testing if the impacts of the biopesticide and the pesticide are context dependent (i.e. soil types, lab vs. field experiments, bulk soil vs. rhizosphere microbiome) and evaluating the functional changes in microbial communities. He closed his speech by suggesting that (1) high-throughput sequencing and bioinformatic analyses allow cost effective analyses of a large number of samples and (2) metabarcoding might be used for ecotoxicological assessment of biopesticides on non-target soil microorganisms. He also pointed out that the method should be adapted and protocols should be standardised (for the different steps of the analyses) prior of starting applying metabarcoding for risk assessment.

The use of Whole Genome Sequencing in risk assessment: Application to the cold smoked salmon-related listeriosis risk model

by Laurent Guillier, French agency for food, environmental and occupational health and safety, ANSES, France [PPT 11]

In this presentation, the speaker began by introducing the quantitative microbiological risk assessment (QMRA) definition for food pathogens that requires numerical estimates and certain levels of uncertainty. The same approach that applies for regular risk assessment is followed by starting with identification of microbial species and hazard, followed by an assessment of the exposure, including equivalence of concentration and consumption data to determine a dose response and the probability of illness per serving. He continued by stating that there are many sources of variability that influence QMRA and he provided an example on cold smoked salmon-related listeriosis. The exposure variability is the number of listeria cells ingested per portion that can range from 1 cell up to 1 billion cells. He emphasised that there is a strong biological variability when the contamination of the salmon occurs at the production site, as there are certain strains that are able to grow at low temperatures. The presenter also mentioned that dose response variability exists and there is an attempt to identify and include sensitivity of population groups in the analysis. The dose response variability is also influenced by the *Listeria* strains and this is where genetics play an important role. EFSA has established a baseline of sequences for *Listeria* strains and by applying genome wide association studies (GWAS), they are able to find associations between genetic variations and observable traits and establish biomarkers. He then presented two examples on establishing markers/genes of cold adaptation and markers of virulence. After that, the presenter summarised how the GWAS information combined with epidemiological data has been applied for risk assessment of listeriosis in cold-smoked salmon that could allow to inform risk management decisions. Lastly, he concluded that this example provides a proof of concept on integrating WGS data in risk assessment and that potentially the same strategy could be applied for hypervirulent strains after adaptation or refinement.

Activities in development of bioinformatics pipelines for characterisation of foodborne pathogens

by Simon Tausch, German Federal Institute for Risk Assessment, BfR, Germany [PPT 12]

At the start, the presenter explained the mission of the German Federal Institute for Risk Assessment (BfR) as regards food pathogens and that there are currently efforts to establish integrated genomic outbreak investigations in Germany. He pointed out that his team works on innovative technology that could be used for regulatory purposes and that they build in house capacity now. He presented their wet lab and their sequencing activities since 2015 that are constantly increasing. As regards the dry lab methods and capacities, he informed the audience that they use Assembly Based QUality Assessment for Microbial Isolate Sequencing (AQUAMIS) to deal with comparable data. The aim of this workflow is to have in place a fully automated basic analysis and quality control together with comprehensible metrics, containing detailed insights. After that, the presenter showed an example from an assembly report and explained how by having a workflow they are able to deal with cases of contamination of samples under testing. The presenter also referred to BACterial CHARACterization (BacCharac) pipeline that may be of interest for microbials as it provides more detailed species specific analysis. This pipeline provides details in isolates, identifies resistance genes and is handy to look at all the data that is sequenced. In conclusion, the speaker summarised that (1) there is a transition in Germany to investigate outbreaks using WGS technologies, (2) standardisation and harmonisation of data generation and metadata nomenclature is needed to ensure data comparability and (3) accreditation of data generation and analysis is necessary before WGS can replace standard methods.

Summary of Presentations and Discussions

All abstracts are presented in Annex 3. All presentation slides can be found online at: http://www.oecd.org/chemicalsafety/pesticides-biocides/seminar-on-bioinformatics-and-regulation-of-microbial-pesticides.htm.

Summary of Discussions, Ideas and Recommendations for Possible Future Work

Summary

Genome sequencing, bioinformatic tools and databases are routinely used in the research and development sector. Now that these technologies are also entering the area of microbial biopesticides they could potentially enrich the toolbox of evaluators for microbials. But, the potential use of this technology in a regulatory decision process for microbials as pesticides needs to be further investigated.

Bioinformatics combines biology, computer science, information engineering, mathematics and statistics to analyse and interpret biological data.

This Seminar provided a good opportunity to exchange information on experiences that OECD countries and relevant stakeholders have in the area of "Bioinformatics and regulation of microbial biopesticides".

Some commonly used sequencing platforms were presented. The 'German Network for Bioinformatics Infrastructure' is given as an example of a sequencing platform operating at the national level. A sequencing platform in which 22 EU countries work together at international level is called 'ELIXER'.

Currently there is a diversity of bioinformatic search tools and databases on the market. The sequencing read length depends on the instrument and chemistry used. The range of the read length of a short-read sequencing instrument is between 100 and 600 base pairs, while that of a long-read sequencing instrument varies between 10 to 15 kilobases. Short-read technologies are well suited for high throughput applications. lllumina instruments allow rapid, cheap and accurate whole genome bacterial, fungi and virus analyses. However, short-read technologies usually do not enable the assembly of a complete DNA-molecule. Long-read technologies are not suitable for standard microbial genomics due to the actual costs and high error rates. The choice between short-read *versus* long-read sequencing depends on the goal of the experiment. One is not considered universally superior to the other.

For the handling and analysis of data, different Whole Genome Sequencing (WGS) tools are available. A wide variety of Open Source Pipelines, that are complex assemblies of published open-source programmes accessible and recognized by the scientific community, are available. Examples of commercial software and genomic web portals were presented.

Due to the diversity of bioinformatics tools there is a need to establish a quality system for the WGS process. It is considered crucial that results of the WGS are reproducible.

WGS of microorganisms has become a well-established and routinely used technique in life sciences. For monitoring purposes, it is increasingly used to prevent food poisoning through microorganisms or to trace food contaminations (Cold smoked salmon model). Furthermore, WGS can be used to monitor antibiotic resistance of microorganisms in different environments.

What is the added value of bioinformatic analysis within the current regulatory approach of microbial biopesticides? WGS data as such are meaningless without additional information (e.g. on the biology of the microbial) and further analysis. What information is *nice* to know and what information do we really *need* to know for a proper risk assessment?

In the scope of regulatory affairs, WGS information can be used to address certain data requirements for the registration of microbials, especially in the area of identification, characterization and antimicrobial resistance. We also have to consider the expression level of genes. Only absence of genes can be considered conclusive, e.g. the information that a gene that codes for a toxic metabolite is absent.

Phenotypic properties resulting from the manufacturing process still need to be evaluated and cannot be replaced by WGS information. WGS should not become a standard data requirement.

Metabarcoding workflow was presented as an example of bioinformatic analysis using high throughput sequencing data to improve the ecotoxicological assessment of biopesticides on non-target soil micro-organisms. However, additional research is needed before any final conclusions can be drawn.

Conclusions/Recommendations

The following conclusions and recommendations were discussed as outcomes of the seminar:

- It is considered important to harmonize at the international level the type of genomic data that is required and the way these data should be evaluated.
- The area of microbial biopesticides should learn from what is happening in other sectors, e.g. food and feed additives.
- Training on new methods may be needed as the expertise within regulatory authorities to analyse all the available data may be limited.
- At this stage, it is not yet envisioned that the use of genome sequencing, bioinformatic tools and databases would lead to new data requirements. WGS can be used to address certain data requirements for the registration of microbials, especially in the area of identification, characterisation and antimicrobial resistance. However, further consideration should be given to the possibility of using WGS as a standard data requirement.
- Bioinformatics technology should be used to make the risk assessment more comprehensive.
- Possible case-studies on how bioinformatics can be used to facilitate the registration of microbial biopesticides should be identified.

Annex 1 – Seminar Programme

The 10th Expert Group on BioPesticides

Seminar on "Bioinformatics and regulation of microbial pesticides"

Monday 24 June 2019

OECD, Paris, France, 2 rue André Pascal, 75016 Paris

Chair: Jeroen Meeussen, EU Minor Uses Coordination Facility

9.30 – 10.00	**Introduction** • Purpose and structure of the seminar • Tour de table to introduce participants • Presentation on the OECD and the work of OECD-EGBP and general introduction to the seminar on 'Bioinformatics and regulation of microbial pesticides' *by Jeroen Meeussen* (EU Minor Uses Coordination Facility, Paris, France)
	Introduction (Cont.)
10.00 – 10.30	- Overview of the commonly used sequencing platforms, bioinformatic search tools and databases by *Michel-Yves Mistou* (French agency for food, environmental and occupational health and safety, ANSES, France)
10.30 – 11.00	- Bioinformatics and sequencing tools used in research and development by *Alexander Goesmann* (University of Giessen, Germany)
11.00 – 11.30	**Coffee break**
11.30 – 12.00	- Use of Next Generation Sequencing techniques for characterisation of baculoviruses by *J.T. Wennmann and J.A. Jehle* (Julius Kühn-Institut, Germany)
	Stakeholders' Experience and Perspectives
12.00 – 12.30	- Bioinformatics: Building the cornerstones of Sequence Homology and its use for Biologics [Strengths and weaknesses of bioinformatics and sequencing tools (within the context of how these tools might inform the regulatory process)] by *Scott McClain* (Syngenta Crop Protection) presented by *Greg Watson* (Bayer Crop Science)
12.30 – 13.30	**Lunch break**

13.30 – 14.00	- How bioinformatic and sequencing data might inform the regulatory process (industry perspective) by *Andre Silvanovich* (Bayer Crop Science)
14.00 – 14.30	- How can Whole Genome Sequencing information be used to address data requirements for approval of microorganisms as active ingredients in plant protection products in the EU? by *Rüdiger Hauschild* (Applied Insect Science (APIS), Germany)
	Government Experience and Perspectives
14.30 – 15.00	- Potential value of bioinformatic analysis in regulatory process by *Jürgen Sturma and Beatriz Guerra* (European Food Safety Agency, EFSA, Parma, Italy)
15.00 – 15.30	- How bioinformatic and sequencing data might inform the regulatory process (US regulatory perspective) by *Shannon Borges* (Environmental Protection Agency, EPA, Washington DC, United States)
15.30 – 15.45	**Coffee break**
15.45 – 16.15	- High-throughput sequencing data of microorganisms opens new perspectives for ecotoxicological assessment of biopesticides in soils by *Thierry Heger* (Changins, The University of Applied Sciences Western Switzerland)
16.15 – 16.45	- The use of Whole Genome Sequencing in risk assessment: Application to the cold smoked salmon-related listeriosis risk model by *Laurent Guillier* (French agency for food, environmental and occupational health and safety, ANSES, France)
16.45 – 17.15	- Activities in development of bioinformatics pipelines for characterisation of foodborne pathogens by *Simon Tausch* (German Federal Institute for Risk Assessment, BfR, Germany)
17.15 – 17.30	Summary of the Discussion, Ideas for Follow-up, Recommendations for possible further OECD work (with reference to the seminar outline)
17.30	End of the seminar

Annex 2 – List of Participants

Participants list for the 10th Expert Group on BioPesticides Seminar on Bioinformatics and regulation of microbial pesticides

Allemagne/Germany

Dr. Ann-Kristin DIEDERICH
Federal Office of Consumer Protection and Food Safety (BVL)

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Bioinformatics and Systems Billogy, Justus-Liebig-University Gießen

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Section IV 1.3 - Plant Protection Products
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Ms. Anne SCHAEFER
Federal Institute for Risk Assessment (BfR)

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Federal Institute for Risk Assessment (BfR)

Dr. Sabrina FEUSTEL
Federal Institute for Risk Assessment (BfR)

Australie/Australia

Mr. Alan NORDEN
Registration Management and Evaluation
Australian Pesticides and Veterinary Medicines Authority (APVMA)

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Suisse/Switzerland	Mr. Min HAHN Biotechnology Section Federal Office for the Environment FOEN
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	Mr. Domenico DESERIO DG Health and Food Safety (SANTE)
	Dr. Beatriz GUERRA European Food Safety Authority, EFSA
	Mr. Jeroen MEEUSSEN European Union Minor Uses Coordination Facility
	Mr. Juergen STURMA Pesticides / Regulated Products European Food and Safety Authority (EFSA)
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Annex 3 – Abstracts for Presentations

Presentation on the OECD and the work of OECD-EGBP and general introduction to the seminar

by the EGBP and Seminar Chair, Jeroen Meeussen, European Union Minor Uses Coordination Facility [PPT 1]

In 1961 the Organisation for Economic Co-operation and Development (OECD) was established with a trans-Atlantic and then global reach. Today the OECD has 36 member countries. More than 70 developing and transition economies are engaged in working relationships with the OECD.

OECD is a forum in which governments work together to address the economic, social and environmental challenges of interdependence and globalisation. OECD is also a provider of comparative data, analysis and forecasts to underpin multilateral co-operation.

The OECD work on agricultural pesticides (i.e. chemical and biological pesticides) aims to help member countries improve the efficiency of pesticide control, share the work of pesticide registration and re-registration, minimise non-tariff trade barriers and reduce risks to human health and the environment resulting from their use.

The Expert Group on BioPesticides (EGBP), previously called the BioPesticides Steering Group (BPSG), was established by the WGP in 1999 to help member countries harmonise the biological pesticides assessment and improve the efficiency of control procedures. Biological pesticides involve: microbials, pheromones and other semiochemicals, plant extracts (botanicals) and invertebrates as biological control agents. The first tasks of the EGBP consisted of:

(i) reviewing regulatory data requirements for three categories of biopesticides (microbials, pheromones and invertebrates); and
(ii) developing formats for dossiers and monographs for microbials, and pheromones and other semio-chemicals.

This was achieved in 2004 and resulted in several OECD-publications in the Series of Pesticides (No. 12, 2001; No. 18, 2003 and No. 21, 2004).

The EGBP then decided to concentrate its efforts on science issues that remain as barriers to harmonisation and work-sharing. This resulted in the preparation of a “working document” which does not provide 'mandatory' guidance but being essentially a set of examples/case studies aimed at helping the regulatory authorities. The document is titled: *“Working Document on the Evaluation of Microbials for Pest Control”* and has been published in OECD Series on Pesticides No. 43, 2008.

The report of the *Workshop on the Regulation of Biopesticides: Registration and Communication issues, 15 – 17 April 2008, EPA, Arlington, USA*, is another publication in the OECD Series on Pesticides (No. 44, 2009). This was followed by publications of an "*Issue Paper on Microbial Contaminant Limits for Microbial Pest Control Products"*

(OECD Series on Pesticides No. 65, 2011) and a "*Guidance to the Environmental Safety Evaluation of Microbial Biocontrol*" (OECD Series on Pesticides No. 67, 2012).

More recently the following documents were published:

- Guidance Document: Outline on Pre-Submission Consultations For Microbial Pest Control Products (OECD Series on Pesticides No.81, 2016);
- Report of a Survey on Regulatory and Testing Issues for the Sensitisation Potential of Micro-Organisms: Survey Results (2014) (OECD Series on Pesticides No. 84, 2016);
- Guidance Document on Storage Stability of Microbial Pest Control Products (OECD Series on Pesticides No. 85, 2016);
- Report of a Survey on the Need for Further Guidance on Data Requirements and Updated Test Guidelines to Support the Assessment of Microbial Pesticides (OECD Series on Pesticides No. 87, 2017);
- Guidance Document on Botanical Active Substances Used in Plant Protection Products (OECD Series on Pesticides No. 90, 2017);
- Guidance Document on Semiochemical Active Substances and Plant Protection Products
- (OECD Series on Pesticides No. 93, 2017);
- Guidance Document for the Assessment of the Equivalence of Technical Grade Active Ingredients for Identical Microbial Strains (OECD Series on Pesticides No. 96, 2018);
- Working Document on the Risk Assessment of Secondary Metabolites of Microbial Biocontrol Agents (OECD Series on Pesticides No. 98, 2018).

From 2009 onwards the EGBP started to organise seminars which focus on key issues on biopesticides of interest to OECD governments. Until now the following seminars have been held:

- Seminar on *Identity and Characterisation of micro-organisms*, OECD Series on Pesticides No. 53, 2010;
- Seminar on *The fate in the environment of microbial control agents and their effect on non-target organisms*, OECD Series on Pesticides No. 64, 2011;
- Seminar on *Characterisation and Analyses of Botanicals for the use in Plant protection Products*, OECD Series on Pesticides No. 72, 2012;
- Seminar on: *Trichoderma spp. for the use in Plant Protection Products: similarities and differences*, OECD Series on Pesticides No. 74, 2013;
- Seminar on: *Application Techniques for Microbial Pest Control Products and Semiochemicals: Use Scenarios and Associated Risks*, OECD Series on Pesticides No. 80, 2015;

- Seminar on: *Hazard and Risk Assessment of Secondary Metabolites produced by Microbial Pesticides*, OECD Series on Pesticides No. 89, 2017;
- Seminar on: *Sensitisation Potential of Micro-organisms*, OECD Series on Pesticides No. 91, 2017;
- Seminar on: *Niche Uses of Highly Specific Biocontrol Products*, OECD Series on Pesticides No. 95, 2018;
- Seminar on: *Test Methods for Micro-organisms*, OECD Series on Pesticides No. 100, 2019.

A joint OECD/KemI/EU Workshop on “Microbial Pesticides: Assessment and Management of Risks” took place between the 17th and 19th of June 2013 in Saltsjöbaden, Sweden. The workshop aimed at addressing issues around both agricultural and non-agricultural microbial pesticides and their assessment from a scientific, technical and regulatory perspective. The report of this workshop is published in the OECD Series on Pesticides No 76, 2014.

Overview of the commonly used sequencing platforms, bioinformatic search tools and databases

by Michel-Yves Mistou (French agency for food, environmental and occupational health and safety, ANSES, France [PPT 2]

{abstract not available}

Bioinformatics and sequencing tools used in research and development

by Alexander Goesmann, University of Giessen, Germany [PPT 3]

The 'German Network for Bioinformatics Infrastructure – de.NBI' is a national infrastructure supported by the Federal Ministry of Education and Research (BMBF) providing comprehensive, high-quality bioinformatics services to users in life sciences research and biomedicine. The partners organize training events, courses and summer schools on tools, standards and compute services provided by de.NBI to assist researchers to more effectively exploit their data. In addition, de.NBI coordinates the cooperation of the German bioinformatics community with international bioinformatics network structures such as ELIXIR.

Within this talk, I will introduce the de.NBI network and provide an overview on recent activities of the eight de.NBI service centers. As an example, I will illustrate the BiGi Bioinformatics Resource Center that combines bioinformatics expertise and facilities at Bielefeld and Giessen Universities as required in the field of microbial genome research. Furthermore, I will present the latest developments of the de.NBI Cloud, that offers large-scale storage and computing resources free of charge for academic users. Finally, I will demonstrate how various bioinformatics software applications can be used for microbial genome analysis to support the development of biopesticides.

More details about de.NBI are available on: www.denbi.de

Homepage: www.computational.bio

Use of Next Generation Sequencing techniques for characterisation of baculoviruses

by J.T. Wennmann and J.A. Jehle, Julius Kühn-Institut, Germany [PPT 4]

Baculoviruses (family *Baculoviridae*) are infectious to larval stages of Lepidopteran, Hymenopteran and Dipteran insects, many of which are pests in agricultural, horticultural and fruit production. Based on their high specificity and high virulence to target insects, many baculoviruses are registered and used as biocontrol agents of pest insects.

To discriminate baculovirus isolates and to identify baculovirus genotype mixtures, several molecular tools are available and applied. In the past, the most common analysis applied was based on so-called DNA restriction endonucleases (REN), enzymes that cut the double stranded DNA (dsDNA) genomes of baculoviruses and create a specific pattern of genome fragments that can be visualized by gel electrophoresis. More recently, whole genome sequencing (WGS) techniques became available to decipher the entire genomes of baculoviruses, which provides a high resolution of baculovirus genomes. In addition, WGS techniques allow the detection of genomic point mutations, so-called single nucleotide polymorphisms (SNPs), which can be used to detect specifically certain isolates and genotypes. Based on the genomic positions of SNP and the high redundancy of WGS techniques applied on baculovirus isolates their genetic compositions can be calculated, offering a profound method for analysing the composition of baculovirus isolates.

In this presentation, a brief comparison between WGS-based SNP analysis and the REN-based approach for the characterization of baculovirus isolates is demonstrated on the basis of the Cydia pomonella granulovirus (CpGV), a baculovirus of economic importance for the control of codling moth larvae in orchards. It is shown, how the SNP based method allows the deciphering of homogenous, heterogenous as well as mixed CpGV isolates, offering a qualitative and quantitative tool for analysing the genetic composition of baculovirus isolates.

Bioinformatics: Building the cornerstones of Sequence Homology and its use for Biologics

by Scott McClain, Syngenta Crop Protection and presented by Greg Watson, Bayer Crop Science [PPT 5]

Biologics and their development as useful commercial products relies on characterising many of the genomic based components of a biologic's source organism. Bioinformatics can play a number of roles in characterising an organism's genome and potential biologics derived from that genetic repertoire. Bioinformatics has the power to sift and sort through large data sets that come from an organism's gene and proteins, and importantly, compare those data to other annotated data sets. Those other data sets, databases, can provide much of the power in the characterisation process. With the right database, a gene that produces a biologic of interest can be characterised for its evolutionary function and structural similarity with other proteins. The bioinformatic comparative process can support identifying key features such as mode of action, catalytic functional domains of a protein, etc. From there, questions regarding safety can be examined e.g., does this biologic have the potential to act as a mammalian toxin? Taken together, bioinformatics is best used when focused questions can be used to reduce large genomic/proteomic comparative data sets down to refined databases that are well annotated.

How bioinformatic and sequencing data might inform the regulatory process (industry perspective)

by Andre Silvanovich, Bayer Crop Science [PPT 6]

The development of new sequencing platforms combined with continuous improvement of existing sequencing platforms has resulted in improved quality and steadily decreasing cost. While whole genome sequence data is highly informative in many contexts, its value as a tool for hazard identification is severely limited. Once collected, additional processing is required to make sequencing data meaningful. This processing includes chromosome assembly, gene identification and annotation of protein function. While the software tools that perform processing are robust and fit for purpose, annotation of protein function, is still only a prediction whose verification requires lab-based studies. To better understand the current state of prediction and verification, information from EMBL and annotations from a protein sequence found in GenBank were examined. May 2019 publication statistics reveal the >99% of ~156 million protein sequences in EMBL are predictions with no lab-based verification of function. Moreover, even for identical protein sequences, databases are replete with contradictory, erroneous and incomplete functional annotations. The collection of sequence data and annotation with predicted function, has vastly outpaced the verification of those predictions. While it's feasible to use WGS for hazard identification in instances that are underpinned by lab work such as ARM identification or taxonomic assignment, the use of WGS/annotation as a survey to identify as of yet unidentified hazard is of no value.

How can Whole Genome Sequencing information be used to address data requirements for approval of microorganisms as active ingredients in plant protection products in the EU?

by Rüdiger Hauschild[1], Agata Jakubowska[2], Gianpiero Gueli Alletti[1], Jacqueline Süß[1]

[1]APIS Applied Insect Science GmbH, Kurze Straße 3, 21682 Stade, Germany, [2]Applied Insect Science S.L., Pza. Alfonso el Magnánimo nº3, 1º-A, 46003 Valencia, Spain. [PPT 7]

The introduction of Whole Genome Sequencing (WGS) in the beginning of the 21th century revolutionised research and industry, including the field of public health and food safety. As part of these techniques, WGS of microorganisms has become a well-established and routinely used technique in life sciences. For monitoring purposes, it is increasingly used to prevent food poisoning through microorganisms or to trace food contaminations. Furthermore, WGS can be used to monitor antibiotic resistance of microorganisms in different environments. However, such uses depend on the availability of large databases with appropriate information on a high number of strains of the same species.

In the scope of regulatory affairs, WGS information can also be used to address some data requirements for the approval of microorganisms in plant protection in the EU under

Regulation (EC) 1107/2009. However, some requirements can also be addressed using specific DNA sequence information. Other potential uses are limited to date by the absence of large databases or sufficient information on gene identity. Concerning microbial metabolites which may represent a concern for human health or the environment, WGS information is of limited use because only absence of the genes involved in the biosynthesis of such genes is informative. Phenotypic properties resulting from the manufacturing process still need to be evaluated and cannot be replaced by WGS information. A crucial point is the appropriate interpretation and presentation of WGS information in the dossier. Examples for data requirements will be presented in the talk.

EFSA WGS activities for Food Safety: Potential value of bioinformatic analysis in regulatory process including pesticides

by Jürgen Sturma and Beatriz Guerra, European Food Safety Agency, EFSA, Parma, Italy [PPT 8]

EFSA is interested in the use of the data generated by new sequencing technologies (e.g. whole genome sequencing, WGS) and other OMICs for Food Safety and Public Health Protection. These data can be used for outbreak detection and investigation; detection, characterization and surveillance of emerging pathogens; monitoring of antimicrobial resistance and also for applications of regulated products (characterization of microrganisms, detection of virulence and resistance genes, genetic modifications, etc): e.g. feed additives, food enzymes, food additives, novel foods and pesticides. In GMO applications WGS can be the method of choice by the applicant for the molecular characterisation of the GM event, the number of the inserts and demonstration of genetic stability and integrity; these data are assessed in EFSA for the quality of the methodology and datasets. Several activities dealing with WGS are currently on going in different EFSA Units. These include, among others, mandates received from the European Commission related to foodborne microorganisms, rapid outbreak assessments, guidelines and technical notes to support applicants for regulated products. Technology developments make it adequate to use WGS to characterise microorganisms, also for those used as biopesticides. EFSA is preparing to require, collect and deal with WGS data and WGS-based information, taking into account that for different needs, similar/different bioinformatic tools are needed.

How bioinformatic and sequencing data might inform the regulatory process (US regulatory perspective)

by Shannon Borges, Environmental Protection Agency, EPA, Washington DC, United States [PPT 9]

{abstract not available}

High-throughput sequencing data of microorganisms opens new perspectives for ecotoxicological assessment of biopesticides in soils

by Thierry Heger[1], Sofia Pereira[1], Bertrand Fournier[1] and Chantal Planchamp[2]*

[1] The University of Applied Sciences Western Switzerland, CHANGINS, [2]Swiss Federal Office fo the Environment (FOEN)[2] [PPT 10]

Soil microbial organisms are extremely diverse and play an important role in soil ecosystem functions. Yet, little is known about the impact of biopesticides on the major groups of soil microorganisms (bacteria, fungi and protists). This lack of information is largely due to methodological limitations of classical molecular approaches. In this talk, we will present the results of a recent study where a metabarcoding approach was used to test the impact of a fungal biopesticide on bacteria, fungi and protists in mesocosms. Our results illustrate how high-throughput sequencing and bioinformatic analyses have opened up new opportunities to improve ecotoxicological assessment of biopesticides on non-target soil microorganisms.

The use of Whole Genome Sequencing in risk assessment: Application to the cold smoked salmon-related listeriosis risk model

by Laurent Guillier, French agency for food, environmental and occupational health and safety, ANSES, France [PPT 11]

{abstract not available}

Activities in development of bioinformatics pipelines for characterisation of foodborne pathogens

by Simon Tausch, German Federal Institute for Risk Assessment, BfR, Germany [PPT 12]

Foodborne disease outbreaks cause thousands of infections per year in Germany alone, leading to severe financial damage, hospitalizations and even fatal cases. To curb these numbers, continuous surveillance of foodborne pathogens along the product chain are necessary. Furthermore, linking human outbreaks to their food, feed or animal source is vital to stop foodborne outbreaks in reasonable timeframes.

To tackle these problems, the German Federal Institute for Risk Assessment has established a joint sequencing and bioinformatics facility with a focus on WGS-based outbreak detection and surveillance. Having sequencing machines of all major platforms at hand enables the development and implementation of highly integrated, automated and sound workflows for pathogen characterization and outbreak detection. Besides these official duties, the Study Centre for Genome Sequencing and Analysis is part of various national and international research projects, either as a service facility to groups from related fields or in autonomous projects. This allows to stay at the cutting edge of the field and detect new trends in a very early stage which can then be translated into everyday regulatory use.

This presentation will show some example workflows for integrated genome assembly, quality control and characterisation from WGS data and their impact on the German surveillance systems. Besides that, a glimpse into some of our research projects with potential influence to biopesticide related research will be given.