EFSA 7th ROUNDTABLE WITH INDUSTRY ASSOCIATIONS

15 September 2020

# EFSA statement on the requirements for whole genome sequence analysis of microorganisms intentionally used in the food chain

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Trusted science for safe food

# Aim of the presentation

*Status of the document:*

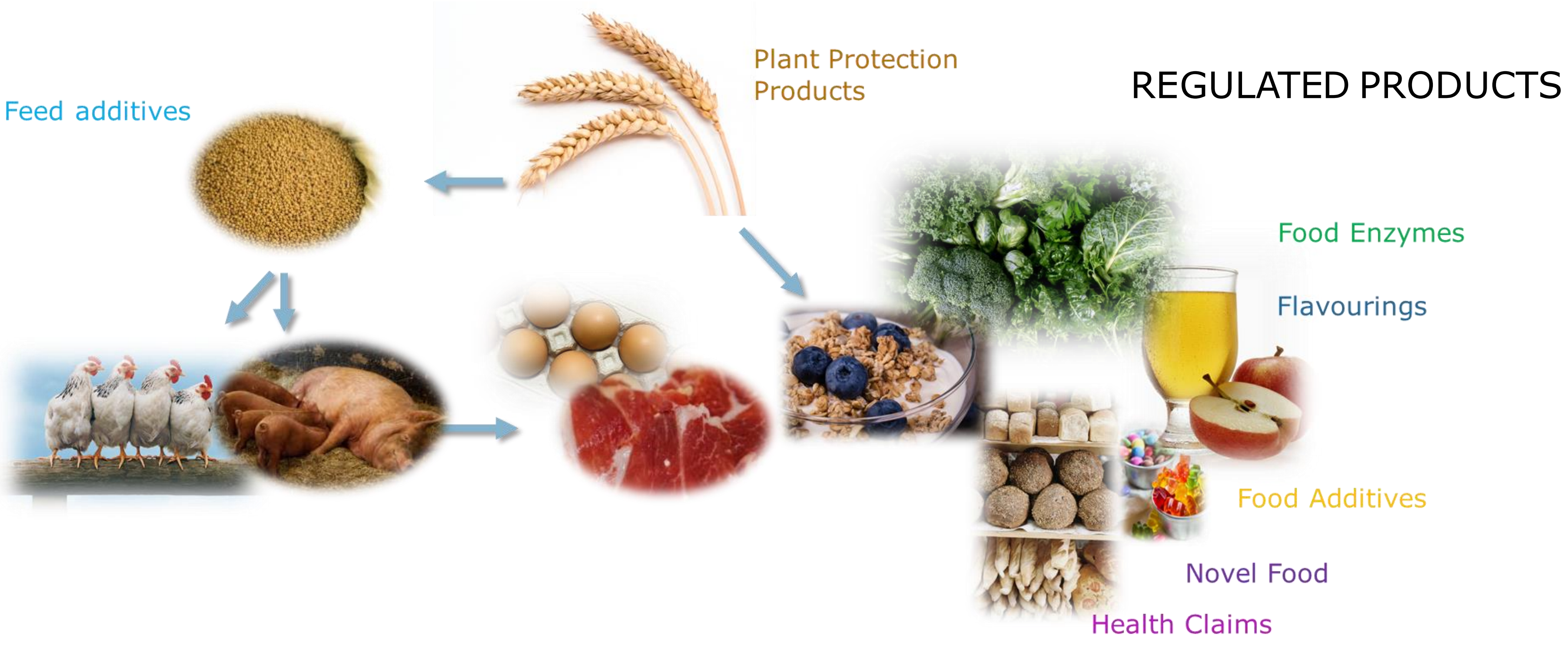
- Endorsement for public consultation by the **Scientific Committee** 4th of December 2019
- Public consultation period finished on 28th of February 2020
- On-going discussion of the comments received and finalisation of the document

Aim of the presentation:

Explain the main features of the document and provide an update on the work done after the public consultation

# Introduction

Use of **Whole Genome Sequence** based data for the **risk assessment** of **microorganisms** (used as such or as production organisms) that are **intentionally** used in the food chain



Plant Protection Products

REGULATED PRODUCTS

Feed additives

Food Enzymes

Flavourings

Food Additives

Novel Food

Health Claims

Genetically Modified Microorganisms Food/Feed

# Introduction

Risk assessment of microorganisms may require:

- Species (and strain) **identification**
- Characterisation of **genetic modification(s)** (where relevant)
- Characterisation of the strain in terms of **resistance to antimicrobials**, **virulence** and **toxigenicity**

# Introduction

Use of Whole Genome Sequence based data:

- For **FEED additives and FOOD enzymes** whole genome sequencing (WGS) and WGS-based data analysis is a requirement for the characterization of bacterial and yeast strains (recommended for filamentous fungi)
- **Guidance of the NDA** Panel offers the possibility to use the WGS to identify/characterize bacteria and yeasts.

Applicants can freely choose the methodology, some indications on what to report and how to use the data are given in the guidances.

The assessment is based on the reports provided by the applicant.

# Introduction

Stakeholders' point of view:

November 2018 – ad hoc meeting with stakeholders regarding the FEEDAP Guidance and the use of WGS based data[1]

- Powerful tool but very challenging
- Current lack of "best practices" how to appropriately use WGS data in risk assessment – more guidance is needed

**REPRO Directorate** started a work to complement the currently available Guidance and to be a reference for new guidance documents

Repro and RASA Units – Experts WG – Scientific Committee

[1] Available at: https://www.efsa.europa.eu/en/events/event/181120-0

# Scope of the Statement

To provide advice to applicants on **how to describe the analysis and results** of WGS-based characterisation of microorganisms, and, where relevant, to provide indications on **how to perform it and any quality criteria/threshold** that should be provided/reached

- Does not establish the need to generate WGS based data for the risk assessment of a given product
- Complement to the sectoral guidance documents
- Aiming at harmonisation in conduct, **reporting** – assessment

https://www.efsa.europa.eu/sites/default/files/consultation/consultation/consultation_EFSA-Statement-WGS-microorganisms.pdf

# STATEMENT - Sections

- Sequencing and related points

  3.1 Microorganism and DNA Extraction
  3.2 Sequencing and data quality control
    3.2.1. Library construction
    3.2.2. Sequencing strategy and quality control
  3.3. Assembly and annotation
  3.4. Reference-based read mapping

- Use of WGS data for the risk assessment

    3.5.1. Identification of the microorganism
    3.5.2. Identification of genes of concern

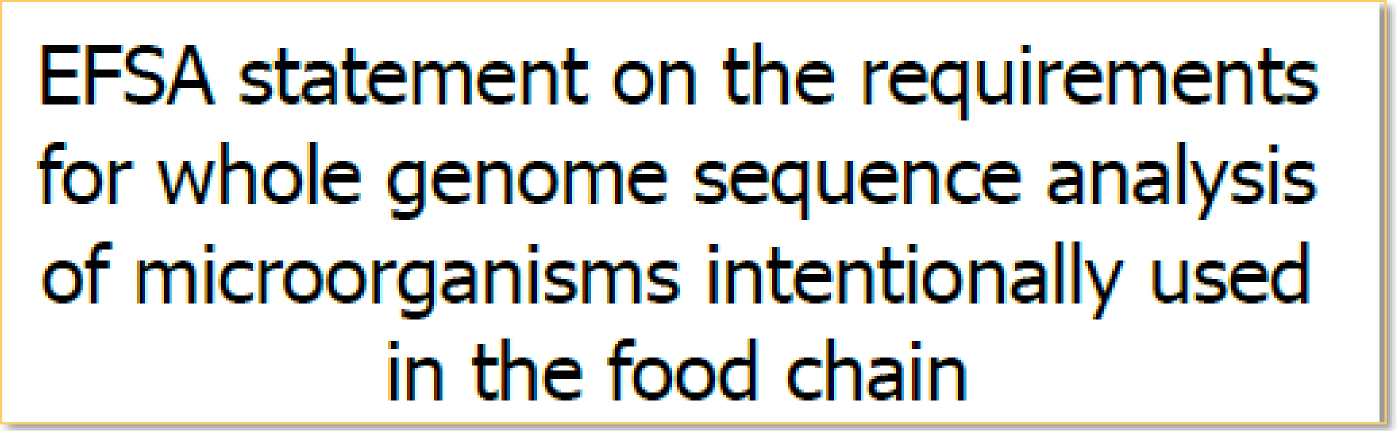
How to report how it was done

Recommendations in some steps

Quality criteria/threshold

- Other: raw data, data formats and check list

# Public Consultation



EFSA statement on the requirements for whole genome sequence analysis of microorganisms intentionally used in the food chain

- Total number of **contributors**: 37 contributions - Private companies, associations (feed/food and plant protection products), public institutions, personal capacity
- Total number of 189 entry comments (general, technical or risk assessment nature

The next slides provide a summary of some of the main comments received (it is not a comprehensive presentation) and note that actions are under discussion

# Comments - General

**C:** Is WGS based data required for any assessment of regulated products where a microorganism is used/present? When should an applicant follow the document?

The need for WGS based data should be established in the relevant Regulations/Guidance documents. Applicants can follow the document when there is the need to generate data based on WGS in any of their application dossiers

**C:** Document focused on bacteria, yeasts and filamentous fungi

Larger body of knowledge and number of applications. The same principles would apply for other taxonomical groups

To consider some modifications in the statement addressing these points

Main areas commented, presented as examples of comments received. Actions under discussion.

# Comments - General

**C:** Document focused on short-read sequencing technologies

Applicants can freely choose the sequencing technology used. Some passages of the statement could lead to this misunderstanding

The statement may need clarity in the text at this regard

**C:** The document lists the parameters to be reported on how the work was done: too many items or too few, some may not be available to the applicant when the work is commissioned

There is the need to know how the work was done, specially when the methodology may impact the quality of the data

Keep good balance between the nice to know and need to know

Main areas commented, presented as examples of comments received. Actions under discussion.

# Comments - General

**C:** The statement details the raw data and formats to be submitted to EFSA. Will these data be treated as confidential by EFSA?(especially for the microorganisms used as production strains). Comments received also on the suitability of the data formats proposed.

Sequencing reads are considered the raw data in this case and should be submitted. Confidentiality requests can be submitted in accordance with Regulation (EC) No 178/2002 and the applicable sectoral legal framework. For applications submitted after 27th March 2021 the Regulation (EU) No 2019/1381 (Transparency Regulation) becomes applicable, and confidential status may be granted by EFSA only to certain elements of applications dossiers submitted by applicants, upon their duly motivated request and consideration of a verifiable justification provided therein. This is without prejudice to the relevant provisions of the sectoral legislation as amended by the Transparency Regulation.

Data formats and some of the data requested may be modified

Main areas commented, presented as examples of comments received. Actions under discussion.

# Comments – Quality

The statement set some quality criteria and thresholds. For instance, the **sequencing** should reach a minimum average read depth of 50x, for the **assembly** the number of contigs for bacteria should be<500.

**C:** Quality parameters and thresholds, how were they established, need for some of the criteria or new criteria proposed, thresholds too high/too low, thresholds to be set at species level

Set based on <u>current</u> experience, considering the currently available technologies and the main taxonomic groups for which applications are received. Limited to the <u>most relevant ones</u> and with thresholds that may suit for most of the cases. Not possible to set thresholds at species level (e.g., N50)

To consider the proposals made regarding new parameters, modification of the already requested ones (including thresholds) in order to better consider all technologies available and current knowledge

Main areas commented, presented as examples of comments received. Actions under discussion.

# Comments – Sequences of concern

**C:** What should be reported and how to assess the data, how the thresholds were set (basis and levels), limitations of the approach (also regarding the databases), need for improvement in the wording and instructions

The WGS data should be interrogated for the presence of sequences of potential concern (e.g., AMR, toxigenic pathways) and all hits above the thresholds proposed should be reported. The thresholds were set considering the variety of microorganisms that may be assessed, experience will help in future.

The statement describes how to generate the data for the risk assessment and how to report it, the risk assessment is not in its scope. The experts will assess the data during the risk assessment.

The statement may need to improve clarity on what needs to be checked and reported, consideration of the comments done regarding the thresholds is needed.

Main areas commented, presented as examples of comments received. Actions under discussion.

# Work is on-going

**On-going** work:

- Careful evaluation of the comments received - answers
- Discuss the need to modify the document and agree on the modifications

**Expected finalisation:** Fourth Trimester of 2020.

Stay tuned

Thank you, questions?

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