AD HOC MEETING WITH INDUSTRY REPRESENTATIVES ON CELL CULTURE-DERIVED FOODS AND FOOD INGREDIENTS

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PRESENTATION OUTLINE

- Feedback from EFSA Scientific Colloquium 27
- Case Study 1 - Apple fruit cell culture biomass
- Case Study 2 - Human-identical milk oligosaccharides
- Colloquium’s input to the Update of the Novel Foods Guidance
- Support to applicants
- Q&A

FEEDBACK FROM EFSA SCIENTIFIC COLLOQUIUM 27 & NF CASE STUDIES

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EFSA's SCIENTIFIC COLLOQUIUM 27

EFSA's Scientific Colloquium 27 "Cell culture-derived foods and food ingredients"

11 May 2023, 09.00 - 12 May 2023, 12.30 (CEST)

Brussels, Belgium and online

Share:

CELL CULTURE-DERIVED FOODS

PRECISION FERMENTATION

- Sectors in the agri-food system
- State of the art of relevant concepts, technologies, and derived products
- Emerging safety and methodological aspects and their impact on EFSA's risk assessment approaches

https://www.efsa.europa.eu/en/events/efsas-scientific-colloquium-27-cell-culture-derived-foods-and-food-ingredients

BACKGROUND & SUB-THEMATIC AREAS FOR DISCUSSION

BO1& BO2

Food safety hazards associated to cell culture-derived foods of animal or plant origin

- Use of animal or plant-derived cells towards ensuring a safe and consistent product
- Bioreactors, culture media and their components
- Scaffolding structures – properties & types
- Nutritional information & the concept of nutritionally disadvantageous
- Toxicology & Allergenicity aspects

BACKGROUND & SUB-THEMATIC AREAS FOR DISCUSSION

Emerging safety and methodological aspects associated to PF (e.g., **knowledge & methodology gaps, development needs**) and their impact on **EFSA's risk assessment** approaches were discussed with relevant experts and stakeholders

BO3

New developments on engineered microbial cell factories: considerations for their safety assessment

BO4

Development needs for the safety assessment of food ingredients derived from precision fermentation (PF)

Cell culture-derived foods

STAKEHOLDERS' FEEDBACK

- **Chemical and microbiological hazards** are to be thoroughly considered for cell culture-derived foods, as they can potentially come from the **production process**.
- Use of **immortalized cell lines vs primary cells** (recurring biopsies and isolations): do not necessarily lead to a final product with the same degree of **consistency**.
- The product of a small/medium **scale production** will not necessarily be **representative** of what will be produced when scaling up the process.
- The **medium, its components and also scaffolds**, if used, but also contact materials could be **sources for microbiological and chemical contaminations and residuals**.
- The **replacement of Fetal Bovine Serum (FBS)** raised questions about **permissible substances** and the respective safety considerations.
- **Phenotypic and genetic stability of cells** : to be tested throughout the different production process steps.
- Antinutrients from plant-derived materials (e.g., culture media, components or scaffolds), may be present in CCDF

STAKEHOLDERS' FEEDBACK

- **Untargeted analyses** (-omics) of the media after harvesting the biomass could help to understand further the toxicological properties of the production process (components, materials, by-products). The implementation of such analyses is currently challenging.
- **Allergenicity** due to new proteins produced (different genes expressed), components, scaffolds.
- **Comparative approaches** of cell culture-derived foods versus their conventional counterparts **regarding residuals**, such as insulin, as well as regarding the **nutritional composition** could be a valuable tool when assessing the safety of cell culture-derived products.
- The need for **toxicological studies** may be reduced through the c**omprehensive characterization** of the composition and the production process and the **comparison of residual substance concentrations** with conventional comparators. The TTC approach could be useful for some substances present in small quantities .

APPLE CELL CULTURE-DERIVED BIOMASS - 1

Cell culture-derived biomass from a Swiss Apple variety as an ingredient for food supplements

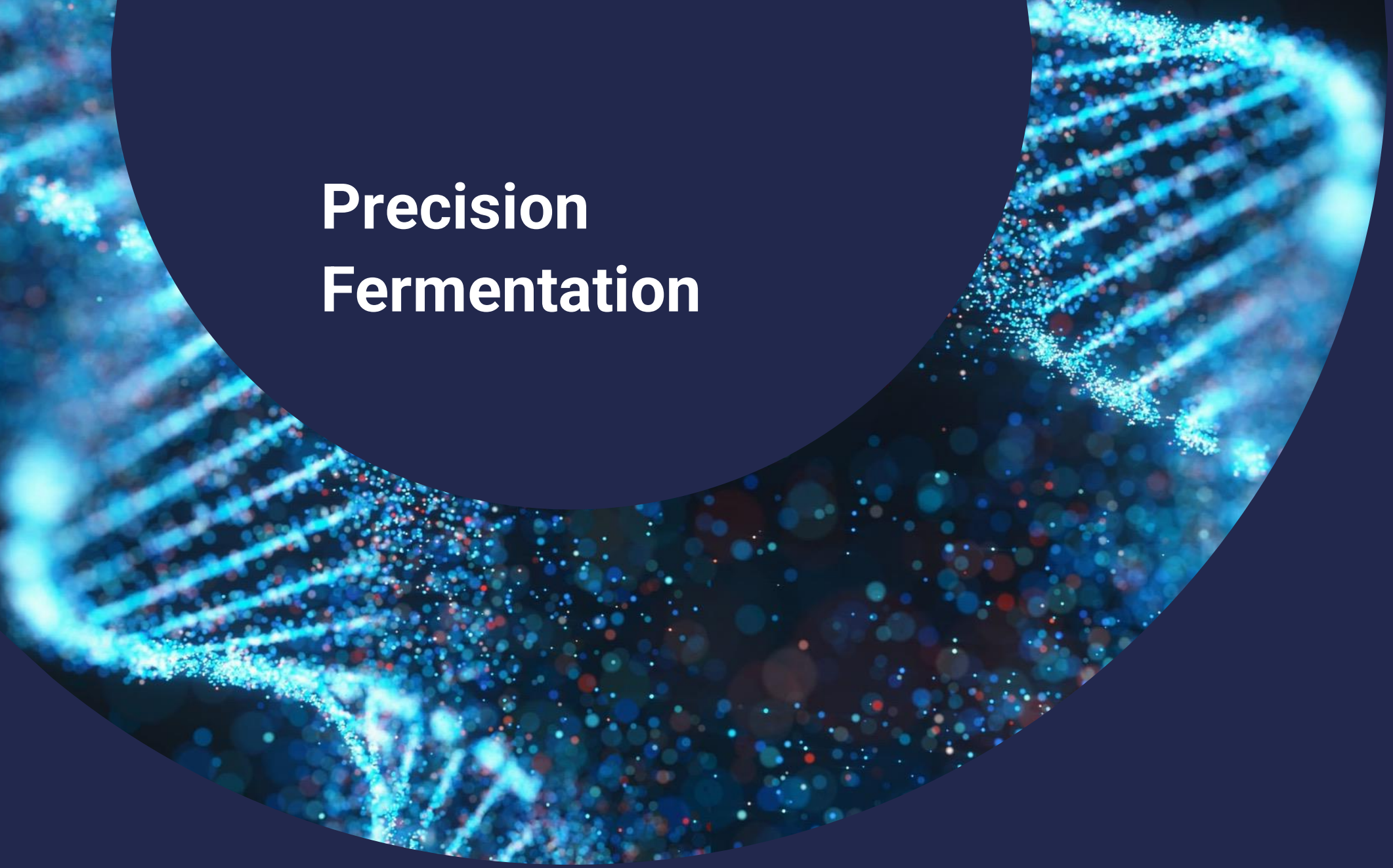
Despite the very low daily intake (0.15 mg/day) some general and CCDF lessons can be learned

- Applicant provided composition data for a **mixture with 98.5 % Isomaltulose** and also performed tox studies with that mixture. All these data were found not useful by the Panel and applicant had to perform new analyses on the biomass (without isomalt) and also perform new *in vitro* genotoxicity studies.
- Applicant performed also a **90 d subchronic tox rat study** with the mixture, but no information on the composition in the study report. Panel did not ask for a new 90 d study, because of low intake, source, detailed compositional data and comprehensive description of the production process.
- The applicant had to **redo almost the entire dossier** because they interpreted that the NF is the item mixed with isomalt.

https://www.efsa.europa.eu/en/efsajournal/pub/8065

APPLE CELL CULTURE-DERIVED BIOMASS - 2

- Panel experts want information on the **production process in all details** including CoA for each of the compounds added to the cultivation medium and also on **materials who have contact** with the cells.
- Panel experts applied **comparative assessment** with apple regarding composition and expressed proteins (proteomics analyses requested), in part because applicant's claim that the source has a history of safe use.
- Composition and expression profile of the apple cell culture biomass **is very different to apple**, nevertheless, Panel came to positive conclusions because of the detailed information on the composition and production process.
- Because of the **negligible daily intake (0.15 mg per day),** Panel did not perform a more detailed nutritional assessment.
- **Proteomics** showed hundreds of proteins which were not detected in the apple and which may be allergic.
- Dedifferentiated plant cells lose function (e.g. production of secondary plant metabolites).



Precision Fermentation

STAKEHOLDERS' FEEDBACK

- PF does not necessarily imply a **major disruption** in the current approaches for RA – **Safety-by-design** would allow to reduce safety issues in relation to the chassis
- **Phenotypic** data (next to genomic data) may still be relevant for the RA of the chassis
- **Strain stability** (genetic traits) is not relevant for the RA of GMM categories 1-3
- Sufficient knowledge about the **metabolism** of the host strain is needed when introducing new metabolic pathways to predict possible adverse effects and optimise production
- Hazards associated to GMMs are **independent of the GM technique**. Off-target effects associated to, e.g., NGTs may be discarded via **toxigenicity/ pathogenicity** testing
- **QPS** status (strain *vs.* species) could also be extended to GMMs generated by **NGTs**
- **HGT** is relevant when genes of potential concern are present (need for safety assessment of **newly introduced sequences**); Proposal for relative quantification of **recombinant DNA *vs.* total DNA**
- Cost reduction and standardisation will boost the routine use of **OMICS** (other than genomics) in RA

*RA: Risk assessment; GMMs: Genetically modified microorganisms; NGTs: New genomic techniques; QPS: Qualified presumption of safety; HGT: Horizontal gene transfer

STAKEHOLDERS' FEEDBACK

- **Comparative approach** could be followed when native counterparts exist
- **Acceptable level of identity/similarity** between native & recombinant products : consensus needed (RA & RM)
- **Post-translational modifications** (product integrity and/or protein function)
- **Harmonisation** of methodology to assess the fate in the GI tract (i.e., ADME, bioavailability)
- Classical tox studies might not be needed for the RA of macro-nutrients (e.g., proteins) – **NAMs** to be integrated in the RA
- **Allergenicity:** WoE approach – Sufficient for products similar to native substances

New-to-nature products

- Concerns for allergenicity: guidance from ICH guidelines for biotechnology products (pharmaceuticals)
- Imbalanced nutrition, e.g., by altering bioavailability

*GI: Gastrointestinal tract; ADME: Absorption, distribution, metabolism and excretion; NAMs: New approach methodologies; WoE: Weight of evidence

HUMAN-IDENTICAL MILK OLIGOSACCHARIDES (HiMOs)

Growing interest in HiMOs

Potential role in infants' health (sci. literature)

17 EFSA Scientific Opinions (PF)

Chemical and structural equivalence to HMOs

Characterisation of production process

Identification of GM production strain

Composition, contaminants, stability

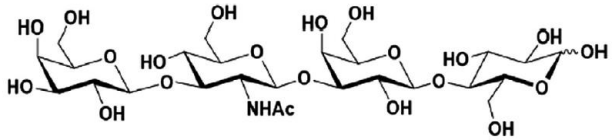
Not nutritionally disadvantageous

Comparison with HMOs intake (human milk)

Tiered toxicological approach

*HMOs: Human milk oligosaccharides

HiMOs – IDENTITY



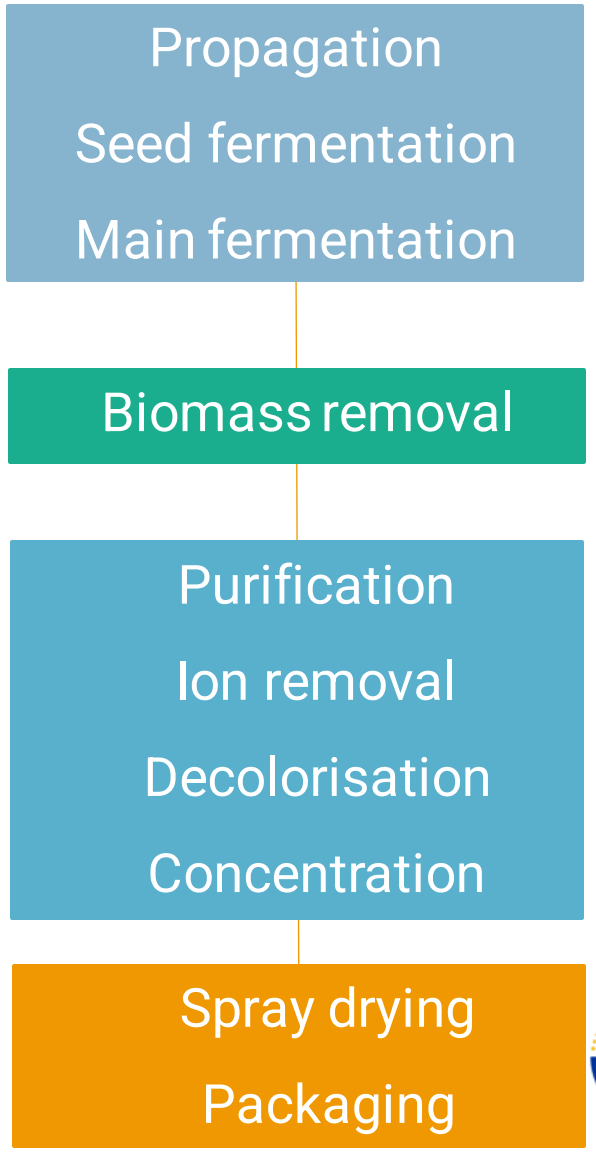
- Food with a **new or intentionally modified molecular structure** where that structure was not used as, or in, a food within the Union before 15 May 1997*
- Food consisting of, isolated from or produced from **microorganisms**, fungi or algae*
- Characterisation of the **NF source**, e.g., D-glucose as carbon & energy source, D-lactose as substrate

- **Chemical identity**: IUPAC name, CAS number, molecular weight, molecular formula, molecular structure
- **Chemical & structural equivalence** to the HMO counterpart in human milk
  - Mono- and two-dimensional NMR spectroscopy (e.g., ^{1}H, ^{13}C, NOESY)
  - Mass spectrometry (e.g., LC-MS/MS)
  - Chromatography (e.g., HPAEC-PAD)

*Regulation (EU) 2015/2283

HiMOs – PRODUCTION PROCESS

- Fermentation by **GMMs**, e.g., *Corynebacterium glutamicum, Escherichia coli*
- **Characterisation** of GM production strains
  - Taxonomic ID; International culture collection
  - Genetic modification
  - Genes of potential concern
  - Antimicrobial production
- No viable cells and recombinant DNA from production strains in the NF
- By-products, impurities, residual solvents



HiMOs – ANTICIPATED INTAKE

- Definition of a **representative (natural) concentration** of the HMO in human milk, based on scientific literature (mean of means and max mean concentrations)
- Estimation of the **highest natural daily intake of the HMO** (per kg body weight) from human milk in infants
- Comparison with the **high daily intake of the NF/HiMO** (per kg body weight) **from infant formula** in infants up to 16 weeks of age
- Comparison with the **maximum P95th daily intake of the NF/HiMO** (per kg body weight) from the **proposed conditions of use**
- Consumption of the NF that does not exceed the highest natural intake is considered as **safe**

COLLOQUIUM'S INPUT TO THE UPDATE OF THE NOVEL FOODS GUIDANCE

Ermolaos Ververis & Estefanía Noriega Fernández

Nutrition & Food Innovation Unit

EUROPEAN FOOD SAFETY AUTHORITY

NF GUIDANCE UPDATE: NEXT STEPS – INDICATIVE TIMELINES

Mandate https://open.efsa.europa.eu/question/EFSA-Q-2023-00442

NF GUIDANCE UPDATE

Disclaimer

- In the following slides, **points discussed** during the colloquium among the participants and are identified by EFSA as **potentially relevant** to the update of the EFSA Guidance on the preparation and submission of an application for authorisation of a novel food in the context of Regulation (EU) 2015/2283, are presented.

- The presentation of these points & their potential inclusion in the future NF Guidance is without prejudice to the final opinion of the NDA Panel.

Comment

- The current Guidance in place provides adequate information on the preparation and submission of applications for authorisation of novel foods and food ingredients derived from cell or tissue cultures.
- Novel food ingredients derived from precision fermentation and plant cell culture-derived foods have been already assessed by EFSA.

NF GUIDANCE UPDATE: CELL CULTURE-DERIVED FOODS

Identity

Existing points to be reinforced

"Foods consisting of, isolated from or produced from cell culture or tissue culture derived from animals, plants, fungi or algae"

- Specific requirements for **two clusters**: (animals) & (plants, fungi or algae)
- Extended requirements for the **identity of primary cells & cells from established cell lines**
- Information on **animal & herd health**
- **Cell bank**-related requirements

New points to be introduced

- Guidance on NF **nomenclature**
- Which is the NF: when **non-novel ingredients** are considered part of the NF?

NF GUIDANCE UPDATE: CELL CULTURE-DERIVED FOODS

Production Process

Existing points to be reinforced

- General provisions (e.g., requirements for input materials, food contact materials)
- Additional considerations

New points to be introduced

- Considerations regarding **specific production process steps**
- Considerations regarding **specific NF categories** (e.g., cell culture practices (GCCP)

Compositional data

Existing points to be reinforced

- **Matrix-related** analytical methods
- How to address compositional **variability**
- **Stability testing**: enhanced content, fate of the NF in the proposed-for-use matrices

New points to be introduced

- Considerations on **representative sampling**

NF GUIDANCE UPDATE: CELL CULTURE-DERIVED FOODS

Nutritional information

Existing points to be reinforced

- Further explanation of what is "**nutritionally disadvantageous**"
- Considerations about "**replacement of other foods**"
- **Comparative** approach

New points to be introduced

- Considerations about "**replacement of protein sources**"

Toxicological information & Allergenicity

Existing points to be reinforced

- Additional input on the implementation of the **tiered toxicity testing, read-across, TTC**

New points to be introduced

- Allergenicity testing requirements for **specific NF clusters**

Specifications, History of use, Proposed uses, ADME: No input from the colloquium

NF GUIDANCE UPDATE: PRECISION FERMENTATION

- **Microorganisms (MOs):** Bacteria, yeasts, filamentous fungi
- **Roles**: MOs as NF (viable/non-viable), production strains or source of food enzymes
- Only **GMM categories 1 and 2** under the remit of the NF Regulation
- **Scientific requirements** for the characterisation of MOs (EFSA FEEDAP Panel, 2018; EFSA, 2021)
  - Unambiguous **taxonomic ID (species)**; Deposition in an international culture **collection** (accession No.)
  - Purpose, characterisation & structure of **genetic modification(s)**
  - **Genes of potential concern**: Acquired AMR, toxigenicity and pathogenicity
  - **Antimicrobial** production
  - Specific **WGS** data formats to allow reanalysis during the risk assessment
  - Evidence of absence of **viable cells** of the GM production strain in the NF
  - Evidence of presence of **DNA** from the GM production strain in the NF

NF GUIDANCE UPDATE: PRECISION FERMENTATION

- **EFSA guidance on the RA of MOs intentionally added to the food chain**:
  - EFSA FEEDAP Panel (2018) + EFSA WGS Statement (2021)
  - **Under discussion:** Characterisation of microalgae/protists, viruses, GMM cat 4
  - **Under discussion**: Acquired AMR, HGT, NGTs
- EFSA statement on how to interpret the QPS qualification on "**acquired AMR genes**" (adopted on **27/09/2023**)
- Update of EFSA statement on **WGS analysis** (public consultation by **Q1 2024**; finalisation by **Q2 2024**)
- EFSA opinion on "New **developments in biotechnology** applied to microorganisms" (NGTs applied to GMM categories 3 & 4) (by **Q2 2024**)

NF GUIDANCE UPDATE: PRECISION FERMENTATION

- Qualified Presumption of Safety (**QPS**) (EFSA BIOHAZ Panel, 2023):
  - Unambiguous identification at **species** level
  - **Qualifications** to be tested at strain/product level
  - Extension to **GMMs** if QPS parental/recipient strain (species) + No safety concerns from GM
- Techniques/conditions for the **removal or inactivation** of MOs (when appropriate)
- Full reference to (GM) microbial production strains in the **specifications** (not confidential)
- **Toxic metabolic reactions** triggered by MOs under certain production conditions
- **Genotoxicity** approach (cell lysate with proof of cell lysis and supernatant) – **Under discussion**
- **Form** for applicants to provide data in a structured manner – **Under discussion**

SUPPORT TO APPLICANTS

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efsa
EUROPEAN FOOD SAFETY AUTHORITY

PRE-SUBMISSION ACTIVITIES

efsa European Food Safety Authority | connect

Register on Connect.EFSA

Notification of studies

Potential applicants must notify studies commissioned or carried out as of 27 March 2021 before the study starting date

(Art.32b General Food Law, mandatory)

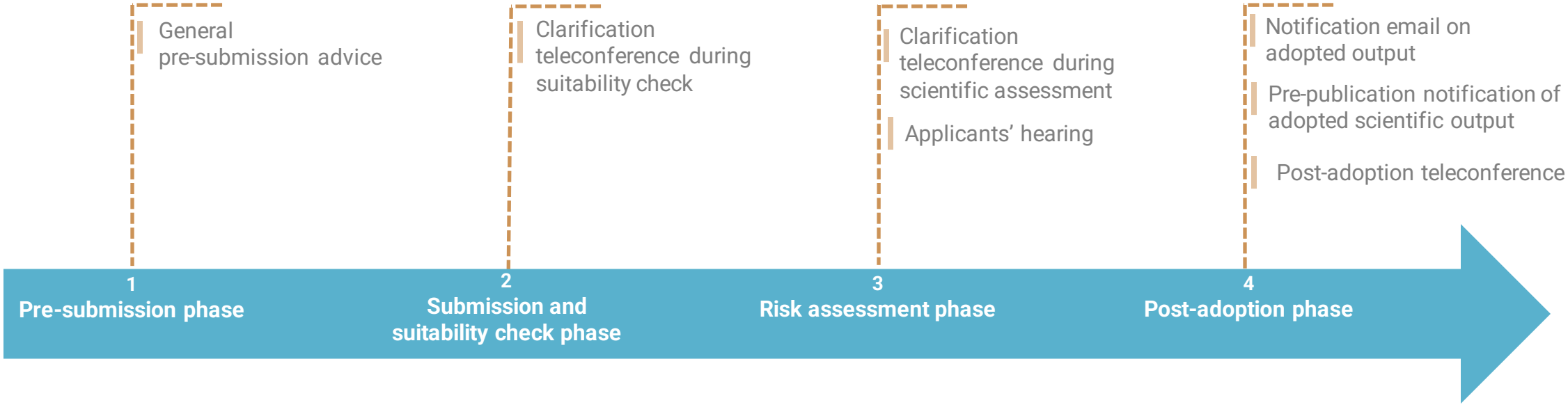
General pre-submission advice

Upon request of the potential applicant, EFSA provides advice on the rules and content for a future application

(Art.32a General Food Law, optional and recommended)

SUPPORT INITIATIVES FOR APPLICANTS

➢ EFSA's Catalogue of support initiatives



Support initiatives provided throughout the life-cycle of applications

- EFSA Info session on applications
- Roundtable with industry associations
- Ad-hoc meeting with an industry association
- Scientific workshop/conferences
- EFSA webinar
- Ask EFSA a Question & follow-up phone calls
- Dedicated support to SMEs

SUPPORT INITIATIVES FOR APPLICANTS

Join our LinkedIn group:

"EFSA support to applicants"

A space where you will find:

- Information and support materials
- Updates on the developments and progress of IT tools and platforms
- Alerts on new training material and upcoming events
- Clarifications to the most frequently asked questions received by applicants
- A space for interaction with your peers

https://www.linkedin.com/groups/9083910/

SUPPORT INITIATIVES FOR APPLICANTS

Mark your calendars for the upcoming **Webinar on novel food applications!**

26 October 2023

📢 Are you a representative of a food business operator, SMEs, research center, university developing novel food products, or their consultant?
💡 Do you want to better understand the procedure for a novel food application and support initiatives we offer? Interested in hearing about the most common issues identified during the suitability check?

Join us for this exciting event! Share your user experience and ask questions! 🙋‍♀️🙋

Don't wait and register by 📅 24 October 2023 at this link 👇 https://lnkd.in/dKVbhYmu

In case of late registration requests, please contact events@efsa.europa.eu

The recording of the webinar will be available in EFSA webpage

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