EFSA GMO PANEL RENEWAL 2024-2029

EUROPEAN FOOD SAFETY AUTHORITY

SELECTION AND APPOINTMENT TIMELINES

Call publication
1 Feb-17 Apr '23

Apr-Jul '23
Evaluation of applications

Declarations of Interest (DoI)
Jul-Dec '23

Mar '24
MB Appointments

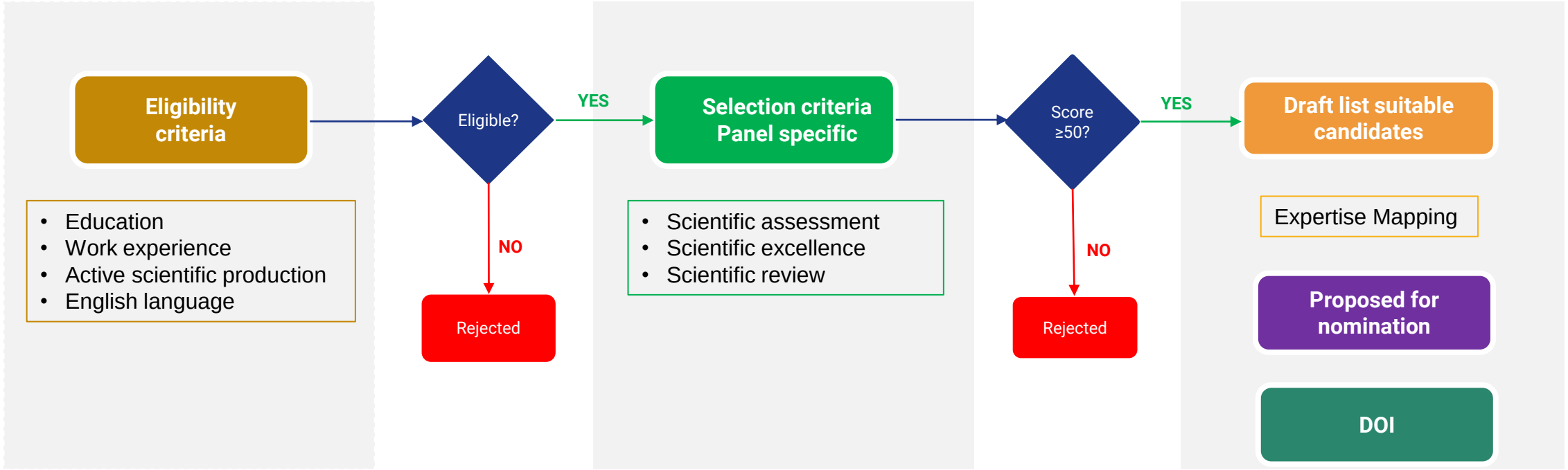
Start new mandates
1 Jul '24

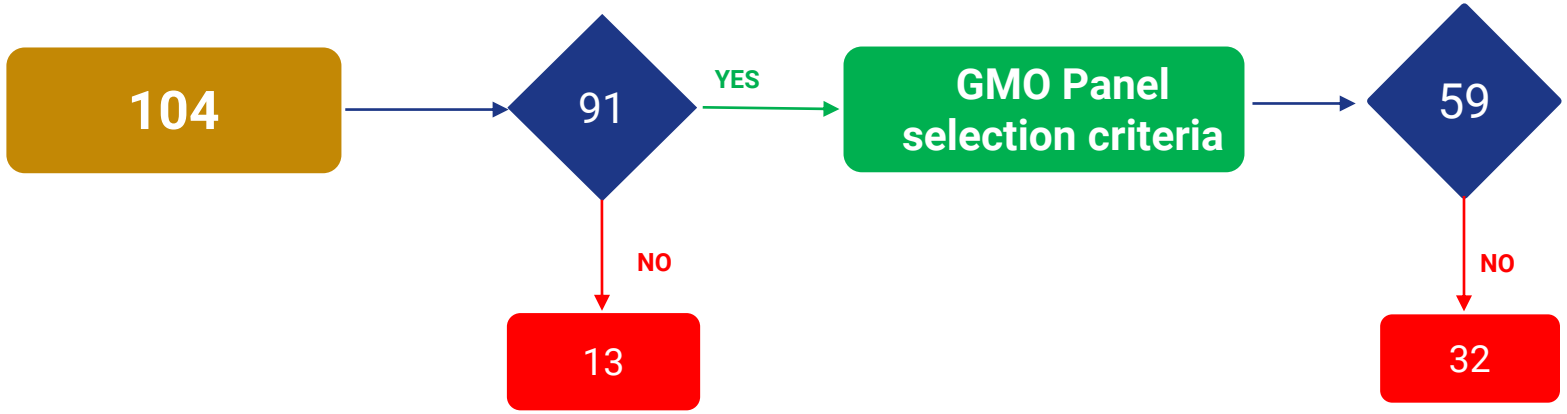




Report on the Outcome of the Call for Expressions of Interest for Membership of EFSA Scientific Panels and the Scientific Committee 2023

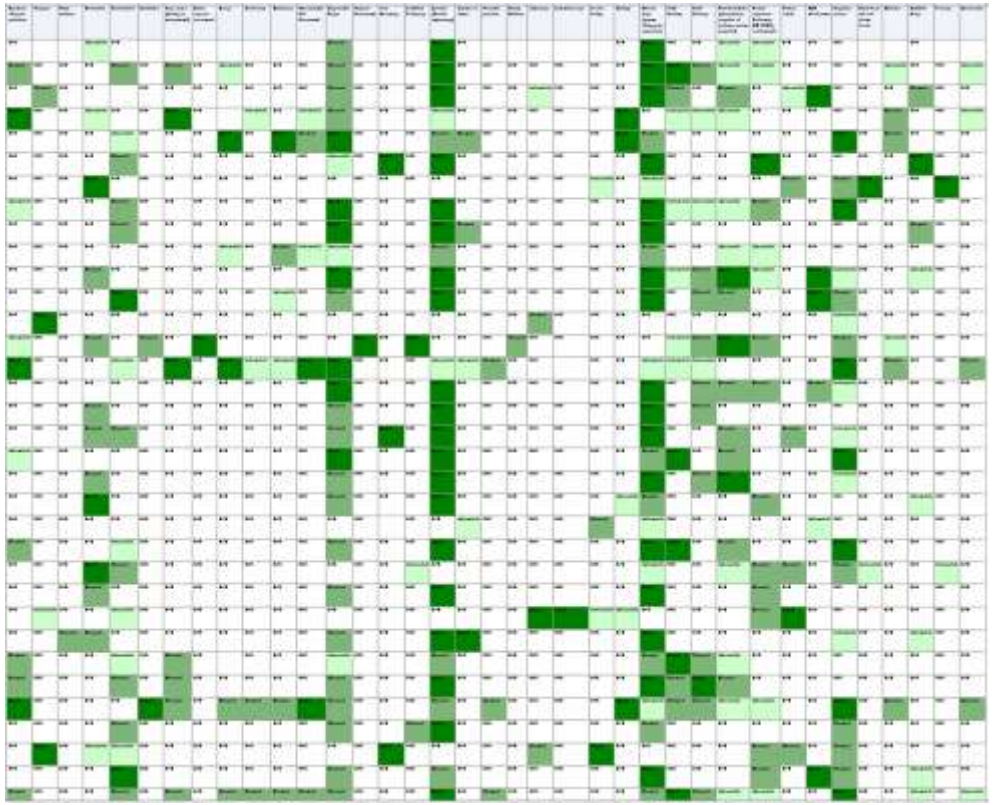
SELECTION PROCESS AND CRITERIA





SELECTION PROCESS

59



YES

18

NO

1

17

Draft list suitable candidates

Expertise Mapping

Proposed for nomination

DOI

EFSA GMO PANEL 2024-2029

Expert	Main expertise	General GMO area
Francisco Barro	Plant genetics, genome editing, plant biochemistry, plant breeding, allergology	Molecular characterisation
Josep Casacuberta	Genetic engineering, gene expression, bioinformatics, regulatory science	Molecular characterisation
Pilar Cubas	Plant genetics, genome editing, plant biochemistry, plant physiology	Molecular characterisation
Jean-Luc Gallois	Plant genetics, genome editing, RNAi, plant biochemistry, virology	Molecular characterisation
Fabien Nogué	Genetic engineering, genome editing, gene expression, regulatory science	Molecular characterisation
Alan Schulman	Plant genetics, plant breeding, OMICs, bioinformatics	Molecular characterisation
Albert Braeuning	Biochemistry, short-term and sub-chronic toxicity, toxicology	Food & feed
Michelle Epstein	Allergology, immunology, protein safety, animal testing	Food & feed
Thomas Frenzel	Dietary exposure, exposure assessment, food/feed technology, plant biochemistry	Food & feed
Frits Koning	Immunology, immunotoxicology, protein safety	Food & feed
Javier Moreno	Allergology, food microbiology, protein safety*, in vitro* testing	Food & feed
Giovanni Savoini	Animal nutrition, exposure assessment,	Food & feed
Ruud de Maagd	Genome editing, ERA, plant breeding, plant physiology, plant biochemistry	CompERA
Antoine Messéan	Agronomy and plant production, crop science, ERA, PMEM, modelling, regulatory science	CompERA
Christoph Tebbe	Ecology, microbiology, ERA, horizontal gene flow, regulatory science	CompERA
Eve Veromann	Ecology, entomology, pest control, ERA	CompERA

EFSA GMO PANEL 2024-2029

Expert
Francisco Barro
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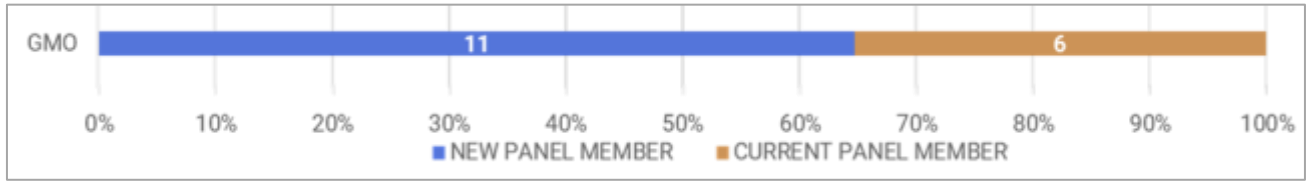
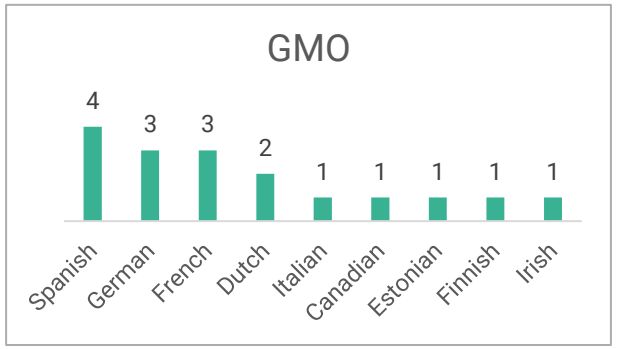
Ruud de Maagd
Antoine Messéan
Christoph Tebbe
Eve Veromann

Turnover



Geographical distribution



ONBOARDING FRAMEWORK - PHASES

Prepare

April - June 2024

Appointment to the start of mandate

- Welcome video
- Information materials
- Tutorial sessions
- Webinar

Arrive

July 2024

Inaugural Event

- General introduction (half day)
- Plenaries (1-2 days)
- Panel Chairs elections

Perform

July - December 2024

First 6 months of mandate

- Tutorial sessions
- Training
- Webinars
- SC Chair election

NEXT GMO PLENARIES

Date 2024	Type
2-3 July	Inaugural Parma
2-3 October	Online
13-14 November	OPEN Parma

NGTs applied to animals for agri food and feed uses

GMO Network

30-31 May 2024

Michele Ardizzone
Scientific officer, EFSA - NIF Unit

EUROPEAN FOOD SAFETY AUTHORITY

Topics of the presentation

1. EC Mandate on NGTs applied to animals

2. GMA-NGT WG activities

3. Focus on horizon scanning

Introduction - 40 years of genetical engineered farmed animals

1985 - transgenic pig – copies of human growth hormone genes in pronuclei of newly fertilized eggs

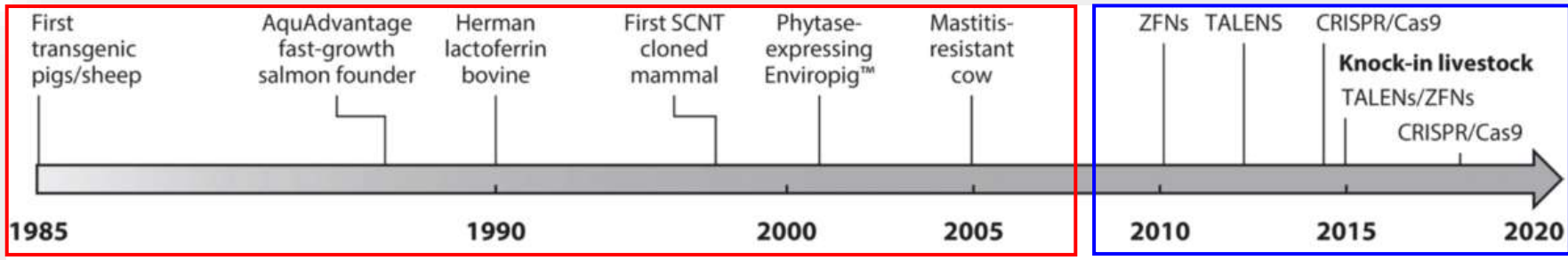
1990 - transgenic bovine – copy of human gene coding for lactoferrin in embryonic cells

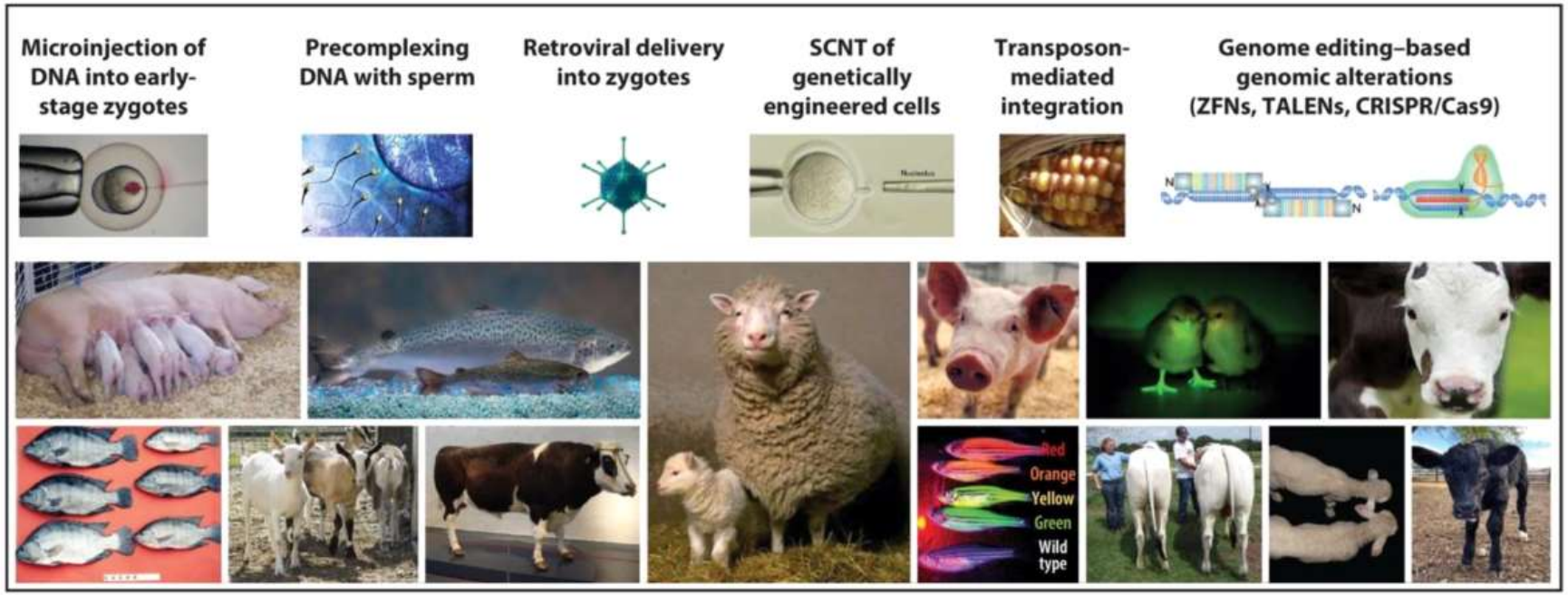
early 2000 - transgenic pig - construct with a promoter expressed in murine parotid gland and *E.coli* phytase gene in embryonic cells

2005 - transgenic cow – insertion of genetic code to express lysostaphin (natural antimicrobial protein)

transgenic random modifications

target modifications





1. the EC Mandate

EC mandate M-2018-0205 - Terms of Reference for GM animals (I)

Part I) Knowledge gathering on known cases of animals (and their food and feed products) obtained by new developments in biotechnology

1. identify animals and their products obtained by new development in biotechnology described since 2001 including their traits and uses
2. list the techniques and modifications used, including explanation of relevant terminology
3. identify animals and their products developed since 2001 that are subject to authorisation procedures by international authorities, and the corresponding available risk assessments (e.g. opinions, guidances, authorizations) that exist
4. collect per case the data and information relevant for risk assessment, and structure it according to the EFSA guidances

EC mandate M-2018-0205 - Terms of Reference for GM animals (II)

Part II) Opinion on potential novel hazards/risks from new developments in biotechnology applied to current and near market animals and adequacy of the current EFSA risk assessment guidance, covering all aspects of molecular characterisation, food feed safety & welfare, and environmental impact.

The expected outcome of this activity will be an opinion which:

1. identifies, where possible, novel potential hazards and risks which new developments in biotechnology applied to current or near market animals could pose for humans, animals and the environment compared to conventional breeding or established techniques of genetic modification.
2. determines whether the existing guidelines for risk assessment of genetically modified animals are applicable, fully or partially, adequate and sufficient to risk assess new developments in biotechnology applied to animals.
3. in case existing guidelines for risk assessment are considered not applicable, partially applicable, not adequate or not sufficient, identifies on which specific areas and aspects existing guidelines should be updated, adapted or complemented.

EC mandate M-2018-0205 – which animals are within the scope

1. NGTs applied to animals for food & feed purposes → **In the scope**

- ❑ mammals, poultry, fish
- ❑ insects ?

2. NGTs applied to animals for agricultural uses

- ❑ sterile mosquitos through non-gene drive applications (environmental control) → **In the scope**
- ❑ silkworm with improved quality of silk (quality of products)

… except gene drive application

3. NGTs applied to animals for biomedical research → **Out of the scope** … but Galsafe® pig *docet* ….

2. the GMA-NGT WG activities

EC Mandate: workflow and components

Dec 2022 → Jul 2023 → To date → end 2024/early 2025 → Jun 2025

EC mandate delivered to EFSA

Scientific opinion → Kick-off meeting of the GMA-NGT WG

Seven meetings of the GMA-NGT WG

Horizon scanning → Knowledge gathering report on NGTs in farmed animals

(Van Eenennaam, 2023) See next

Public consultation on the final draft scientific opinion

GMO Panel adoption of the scientific opinion

efsa JOURNAL

Regular updates to GMO network on the progress of mandates

Knowledge gathering from the scientific community

- TARC conference XIV Aug 2023
- Steering Committee of International Workshop on Regulatory Approaches for Agricultural Applications of Animal Biotechnologies - Global web conference Aug 2024
- Environment Agency Austria - Workshop on Risk assessment and sustainability of GM algae and GM microorganisms - June 2024
- EFFAB & FABRE TP annual meeting - Regulatory & ethical dimensions of Gene Editing - June 2024 (EFSA & EC)

Horizon scanning

Composition of the GMA-NGT working group

Comparative analysis - Thomas Frenzel
- ❑ Principle of substantial equivalence
- ❑ Criteria for the selection of the comparator(s)
- ❑ Comparative analysis of pheno-compo endpoints

Molecular characterisation - Fabien Nogué, Simon Lillico, Mike McGrew, Anna Wargelius
- ❑ EGT and NGT: general principle & applied to GM animal breeding

GM Food & Feed safety - Giovanni Savoini, Javier Moreno, Robin Ornsrud
- ❑ Toxicology general principles & applied to GM animal breeding
- ❑ Allergenicity general principles & applied to GM animal breeding
- ❑ Nutrition general principles & applied to GM animal breeding

Environmental risk assessment - Leslie Firbank, Debora Glandorf
- ❑ ERA general principles & applied to GM animal breeding

Animal welfare – Mette Herskin
- ❑ AHAW general principles & applied to GM animal breeding -

The scientific opinion – work in progress

What on the plate for discussion?

Interpretation of ToRs
Overview of new developments in biotechnology applied to animals
Definitions applicable to the scope of the mandate
Transability of familiar concepts for NGT crop & plants to NGT animals
Criteria applicable to the selection of appropriate comparator(s) for NGT animals
Methodologies for animal welfare risk assessment in at EFSA

Methodology aspects and assessment of:

- ❑ novel potential hazard and risk identification
- ❑ adequacy and sufficiency of EFSA GMO & AHAW Panel (2012)
- ❑ adequacy and sufficiency of EFSA GMO Panel (2013)

Selection of case studies to test assessment of adequacy and sufficiency of EFSA guidances

- ❑ hypoallergenic cow's milk
- ❑ hypoallergenic chicken's eggs
- ❑ increase of skeletal muscle mass and reduced body length in fish
- ❑ increased resistance to pathogens pig
- ❑ improved growth performance and resistance to pathogens pig
- ❑ hornless dairy cattle
- ❑ sterile fish
- ❑ sterile insect **??? Lack of known cases of NGTs in edible insects**

3. Horizon scanning

Knowledge gathering report - *Van Eenennaam (2023)* - methodology

External Scientific Report
APPROVED: 31 July 2023
doi: 10.2903/sp.efsa.2023.EN-8311

New Genomic Techniques (NGT) in animals and their agri/food/feed products

Alison L. Van Eenennaam

Abstract

This report presents a review of the commercial and pre-commercial stage applications of new genomic technologies (NGT) applied to farm animals and their agri/food/feed products. Additionally, a literature review was performed to compile a comprehensive listing of peer-reviewed research and development stage gene edited animals for food and agricultural applications. A total of 195 publications resulting in live animals were compiled. To date, several developed or ongoing research applications have been authorized for commerce, or judged to be "non-GMO" hence conventional, in at least one country including knockout tiger pufferfish and red sea bream in Japan; tilapia, cattle, pigs and horses in Argentina; cattle and tilapia in Brazil; and two gene-edited cattle were granted enforcement discretion in the United States meaning their products can enter the food supply. One application, the targeted *exon* deletion of a gene resulting in porcine respiratory and reproductive syndrome virus resistance in pigs is formally in the precommercial stage. There are proof-of-concept applications in multiple food species testing gene targets for traits of commercial interest. The most common trait category targeted was meat and fibre yield (31%), followed by reproduction (24%), biotic stress (18%), multiple traits (7%), colour (6%), production of hypoallergenic products (5%), product quality (4%), abiotic stress (1%), and other (4%). The majority of these were SDN-1 applications using Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR)/Cas9 to introduce small insertions and deletions to inactivate a gene. The large number of applications focused on reproduction is due in part to interest in both single-sex offspring in numerous industries (e.g. females in the case of egg production), and infertility coupled with germline complementation chimeras (where germline-competent donor cells are used to replace the germline of an otherwise sterile host of a different genetic background) in multiple species including finfish, chickens, cattle, goats, and pigs.

Key words: (new genomic techniques, CRISPR/Cas, genome editing, livestock, farmed animals)

Question number: EFSA-Q-2023-00534
Correspondence: nif@efsa.europa.eu

External Procurement awarded to prof. Alison Van Eenennaam
Animal genomics & Biotechnology, UC Davis, US

Review of applications of NGTs applied to animals involved in the production of agri/food/feed products.

Data collected from multiple sources:
- ❑ literature review (English language)
- ❑ grey literature (information publicly available online)
- ❑ survey (public and private scientists working in the field)
- ❑ consultation with experts in the field

JRC report on "Current and future market applications of new genomic techniques" used as a basis (Parisi Rodríguez-Cerezo 2021).

Knowledge gathering report - *Van Eenennaam (2023) - results*

NGT Animals

Appendix B – Table of peer-reviewed NGT animal applications

Table of peer-reviewed NGT applications broken down by trait category and targeted animal. New genomic techniques (NGTs) animals and their agri/food/feed products at the commercial stage are marked with a white background and bold font. Pre-commercial stage applications associated with a peer-reviewed publication are marked with a white background and underlined.

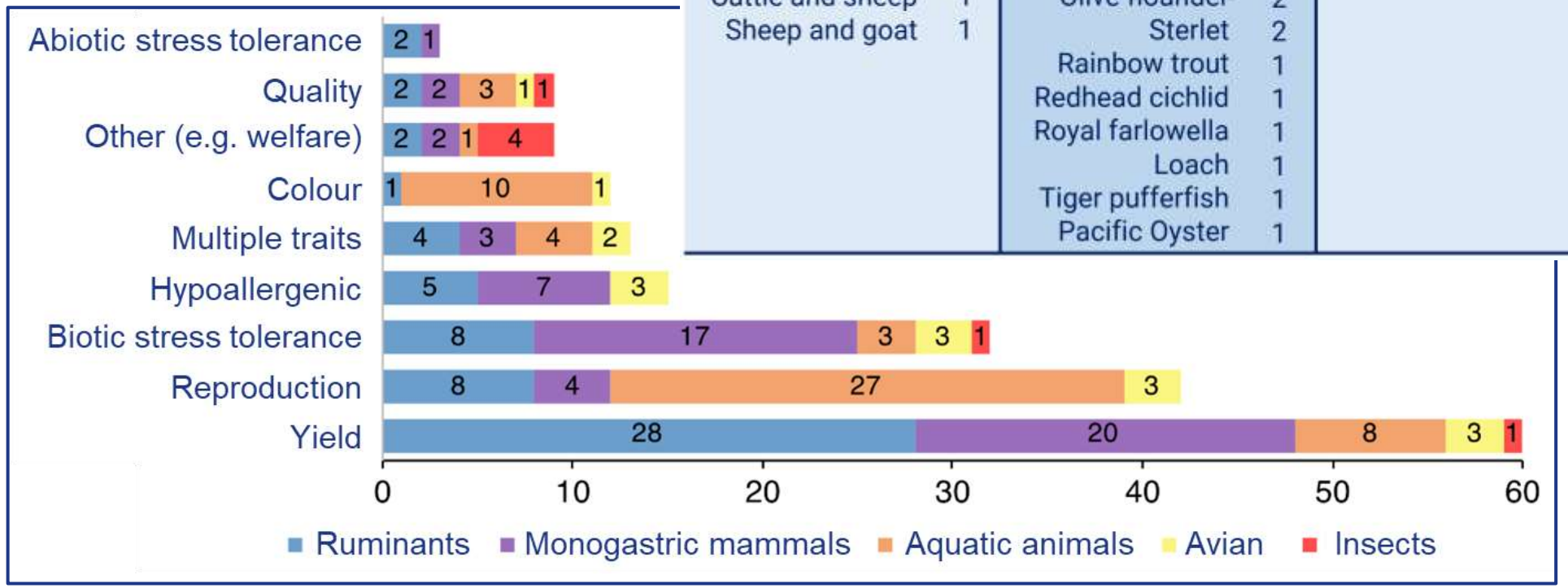
Purpose	Targeted animal	Method	SDN	CLONE	Methodology	Gene	Significance	References
Biotic Stress	Cattle	TALEN	3	YES	Knock in	Mouse SP110 (nuclear body protein)	Increased resistance to tuberculosis	(Wu et al., 2015)
Biotic Stress	Cattle	CRISPR/Cas9	3	YES	Knock in	Natural resistance-associated macrophage protein-1 (NRAMP1)	Increased resistance to tuberculosis	(Yuan et al., 2021)
Biotic Stress	Cattle	CRISPR/Cas9	3	YES	Knock in	Natural resistance-associated macrophage protein-1 (NRAMP1)	Increased resistance to tuberculosis	(Gao *et al.*, 2017b)
Biotic Stress	Cattle	ZFN	3	YES	Single amino acid substitution	CD18 (a signal peptide present on the surface of cattle leukocytes)	Increased resistance to damage from *Mannheimia haemolytica* leukotoxin	(Shanthalingam et al., 2016)
Biotic Stress	Cattle (Gir)	CRISPR/Cas9	3	YES	Amino acid substitution	BVDV binding domain of bovine CD46	Increased resistance to Bovine Viral Diarrhoea Virus	(Workman et al., 2023)
Biotic Stress	Cattle	CRISPR/Cas9	1	NO	Knock in	PRNP (prion protein)	Resistant to prion diseases	(Park et al., 2020)
Biotic Stress	Cattle (Japanese black cattle)	CRISPR/Cas9	3	YES	Substitution of single mutated nucleotide with the correct nucleotide	Isoleucyl-tRNA synthetase	Prevention of Isoleucyl-tRNA synthetase syndrome (Low birth rate, poor sucking, weakness)	(Ikeda et al., 2017)
Biotic Stress	Cattle	ZFN nickase	3	YES	Knock in	lysostaphin	Secretion in milk, against Staphylococcus aureus infection	(Liu *et al.*, 2013)

List of ~ 190 peer-reviewed papers on NGTs animals and their agri/food/feed products categorised by:

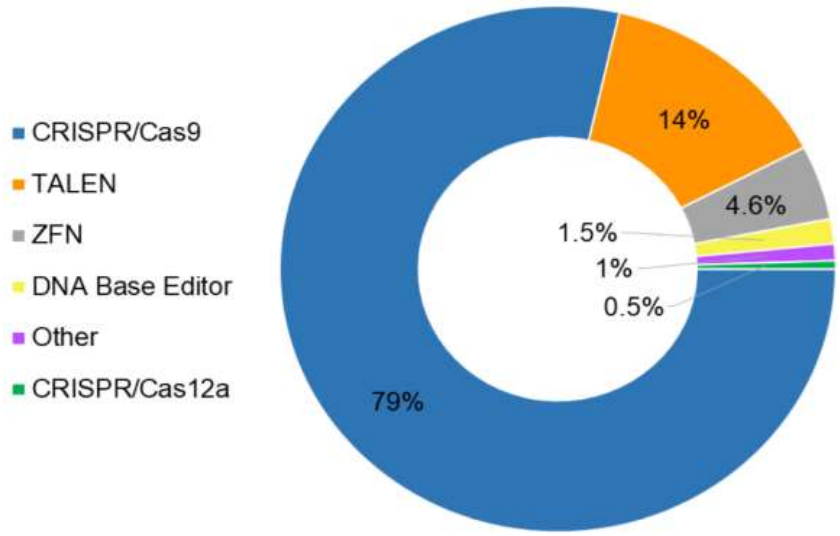
- ❑ Trait purpose category slide #7
- ❑ Animal species slide #7
- ❑ Gene editing tool & technique slide #8
- ❑ Worldwide-based stage of development (commercial, pre-commercial, R&D) slide #9

NGTs - *traits & animal species*

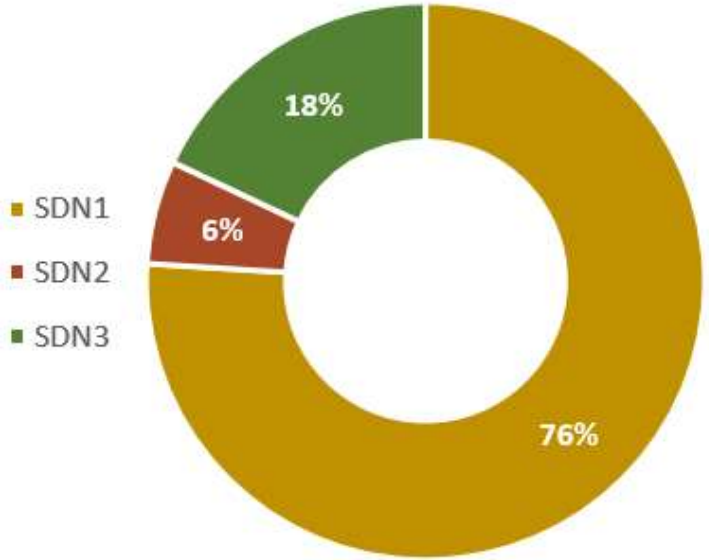
ANIMAL SPECIES							
113 mammals		**53 aquatic**		**15 poultry**		**7 insects**	
Pig	52	Tilapia	19	Chicken	12	Honeybee	4
Bovine/cattle	22	Catfish	10	Chicken and duck	1	Silkworm	3
Sheep	17	Salmon	7	Quail	2		
Goat	16	Carp	5				
Rabbit	4	sea bream	2				
Cattle and sheep	1	Olive flounder	2				
Sheep and goat	1	Sterlet	2				
		Rainbow trout	1				
		Redhead cichlid	1				
		Royal farlowella	1				
		Loach	1				
		Tiger pufferfish	1				
		Pacific Oyster	1				



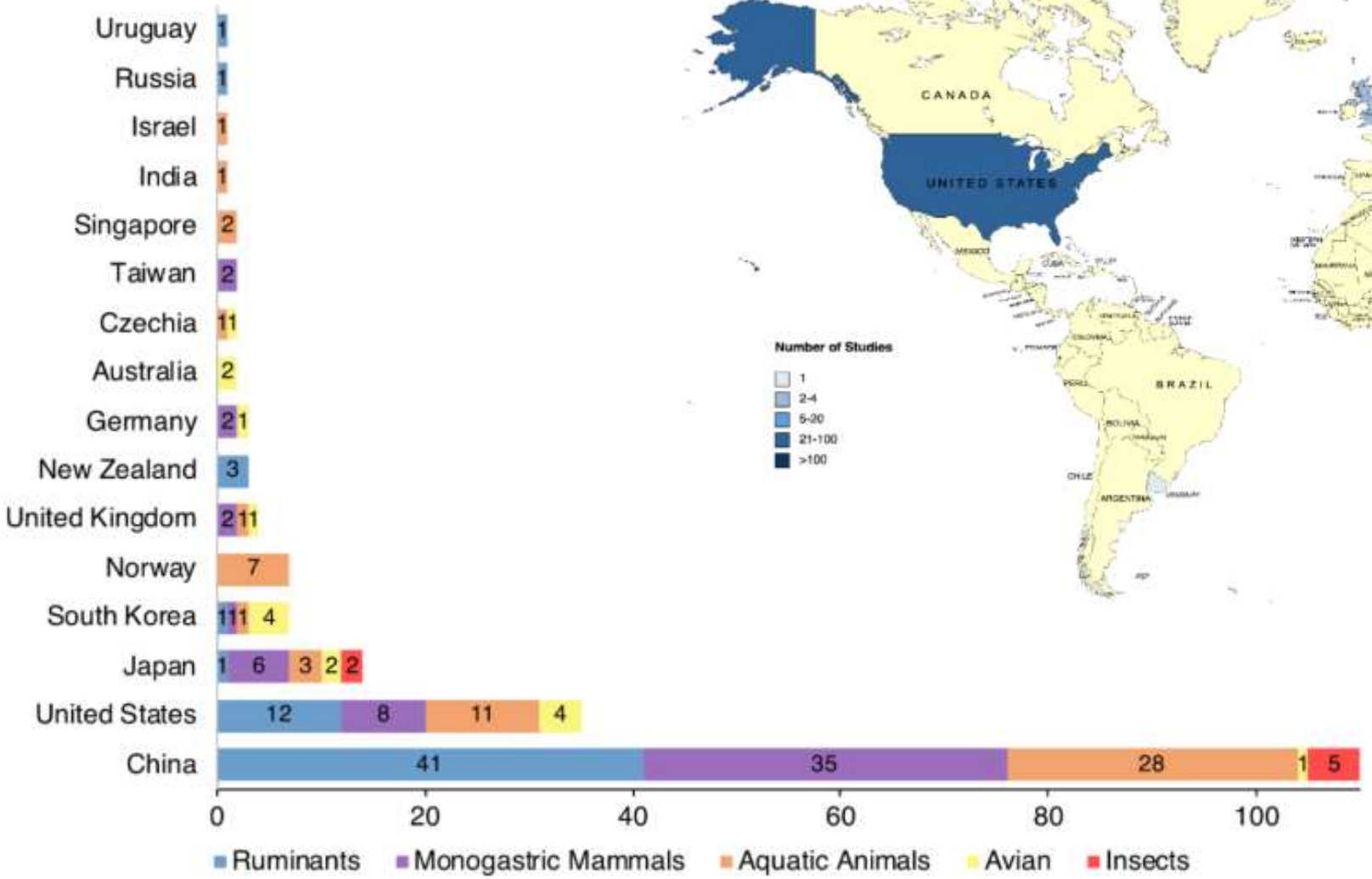
NGTs - *tools & techniques*



Tools		Technique	
CRIPSR/Cas9	~150	SDN1	~140
TALEN	~25	SDN2	~11
ZFN	~10	SDN3	~33
others			



NGTs - *geographical distribution*



Geographical distribution of first author on peer-reviewed publications on NGT animals for agri food and feed uses

Animal category breakdown by country on peer-reviewed publications on NGT animals for agri food and feed uses

NGTs – *development stage*

Commercial stage GM farm animals
- *AquAdvantage salmon* slide #18
- *GalSafe® pig*

Commercial stage NGT farm animals
- *22ndCentury Sea Bream* slide #19
- *22ndCentury Fugu*
- *"Samson" heavy muscled cattle* slide #20
- *PRLR-SLICK cattle*

Pre-commercial stage NGT farm animals
- *Holstein breed cattle* slide #21
- *Red Angus breed cattle* slide #22
- *Tilapia FLT-01*
- *Landrace, Large White, Duroc breed pig* slide #23

Research & Development stage NGT farm animals
- *over 180 papers*
- *mammals, poultry, fish, invertebrates (e.g. insects, oyster)*

Back up Slides

NGT applications in animals currently marketed in at least one country worldwide

NGT applications in animals ready to be commercialized in at least one country worldwide, but not yet on the market

NGTs applications in animals at a proof of concept stage

Commercial stage - GM fish & pig for “agri food and feed uses”

nature

First genetically engineered salmon sold in Canada

Emily Waltz *Nature* **548**, 148 (2017)

US firm AquaBounty Technologies says that its transgenic fish has hit the market after a 25-year wait.

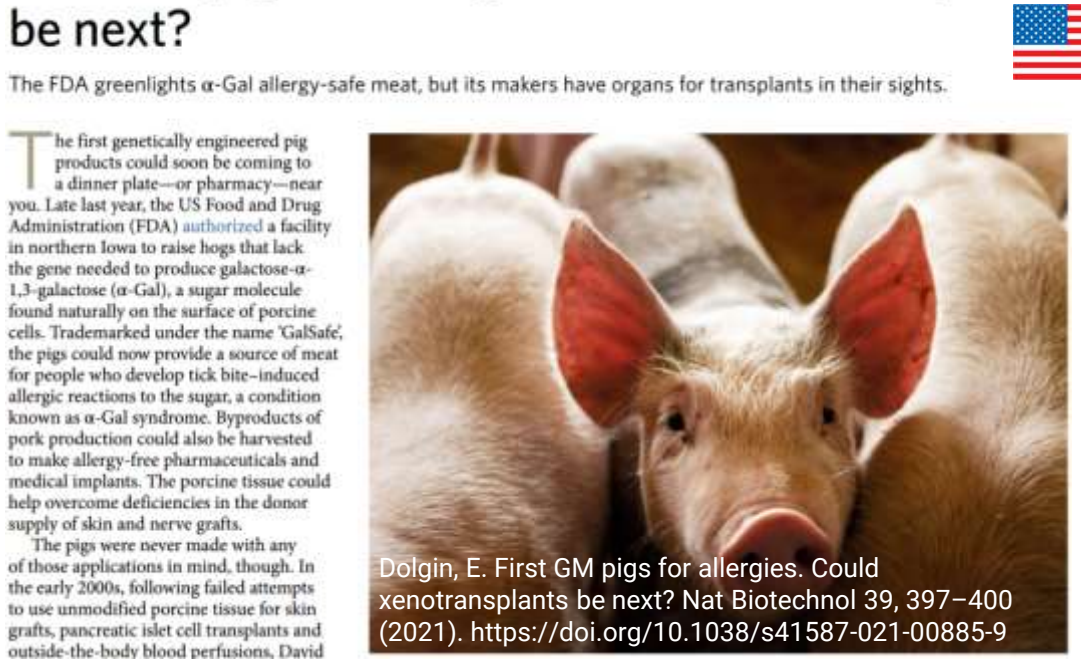
GM AquaBounty salmon: about twice the size of its wild kin (same age)

Waltz, E. First genetically engineered salmon sold in Canada. *Nature* 548, 148 (2017). https://doi.org/10.1038/nature.2017.22116.

- Common name: Atlantic Salmon (Salmo salar)
- Trade name: **AquAdvantage salmon**
- Company: AquaBounty (USA)

- Trait: Fast growth due to expression of Pacific Salmon growth hormone gene

- Approvals: USA (2015); Canada (2016); Brazil (2021)



First GM pigs for allergies. Could xenotransplants be next?

The FDA greenlights α-Gal allergy-safe meat, but its makers have organs for transplants in their sights.

The first genetically engineered pig products could soon be coming to a dinner plate—or pharmacy—near you. Late last year, the US Food and Drug Administration (FDA) authorized a facility in northern Iowa to raise hogs that lack the gene needed to produce galactose-α-1,3-galactose (α-Gal), a sugar molecule found naturally on the surface of porcine cells. Trademarked under the name 'GalSafe', the pigs could now provide a source of meat for people who develop tick bite–induced allergic reactions to the sugar, a condition known as α-Gal syndrome. Byproducts of pork production could also be harvested to make allergy-free pharmaceuticals and medical implants. The porcine tissue could help overcome deficiencies in the donor supply of skin and nerve grafts.

The pigs were never made with any of those applications in mind, though. In the early 2000s, following failed attempts to use unmodified porcine tissue for skin grafts, pancreatic islet cell transplants and outside-the-body blood perfusions, David

Dolgin, E. First GM pigs for allergies. Could xenotransplants be next? Nat Biotechnol 39, 397–400 (2021). https://doi.org/10.1038/s41587-021-00885-9

- Common name: Pig (Sus scrofa)
- Trade name: **GalSafe® pig**
- Company: Revivicor Inc. (USA)

- Trait: Knockout of glycoprotein galactosyltransferase alpha-1,3 (GGTA1) gene

- Approvals: USA (2020)

Commercial stage - NGT fish for “agri food and feed uses”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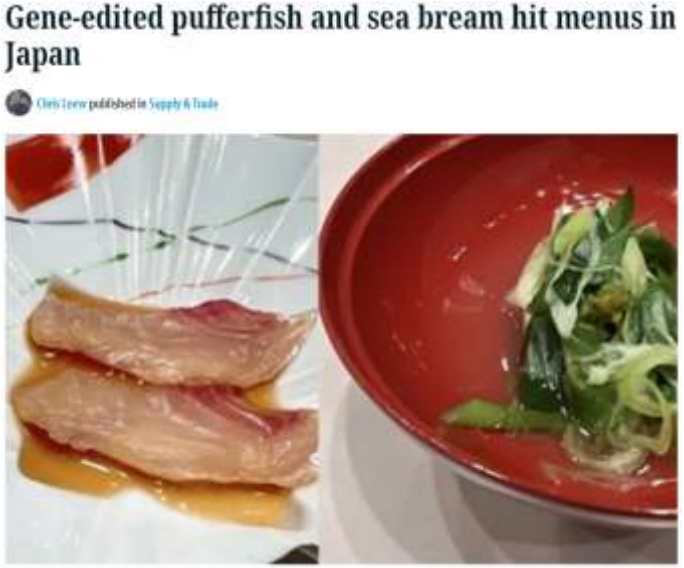
Science »

Kyoto firm puts genome-edited tiger puffer on the table

A 2-year-old genome-edited tiger puffer, top, and a conventional fish

https://japannews.yomiuri.co.jp/science-nature/science/20211101-1725/


Gene-edited pufferfish and sea bream hit menus in Japan

https://www.seafoodsource.com/news/supply-trade/japan-s-government-taking-positive-stance-on-gene-editing-fish


https://www.fishfarmingexpert.com/gene-edited-bream-japan-kindai-university/first-gene-edited-fish-goes-on-sale-in-japan/1261224

A gene-edited red bream. The fish is said to produce 20% more meat. Photo: Regional Fish Institute.

First gene-edited fish goes on sale in Japan

A Japanese start-up has notified the country's health ministry of its plan to market genome-edited fish with thicker meat, website NHK World-Japan reports. The firm began taking orders for trial sales last Friday.

- ❑ Common name: Tiger pufferfish (Takifugu rubripes)
- ❑ Trade name: **22nd Century Fugu**
- ❑ Company: Regional Fish (Japan)

- ❑ Trait: Knockout of four leptin receptor genes that control appetite, boosting their appetite and weight gain

- ❑ Approval: Japan (considered non-GMO, 2021)

- ❑ Common name: Red sea bream (Pagrus major)
- ❑ Trade name: **22nd Century Sea Bream**
- ❑ Company: Regional Fish (Japan)

- ❑ Trait: Knockout of myostatin gene: increased yield relative to conventional sea bream, and improved feed utilization efficiency

- ❑ Approvals: Japan (considered non-GMO, 2021)

Commercial stage - NGT cattle for “agri food and feed uses”

- Common name: Cattle (Bos taurus) Nelore breed
- Trade name: “Samson” heavy muscled
- Company: Acceligen (USA)

- Trait: myostatin (MSTN) gene knockout using TALENs to increase muscle fibers resulting in higher carcass yield, leaner meat, and increased beef tenderness

Approvals:

- Brazil - Parecer Técnico n. 7520/2021 (CTNBio): the animal does not possess recombinant DNA/RNA sequences and as such is not considered a GMO

- Common name: Cattle (Bos taurus) Angus breed
- Trade name: PRLR-SLICK cattle - Male (Slick04) and female (Slick03)
- Company: Acceligen (USA)

- Trait: prolactin receptor (PRLR) gene edit using CRISPR/Cas9 resulting in short hair which improves heat-tolerance trait reducing heat stress.

Approvals:

- Brazil - Parecer Técnico n. 7865/2022 (CTNBio): the animal does not possess recombinant DNA/RNA sequences and as such is not considered a GMO

- USA - Enforcement Discretion 2022 (FDA) of IGA: Low-risk for marketing of products from Male (Slick04) and female (Slick03); IGA equivalent to naturally occurring mutations as in conventional cattle with HoSU

Pre-commercial stage - NGT cattle for “agri food and feed uses”

- Common name: Cattle (Bos taurus) Holstein breed
- Trade name: Still in development
- Company: Acceligen (USA) / Kheiron S.A. (Argentina)

- Trait: Celtic allele & prolactin receptor (PRLR) gene edit using TALENs, resulting in hornless & short hair which improves heat-tolerance trait reducing heat stress

Countries approached/considered for regulatory evaluation:

- Argentina – Indicative response (CONABIA 2020): the animals to be obtained will not have inserted foreign DNA sequences, and as such a “new combination of genetic material”. For this reason they would be considered non-GMO

- Common name: Cattle (Bos taurus) Red Angus breed
- Trade name: Still in development
- Company: Acceligen (USA) / Kheiron S.A. (Argentina)

- Trait: prolactin receptor (PRLR) gene edit resulting in short hair which improves heat-tolerance trait reducing heat stress

Countries approached/considered for regulatory evaluation:

- Argentina – Indicative opinion (CONABIA 2020): the animals to be obtained will not have inserted foreign DNA sequences, and as such a “new combination of genetic material”. For this reason they would be considered non-GMO

(Pre ?) Commercial stage - NGT fish for “agri food and feed uses”

- Common name: Tilapia (Oreochromis niloticus)
- Trade name: Tilapia FLT-01 "Extra fillet"
- Company: AquaBounty (USA)

- Trait: myostatin (MSTN) gene knockout resulting in an increased muscle mass, with a greater weight and yield of the fillet, in comparison with its counterpart without editing

Countries approached/considered for regulatory evaluation :

- Argentina (Considered as non-GMO by CONABIA, 2018)

- Brazil - Parecer Técnico n. 6527/2019 (CTNBio): the animal does not possess recombinant DNA/RNA sequences and as such is not considered a GMO

(Pre ?) Commercial stage - NGT pig for “agri food and feed uses”

- Common name: Pig (Sus scrofa domesticus) Landrace/Large White/Duroc
- Trade name: Same as current commercial line names
- Company: Genus (UK)

- Trait: CD163 Exon 7 deletion to remove protein domain 5 from the expressed protein. Without this protein domain the PRRS virus cannot establish an infection

Countries approached/considered for regulatory evaluation :

- USA: although the CD163 modification could occur naturally, it has never been observed in pigs (no Enforcement Discretion from FDA); therefore, the company has to submit FDA a formal request for approval.

- Colombia: in October 2023 regulators indicated that because the edited pigs from Genus do not involve transgenics, they will treat the swine the same as conventionally bred animals.

- Brazil, Japan, Canada, Mexico, China

Other examples of NGT animals in R&D – see back-up slides

- ❑ ABIOTIC STRESS
- ❑ BIOTIC STRESS
- ❑ WELFARE
- ❑ COLOUR of coat

- ❑ REPRODUCTION STERILITY

- ❑ HYPOALLERGENIC FOOD/FEED PRODUCTS
- ❑ QUALITY OF THE FOOD/FEED PRODUCTS
- ❑ YIELD MEAT

Thank you for your attention

Any question?

BACK UP SLIDES

Details to slide 17

Adaptation to climate conditions

Animal	Characteristic	Gene of interest	Tool	Technique	Reference
Pig	Improvement of thermogenic capacity	UCP1 gene	CRISPR/Cas9	SDN3	(Zheng et al., 2017)
Cattle	Diluted coat color as a potential adaptation to climate change	PMEL gene (pre-melanosomal protein 17 gene)	CRISPR/Cas9	SDN1	(Laible et al., 2021)
Cattle	Slick hair coat for improved thermotolerance	Prolactin receptor (PRLR)	CRISPR/Cas9	SDN1	(Rodriguez-Villamil et al., 2021)

Increased resistance to pathogens

Animal	Characteristic	Gene of interest	Tool	Technique	Reference
Pig	Increased resistance to transmissible gastroenteritis virus (TGEV) and Porcine epidemic diarrhea virus (PEDV)	Amino peptidase N receptor (ANPEP)	CRISPR/Cas9	SDN1	(Whitworth et al., 2019)
Cattle	Increased resistance to Bovine Viral Diarrhoea Virus	BVDV binding domain of bovine CD46	CRISPR/Cas9	SDN3	(Workman et al., 2023)
Cattle	Increased resistance to damage from Mannheimia haemolytica leukotoxin	CD18 (a signal peptide present on the surface of cattle leukocytes)	ZFN	SDN3	(Shanthalingam et al., 2016)

Details to slide 17

Welfare aspect

Animal	Characteristic	Gene of interest	Tool	Technique	Reference
Pigs	Avoid puberty-derived boar taint and aggressiveness, castration free	KISS1	CRISPR/Cas9	SDN2	(Flórez et al., 2023)
Bovine	Production of hornless dairy cattle	POLLED allele	TALEN	SDN2	(Carlson et al., 2016)

Color

Animal	Characteristic	Gene of interest	Tool	Technique	Reference
Quail	Alteration of coat color pattern	melanophilin (MLPH) gene	CRISPR/Cas9	1	(Lee et al., 2019b)
Atlantic Salmon	Pigmentation - melanin reduction	Pmel17	CRISPR/Cas9	1	(Edvardsen et al., 2014)

Details to slide 17

Hypoallergenic eggs

Animal	Characteristic	Gene of interest	Tool	Technique	Reference
Chicken	Production of hypoallergenic eggs	Ovalbumin (OVA) and ovomucoid (OVM) egg white genes	CRISPR/Cas9	SDN1	(Oishi et al., 2016)
Chicken	Production of hypoallergenic eggs	Ovalbumin (OVA)	TALEN	SDN1	(Park et al., 2014)
Chicken	Production of hypoallergenic eggs	ovomucoid (OVM)	TALEN	SDN1	(Ezaki et al., 2023)

Hypoallergenic milk

Animal	Characteristic	Gene of interest	Tool	Technique	Reference
Bovine	Production of hypoallergenic milk	Beta-Lactoglobulin (BLG)	CRISPR/Cas9	SDN1	(Singina et al., 2021)
Bovine	Production of hypoallergenic milk	Beta-Lactoglobulin (BLG)	TALEN	SDN2	(Wei et al., 2018)
Bovine	Production of hypoallergenic milk	Beta-Lactoglobulin (BLG)	ZFN	SDN1	(Yu et al., 2011)
Goat	Production of hypoallergenic milk	Beta-Lactoglobulin (BLG)	CRISPR/Cas9	SDN1	(Zhou et al., 2017)
Bovine	Production of hypoallergenic milk	LacS gene (β-glycosidase produced from Sulfolobus solfataricus	TALEN	SDN3	(Su et al., 2018)

Hypoallergenic meat → several papers but …

Details to slide 17

Animal	Characteristic	Gene of interest	Tool	Technique	Reference
Pig	Decrease of n-6PUFAs/n-3PUFAs ratio	fat-1 gene from Caenorhabditis elegans	CRISPR/Cas9	SDN3	(Li et al., 2018)
Pig	Enhanced oxidative fiber formation and intramuscular fat deposition	PPARγ	CRISPR/Cas9	SDN3	(Gu et al., 2021)
Sheep	melatonin-enriched milk	AANAT/ASMT	CRISPR/Cas9	SDN3	(Ma et al., 2017)
Sheep	Yellow fat color	beta-carotene oxygenase 2 (BCO2)	CRISPR/Cas9	SDN1	(Niu et al., 2017)
Chicken	Reduce of lipid content and fat deposition	G0/G1 switch gene 2 (G0S2)	CRISPR-Cas9	SDN1	(Park et al., 2019)
Atlantic salmon	Increased content of linoleic acid	fatty acyl desaturases (fads2)	CRISPR/Cas9	SDN1	(Datsomor et al., 2019a)
Atlantic Salmon	Inhibits elongation of polyunsaturated fatty acids	fatty acyl elongases (evovl2)	CRISPR/Cas9	SDN1	(Datsomor et al., 2019b)
Channel catfish	Improved n-3 Fatty Acid Content	Elovl2 transgene isolated from masu salmon (Oncorhynchus masou) driven by a carp β-actin promoter	CRISPR/Cas9	SDN3	(Xing et al., 2022)

Details to slide 17

Animal	Characteristic	Gene of interest	Tool	Technique	Reference
Pig	Muscle hypertrophy	MSTN	CRISPR/Cas9	1	(Li *et al.*, 2020b)
Pig	Muscle hypertrophy	MSTN	ZFN	1	(Bi et al., 2020)
Pig	Muscle hypertrophy	MSTN	ZFN	1	(Qian et al., 2015)
Pig	Increased skeletal muscle mass	Follistatin	CRISPR/Cas9	1	(Li *et al.*, 2021)
Pig	Improved lean meat percentage	Insulin-like growth factor 2 (IGF2)	CRISPR/Cas9	1	(Liu *et al.*, 2019b)
Pig	Muscle hypertrophy	FBXO40*	CRISPR/Cas9	1	(Zou et al., 2018)
Sheep	Muscle hypertrophy	MSTN	CRISPR/Cas9	1	(Guo *et al.*, 2023)
Sheep	Muscle hypertrophy	MSTN	TALEN	1	(Li *et al.*, 2016a)
Sheep	Muscle hypertrophy	MSTN	CRISPR/Cas9	1	(Zhou et al., 2022)
Goat	Muscle hypertrophy	MSTN	CRISPR/Cas9	1	(He et al., 2018)
Quail	Muscle hypertrophy	MSTN	CRISPR/Cas9	1	(Lee *et al.*, 2020b)
Sea bream	Muscle hypertrophy	MSTN	CRISPR/Cas9	1	(Sun *et al.*, 2020)
Sea bream	Muscle hypertrophy	MSTN	CRISPR/Cas9	1	(Kishimoto *et al.*, 2018)
Olive flounder	Muscle hypertrophy	MSTN	CRISPR/Cas9	1	(Kim *et al.*, 2019)

Details to slide 17

Animal	Characteristic	Gene of interest	Tool	Technique	Reference
Atlantic Salmon	Germline ablated	dnd	CRISPR/Cas9	SDN1	(Wargelius *et al.*, 2016)
Atlantic Salmon	Germline ablated and targeted single nucleotide replacements (SNR) in F0	dnd	CRISPR/Cas9	SDN2	(Straume *et al.*, 2021)
Channel catfish	Sterilize channel catfish	Follicle stimulating hormone	TALEN	SDN1	(Qin *et al.*, 2023)
Channel catfish	Sterilize channel catfish	luteinizing hormone (cgbb)	ZFN	SDN1	(Qin *et al.*, 2016)
Sterlet	Germline ablation	dnd1	CRISPR/Cas9	SDN1	(Baloch *et al.*, 2019)
Nile tilapia	arrests oogenesis causing infertility	Foxh1	CRISPR/Cas9	SDN1	(Tao *et al.*, 2020)
Rainbow trout	Germline ablated for use as germ cell transplantation	dnd	Not specified	NO	(Fujihara *et al.*, 2022)

17th GMO Network meeting,
May 2024

GMM NGT MANDATE

Dafni Maria Kagkli

efsa
EUROPEAN FOOD SAFETY AUTHORITY

MANDATE NEW DEVELOPMENTS IN BIOTECHNOLOGIES APPLIED TO MICROORGANISMS OF CAT 3 AND 4 (M-2022-00146)

- ✓ Horizon scanning report July 2023
- ✓ WG scientific opinion Jan 2023-Feb 2024
- ✓ Panel endorsement before PC Feb 2024
- ✓ Public consultation February 2024- April 2024

Adoption GMO Panel June 2024

ENGAGEMENT OF THE GMO NETWORK DURING THE MANDATE

15th Meeting of the GMO Network (June 2023)

- Results of the horizon scanning and EFSA call for data
- Input on several WG questions (guidances, risk assessments ongoing, WGS, QPS, etc)

16th Meeting of the GMO Network (December 2023)

- Status of the opinion before the endorsement by the GMO Panel and public consultation
- Discussion and responses provided to the MS
- Invitation to participate to the public consultation

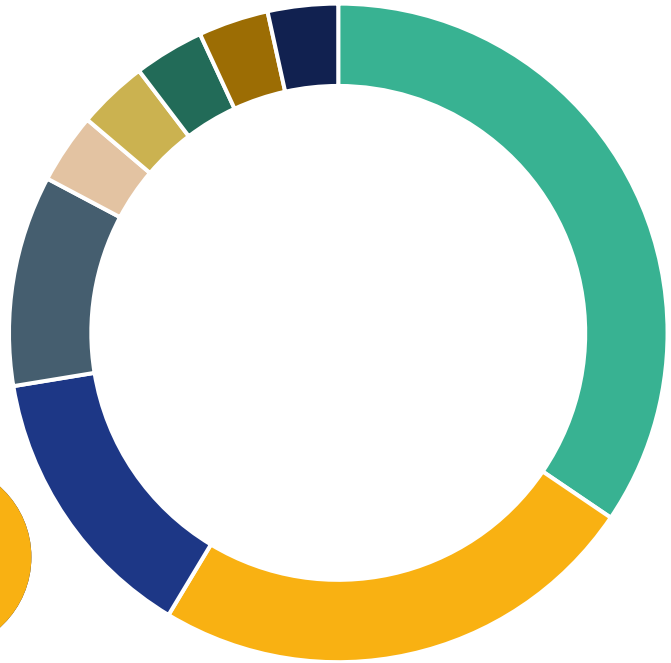
17th Meeting of the GMO Network (June 2024)

- **Status after the public consultation**

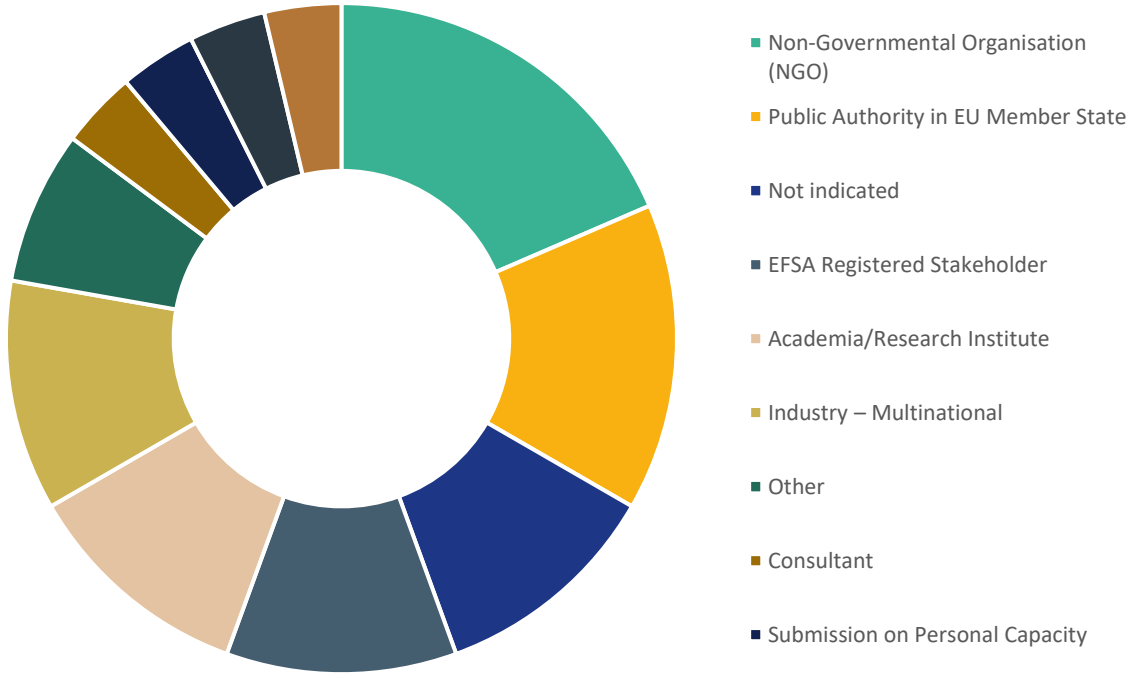
PUBLIC CONSULTATION RESULTS

- ✓ Public consultation February 2024- April 2024
- ✓ Three hundred ninety-eight (398) comments received

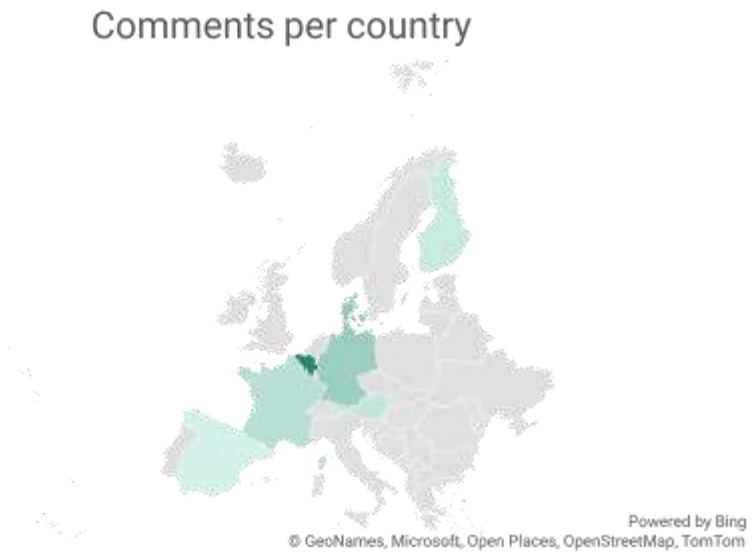
Stakeholders per country



Organizations



COMMENTS PER COUNTRY- PUBLIC AUTHORITIES CONTRIBUTION



Stakeholder	Category	Country
Federal Agency for Nature Conservation	Public Authority in EU Member State	Germany
German Federal Institute for Risk Assessment (BfR)	Public Authority in EU Member State	Germany
German Federal Office of Consumer Protection and Food Safety (BVL)	On behalf of affiliation/organisation	Germany
Danish Veterinary and Food Administration and Danish Agricultural Agency	Public Authority in EU Member State	Denmark
Biosafety Advisory Council	Public Authority in EU Member State	Belgium

Note. One Member State submitted comments after the closure of the public consultation. These comments were considered but will not be responded individually.

DETAILS ON COMMENTS PER SECTION

Chapter	Number of comments
1.2 Definition of new developments in biotechnology for the **Terms of Reference**	14
1.3 **Interpretation** of the Terms of Reference	18
1.4 General **outline of risk assessment** for genetically modified microorganisms	16
2.4 Selection and description of the **case studies**	15
3 Assessment	6
3.1 ToR1: Novel potential hazards and risks that new developments in biotechnology applied to microorganisms could pose for humans, animals and the environment	11
3.1.1 AQ1. What are the new **techniques/approaches** developed since 2001 (namely, new developments in biotechnology) which could be applied/are applied to microorganisms?	**34**
3.1.2 AQ2. Are there any **novel hazards** that these new developments in biotechnology applied to microorganisms could pose to humans, animals and the environment, as compared to ...	10
3.1.3 AQ3. Are there any **novel risks** that these new developments in biotechnology applied to microorganisms could pose to humans, animals and the environment, as compared to ...	8

Chapter	Number of comments
3.2 TOR2: Applicability and sufficiency of the existing guidelines for risk assessment of GMM to risk assess new developments in biotechnology applied to microorganisms	1
3.2.1 AQ1 and AQ2. What kind of GM microorganisms and GM microbial products within the EFSA remit have been identified and can be expected in **the next 10 years that** were developed ...	15
3.2.2 AQ3. Which are the **existing guidelines** to be used for the risk assessment of these GMMs?	11
3.2.3 AQ4. Are the **existing guidelines for risk assessment applicable, fully or partially, and sufficient for the risk assessment** of GMMs generated with the use of the new developments in biotechnology?	**148**
3.3 ToR3: In case existing guidelines for risk assessment are considered not applicable, partially applicable or not sufficient, to identify on which aspects existing guidelines should be updated, ...	3
3.3.1 AQ1. Which **aspect (if any) of existing guidelines** should be updated, adapted, or complemented?	8
3.3.2 AQ2. What **recommendations** can be formulated for future guidance updates?	**58**
3.3.3 Future recommendations	17

CLARIFICATIONS FOLLOWING THE PC- SECTIONS 1.3-1.4 AND 2

- Explanations on the scope and clarifications on the interpretation of the Terms of Reference
- Better explanation of term “sufficient” and “adequate”
- Better explanation of the outline of the risk assessment for GMMs
- Improvements and clarifications on the case studies table
- Clarification that the selected case studies are indicative to be able to address the ToRs and not exhaustive
- Alignment of language for NGT-Ms

CLARIFICATIONS FOLLOWING THE PC- TOR1 IDENTIFY NOVEL POTENTIAL HAZARDS AND RISKS

- 3.1.1.1 Addition of gene-drive like systems
- 3.1.2 Addition of the following phrase: "*Apart from some exceptions, like gene-drive like systems, the introduced CRISPR-Cas system should be removed during the process of modification when using NGTs. If present, the potential new modifications need to be assessed on a case by case basis*."

CLARIFICATIONS FOLLOWING THE PC- TOR2 APPLICABILITY AND SUFFICIENCY OF EXISTING GUIDELINES

- 3.2.1 Explanation of the 10-year interval considered for the case studies
- 3.2.3 Clarifications and re-drafting of several parts of the Opinion, namely microbial characterisation, gut microbiome, allergenicity, horizontal gene transfer

CLARIFICATIONS FOLLOWING THE PC- TOR3 WHICH ASPECT (IF ANY) OF EXISTING GUIDELINES SHOULD BE UPDATED, ADAPTED, OR COMPLEMENTED?

Area of risk assessment	Applicable guidance exists (see section 3)	Recommended updates	NGT specific update
Comparative assessment	EFSA GMO Panel, 2011	Expand the definition of comparator: inclusion as comparator of microorganisms not previously used in the food and feed chain (no history of safe use)	None
Microbial characterisation	EFSA GMO Panel, 2011; EFSA FEEDAP Panel, 2018; EFSA CEP Panel, 2021	Inclusion of protists/microalgae/viruses Antimycotic resistance of viable yeasts and fungi	Assessment of the presence/absence of the CRISPR-Cas system intentionally introduced
Production process	EFSA GMO Panel 2011, EFSA FEEDAP Panel 2018, EFSA CEP Panel 2021, EFSA ANS Panel, 2012, EFSA FAF Panel, 2021	Inclusion of protists/microalgae/viruses	None
Compositional analysis	EFSA GMO Panel 2011	None	None
Toxicological assessment	EFSA GMO Panel 2011	Inclusion of in silico and in vitro methods to replace animal studies	None
Gut microbiome	EFSA GMO Panel 2011, EFSA FEEDAP Panel 2018	The setting of suitable endpoints and the development of validated methodologies are recommended to assess effects on the gut microbiota	None

CLARIFICATIONS FOLLOWING THE PC- TOR3 WHICH ASPECT (IF ANY) OF EXISTING GUIDELINES SHOULD BE UPDATED, ADAPTED, OR COMPLEMENTED?

Area of risk assessment	Applicable guidance exists (see section 3)	Recommended updates	NGT specific update
Allergenicity	EFSA GMO Guidance, 2011	Expand on adjuvanticity and potential methodologies (when available) to assess it	None
Nutritional assessment	EFSA GMO Panel, 2011, EFSA FEEDAP Panel 20XX	None	None
Exposure assessment	EFSA GMO Panel 2011	Address primary and secondary exposure for all uses and microorganisms under the remit of EFSA	None
ERA	EFSA GMO Panel 2011	Inclusion of all uses and microorganisms under the remit of EFSA Detail all areas of risk as per Directive 2001/18/EC	None
HGT	EFSA GMO Panel 2011	Consideration of cases in which the HGT may not be needed	None
PMEM	EFSA GMO Panel 2011	Include fit-for purpose approaches to monitor for potential adverse environmental effects Broaden scope to include other uses other than food/feed uses under the remit of EFSA Considerations of cases in which PMEM may not be needed based on the ERA	None

CLARIFICATIONS FOLLOWING THE PC- TOR3 WHICH ASPECT (IF ANY) OF EXISTING GUIDELINES SHOULD BE UPDATED, ADAPTED, OR COMPLEMENTED?

- Addition of 3.3.3 Recommendations for additional guidance

In the case of GMMs, including the NGT-Ms, developed to contain engineered gene drive or similar technologies designed to bias-and therefore speed up- the transmission of certain genetic elements in a target population, additional guidance is recommended to be developed.

- Merging the future recommendations with ToR3.

CONCLUSIONS

- EFSA GMO Panel concludes that none of the EFSA guidances are “fully applicable” but they are “partially applicable”

- The EFSA GMO Panel also notes that “not sufficient” does not imply that more requirements are needed for the risk assessment of NGT-Ms; on a case-by-case basis less requirements may be needed.

- Possible hazards relate to the genotypic and phenotypic changes introduced in the microorganism, not to the method used for its modification.

- The GMM NGT WG made recommendations for updates not exclusive to the assessment of NGT-Ms

RECOMMENDATIONS

- The microbiological risk assessment approach should therefore be based on the strain/product itself, independently of the method used to alter genotypic or phenotypic characteristics.

- It is therefore recommended that any new guidance should take a common risk assessment approach for strains/products derived from or produced with microorganisms obtained with conventional mutagenesis, EGTs or NGTs.

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Some recent trends in agri-food biotechnology and their possible implications for food & feed safety, regulation, and enforcement

(results of desk research at WFSR, Netherlands)

NLD delegation

EFSA GMO MS network meeting, May 2024

Schedule

- Background
- Highlights from the reports
- Overarching conclusions

Disclaimer: the views presented here are those of the authors of the presentation and featured reports and may not represent those of the sponsors from the Dutch government

Background

"Analysis and evaluation of new risks to the food chain and animal feed production"

- Perennial activity
  - Part of our institute's "statutory tasks"
  - Funded by the Dutch Ministry of Agriculture, Nature and Food Quality
  - Each year, recent developments in biotechnology are reviewed
    - Technological progress
    - Implications for:
      - Safety assessment
      - Detection and traceability
      - Regulation

Highlights: Plant molecular farming

Production of animal proteins in plants for different food purposes:

- Enzymes (*e.g.,* chymosin)
- Animal protein analogues (*e.g.,* casein)
- Feed additives
- Cell culture media additives (*e.g.,* growth factors)
- Functional proteins (*e.g.,* heme proteins)

Highlights: Plant molecular farming

Possible risks: minor concerns overall

- Health:
  - To be assessed pre-market as transgenic products
  - Concern over allergy risks if commingled with mainstream host crop
- Commingling:
  - Depends on procedures and rules in countries exporting host commodity
    - Little concern, *e.g.,* example of amylase-producing maize (proxy)
  - Accidental exposure for other countries growing molecular farming crops
    - Marginal exports and stringent GMO approval procedures

Highlights: Random mutagenesis innovation

Innovative mutation breeding methods:

- Random mutagenesis
  - Space breeding (cosmic radiation)
    - Different range, combination with microgravity and temperature
      - Mainly causes point mutations
  - Ion particle beams
    - Higher energy transfer, more clustered DNA damage
      - Indels, point mutations and chromosome rearrangements
      - By-stander effects on non-affected cells
- Genomic selection
  - TILLING (Targeting Induced Local Lesions In Genomes)
    - Combination of chemical mutagenesis and genomic screening, more efficient

Highlights: Random mutagenesis innovation

Conclusion:

- Space breeding and ion beams:
  - More clustered DNA damage
    - Co-segregation of clustered mutations more likely
      - Breeding practice as “safety net”: multiple backcrosses, discard off-types, molecular characterization
  - Frequencies of mutations may differ, yet no new types of mutation are found
- All random techniques:
  - Off-target mutations more likely than for targeted mutagenesis
    - Insertion of vector DNA a theoretical possibility for vector-based forms of targeted mutagenesis

Highlights: GMM biomass valorization

Recent developments:

- GMMs use for bioethanol production
  - GM yeast strains increasingly used
    - Higher yield, tolerance to stressors, reduced by-products
    - Cellulosic substrates (crop residue etc.)
  - Forty-two strains of GM yeast identified
- Gas fermentation using GMMs
  - Commercial non-GM strains for conversion of CO_2, CO, CH_4, H_2, and NH_3 into EtOH & feed
  - Synthetic biology applied for production of other chemicals besides ethanol

Considerations:

- GM feed approval required
- Non-EU nations may not specifically consider the presence of GM DNA as criterion for categorization as “GM”
  - Nature of modifications not of concern

Highlights: Null-segregants in plant breeding

Definition:

- Non-transgenic progeny of transgenic plants that lost the trait through segregation

Potential fields of application:

- Early flowering
- Reverse breeding
- Double haploids
- Synthetic apomixis
- Seed Production Technology
- RNA-dependent DNA methylaion

Considerations:

- Status as GM or non-GM varies across the globe
- Detection and traceability challenging
- No accounts or plausible theories of possible health impacts

Examples of other topics considered in the past

- Biotechnology applied to animal production:
  - Pigs
  - Cattle
  - Fish
- Updates (recurrent) on new genomic techniques
- Microbial biotechnology
  - Microalgae
  - Bacteriophages
  - Yeasts/fungi

Overall conclusions

- Ongoing biotechnological developments in crops, microorganisms and livestock used for food and feed
- So far, no major issues identified for current and near-future applications
  - Traceability/detectability not always feasible
- Need to keep track of developments

Acknowledgement

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IMPROVEMENT OF PMEM PLANS FOR IMPORT AND PROCESSING APPLICATIONS

Ana Martín Camargo (EFSA, NIF)

GMO Network

30/05/2024

EUROPEAN FOOD SAFETY AUTHORITY

BACKGROUND

- Directive 2001/18/EC includes an obligation for notifiers to implement a Post-Market Environmental Monitoring (PMEM) plan
- EFSA gives its opinion on adequacy of scientific rationale of the PMEM plan
- Adoption and implementation of the PMEM plan are a risk management issue, and therefore outside the remit of EFSA

BACKGROUND

- The objectives of a PMEM plan are:

A. **Case-Specific Monitoring** to confirm that any assumptions regarding occurrence and impact of potential adverse effects of the GMO identified in the ERA are correct

B. **General Surveillance (GS)** to identify occurrence of adverse effects of the GMO or its use unanticipated in the ERA (mandatory for each application)

- In case no risks or significant levels of critical uncertainty are identified in the ERA, then a PMEM plan consists only of GS
- For I&P applications, GS is commonly composed of observations by those directly involved in handling and processing of the GM crop and monitoring of ongoing research and development and scientific literature

BACKGROUND

- Member States recurrently comment on the lack of elaboration of PMEM plans – Methodology proposed for General Surveillance needs more detail

Austria	Fed.Ministry_Soc.Affairs/Health	II.6.3 General Surveillance (strategy, method)	General surveillance for unanticipated adverse effects The proposed general surveillance for unanticipated adverse is not sufficiently elaborated and should be amended regarding the following elements: • Elaboration of a detailed monitoring methodology (e.g. parameters, specific information). • Identification of existing national institutions and operators involved in GS in individual Member States and evidence for their commitment to GS activities. • Assignment of clear responsibilities and concrete
Germany	BVL (German CA)	II.6.3 General Surveillance (strategy, method)	The monitoring plan does not relate the monitoring activities to relevant protection goals. Even more it is not described which routine observations (including parameters or monitoring characters) are carried out in relation to the protection goals. Only reporting on 'any unanticipated effect' is solely not an appropriate parameter, because it already anticipates an evaluation. This evaluation process should be based on a distinct set of parameters and a scientific sound data analysis. It is requested that the applicant specifies in detail, how and which information will be pro-actively queried, gathered, and how they will be evaluated. In addition, it might be useful to integrate
Germany	BfN	II.6 Post-Market Environmental Monitoring Plan (PMEM)	The scope of this application is for import, processing and all uses for food and feed. The applicant provided an environmental monitoring plan, which remains very general. The monitoring plan has to be elaborated in more detail in order to meet the following requirements: • Provision of a fully specified list of monitoring parameters. • Application of standardized sampling methodologies: A basic prerequisite for comparing GMO monitoring data is the use of appropriate standard detection or analytical methods. Several standards specific for GMO monitoring are provided by the Association of German Engineers (VDI). They are available under http://www.vdi.eu/engineering/vdi-standards/. • Elaboration of a sampling concept. • In case of monitoring data being collected by external people or institutions other than the applicant, binding agreements/contracts with third parties are requested which clearly determine what data are provided and how these data are made available

BACKGROUND

- The WG of the GMO Panel dealing with the ERA has discussed the adequacy of the methodology proposed in PMEM plans
- WG considered that PMEM plans suggested by applicants are proportionate to the scope of the application, but they lack transparency
- EC supported EFSA's initiative to start a dialogue with MS & applicants to clarify how PMEM plans are practically implemented
- PMEM plans are at the border between risk assessment and risk management, so coordination between risk assessors and risk managers is needed

ONGOING ACTIVITY

- CompERA WG considered to complement the current PMEM plan proposed by applicants with additional documents to:
  1. Increase transparency on the actual monitoring activities implemented by applicants by providing more detail on the methodology proposed for General Surveillance
  2. Identify the locations in the EU where exposure to GM material is more likely (e.g. main transportation hubs or processing plants dealing with import/processing of GM plants)
  3. Identify the steps in the processing of the GM plant material when exposure to the environment and environmental harm are more likely
- CompERA WG considered that the identification of areas with higher potential exposure to GM material was unfeasible and sensitive
- CompERA WG agreed to recommend applicants to **provide further information on the monitoring activities proposed to detect unanticipated adverse effects and their expected outcome**

PROCESS SO FAR

Jan 2024

CompERA WG makes public a set of **recommendations to applicants** for the preparation of PMEM plans (Annex I webminutes January 2024)

Feb 2024

EFSA **presents the proposal to MS at PAFF meeting**

Apr 2024

Start of **discussions with applicants and operators**

RECOMMENDATIONS OF THE COMPERA WG

Jan 2024

CompERA WG makes public a set of **recommendations to applicants** for the preparation of PMEM plans (Annex I webminutes January 2024)

Annex I.

Recommendations for the preparation of PMEM plans for applications for import, processing and all food and feed purposes

Directive 2001/18/EC (EC, 2001) introduces an obligation for notifiers to implement a post market environmental monitoring (PMEM) plan to confirm that any assumption regarding the occurrence and impact of potential adverse effects of the GMO or its use in the ERA are correct, and to identify the occurrence of adverse effects of the GMO or its use on human health or the environment which were not anticipated in the ERA.

It is worth reminding that the adoption of the final PMEM plan and its implementation is a risk management issue and thus falls outside the remit of EFSA. However, the GMO Panel gives its opinion on the adequacy of the scientific rationale of the PMEM plans proposed by applicants.

During the risk assessment of GM events for food and feed uses (submitted under Regulation (EC) No 1829/2003), the GMO Panel considers the scientific comments provided by Member States. The CompERA WG of the GMO Panel has discussed previously the need to update PMEM plans (see minutes of 19 September 2017, 24 October 2017, 29 November 2022, 26 January 2023, 14 March 2023, 25 April 2023, 11 May 2023, 27 June 2023, 26 September 2023, 28 November 2023) and shares the concerns raised by different Member States on a lack of detail of the methodology proposed by applicants for the implementation of PMEM plans.

The CompERA WG therefore recommends applicants to provide detail on the methodology proposed for the General Surveillance of PMEM plans by clearly stating the specific monitoring activities proposed and their expected outcome.

RECOMMENDATIONS OF THE COMPERA WG

Suggested examples

Hereunder, applicants are provided a non-exhaustive list of examples of measures proposed for the implementation of General Surveillance of PMEM plans.

Considered route of exposure: Accidental release of viable GM material during import, handling, storage and processing.

Affected area(s) of concern: Persistence and invasiveness; vertical gene transfer.

Operational strategy: Modern systems for handling seeds

Description of actions: Monitoring the handling areas to identify spilled seeds/grains; cover with protective systems storage/transport containers by various means; cleaning equipment/handling areas in event of spillage.

Expected outcome: Prevent accidental release of GM seeds/grains.

Operational strategy: Inspection of ports, storage and processing facilities for target feral plants.

Description of actions: Indicate operators in charge of the inspections; ensure personnel carrying out the inspections receive adequate training to identify target species and have basic knowledge on their biology and trait identification.

Expected outcome: To identify potential feral GM plants.

Operational strategy: Monitoring of target feral plants (in ports, and storage and transformation facilities)

Description of actions: To report monitoring strategy including inspected places, frequency and recording methodology of the observations. The strategy should

RECOMMENDATIONS OF THE COMPERA WG

Feb 2024 EFSA **presents the proposal to MS at PAFF meeting**

- **Member States supported the proposal** to request more detail on the methodology of the PMEM plan

- **Considerations** on:
  - How would applicants provide additional information in the PMEM plan, as an annex?
  - How much detail would be needed?
  - Should more detail be requested for all crops or only for oilseed rape?
  - Should this additional info be crop-specific?
  - Can the same set be annexed for a group of crops?
  - Need for balance between being very prescriptive vs. keeping flexibility
  - Importance to confirm that activities declared are carried out – risk managers should carry out inspections

RECOMMENDATIONS OF THE COMPERA WG

Apr 2024 Start of **discussions with applicants and operators**

- First discussion held on 18 April 2024 – attended by representatives from CropLife Europe, other applicants and the operator networks FEDIOL and COCEREAL
- **More discussion is needed**

Q&A

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EUROPEAN PARLIAMENT MANDATE TO EFSA ON THE ANSES REPORT ON THE EC-CATEGORY 1 NGT PLANT PROPOSAL

Nikoletta Papadopoulou
Team Leader NIF
GMO Molecular Characterisation

EUROPEAN FOOD SAFETY AUTHORITY

EP MANDATE TO THE EFSA GMO PANEL

- The European Parliament on 22 February 2024 requested EFSA in accordance with Article 29 of Regulation 178/2002, **to deliver a scientific opinion on the analysis by ANSES on Annex 1 of the EC proposal** for a regulation on plants obtained by certain NGTs and their food and feed, and amending Reg (EU) 2017/625 (NGTs plants proposal)
  - ANSES opinion published January 2024
  - EC proposal and Annex 1 criteria as published 5th July 2023

ANSES OPINION

An opinion of the French National Agency for Food, Environmental and Occupational Health and Safety (ANSES) was published on 24 January 2024 (ANSES, 2023) providing an analysis of Annex I equivalence criteria for Category 1 NGT plants, of the EC proposal for plants obtained by NGTs.

The ANSES opinion calls for clarifications on:

1. several **aspects and definitions of the Annex I** of the EC proposal.
2. **the scientific basis of the equivalence criteria** included in Annex I of the EC proposal.
3. the **potential risks of NGT plants** falling under Category 1.

WORKING GROUP AND TIMELINE

WG: Cross Cutting working group of the GMO Panel

Chair (EFSA): Franco Neri

Experts: Josep Casacuberta, Javier Moreno, Ewen Mullins, Fabien Nogue, Nils Rostocks

EFSA staff: Ana Afonso, Paolo Lenzi, Nikoletta Papadopoulou (coordinator), Tommaso Raffaello

Scientific opinion to be delivered by end of July 2024

1. 13/3 GMO panel – EP mandate and ANSES opinion was presented
2. 11/4 GMO 1st Cross-cutting WG on the EP mandate – review ANSES opinion/ structure of draft opinion
3. 2/05 GMO 2nd Cross-cutting WG on the EP mandate
4. 15/5 GMO panel discussed draft opinion
5. 27/05 GMO 2nd Cross-cutting WG on the EP mandate- ANSES as hearing experts for potential clarifications.
6. **19-20 June: proposed adoption at the GMO Panel meeting**

STRUCTURE OF THE EFSA OPINION

Abstract

Keywords

Summary

1. Introduction/Background

1.1. Request by the EP

2. Data and Methodology

3. Assessment

3.1 Terms used in the EC proposal and the criteria on Category 1 NGT plants in Annex 1 of the EC proposal that ANSES considers as requiring clarification

3.2 Scientific basis of the equivalence criteria included in Annex I of the EC proposal.

3.3. Potential risks of NGT plants falling under Category 1

7. Conclusions

8. Recommendations

9. References

DATA AND METHODOLOGY

- The cross-cutting WG took into account **all relevant scientific considerations from the published EFSA Opinions on targeted mutagenesis** (including site-directed nuclease (SDN) type 1, 2 and 3, and oligonucleotide directed mutagenesis), cisgenesis and intragenesis (EFSA GMO Panel, 2012a,b, 2020, 2022a, b) to support the development of this Scientific Opinion.
- Past Scientific Opinions were subject to open **consultation** and extensive public scrutiny as clarified in the EFSA acceptance letter to the EP mandate; therefore another public consultation on this opinion was not deemed necessary.
- A **protocol** to plan the scientific assessment methodologies was deemed unnecessary for this mandate.
- **The WG considered the ANSES's analysis, conclusions and questions, and provides clarifications.**

ANSES REQUESTS CLARIFICATION ON ASPECTS AND DEFINITIONS OF THE EC PROPOSAL ANNEX 1

DEFINITIONS (3.1)

- Lack of clarity in the definition of the "breeders' gene pool", and the use of the expression "genetic information", which needs to be clarified.
- The term “cisgenesis” contained in Article 3 of the proposed regulation requires to be explained further.
- The proposed regulation does not provide a definition of the conventional breeding techniques considered in this proposal.
- Definition of “targeted site” is not clear enough and ANSES makes a proposition

ANSES REQUESTS JUSTIFICATION ON THE SCIENTIFIC BASIS OF ANNEX 1

SCIENTIFIC BASIS OF THE CRITERIA (3.2)

- ANSES emphasizes that the size of the modification does not provide any information on its functional consequences. The limit of 20 nucleotides maximum has no biological significance
- ANSES concludes that deletions observed in conventional plants are more often close to a kilobase. Whatever their size, the functional consequences of these deletions should be characterized
- ANSES believes that targeting the cisgenic sequence at the site of orthologous sequence (SDN2 like) would make it possible to avoid potential position effects associated with a new insertion site
- ANSES believes that, as for the deletion criterion, criterion for inversion (no size limit), is not justified in view of the literature on pan-genome analyzes

CONCERNS EXPRESSED IN THE ANSES REPORT FOR POTENTIAL RISKS OF NGT-1 PLANTS

POTENTIAL RISKS (3.3)

- Number of 20 modifications in total chosen as a maximum accepted for an NGT-1 plant to be considered equivalent to a conventional plant is not justified
- ANSES recommends that predicted off-targets should be eliminated as much as possible from NGT plants or evaluated if they cannot be eliminated
- ANSES requests a clarification regarding the consideration of genetic modifications (including non-predicted off-targets), generated in the rest of the genome

ASSESSMENT OF THE ANSES OPINION BY THE GMO PANEL WG

1. The cross-cutting WG considered the ANSES's analysis, conclusions and questions. On several aspects and definitions, **the WG provides clarifications on terms used and citations to existing definitions** from the EC proposal and the EFSA GMO Panel opinions.

2. The WG clarifies that the criteria proposed in the Annex 1 of the EC proposal were developed **to determine whether a given NGT plant is equivalent to conventional bred plants** (including plants obtained by random mutagenesis). Given the data available in the scientific literature (EC, 2023), **this number is a conservative threshold**.

3. The criteria are not meant to define levels of risks. EFSA would like to remind that with respect to the potential risks of NGT plants, **the EFSA GMO Panel in its past opinions did not identify any additional hazard associated with the use of NGTs compared to conventional breeding techniques.**

DRAFT CONCLUSIONS

- With regards to the definition of the target site, this may need to be clarified in future texts.

- With respect to all equivalence criteria, the EFSA GMO Panel considers that the available scientific literature shows that plants containing the types and numbers of genetic modifications used as criteria to identify NGT-1 in the NGT plants proposal do exist as the result of spontaneous mutations or random mutagenesis, and therefore considering them as equivalent to conventional-breed plants is scientifically justified.

- The equivalence criteria described in the NGTs plant proposal are not meant for the safety assessment of Category 1 NGT plants, but they rather allow to classify NGT plants as equivalent (or not) to conventional bred plants with respect to the similarity of the type of genetic modifications and the similarity of the type of risks.

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ANSES' COLLECTIVE APPRAISAL ON RISK EVALUATION OF NGT PLANTS

Mandate 2021-SA-0019

1. Context and introduction

a. Mandate

Commission's study on NGTs

(April 2021)

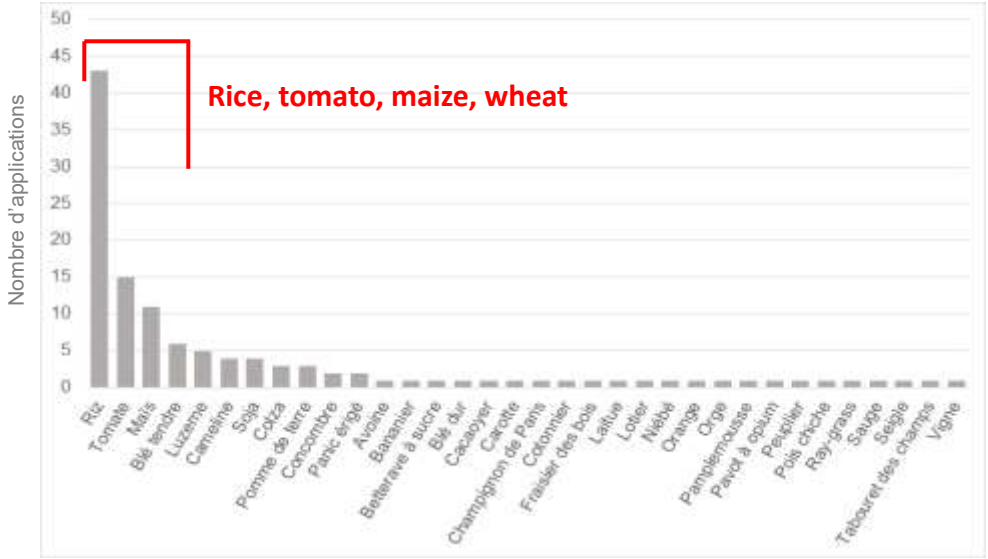
JOINT MANDATE TO ANSES BY MINISTRIES IN CHARGE OF AGRICULTURE AND ECOLOGY

WORKING GROUP LAUNCHED IN OCTOBER 2022

Determine wether **adaptations** should be introduced in the regulatory requirements for the purpose of **health and environmental risk assessment** concerning plants obtained by **targeted mutagenesis**

Analyse the **socio-economical context and issues** of the introduction of NGTs

b. Most common applications



Most NGT applications are performed using a CRISPR-Cas system, and consist of insertions / deletions of a few base pairs.

WORK FOCUSED ON TARGETED MUTAGENESIS USING CRISPR-CAS

Screening of applications using **CRISPR-Cas**

Number of application for different species, some of them being significantly different compared to transgenesis obtained plants

2. Health and environmental risk assessment of NGT plants

a. Molecular characterization of NGT plants

GOAL

Understand the nature, frequency and determinants of undesired effects occuring both on- and off-target, in order to propose adapted guidelines to characterize plants obtained by targeted mutagenesis

METHOD

Systematic litterature reviews

a. Molecular characterization of NGT plants

Analysis of original articles published between 2021 and june 2023

Biased approaches used in 78% of the published works, unbiased in 18 %, combined approaches in 4 %

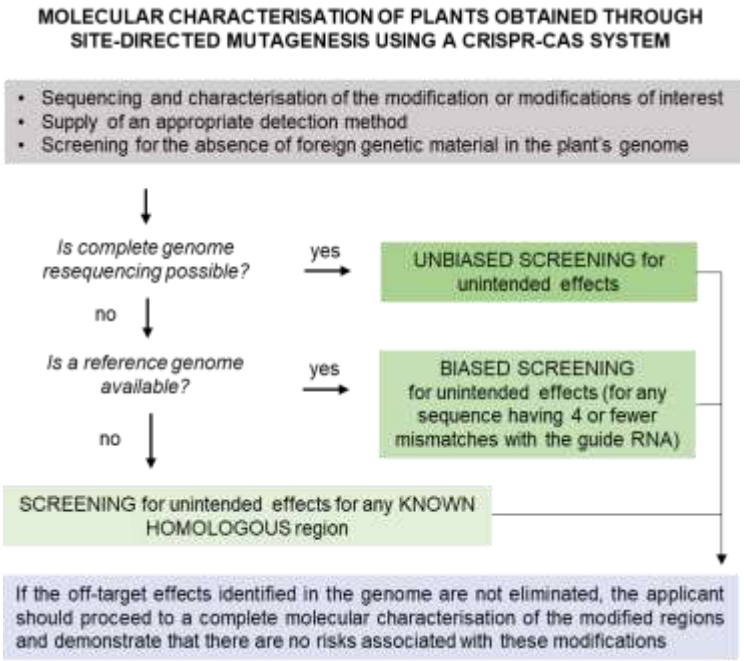
In total, undesired effects identified in 34 % of the publications

Among 837 sequences analysed to identify undesired effects (off-target), using a biased approach, occurrence of the undesired effect is identified for **7 % of the sequences**

For most cases, **short deletions or insertions**

Number of mismatches between the gRNA and the off-target sequence: **lower or equal to 3**

a. Molecular characterization of NGT plants



Recommandations based on feasability of genome sequencing

Technical requirements detailed in the opinion

b. Health and environmental risks – analysis of the current assessment framework

GOAL

Analyse the applicability and pertinence of the current requirements for the assessment of GMOs in view of their application to plants obtained via targeted mutagenesis

METHOD

Step by step analysis of the current assessment guidelines

b. Health and environmental risks – analysis of the current assessment framework

Experts conclude that the currently applicable references are only partially adapted to the assessment of plants obtained by targeted mutagenesis

In particular, requirements regarding expression of new proteins are not directly applicable and the analysis of the risk of gene transfer to micro-organisms is of low pertinence

Experts moreover consider that technical difficulties might appear to perform certains studies

c. Health and environmental risks – literature review and case study

GOAL

Identify health and/or environmental risks associated to plants obtained by targeted mutagenesis (using CRISPR-Cas)

METHOD

Systematic literature review

Study of 12 cases representing the diversity of possible applications of CRISPR-Cas

c. Health and environmental risks – litterature survey and case studies

3 database searched (Scopus, Pubmed, CAB Abstracts) :

- 296 unique referencies identified
- 13 selected references

→ **Only reviews were retrieved (no original article)**

Experts conclude that new risks for heath and/or the environment that could be associated with plants obtained by targeted mutagenesis would be mainly linked to :

- **obtention of genotypes that cannot be achieved by other selection techniques**
- **new species and characters might be modified thanks to CRISPR-Cas, compared to plants obtained via transgenesis (modification of more invasive plants, or easier modifications of the composition)**
- **a potentially significant increase of cultivated surfaces bearing varieties with the same modified character**

Experts also recall that part of the known risks associated with GMOs plants are still valid for the one obtained with CRISPR-Cas

c. Health and environmental risks – litterature survey and case studies

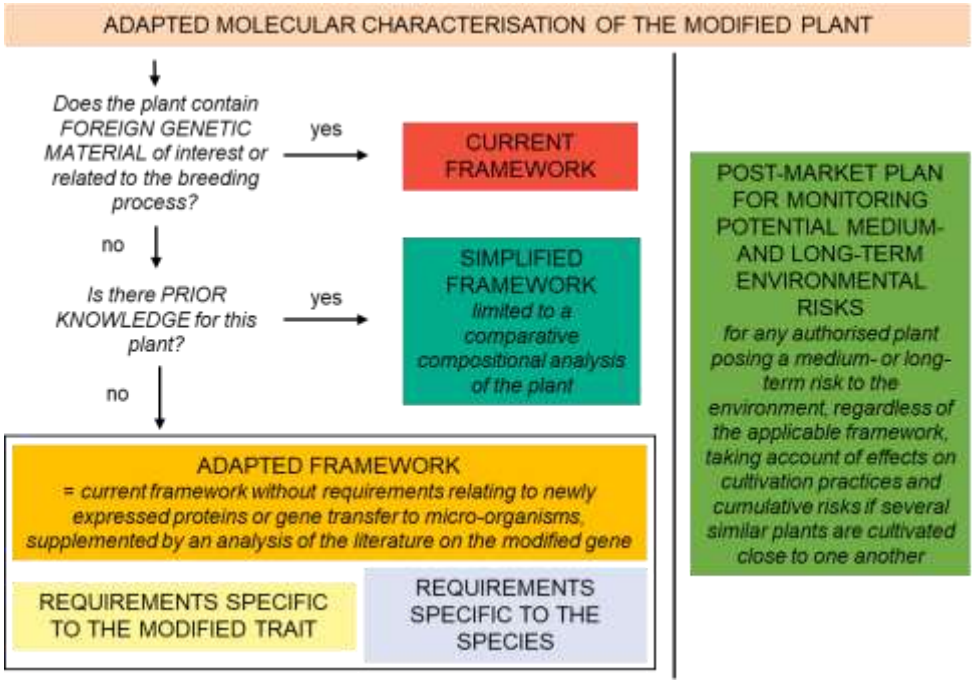
Experts consider that certain types of potential risks are recurrent :

- **unexpected modification of the plant composition**
- **environmental risks in the medium and long terms**

Experts also conclude that in certain cases, use of CRISPR-Cas for targeted mutagenesis allows to replicate known phenotypes, by acting precisely on one or few well defined genes, and that a lower level of risk might be associated to these plants

In conclusion, experts recommend a graded approach for assessment of plants obtained by targeted mutagenesis, on a case by case basis

d. Proposal for a new framework



Recommandations based on the predicted risk level

Recommandation of a reinforced post-market monitoring plan

3. A word on socio-economic impacts

A word on socio-economic impacts

Advantages (modification *vs* status quo) :

- More supportive to NGTs development
- Lower « cost to market » and costs of coexistence for NGTs sectors
- Possible positive impacts on competitiveness of european agriculture
- …

Disadvantages (modification *vs* status quo) :

- Possible effects on costs and credibility of non-NGTs sectors (e.g. organic)
- Lower freedom for consumers choices
- Potential issues for public engagement (with respect to their views on technologies introduction)
- …

The WG recommended

- **A post-authorization monitoring plan to help control the effects of the development of NGT plants (market powers, level of concentration, value-sharing, etc.)**
- **A system to ensure their traceability and control**
- **The decisions on the development and management of NGT innovations should be considered as social choices that cannot be based solely on scientific and socio-economic arguments – these social choices should be expressed, structured and governed in a democratic way**

4. Conclusion and perspectives

Conclusion and perspectives

- **Current ex-ante assessment framework only partially adapted**
  - **Proposal for a revised assessment, with a graded approach (current / simplified / adapted framework)**
  - **Recommendation for a case by base analysis, according to the proposed framework**
- **Importance of the post market surveillance**
  - **Broader scope (not limited to undesired effects or risks aspects)**
- **Socio-economic analysis**
  - **Wide range of socio-economic issues**
  - **Importance to take account for social choices in future decisions**

- **Need to develop jointly the future framework and technical guidelines**
  - **Following the regulatory decisions, in close cooperation with Efsa and other countries agencies**

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THANK YOU FOR YOUR ATTENTION

UPDATE FROM THE SUBGROUP ON NGTS

17th GMO Network – Brussels – 30-31 May 2024

EUROPEAN FOOD SAFETY AUTHORITY

BACKGROUND

WHY and HOW:

1. 89th Advisory Forum meeting (October 2023)
2. 90th Advisory Forum meeting (November – December 2023)
3. GMO Network meeting (December 2023)
4. 91st Advisory Forum meeting (March 2024)
5. Establishment and first meeting of the Subgroup on NGTs

BACKGROUND

89th Advisory Forum meeting (October 2023)

- EFSA presented an overview of the relevant discussions on NGTs that took place at the GMO Network meetings in the last three years

90th Advisory Forum meeting (November – December 2023)

- Risk assessment of plants developed using **new genomic techniques presents challenges**
- The goal is to achieve **wide engagement and acceptance of future RA guidance and address challenges consciously**, ensuring effective communication and understanding among Member States throughout the process.

GMO Network meeting (December 2023)

- AF and the GMO network of Member States welcomed EFSA's proposal to set up a sub-group of the GMO network on NGTs to foster knowledge sharing and jointly address the risk assessment challenges

91st Advisory Forum meeting (6-7 March 2024)

- Fast track process to setup the Subgroup on NGTs (ToR and nominations)

TERMS OF REFERENCE (LINK)

Main Objective of the Subgroup on NGTs:

- The main objective of the Subgroup on NGTs is **to foster knowledge sharing** on the development of NGTs, their application to plants, animals and microorganisms **and jointly address the risk assessment and monitoring challenges** specifically linked to NGTs applied to plants, animals and microorganisms.

…And from the 91st AF meeting:

- **the subgroup will serve as a consultative body for EFSA's Working Groups and Panel**

CASE STUDIES DISCUSSION

- 3 groups (2 on-site and 1 online)
- 2 case studies

Questions to be answered:

1. What are the **RA challenges for MC, COMPERA and FF areas** using current RA requirements?
2. What are the **RA areas that need further development and/or elaboration** when assessing NGT plants?

Case study 1 (Sánchez-León S, et al., 2018)

Crop: common wheat

Intended trait: reduced α-gliadin content

Technique: CRISPR/Cas9 construct targeting >30 *Gli*-2 loci. The CRISPR/Cas9 produced indels at the target loci obtaining a knock-down of the α-gliadin.

The CRISPR/Cas9 cassette will not be present in the final wheat.

Case study 2 (hypothetical)

Crop: durum wheat

Intended trait: leaf rust (Lr) resistance

Technique: 10 CRISPR/Car9 targeting 10 endogenous *Lr* gene. The endogenous genes will be replaced with Lr genes from a wild relative conferring broader resistance. In addition, 15 endogenous 'susceptibility genes' were disrupted via CRISPR/Car9 approach to promote a durable resistant phenotype.

The CRISPR/Cas9 cassette will not be present in the final wheat.

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GM PLANTS PROTEIN SAFETY MANDATE PRESENT AND FUTURE RISK ASSESSMENTS

Network GMO
Antonio Fernandez
Scientific Officer
May 2024

EUROPEAN FOOD SAFETY AUTHORITY

CURRENT STATUS

Survey

Open until 18th Feb 2024

Public consultation

End November 2024 – mid January 2025

Adoption of the Scientific Opinion

Middle 2025

Time to act by all parties involved

EFSA Developmental projects (GMO-15 cluster) plus potential OECD workshop

PROTEIN SAFETY – RISK ASSESSMENT REQUIREMENTS AT PRESENT

Protein safety = protein toxicity and allergenicity

2003-2009

<u>Codex 2003/2009 defined the principles for the assessment</u>

- Main information considered:
  1. Knowledge on the source/protein – HoSU
  2. Bioinformatics analysis
  3. *In vitro* studies
  4. *In vivo* studies

EFSA GMO PANEL MANDATE

Scientific Opinion reflecting on current practice, challenges and future opportunities of protein safety in GMOs

1. Lessons learned from experiences in the assessment of newly expressed proteins in the last 20 years, including more recent complex cases

2. Building on the experience and issues identified, develop a critical appraisal of new methodologies available with the potential to be used as complementary/ alternative testing strategies to current methodologies described in legal frameworks

3. Road map for future implementation of such complementary/alternative methods in risk assessment strategies

4. Recommendations for further research to address methodological development needs

DEVELOPMENT NEEDS

Weight of evidence approach Protein safety

- History of safety use
- Protein characterisation
- Mode of action
- Stability
- Source organism
- Phylogeny
- Structural/Functional similarity to known proteins
- Similarity to known toxins/allergens
- Fate in the gastrointestinal tract
- Interaction between proteins
- Others

Toxicological assessment

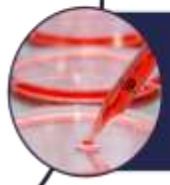
Tiered approach using *in vivo* studies only if concerns identified

Allergenicity assessment

Ranking of allergens[2] and post-market monitoring

New Approach Methodologies (NAMs) [3]

In silico tools: information on the derived structure of the novel protein



In vitro testing: stability tests could better inform about the fate of the novel proteins during processing, storage and after digestion in the gastrointestinal tract

EUROTOX 2023 – Toxicology letters – https://toxlet-384-s1.elsevierdigitaledition.com/

[2] EFSA GMO Panel, 2022. Scientific Opinion on development needs for the allergenicity and protein safety assessment of food and feed products derived from biotechnology. EFSA Journal 2022;20(1):7044

[3] Cattaneo et al., 2023. Implementing New Approach Methodologies (NAMs) in food safety assessments: Strategic objectives and actions taken by the European Food Safety Authority. Trends in Food Science & Technology, 133:277-290

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PROTEIN SAFETY MANDATE: SURVEY

- Survey: replies covering industry, academia, public authorities and NGOs

Current situation

- Actual WoE identifies no/few safety concerns
- *In vivo* studies contribute least (industry) and most (academia/authorities)
- Gaps: Complex cases (intractable, multiple NEPs), NEPs interaction, minimise animal use
- Gaps to be addressed with NAM validation

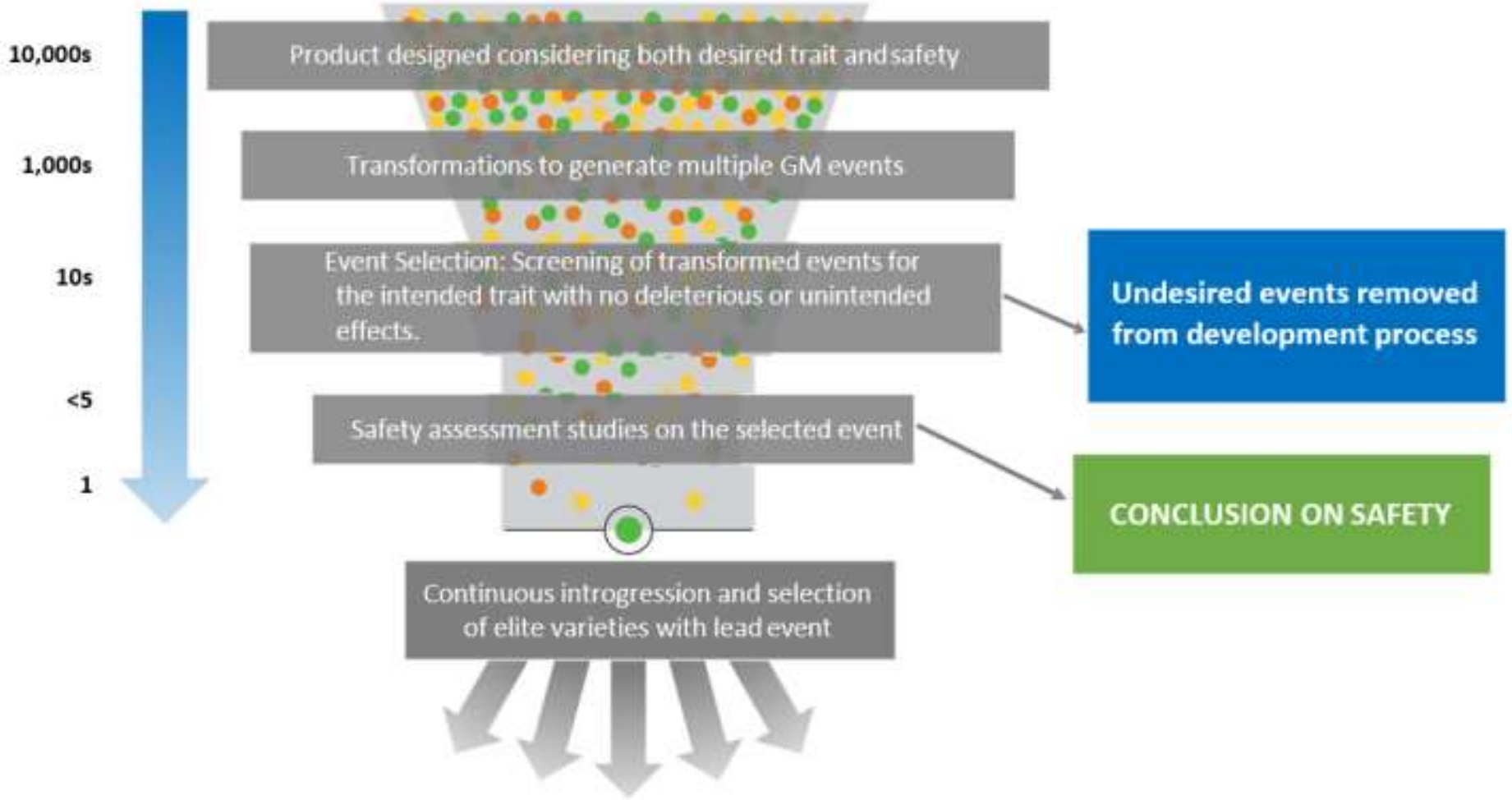
Complementary methodology

- *In silico* (GO, protein structure), *in vitro* (cell lines, organ systems)
- To be interpreted as WoE
- To be introduced to address specific questions, supplement
- Lack of databases, lack of validation and at-hand resources

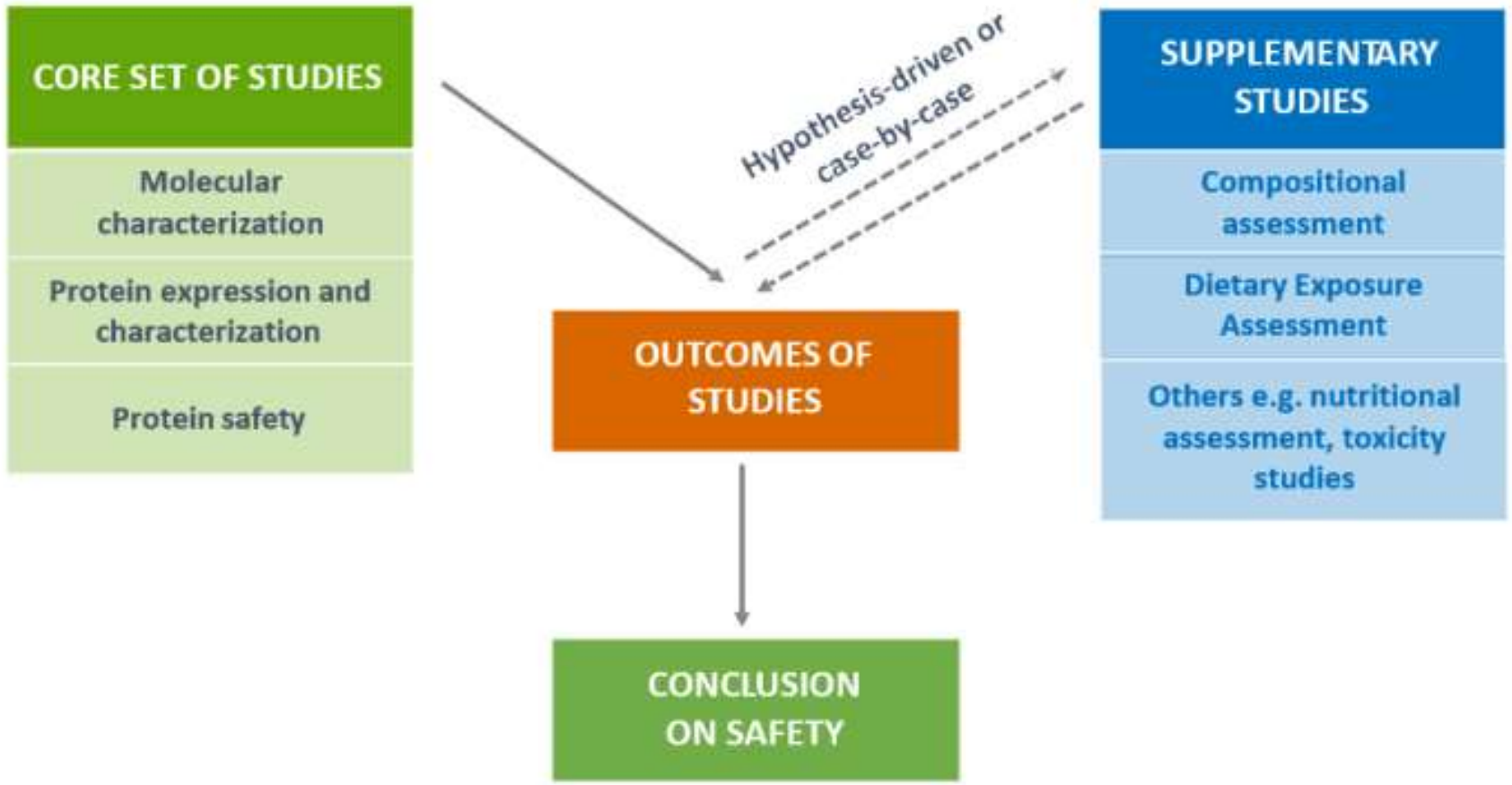
Proposed future approach

- "Core studies": protein expression and characterization, HoSU
- "Supplementary": hypothesis-driven; *in vitro,* exposure
- *In vivo* animal studies are also supplementary – only upon hypothesis

WATERS ET AL 2021



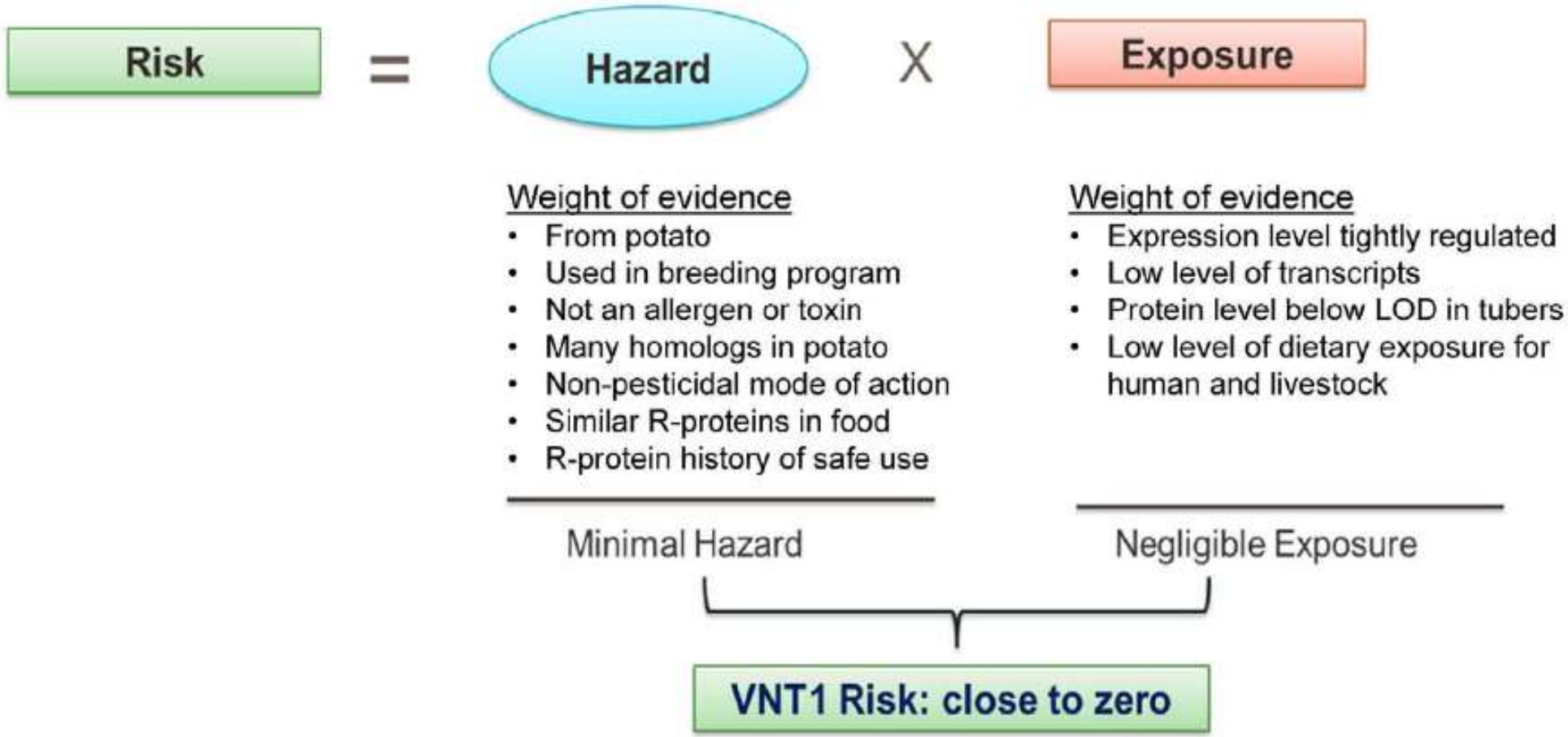
WATERS ET AL 2021 / BRUNE ET AL 2021



ROPER ET AL 2021

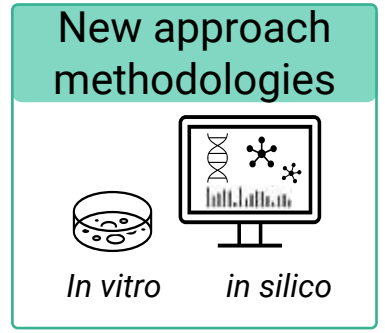
- A stepwise approach is recommended to evaluate the safety of NEPs taking the totality of information into account
- Core studies
  - HoSU of the NEP – demonstration of prior human/animal consumption or closely related proteins
  - No need for any specific toxicity or allergenicity testing in cases where both the plant and proteins expressed in the GM plant have a history of safe consumption by humans and animals – reference to EFSA guidance 2011
  - HoSU structural and/or functional similarity and exposure to other endogenous proteins
  - **The appropriate methods for establishing this similarity need to be determined on a case-by-case basis**
  - Bioinformatics results should be regarded as guiding rather than predictive
  - **Intestinal epithelial cell line monolayers from rodents and humans have been investigated** to evaluate the effects of known hazardous proteins, including ricin and PHA-E

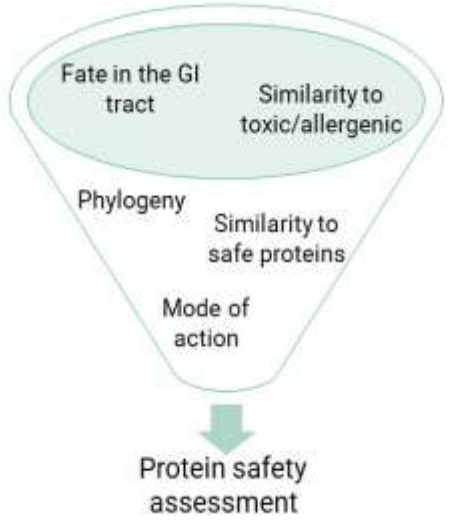
HABIG ET AL 2018



STEPWISE APPROACH IN THE PROTEIN SAFETY ASSESSMENT

- Protein vs simple chemical safety assessments
- Comparative approach as baseline–HoSU, familiarity, knowledge on proteins
- What is considered safe?
- What is considered a hazard in protein safety?
- Structural/functional similarity; but how similar is similar?
- How can evidence of consumption of a protein or source be established?
- Is there a need or possible to have additional thresholds/cut-off values (e.g. bioinformatics)? Validation?
- Is *in vitro* testing ready to be used when needed?
- How can exposure be considered in protein safety – WoE?





NEXT STEPS

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End November 2024 – mid January 2025

Adoption of the Scientific Opinion

Middle 2025

Time to act by all parties involved

EFSA Developmental projects (GMO-15 cluster) plus potential OECD workshop

PROTEIN SAFETY ASSESSMENTS

Thank you very much!!!!

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EUROPEAN FOOD SAFETY AUTHORITY

ASSESSMENT OF SOY LEGHEMOGLOBIN PRODUCED FROM GENETICALLY MODIFIED *KOMAGATAELLA PHAFFII* , UNDER REGULATION (EC) NO 1829/2003 (APPLICATION EFSA GMO NL 2019-162)

MAY 2024 – GMO NETWORK

R. Schoonjans

EUROPEAN FOOD SAFETY AUTHORITY

INTRODUCTION PRODUCT

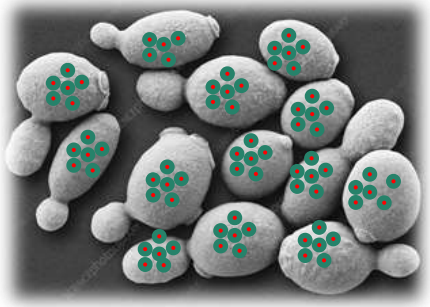
Soy — LBG2 gene

GM Pichia — Production organisms containing LBG2 gene

LegH prep — •Liquid solution

Meat Analogue — 0,8 % containing LegH prep

GM



To be marketed in Europe

ADDITIONAL FOOD ADDITIVE DOSSIER

- Preamble 12 of Reg.1829/2003: "…..on to this authorisation procedure, **food additives** containing, consisting of or produced from **GMOs should fall also within the scope of this Regulation for the safety assessment of the genetic modification**, while the final authorisation should be granted under the procedure referred to in Directive 89/107/EEC.
- *Preamble 13 of Reg. 1829/2003: Flavourings falling within the scope of Council Directive 88/388/EEC of 22 June 1988 on the approximation of the laws of the Member States relating **to flavourings** for use in foodstuffs and to source materials for their production (3) which contain, consist of or are **produced from GMOs** should also fall within the scope of this Regulation for the safety assessment of the genetic modification.*

BACKGROUND: TWO SEPARATE APPLICATIONS

Applicant: Impossible food

FAF Panel

- The **current application**, Soy leghemoglobin derived from *K. phaffii* as a new food additive
- Adopted 15 May.

GMO Panel

- A separate application has been submitted as a **GM food** (EFSA-Q-2019-00651)
- An authorisation under Reg (EC) No 1829/2003 has not yet been granted
- RA opinion to be adopted – clock stopped

SCOPE IN THE DOSSIER AND IN REG. 1829/2003

Soy → LBG2 gene

GM Pichia → Production organisms containing LBG2 gene

LegH prep → •Liquid solution containing rDNA

Meat Analogue → 0,8 % containing LegH prep

1. This Section shall apply to:

(a) GMOs for food use;

(b) food containing or consisting of GMOs;

(c) food produced from or containing ingredients produced from GMOs.

4. The authorisation referred to in paragraph 2 may cover:

(a) a GMO and foods containing or consisting of that GMO as well as foods produced from or containing ingredients produced from that GMO; or

(b) food produced from a GMO as well as foods produced from or containing that food;

(c) an ingredient produced from a GMO as well as food containing that ingredient.

GMM Guidance 2011

Scope Applicant: This application is submitted to gain **authorisation for the use of** soy leghemoglobin (the liquid preparation is referred to as "**LegH Prep**") produced from genetically modified *Pichia pastoris* (*P. pastoris*) **as a flavouring** ("meaty taste") in meat analogue products that will be marketed in the European Union (EU).

RECENT STEPS IN THE GMO DOSSIER (2024)

- January: RA Additional Information 8 pending
- February: (C)RM clarifications ongoing
- February: Decision to continue with the FAF opinion as complete as possible
- March: assessment of Additional information 8
- April: draft FAF opinion went into consultations with all standing GMO Working groups
- May: continuing the GMO opinion and finalisation of Member State comments
  - RM progress notified
  - Adoption FAF opinion
- June WGs: endorsement of the FF sections of the GMO opinion, MC text has been endorsed pending one verification needed

BACKGROUND: COOPERATION BETWEEN FAF AND GMO PANELS

EFSA FAF and GMO Panels established a cooperation to ensure **consistency** and avoid duplication of work

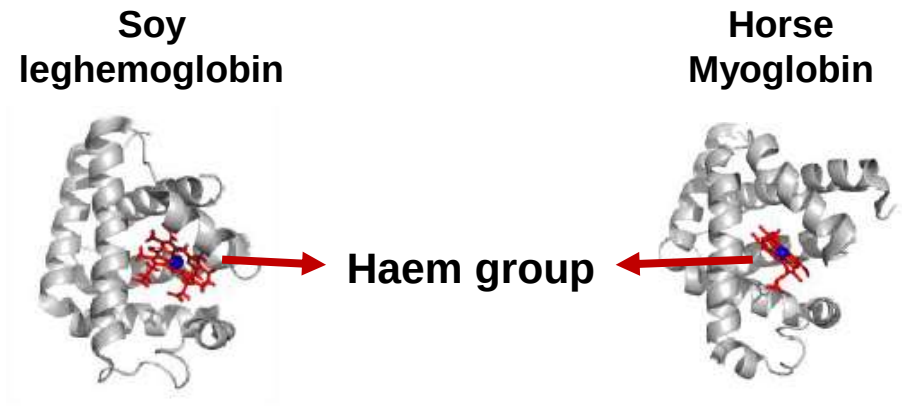
Consultation with the GMO Panel WGs

The Opinion by the GMO Panel on the genetic modification will be finalized **later this year**

A final conclusion on the safety of the GM by the GMO Panel was not be available at the time of the adoption by FAF Panel (May 2024)

FAF OPINION

- Functional component and function in food
- Manufacturing/fermentation process details
- Sections on the production organism and absence of viable cells
  - In line with CEP opinions on enzymes and in line with the information in the FAF dossier
- Product specifications proposed to be linked to final production strain MXY0541
- Exposure estimations in detail
  - Max use level for soy leghemoglobin protein in food set at 0.8% (8000 mg/kg), to be similar to myoglobin content in beef (0.8-1.8%).



Appearance of plant-based burgers without and with LegH Prep

No LegH **Added LegH**

FAF OPINION

- Biological data according to FA legal framework
  - ADME / Fate in the organism: digestion studies
  - Toxicology based on *in vitro* and *in vivo* studies
    - Incl. Subchronic Toxicity Studies and genotoxicity
    - Assessment of a 90-day study drafted together
  - Allergenicity section taken over from the GMO Opinion + finetuned together
    - The product will be labelled “Contains soy” (according to Regulation (EU) No 1169/2011) on the provision of food information to consumers
  - Refers to the GMO Opinion for the safety of the genetic modification

GMO OPINION – WORK ONGOING

- Molecular characterization of the genetic modification details following the GMM Guidance 2011
- Compositional analysis is referring to the FA product specifications that encompassed all details
- FF safety section refers to the FA opinion, except for
  - Nutrition (this is not needed under FA regulation)
  - The conclusion on toxicity will be in correspondence with the scope of the assessment as per pre-able 12: safety of the genetic modification
- ERA - HGT: approach RA is presence/absence of genes of concern
- Confidentiality claims to be taken into account

MC UPDATES - WORK ONGOING

- Method validation at the JRC is ongoing.
- Bioinformatics are updated.

FOOD AND FEED - UPDATES - – WORK ONGOING

- **Production process**
  - Referring to FAF + clarification request on antifoam
- **Product preparation and description**
  - No viable cells, recombinant DNA present, MoA, composition
- **Toxicity (in finalisation)**: NEP + other constituents
  - Additional information on the method and quality checks used for diet preparation
  - Refer to the FAF opinion for the *in vivo* studies
- **Allergenicity**: Section finished, also endorsed by FAF WG and transferred into the FAF opinion
- **Dietary exposure** only for humans; no animal exposure: referring to FAF opinion
  - Part not linked to the genetic modification but linked to the meat analogue comprising the food additive is in the remit of the FAF Panel
- **Nutrition**:
  - Minerals, Heam - Fe upper levels – cooperation with and referring to FAF opinion.

ERA – NO UPDATES

- HGT: approach to RA is no hazard, no risk

LITERATURE SEARCH - UPDATE

Open question in ADR8:

Under the original dossier section C.4.2 Literature Search, comprehensive literature searches were performed to identify publications relevant to the safety of the soy leghemoglobin and P. pastoris. Three files were provided, covering dates from 2017-2018.

The applicant is requested to provide an updated literature search on soy leghemoglobin and production strain, covering the period till now. The updated literature search should comply with the recommendations outlined in EFSA GMO Panel Guidance on microorganisms, 2011 and in line with the EFSA ANS Panel Guidance on the submission for food additives, 2012.

Info received and inspected.


THANK YOU

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