Safety of GM Crops

Bayer Russia Biotechnology Conference

July 2023

Regulatory Agencies assess the safety of the intended and unintended changes in a GMO

- ✓**Is the food / feed safe for humans and animals to consume?**
- ✓**Are the plants safe for the environment?**

> "…a reasonable certainty that no harm will result from intended uses under the anticipated conditions of consumption…"
> *OECD , 1993*

. . . "as safe as" . . .

Approach to Safety Assessment

Underlying assumptions:

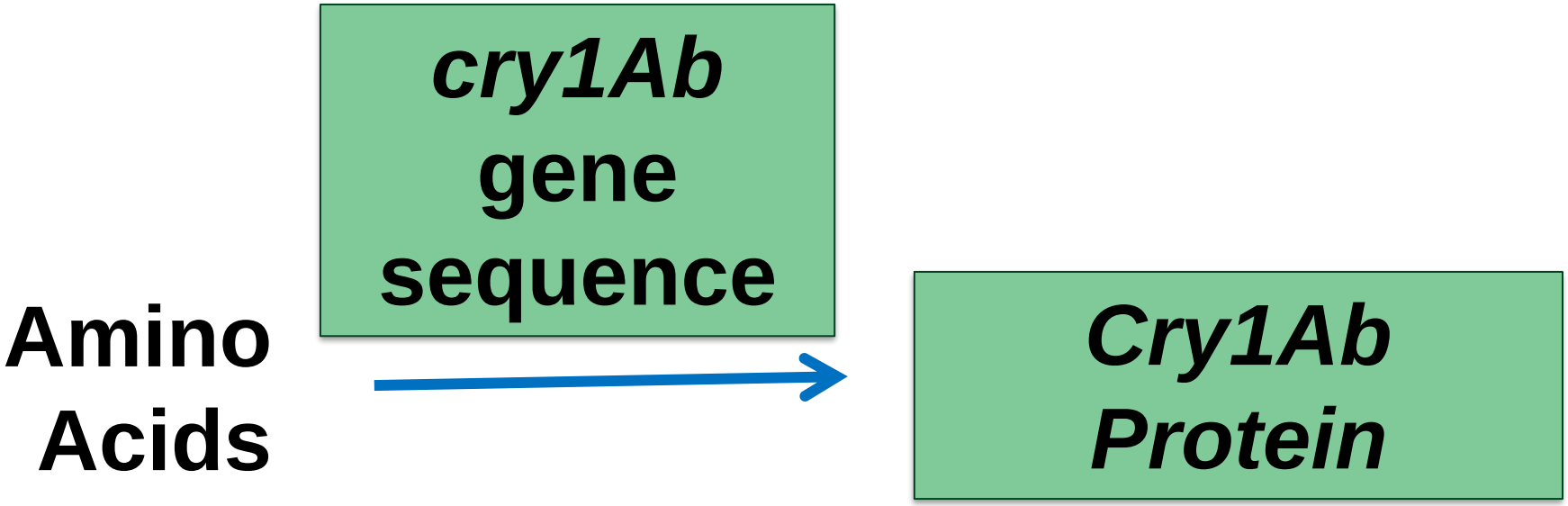
1. Traditionally cultivated crops have gained a history of safe use for the environment, animals, and consumers.

2. These crops serve as a baseline for the environmental and food/feed safety assessment of GM crops.

Comparative Safety Process:

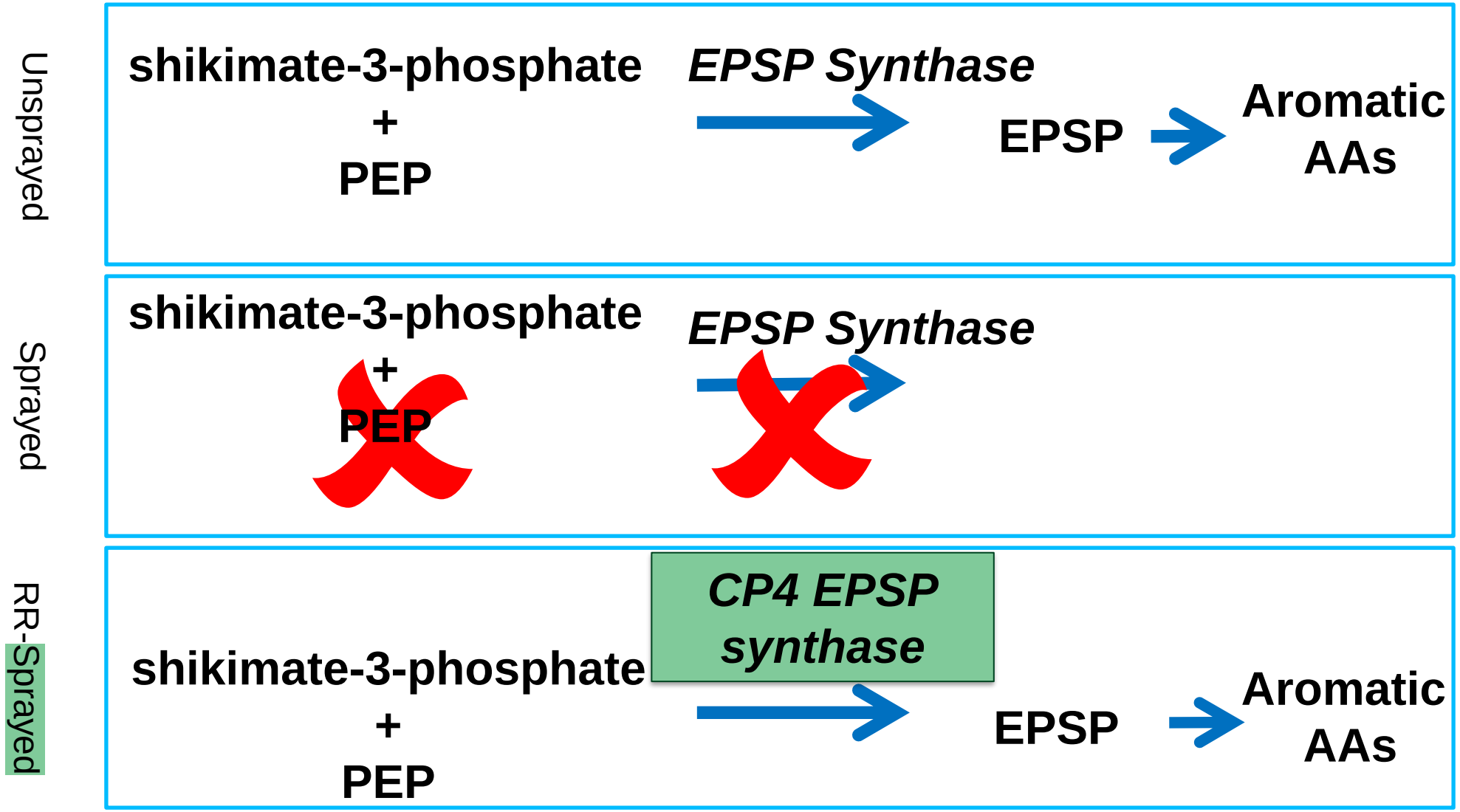
Looks at newly introduced components (DNA, Protein) and assesses any safety impacts of intended or unintended changes in composition.

Example #1. Bt Protein for Insect Resistance

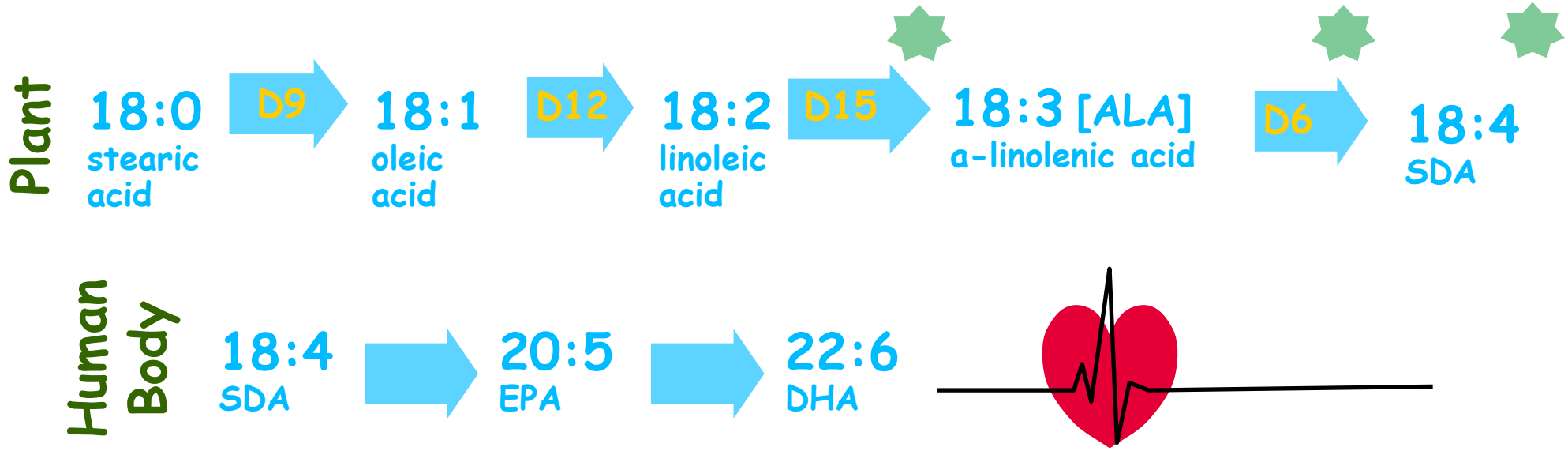


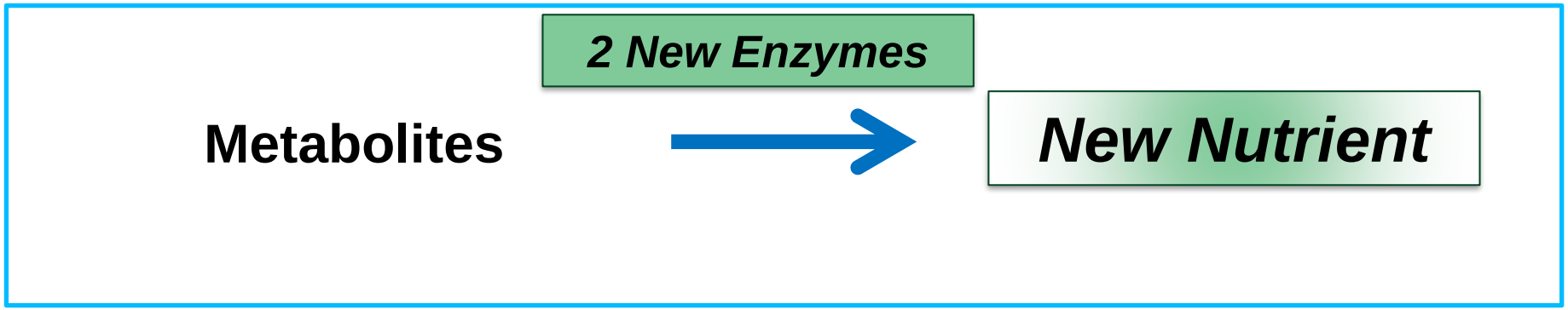
= Different

Example #2. Roundup Ready™ Herbicide Tolerance



Example #3. SDA Soy (VLC ω-3 oil that when part of a diet low in saturated fat and cholesterol, helps maintain a healthy heart)





= Different

A Rigorous, Integrated Process is Used to Assess Safety of All Elements of the Biotech Crop/Food

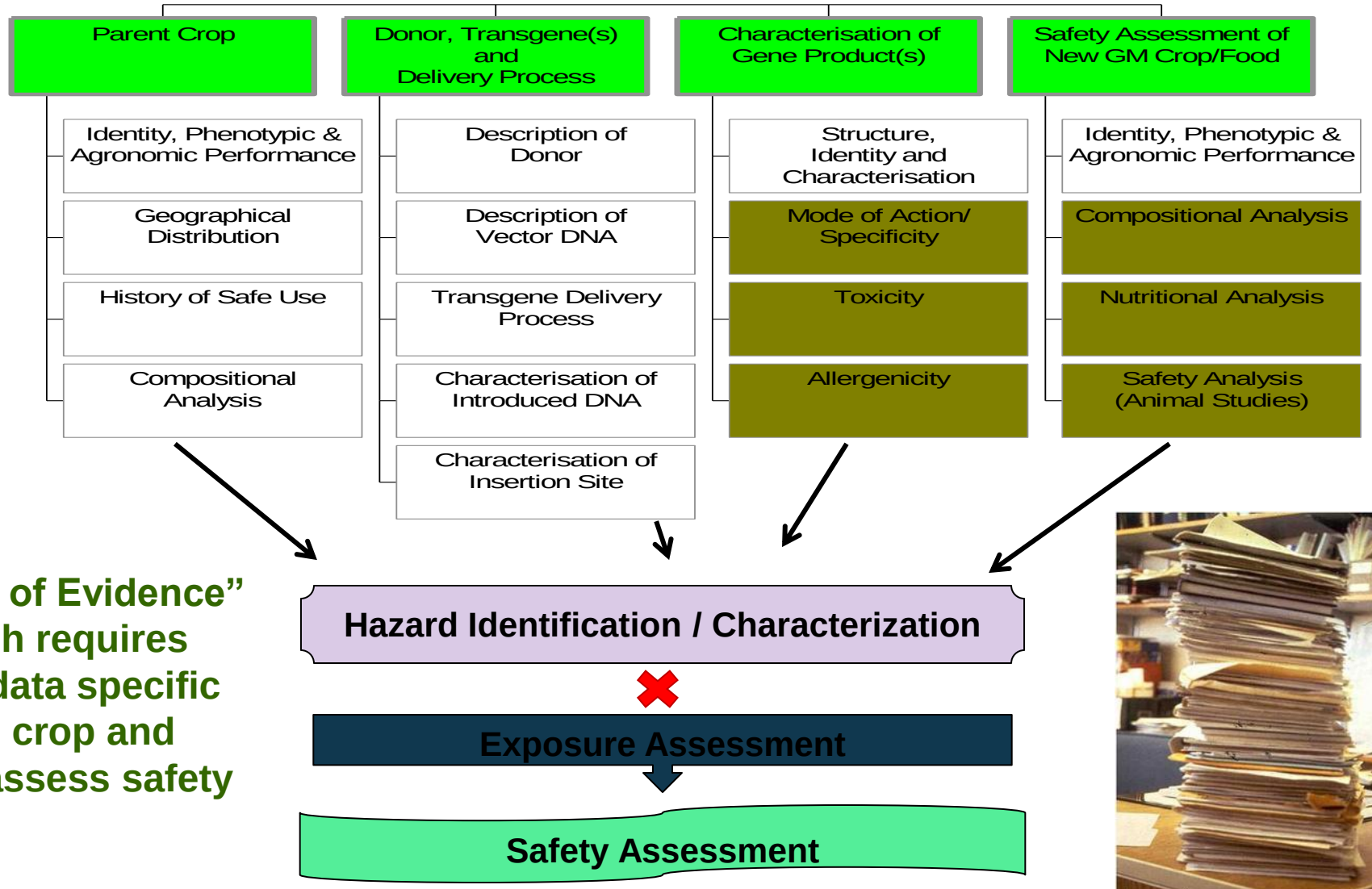


Chart from: Konig et al. (2004).

"Weight of Evidence" approach requires critical data specific for each crop and trait to assess safety

Overall Safety Assessment Approach for Biotech Crops

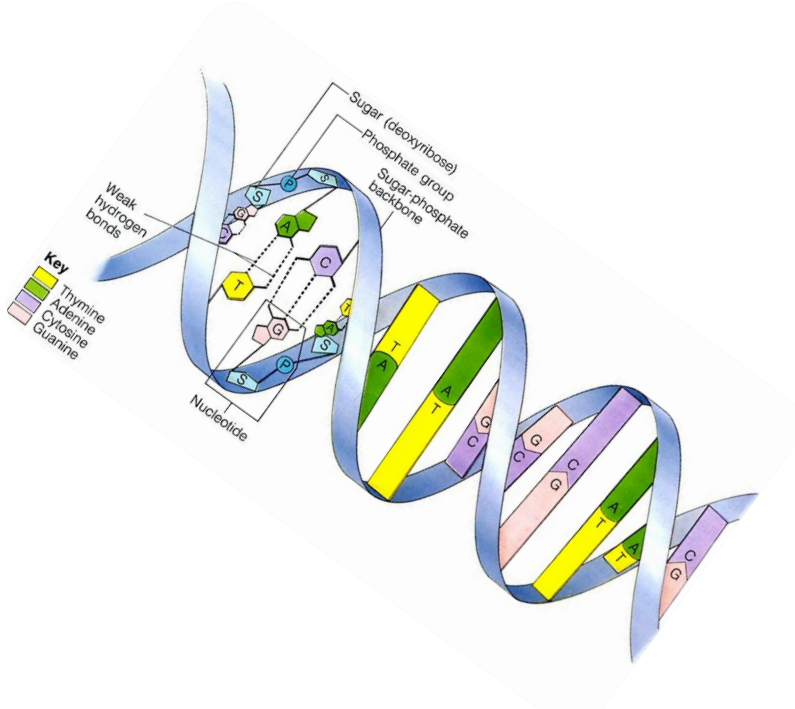
Weight of Evidence

- **Gene(s)**
  - Molecular characterization of inserted DNA
- **Gene Product - Introduced Protein(s)**

  (non-coding RNA for RNAi products)
  - History of safe consumption & source
  - Function / specificity / mode-of-action
  - Levels of introduced protein (non-coding RNA) (exposure)
  - Toxicology / allergenicity
- **Crop Characteristics**
  - Agronomic & Phenotypic
  - Interactions with biotic and abiotic factors
- **Food / Feed Composition**
  - Proximate analysis
  - Key nutrients & anti-nutrients
- **Feeding studies**
  - Livestock nutrition / Performance (e.g. broiler)
  - Subchronic toxicity (90 day rat)

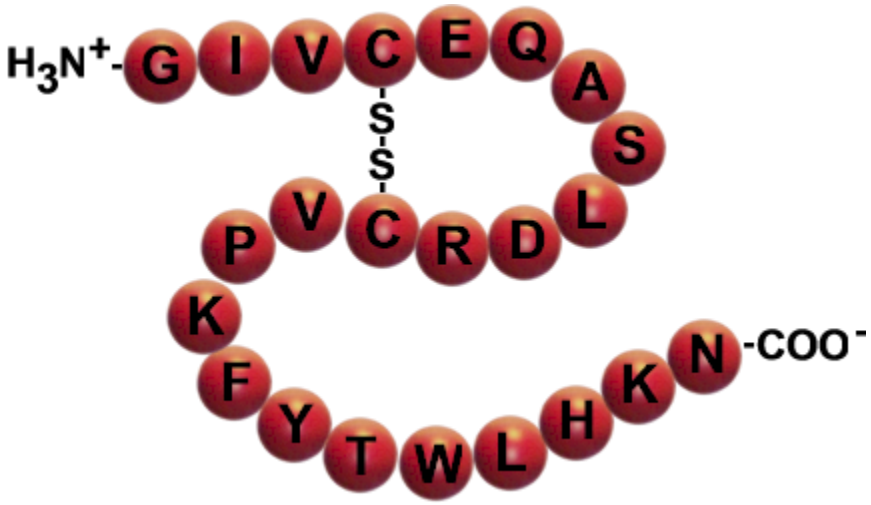
Approach To Food / Feed Safety -- DNA

- **Selection from ≈ 20,000 plants. Screening for unintended phenotypes (same process is done for conventional breeding).**
- **Gene(s)**
  - ✓ **No new nucleotides (As, Cs, Ts and Gs) or structure**
  - ✓ **A new gene sequence is on average 0.0003% of the DNA**
  - ✓ **Source(s)**
  - ✓ **Molecular characterization**
  - ✓ **Insert / copy number / gene integrity**



- The average person in the US consumes 22 mg/d of nucleic acids.
- Of that, 0.0002% is from a transgene.

Approach To Food / Feed Safety -- Protein



- History of safe use and consumption
- Function / mode-of-action
- Allergenicity testing

  Source organism

  Bioinformatics (41M comparisons)

  Digestive fate

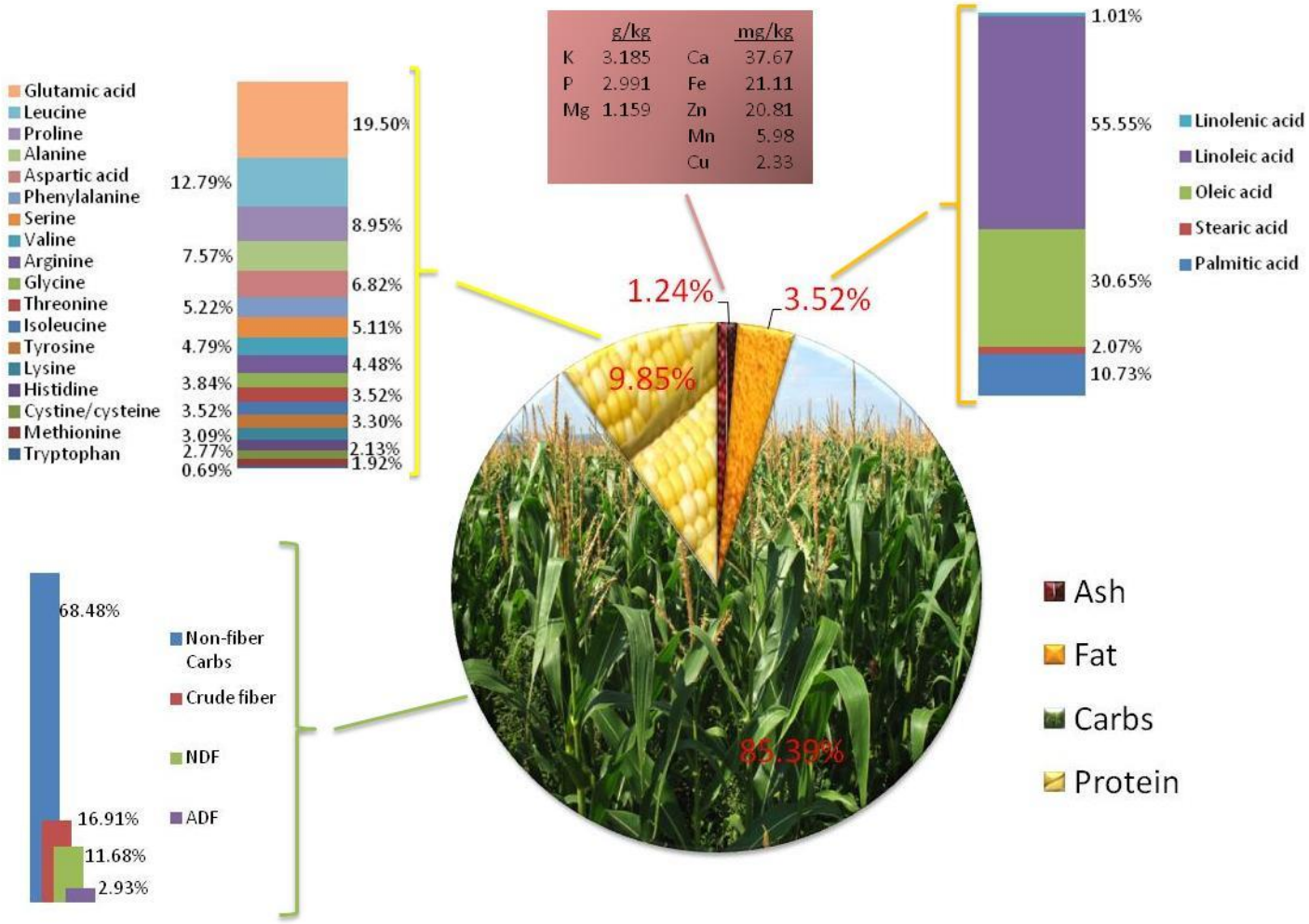
  IgE binding studies (if necessary)
- Toxicity

  Bioinformatics

  Acute Tox

  Other tox studies (Chronic studies generally NOT indicated for DNA, RNA or dietary proteins but MAY be indicated for changes in composition)

Comprehensive field study shows that crop composition is not changed compared to the control

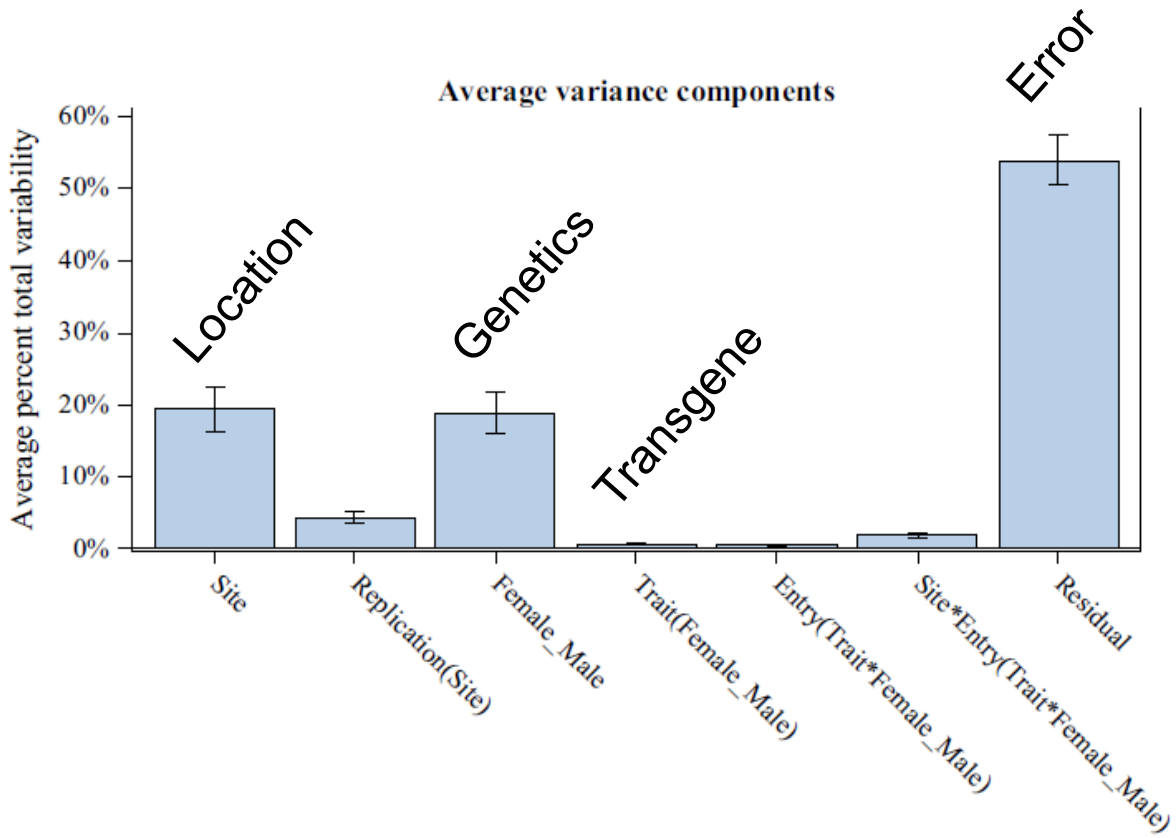


Study Design

- Large replicated plot, multi-site field trial
- 60 – 80 analytes defined by OECD
  - Proximate analysis
  - Key nutrients
- Key anti-nutrients

Composition Differences Mostly Due to Environment and Genetics

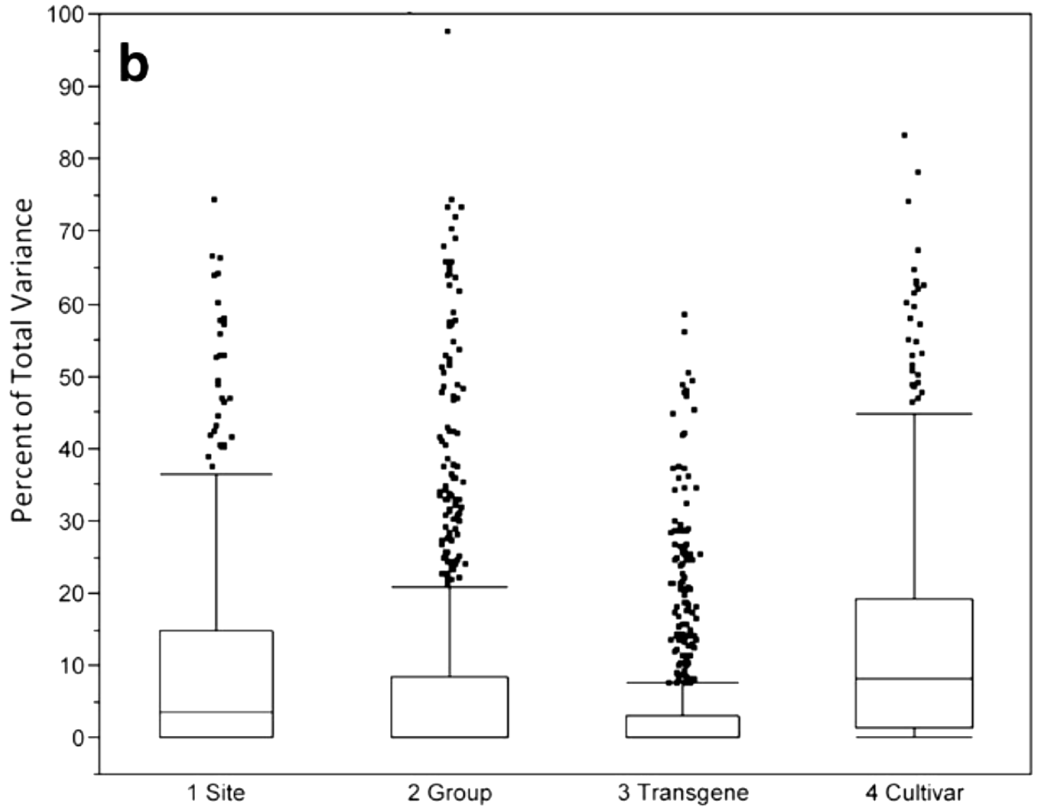
- GM traits:
  - Drought tolerance
  - Herbicide resistance
  - Insect protection
- Sites (2012 growing season):
  - Boone County, IA
  - Sangamon County, IL
  - McClean County, IL
- Composition:
  - Protein, Oil, Starch, AAs, FAs, minerals, tocopherol



Venkatesh et al., 2015. Soc Expt Bio 13:200

Genetics (yield) and environment are the biggest factors in metabolite profiling

- 9 soybean varieties
- Two lineages (Group)
  - 4 earlier lower-yielding varieties
  - 5 later higher-yielding varieties
- 2 locations in IL (50 miles apart)
- GM vs non-GM



Kusano et al. (2015)
Metabolomics 11:261–270

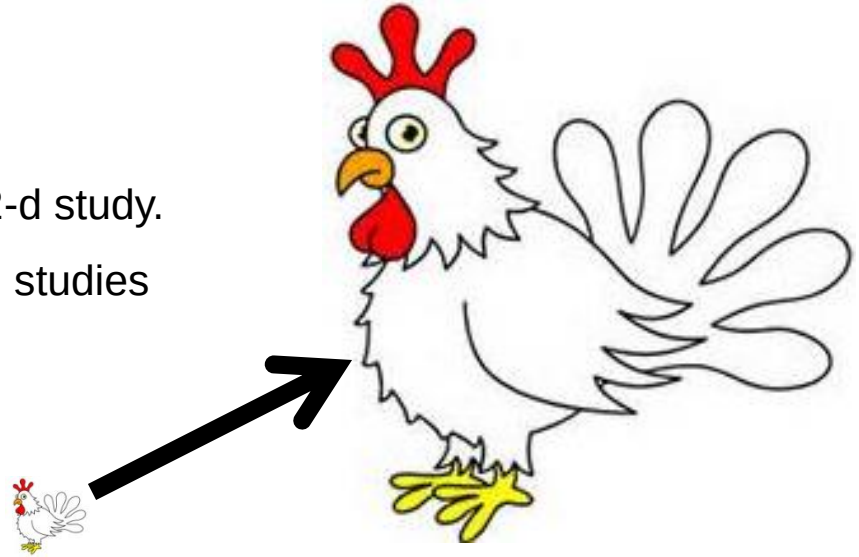
Approach To Food / Feed Safety -- Animal Studies

Toxicology Studies

- Protein/Trait
  - Acute - An acute study might be appropriate since protein toxins tend to act rapidly
- Whole Food
  - At the early stages of GM development and regulatory evaluation, 90-day sub-chronic studies were done as confirmatory; however, these sub-chronic studies became mandatory in EU. EFSA vs EC.

Domestic Animal Studies

- Whole Food
  - Broilers are typically used due to 50X gain in 42-d study.
  - Comprehensive reviews of >400 animal feeding studies conclude that GM crops are wholesome.



Transgenic Proteins are Tested for Humans

Cry1Ab protein was dosed at up to 4000 mg/kg in mice and did not produce oral toxicity (Betz et al., 2000; U.S. EPA, 2010).

A 70 kg person would have to eat 560,000 kg of maize grain to reach a consumption level that was equivalent to the highest dose of Cry1Ab tested.

= 22,000 bushels

Toxicity Studies for GM Crops

INTRODUCED TRAIT/PROTEIN

Acute toxicity studies: An acute oral toxicity study is usually conducted after evidence for the safety of the newly expressed protein already exists. This study might be appropriate since protein toxins tend to act rapidly.

Sub-acute toxicity studies: A 28-day ("sub-acute") repeat dose oral toxicity study may be conducted on a case-by-case basis when there is insufficient history of safe use (HOSU) for the protein. An absence of systemic exposure makes it unlikely that repeated dosing will result in cumulative toxicity.

WHOLE FOOD

Sub-chronic toxicity studies: A 90-day ("sub-chronic") repeat dose oral toxicity study may be conducted on a case-by-case basis when there are biologically meaningful differences between the GM and and its conventional counterpart. Early on 90-day sub-chronic studies were done as confirmatory studies; however, these studies became mandatory due to political pressure.

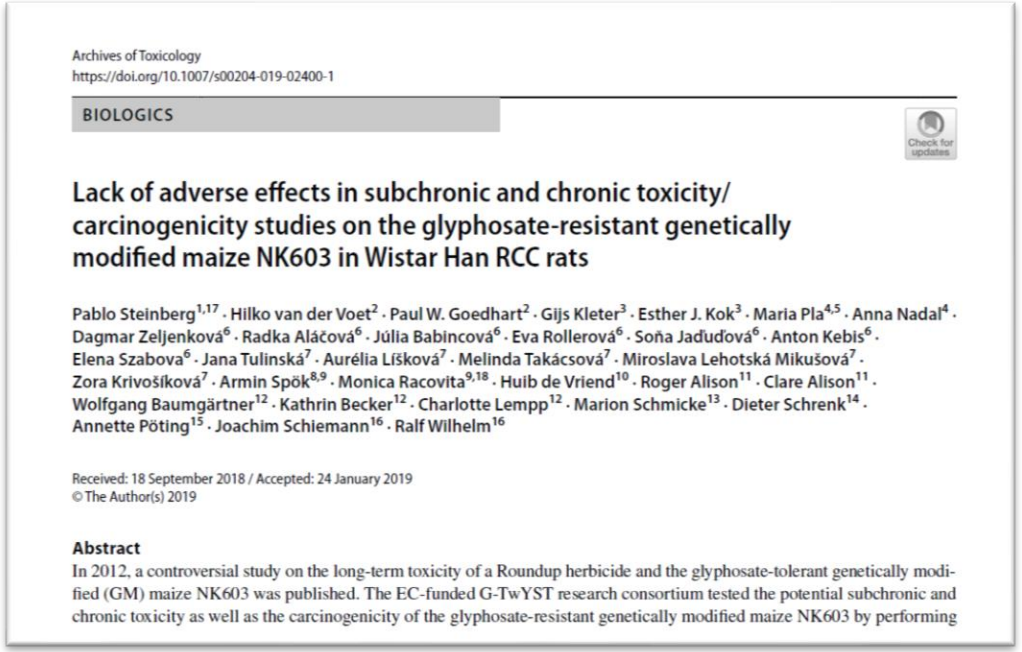
EU-funded tox projects have confirmed no health concerns from GMO crops.

GMO Risk Assessment and Communication of Evidence (GRACE): lasted 3 years, cost €7.8M and consisted of 90-d and 1-yr studies. They concluded…

- *"MON 810 maize has not induced any adverse effect…"*
- *"… did not provide relevant additional information when compared to the 90-day studies."*

Genetically modified plants Two Year Safety Testing (G-TwYST): lasted 4 yr, cost €3.8M and included a 90-day, sub-chronic tox study, a 1-yr study for chronic tox and a 2-yr carcinogenicity study.

- *"It is concluded that no adverse effects related to the feeding of the NK603 maize cultivated with or without Roundup for up to 2 years were observed."*



Archives of Toxicology
https://doi.org/10.1007/s00204-019-02400-1

BIOLOGICS

Check for updates

Lack of adverse effects in subchronic and chronic toxicity/carcinogenicity studies on the glyphosate-resistant genetically modified maize NK603 in Wistar Han RCC rats

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Abstract
In 2012, a controversial study on the long-term toxicity of a Roundup herbicide and the glyphosate-tolerant genetically modified (GM) maize NK603 was published. The EC-funded G-TwYST research consortium tested the potential subchronic and chronic toxicity as well as the carcinogenicity of the glyphosate-resistant genetically modified maize NK603 by performing

GMO90+: lasted 3 years, cost €2.5M was aimed at determining if feeding rats GM maize results in metabolic changes that could be linked to early effect biomarkers. The results have confirmed those of the GRACE project.

Science vs Politics

2010
"The use of animals for scientific studies should have a clear objective and be in line with the "3Rs principle" (Replace, Refine and Reduce)."

EC Directive 2010/63/EU

2011
If the GM plant and derived food and feed have been assessed as compositionally not different from its comparator except for the introduced trait(s), no further studies to demonstrate nutritional equivalence are required.

EFSA Journal 2011; 9(5):2150

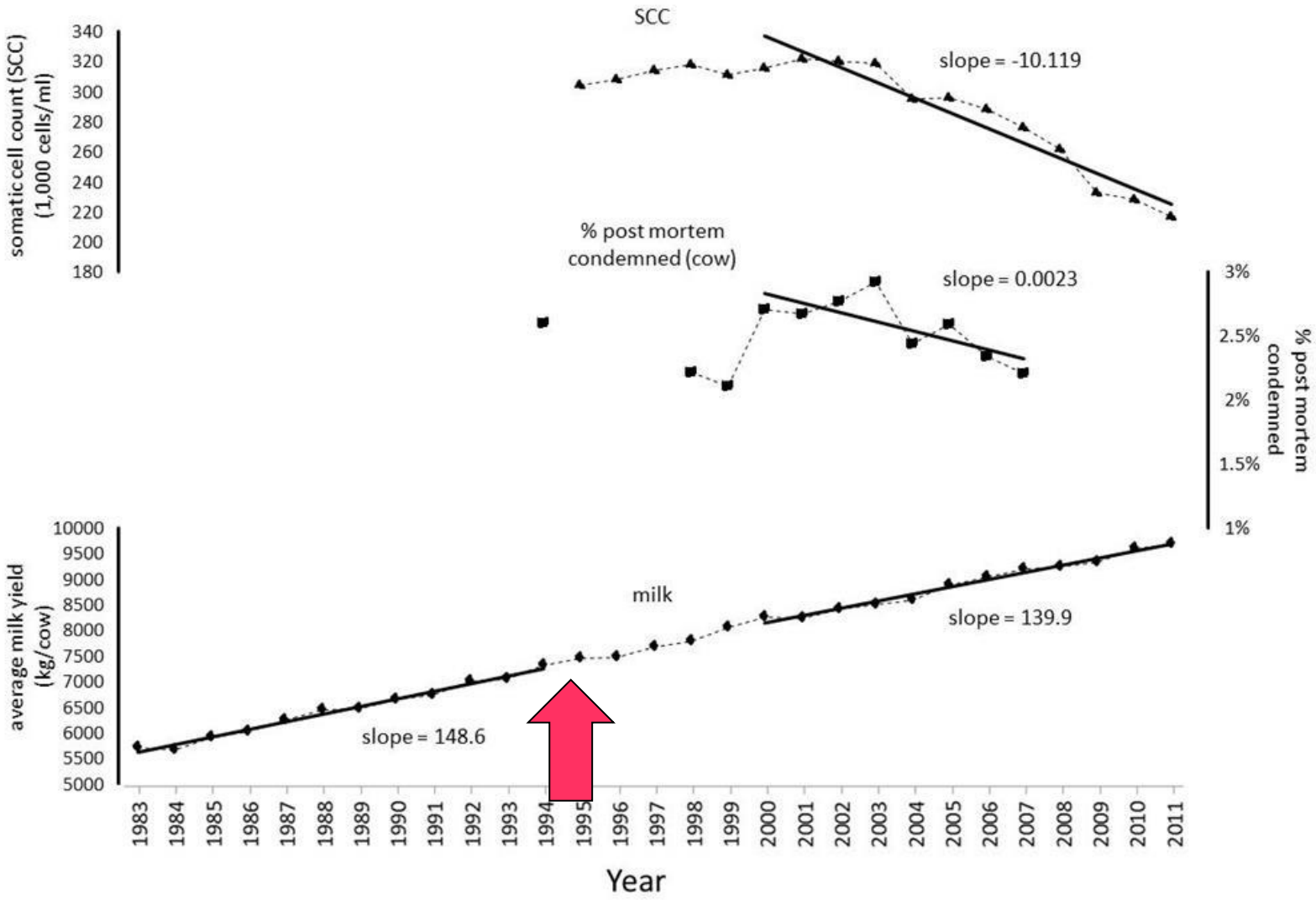
2013
The applicant shall include a 90-day feeding study with whole food and feed in rodents for the assessment of food and feed containing, consisting of or produced from genetically modified

COMMISSION IMPLEMENTING REGULATION (EU) No 503/2013 of 3 April 2013

2018
This series of studies neither delivered a scientific basis for the 90-day animal feeding trial demanded by the European Commission… nor did it indicate that untargeted, extended feeding studies with rats fed GM plant material are of value for a final confirmation of safety.

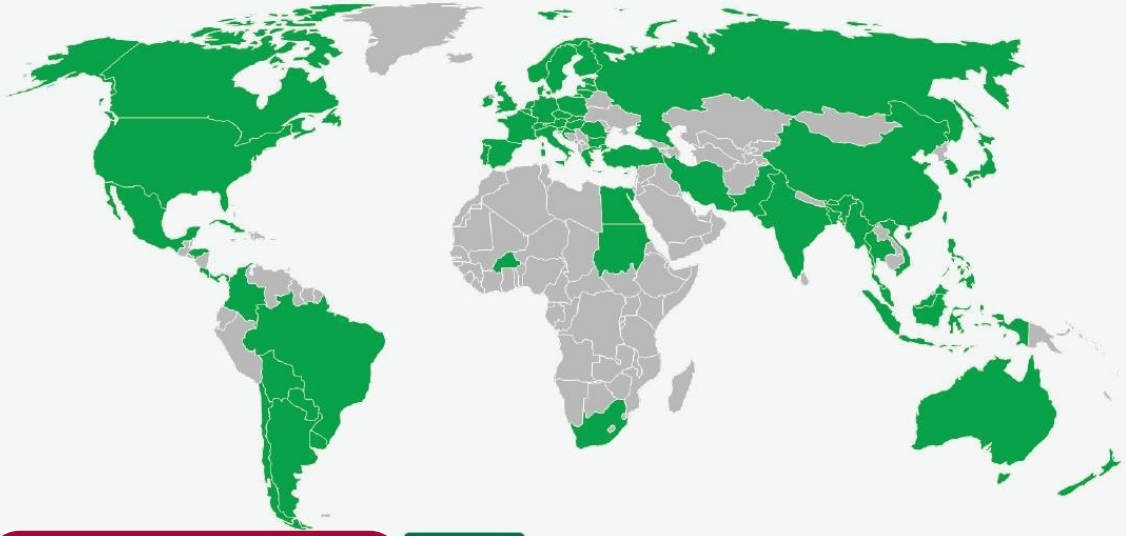
G-TwYST (Archives of Toxicology Jan 2019)

Milk production statistics and somatic cell counts in US prior to and subsequent to the introduction of GE crops in 1996



Van Eenennaam, A. L. and A. E. Young. 2014 . Journal of Animal Science.

STATUS OF APPROVED EVENTS FOR BIOTECH CROPS USED IN FOOD, FEED, PROCESSING, AND CULTIVATION



67 COUNTRIES ISSUED

4,133 REGULATORY APPROVALS
FOR **26** GM CROPS SINCE 1992

1,995 FOOD USE

1,338 FEED USE

800 CULTIVATION

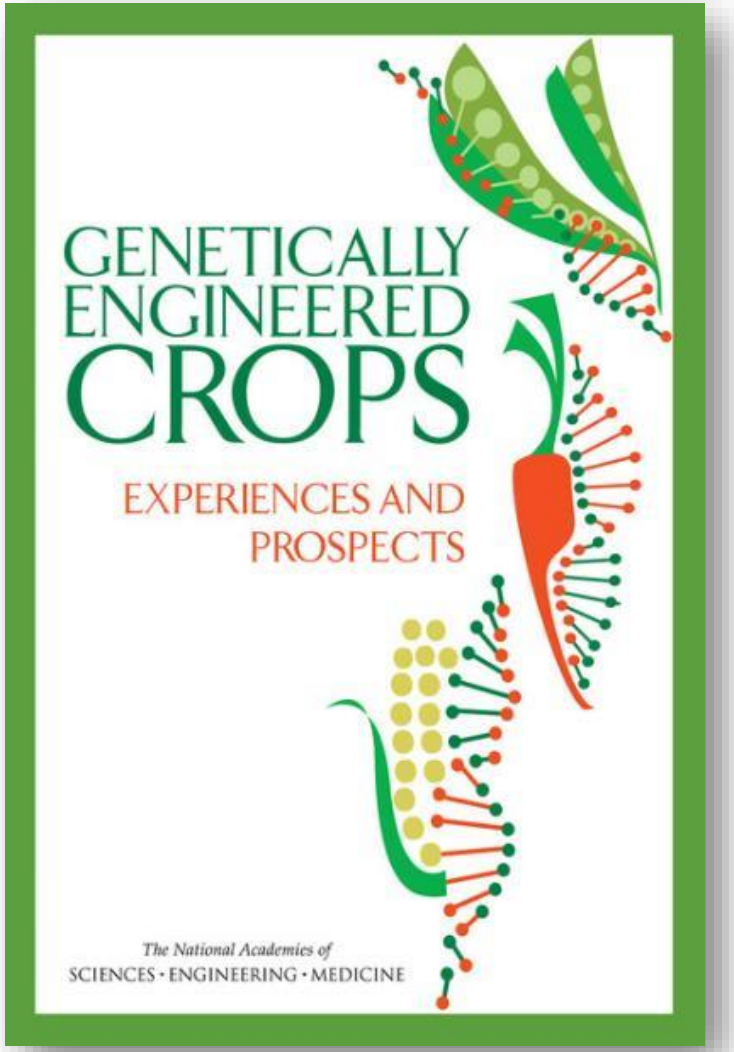
JAPAN HAS MOST NUMBER OF APPROVALS
646 APPROVALS

MAIZE HAS LARGEST NUMBER OF APPROVED EVENTS
232 APPROVED EVENTS IN **30** COUNTRIES

HERBICIDE TOLERANT MAIZE EVENT
NK603 HAS MOST APPROVALS
55 APPROVALS IN **26** COUNTRIES

Source: ISAAA, 2017

The U.S. National Academies of Science conducted a comprehensive review of GM crops…

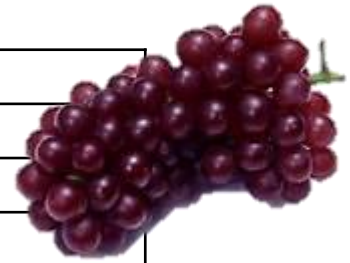


… concluded that there was no evidence of a risk to human health from GM crops compared to conventional crops (NAS, 2016)

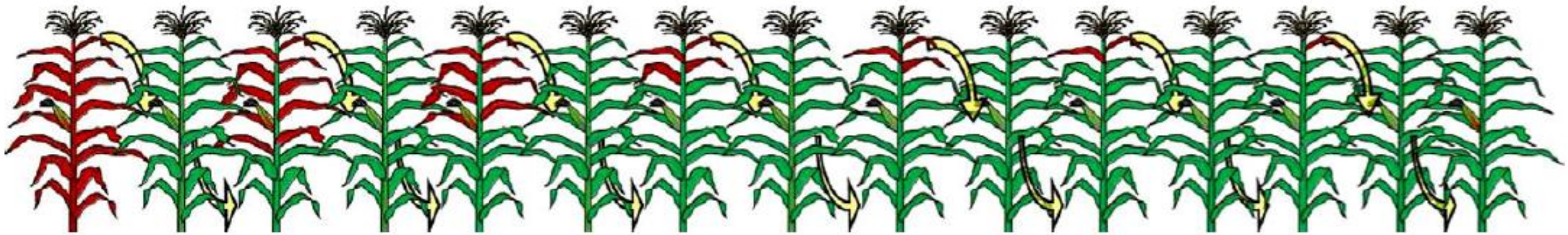
Downlaod Free PDF: https://www.nap.edu/catalog/23395/genetically-engineered-crops-experiences-and-prospects

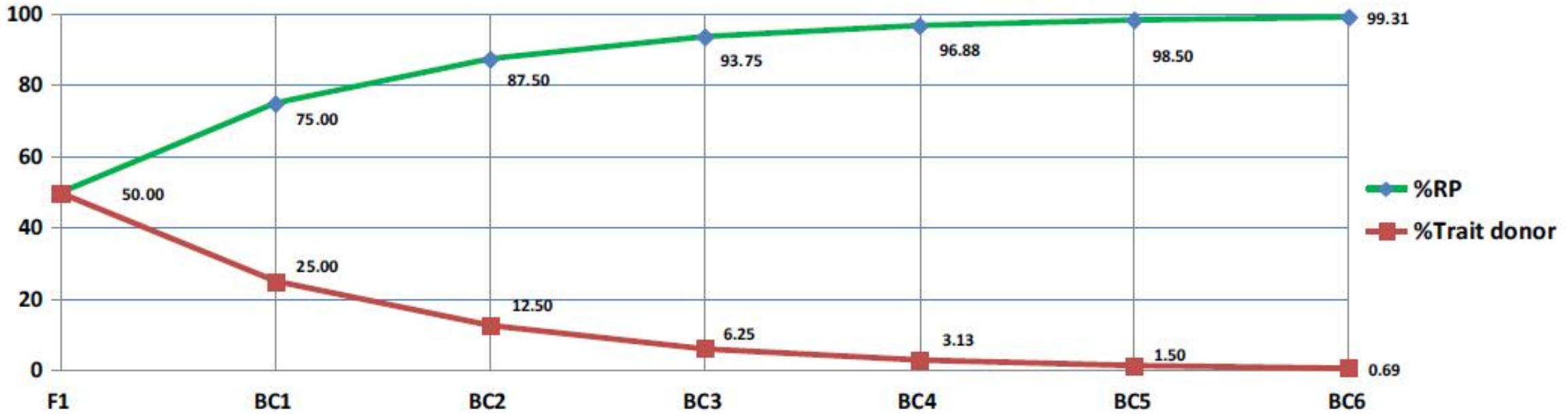
Naturally-occurring genetic changes common in plants

Genetic Change	Resulting Characteristic
Transposable elements (transposons)	Various colors of **grapes, grapefruit, maize**
	> 22K insertions detected in single variety of **soybean**
	> 50 new inserts of a transposon per **rice** plant per generation
	Extreme large size of modern **tomatoes** compared to ancestors
	Round or wrinkled **peas** (Mendel)
Mitochondrial DNA in nuclear DNA	Gain and loss of mtDNA common to **maize** inbred lines
Natural transgenic crops	Expression of several bacterial genes in **sweet potatoes**
Mating with wild relatives	>60 wild relatives have been used for >100 characteristics (80% involve pest or disease resistance) in **13 crops**
Pararetroviruses	Stable viral DNA in **rice** genome
	Stable viral DNA in **tomato** (previously also seen in potato)
Single nucleotide polymorphisms (SNPs)	**Maize** proteins (300-400 aa long) from 2 alleles differ by 3-4 aa
	Maize genome has 55 million SNPs
	Green Revolution gene has 2 SNPs for dwarf **wheat**
	One SNP caused loss of shattering in domestic **rice**
	Tall or short **pea** plants (Mendel)
Presence/Absence/Copy number of genes	856 wild-type **soy** genes absent in cultivated varieties (and >186K DNA insertions/deletions)
	>10^6 SNPs, 30K insertion/ deletions and a few large chromosomal deletions (>18 genes) in 6 elite **maize** varieties
	Copy number variation relates to **soy** cyst nematode resistance



Backcrossing to remove unintended effects





Potential “Hazards” in Foods

agaritine

Glyco-alkaloids

TOXICANTS

tetrodotoxin

cyanide

ANTI-NUTRITIONAL FACTORS

saponins

raffinose

phytates

BIOACTIVES

caffeine

Phyto-estrogens
Polyphenols
Sterols

“CONTAMINANTS”

Mycotoxins

Heavy metals

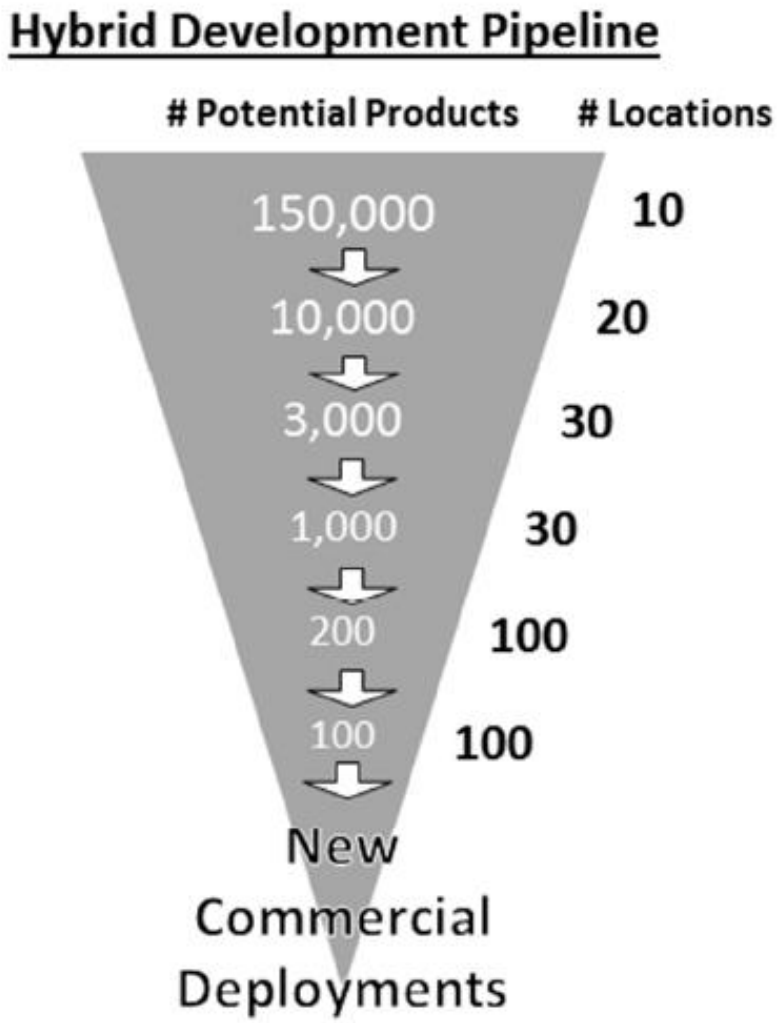
nuts

fish

ALLERGENS

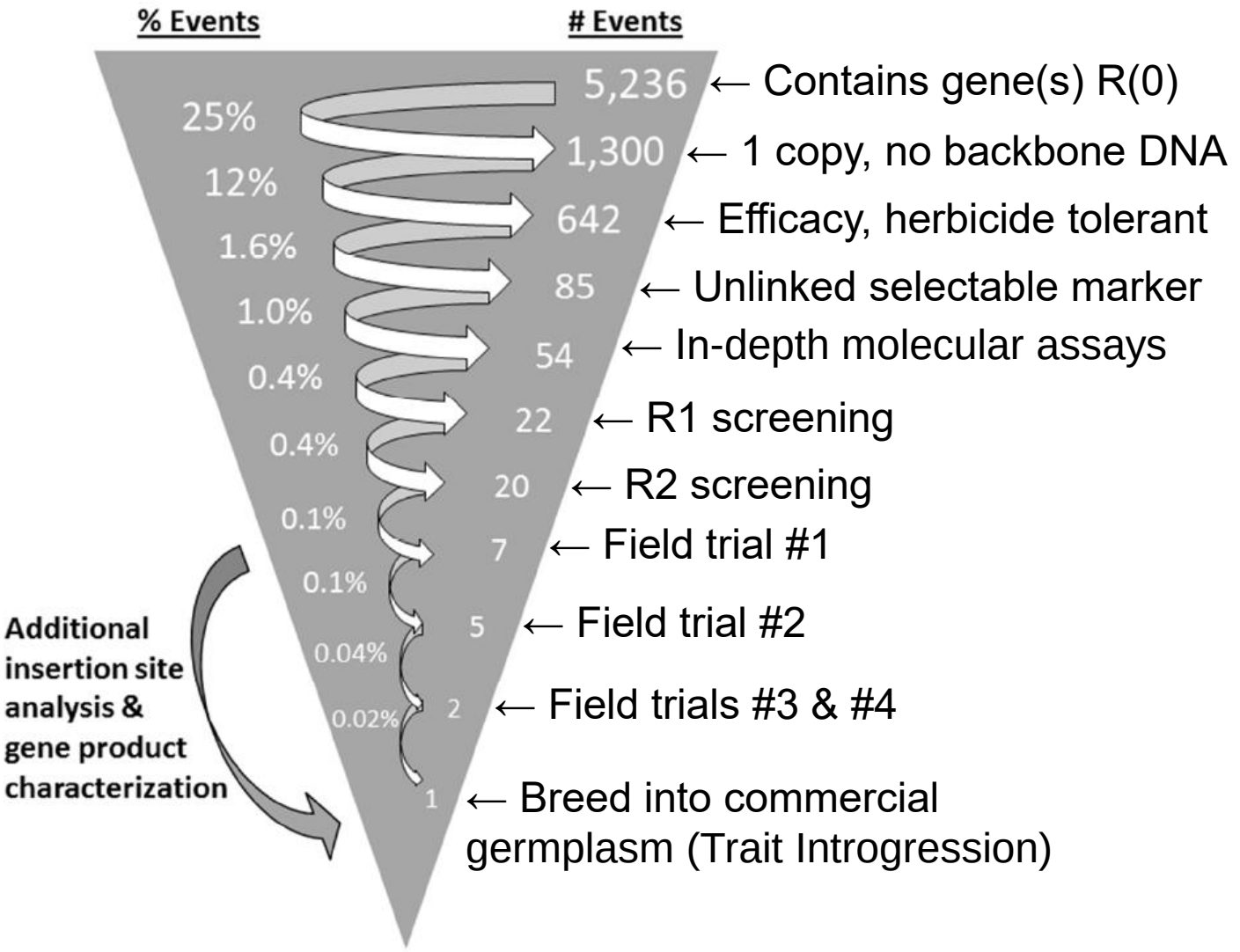
*Kaiser N, et al., (2020). Trends Food Sci Technol **100**: 51–66*

Conventional Breeding. 300,000 plants evaluated to develop new hybrids



Glenn et al., 2017

One event (0.02%) selected to reduce unintended effects



Discard off-phenotypes

Glenn et al., 2017

Commercial scale plant breeding selection practices minimize unintended changes: 300,000 plants

Event Selection

GM Event Selection

10,000

1

Unintended DNA changes tested and eliminated

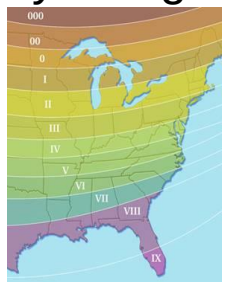
Trait integration

Breeding Desired Traits Into Elite Germplasm

99% of DNA from source varieties is bred out as high yield germplasm is selected

Commercial breeding

Breeding Seed To Grow In Many Geographies



An additional 90-99% selection of germplasm optimal for each region

Unintended genetic changes from breeding may be occuring during plant breeding

Unintended genetic changes from transformation are diminished many orders of magnitude during plant breeding

DNA Changes from Mutation Breeding

- DNA changes
  - 4 bp to 8 kb deletions
  - Inversions of up to 1.5 kb
  - Insertions ~200 bp
  - Frame-shift mutations
  - Premature stop codons
- 2543 known varieties developed from mutation breeding
  - FAO/IAEA database (http://www-infocris.iaea.org/MVD/)

Institute of Radiation Breeding
Ibaraki-ken, JAPAN
www.irb.affrc.go.jp/

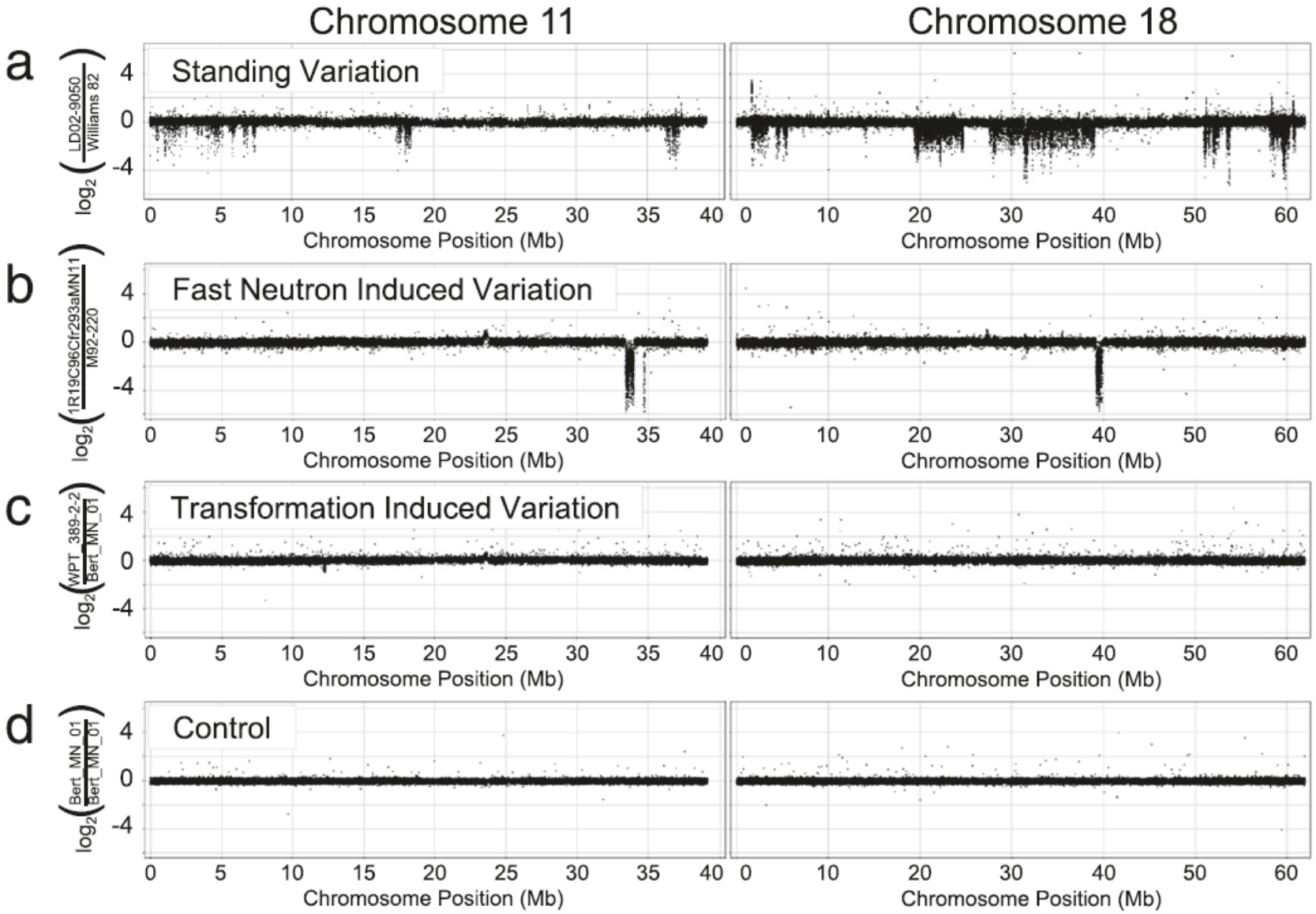
- High-yielding and short barley
- Heat tolerant and early maturing cotton
- Seedless watermelon
- Multiple disease resistances in tomato
- Ruby red grapefruit
- Gold Nijisseiki Japanese pear
- Semi dwarf rice with higher yields
- Virus resistant cocoa plants
- Canola with health fatty acid profile

Genetics > mutagenesis > transgene

Variability between cultivars

Variability from mutagenesis

Variability from Transgene



Anderson et al. BMC Biotechnology (2016) 16:41
DOI 10.1186/s12896-016-0271-z

Summary

- Genetically modified (**GM**) crops have been widely adopted by growers.
- Most commercialized GM crops have input traits that do not change their composition or nutritional value.
- Feeding GM crops to animals does not result in detection of transgenic DNA or their translated proteins in meat, milk, or eggs.
- GM crops help reduce greenhouse gases, decrease agricultural chemical use, and increase farmer incomes.
- GM crops provide better pest protection and weed control, which preserves yields, which prevents expansion of land use.

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Feel free to email any questions to me:

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