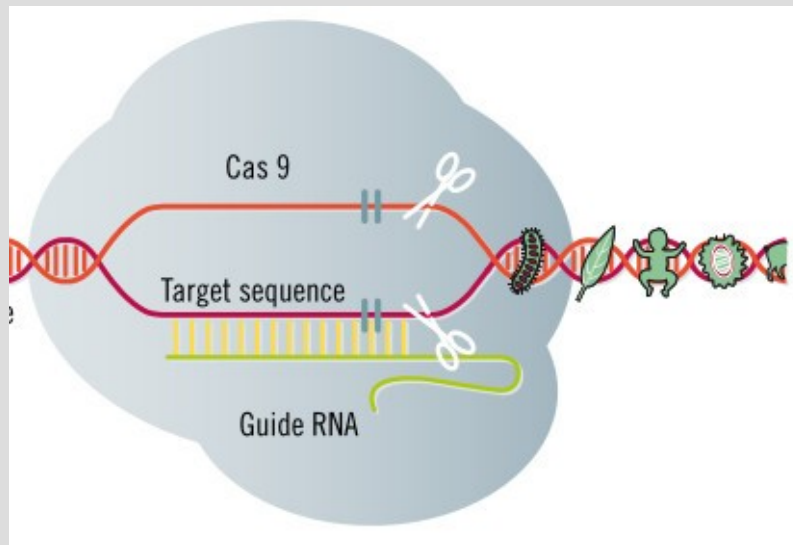


# Synthetic gene technologies & gene editing: Applications in food plants and farm animals



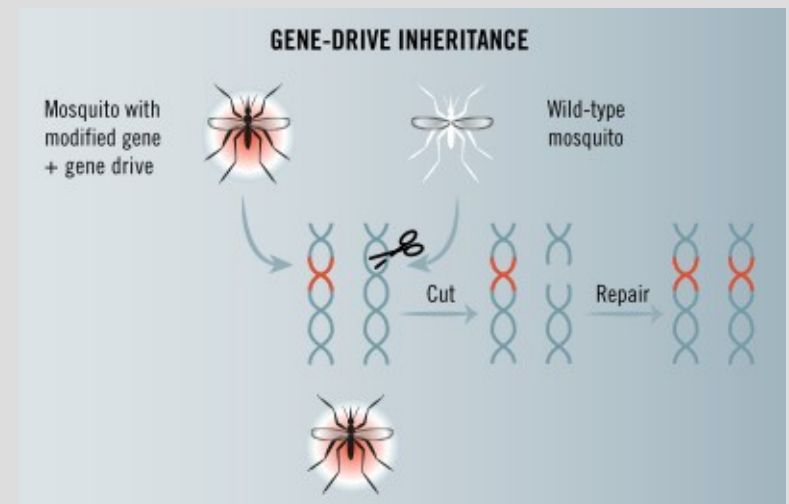
2 March, 2016

Dr. Christoph Then,  
Testbiotech e.V.  
[www.testbiotech.org](http://www.testbiotech.org)

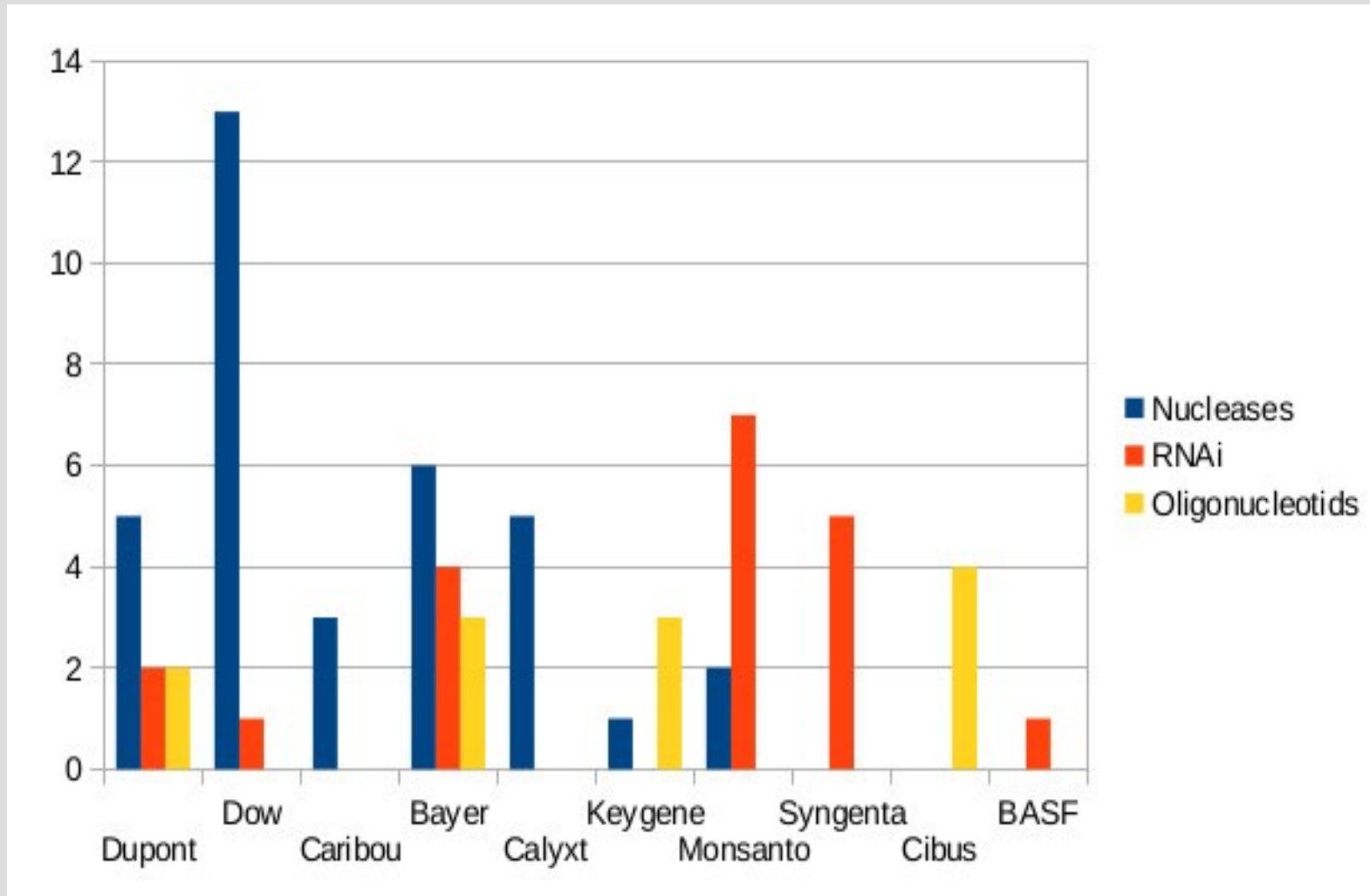
[christoph.then@testbiotech.org](mailto:christoph.then@testbiotech.org)

# Synthetic Gene Technologies ("genome editing")

- > DNA synthesis
- > nucleases such as CRISPR
- > genome editing with oligonucleotids
- > gene drives
- > manipulation of epigenetic regulation



## Companies active in genetic engineering of plants



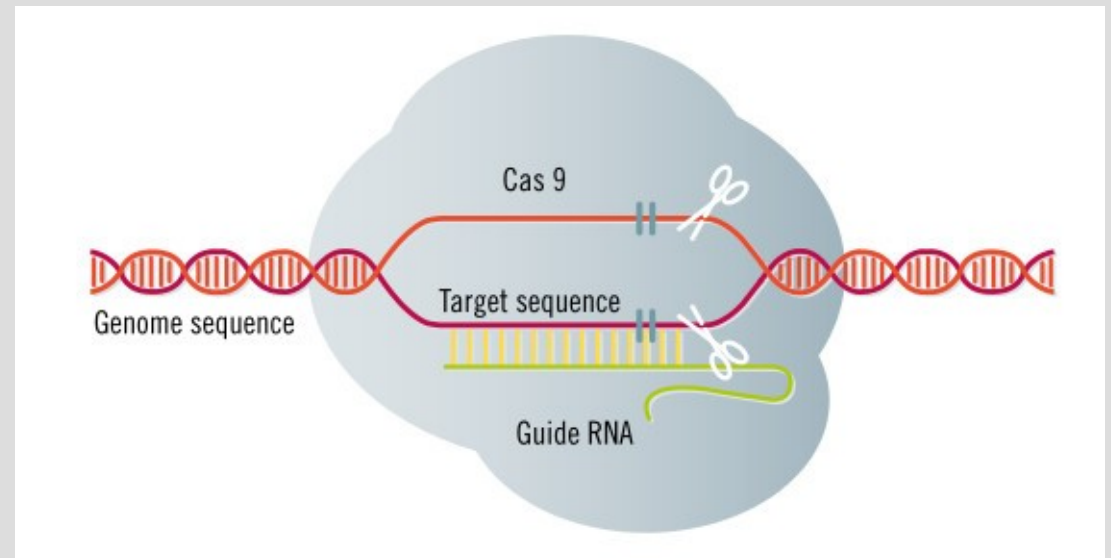
Number of WO patent applications registered at the World Intellectual Property Organisation (WIPO) between 2010 and 2015

## Examples for applications in livestock

Application	Claims
WO2012116274	Usage of nuclease TALEN to increase muscles in cows and pigs
WO2013192316	Usage of nuclease TALEN to increase muscles in cows and pigs and produce cows without horns
WO2014070887	Usage of nuclease TALEN, zinc finger or CRIPSPR to prevent animals from reaching sexual maturation, these animals cannot be used for further breeding.
WO2014110552	Hornless cows by application of methods for gene editing.
WO2014193583	Usage of nucleases to block genes for production of sperm cells.
WO2015168125	Usage of nucleases to create animals with multiple genetic changes.

# Plants: unintended effects from the process necessitate risk assessment

Li et al., 2016: DuPont produces genetically engineered soybeans with the help of CRISPR-Cas



## Plants: unintended effects from the process necessitate risk assessment

Li et al., 2016:

- In a first step the DNA sequence for the production of the nuclease was inserted by random insertion via particle bombardment.
- Consequently, the cells and plants derived from this process showed numerous unintended changes in the genome such as extra copies of randomly integrated DNA needed for producing the nuclease.

## Plants: unintended effects from the process necessitate risk assessment

Li et al., 2016:

- In a second step the enzyme Cas9 was expected to target defined regions in the genome.
- Around half of the targeted sites showed a break in the two strands, in the other half, just one strand of DNA was cut.
- The deletions observed in the targeted sites varied in length, some of them showing additional unintended insertions of DNA.
- In many cases several copies of the intended DNA construct were inserted.
- In most cases the DNA of the nuclease was unintentionally co-integrated at the targeted sites.

## Plants: unintended effects from the process necessitate risk assessment

Li et al., 2016:

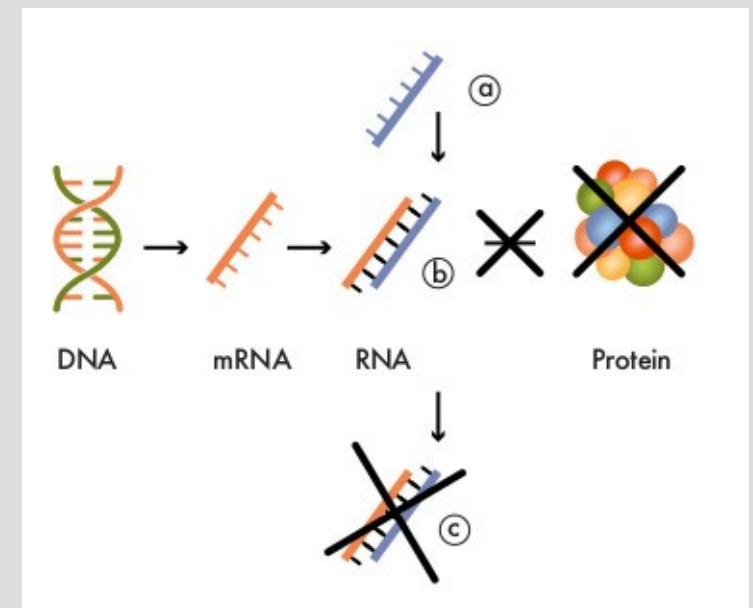
- Surprisingly, only a small number of the plant tissue derived from the genetically engineered cells contained the DNA construct as expected.
- A small number of plants was finally derived. Most of them revealed unintended changes in the structure of the DNA at the targeted site.
- These plants all contained randomly integrated extra DNA.

Finally the plants were crossed further to segregate plants that only would show intended insertions.



## Animals: health risks for animals and humans

Jabed et al. (2012) wanted to create genetically engineered cows to produce milk with a reduced content of beta-lacto-globulin (BLG). This protein is known to cause allergies. The method used was RNA interference.



## Animals: health risks for animals and humans

Jabed et al. (2012)

- In a first step, somatic cell cultures were manipulated with the DNA necessary to produce the relevant miRNA that is intended to interfere with the protein production.
- From these cells, 57 embryos were cloned, but only a single calf was born with the desired genetic information.

## Animals: health risks for animals and humans

Jabed et al. (2012)

- The resulting calf (called 'Daisy') not only showed a much lower content in beta-lacto-globulin in its milk, but also a general change in the composition of all proteins.
- Further, Daisy is not healthy and does not have a tail. The reason for this could not be determined.
- It is known that miRNA can pass the gut and enter the blood stream and could then also interfere with cell regulation at the stage of consumption. It is not unlikely that the artificial miRNA produced in the cows might also be found in the milk and can interfere with cell regulation of the consumers.

# Thank you very much for your attention !

Some recommendations:

- prohibit patents of plants and animals used in agriculture and food production;
- Consider a general ban of genetic engineering in farm animals and products thereof;
- do not allow plants and animals derived from gene-editing to escape EU regulations (such as Directive 2001/18);
- strengthen the precautionary principle;
- establish an international framework for a general ban on genetically engineered organisms that cannot be controlled in their spatio-temporal dimension.