

**Regulatory
approaches to
modern plant
breeding -
the case of
mutagenesis and
new gene editing
technologies**

Summarising conclusions

Europe's plant breeding industry and public plant science research are global leaders in developing advanced plant breeding techniques and providing competitive, high performing, and safe seeds for agriculture and food and feed production. Multiple techniques have been introduced in plant breeding over the past decades with mutagenesis being one of them. Mutagenesis techniques have a long safety record in plant breeding and will become even more helpful when applying them in the most advanced way. Often, these advanced techniques improve and refine traditional breeding methods. This is e.g. the case of new gene editing techniques in relation to mutagenesis. But Europe's leadership will be put at risk if plants generated using such new gene editing techniques would be regulated as GMOs under Directive 2001/18/EC as that would be a major disincentive for innovative research and development of advanced breeding methods, including but not limited to gene editing. It would erode the EU's scientific leadership, stifle its plant breeding innovation and negatively affect the competitive position of its agri-food chain.

New discoveries and advanced understanding of the biology, physiology and genetics of plants and their interaction with the environment, combined with new techniques in the toolbox of the plant breeder, now allow driving plant breeding faster and more targeted towards new innovations. This increase of speed and accuracy is urgently needed to offer sustainable solutions to the grand challenges of ensuring food security for a growing population, improving diets and health, safeguarding the environment and tackling climate change. Europe will fail to respond to these challenges without continuous scientific progress and adoption of advanced plant breeding techniques.

Economically, the application of the burdensome provisions of Directive 2001/18/EC would specifically hinder small and medium enterprises (SMEs) from using gene editing and most likely prevent such companies from ever commercialising resulting innovative products in the EU due to the uncertainty and high costs of the lengthy and politicised regulatory approval process and subsequent marketing conditions.

In practice, regulating plants generated by advanced gene editing as GMOs under Directive 2001/18/EC would lead to large scale requirements for the authorisation and use of these plants and subsequent food/feed products while similar or even identical plants resulting from other forms of mutagenesis are excluded from such requirements. This would be unjustifiable and disproportionate.

Furthermore, as modifications produced by gene editing are comparable to those produced by other mutagenesis techniques or to mutations spontaneously occurring in nature, detection and unambiguous identification of plants produced by gene editing are neither possible nor enforceable in case of imports produced in third countries without specific requirements. Unilateral EU requirements would thus disadvantage EU plant breeding and variety development and result in a relocation of respective activities outside of Europe, to the detriment of the competitiveness of its agri-food chain and contrary to its objective of becoming the world's leading knowledge-based bio-economy.

Mutagenesis

Mutagenesis is a process by which the genetic information of an organism is changed in a stable manner, resulting in a so-called mutation. It occurs spontaneously in nature and has been a driving force of evolution in general. It is also a main element of modern plant breeding as a result of exposure to mutagens such as certain chemicals or radiation since as early as the first half of the 20th century.

Not all mutations are beneficial for a plant. In plant breeding research, mutagenesis is a useful technique for generating mutations and genetic variation that allows the functions of genes and gene products to be examined in detail, e.g. producing proteins with improved characteristics or novel function, as well as mutant strains with useful properties. Through a thorough and extensive screening and selection process, plant breeders finally determine the plants that have acquired the desired set of altered or new useful traits through genetic mutation.

Initially, the ability of certain types of radiation and chemicals to cause mutation was exploited to generate random mutations in plants. Later, more refined innovative techniques were developed to induce specific mutations in a more targeted way.

Induced mutagenesis

Plant breeding generally uses induced mutagenesis to produce large numbers of plants with new genetic characteristics and variation as a base to develop new varieties with useful traits. Still, induced mutagenesis is a rather random process, limiting its efficiency; it requires long processes of identifying useful traits, removing unwanted traits by back-crossing, and finally introducing the desired trait(s) into elite new plant varieties.

The Mutant Variety Database (<http://mvgs.iaea.org/AboutMutantVarieties.aspx>), held by the joint FAO/IAEA program, shows that 3218 varieties are listed worldwide up to now. In the EU, the number is close to a thousand (EU Common Catalogue). These numbers probably undervalue the importance of mutagenesis as only varieties directly obtained from mutagenesis are counted while induced mutations were also introduced into further varieties by traditional crossing.

Mutagenesis derived varieties have been planted and safely consumed in over 175 plant species, including rice, maize, wheat, tomato, squash and soybean¹. There is a long history and acknowledged contribution of mutagenesis to improved crop varieties and mutagenesis is globally considered to continuously contribute to the production of safe, reliable and sustainable crops.

¹ According to AHLOOWALIA et al. (2003), during the past seventy years, more than 2252 mutant varieties have been officially released, most of them based on mutagenesis breeding using ionising irradiation rather than chemicals. Of these, 60% were released from 1985 onwards. C. DORE and F. VAROQUAUX notice that according to the same study of FAO/IAEA, there are around 180 crops involved in such a breeding method.

Regulatory status of mutagenesis

Mutagenesis is specifically excluded from the scope of the relevant EU legislation that regulates a number of genetic modification techniques.

Directive (EC) 2001/18 states in its Annex 1B that “Techniques/methods of genetic modification yielding organisms to be excluded from the Directive, on the condition that they do not involve the use of recombinant nucleic acid molecules or genetically modified organisms other than those produced by one or more of the techniques/methods listed below are: 1) mutagenesis, [...]” .

Consequently, none of the almost 1000 plant varieties currently listed on the EU Common Catalogue and obtained by some form of mutation breeding is falling under the scope of Directive 2001/18 and its definition of regulated GMOs.

This approach is not unique to the EU. Nowhere in the world are mutagenesis derived plant varieties considered to constitute genetically modified organisms requiring specific GM safety assessment and approval.

New gene editing techniques

New gene editing techniques generally allow for a much shorter and thus faster mutagenesis process by immediately and solely targeting an identified, useful trait in a suitable variety. This eliminates the need for the time-consuming back-crossings to breed out a potential multitude of unwanted traits and with that helps to significantly speed up variety development.

Gene editing techniques involve targeted *in vivo* editing of native plant genes. Current techniques adapted for use in plants include Oligo-Directed Mutagenesis (ODM) and several Site-Directed Nuclease techniques such as SDN-1 and SDN-2 (using ZFNs, TALENs, meganucleases, and CRISPR/Cas9).

These gene editing techniques can be used to create products that are comparable, even identical, and indistinguishable from products that could be developed through traditional mutagenesis breeding methods or that are the result of naturally occurring mutations.

Why plants produced using gene editing techniques do not fall under Directive 2001/18

ESA considers a potential classification of the new gene editing techniques as requiring a GMO authorisation under Directive 2001/18 incompatible with both letter and spirit of the legislation.

The changes induced by gene editing techniques can occur naturally, do not result in insertion of foreign DNA into the final product, and do not result in the formation of a new combination of genetic material. Thus, the products resulting from such techniques are not covered by Art. 2(2) of Directive 2001/18/EC.

Instead, gene editing technologies should be considered as encompassed within the term “mutagenesis” as used in and exempted from scope of the Directive as its history of safe use is well-established. The JRC has acknowledged that gene editing techniques are mutagenesis techniques.²

Furthermore, it should be considered that:

- The reason for implementing the recombinant nucleic acid technology regulation in the EU was a concern about gene combinations that are highly unlikely to occur in nature. Gene editing, however, is just a more precise way of producing desired mutations in a native plant gene in comparison to other mutagenesis approaches.
- The precautionary principle applies to cases of scientific uncertainty. It is clear from science that there is less uncertainty related to mutagenesis using new gene editing than with mutagenesis using radiation or chemical mutagens.³
- Recital 17 of Directive 2001/18/EC refers to certain techniques of genetic modifications that have been widely used for decades and have a long safety record. Mutagenesis is such a technique and gene editing is one type of mutagenesis. Therefore, gene editing technologies should be exempted under Directive 2001/18/EC as there is no scientific reason, nor any basis in the Directive, to differentiate between variations of “mutagenesis” techniques, notably because the end results are similar or even identical.
- In order for products to be excluded under Annex I B, they do not involve the use of ‘recombinant nucleic acid molecules’. It is clear this item was specified to exclude insertional mutagenesis methods that result in the introduction of foreign DNA (among others T-DNA mutagenesis) from Annex I B.

There is no explicit definition of ‘recombinant nucleic acid molecules’ in Directive 2001/18/EC but Annex I A, Part 1 (1) provides insight. It sets out three criteria for techniques involving recombinant nucleic acid: (i) the formation of new combinations of genetic material, (ii) the incorporation of recombinant nucleic acid molecules produced outside the organism (via a vector) into the host organism, and (iii) their continued propagation. Gene editing methods induce a change in a native plant gene without introducing recombinant nucleic acid molecules capable of continued propagation in the final product and therefore do not meet the criteria for ‘recombinant nucleic acid molecules’ as meant in Directive 2001/18/EC.

² In the JRC report [63971] it is highlighted that the product cannot be distinguished from other mutagenesis techniques

³ EFSA Journal 2012;10(10):2943.