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Aspects of nutritional assessment of feeds from genetically modified plants (GMP) with output traits including beneficially acting substances

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Introduction

The cultivation of genetically modified plants (GMP) increased from 1.7 million to 81 million hectare from 1996 to 2004 (James 2004). Scientists and farmers, but also consumers, are increasingly asking for nutritional assessment, including safety aspects, of feeds and food from those plants. Substantial equivalence (SE) was created as a framework for the compositional assessment of feeds and food from GMP of the so-called first generation (without substantial changes of composition or without output traits) and is widely accepted. The concept of SE is based on the idea that an existing plant used as food or feed with a history of safe use and known feeding value can serve as a comparator, when assessing the safety and the feed value of a genetically modified plant (OECD 1993). Results of studies with feeds from these plants were recently summarized by Flachowsky et al. (2005).

Feeds with intended beneficial physiological properties such as amino acids, fatty acids, minerals, vitamins and other substances, which are called GMP with output traits or GMP of the second generation, may contribute to higher feed intake of animals and /or improved conversion of feed/nutrients into food of animal origin, associated with lower excretion of nitrogen, phosphorus and other nutrients. A nutritional assessment of these feeds is not adequate on the basis of compositional equivalence, other types of studies are necessary. The objective of this paper is therefore to present some considerations for experimental procedures to assess feeds from GMP with output traits, esp. including beneficially acting substances. More details are recently given by Flachowsky and Böhme (2005).

Studies to assess feeds from GMP with increased contents of beneficially acting substances

Feeds with intended beneficial physiological properties such as amino acids, fatty acids, minerals, vitamins and other substances may contribute to higher feed intake of animals and/or improved conversion of feed/nutrients into food of animal origin

and lower excretion of nitrogen, phosphorus and other nutrients. Most of these substances belong to the group of feed additives used in animal nutrition. Depending on the claim of changes as a consequence of the genetic modification, the experimental designs must be arranged to demonstrate these effects. Various experimental designs are necessary to show the efficiency of changes concerning nutrients or constituents:

- Bioavailability or conversion of nutrient precursors into nutrients (e.g. ß-carotene)
- Digestibility/bioavailability of nutrients (e.g. amino acids, fatty acids, vitamins)
- Efficiency of substances, which may improve nutrient digestibility/availability (e.g. enzymes)
- Utilization of substances with surplus effects (e.g., prebiotics)
- Improvement of sensoric properties/palatability of feed (e.g. essential oils, aromas)

Apart from the intended increase of desirable substances, genetic modification has also proved to cause some side effects, as recently discussed by Cellini et al. (2004). Such secondary changes have to be considered, when GMPs of the second generation are assessed concerning their nutritional value or their safety. Animal studies which serve as the basis of comparative approaches are necessary to answer the questions mentioned. One of the most important questions concerning nutritional assessment of GMP of the second generation is the formulation or the type of controls. In many cases is can be presumed that the isogenic comparator is not available. Therefore a special experimental design must be created to assess the GMP of the second generation. Basic questions such as the optimal species or category of animals, their age, their keeping conditions and the extent and type of measurements have to be considered. More details for animal experimentation (e.g. number of animals, duration of experiments, composition of diets, measurements) are proposed by ILSI (2003, 2004).

Conversion of nutrient precursors

Balance studies with target animal species/categories are necessary to assess the conversion of nutrient precursors (e.g., ß-carotene) into nutrients. At least two groups of animals are necessary for this purpose (Table 1).

Table 1: Proposal for assessing the conversion of nutrient precursors from GMP of the second generation into nutrients (e.g. ß-carotene)

Group	Diet composition	Measurements
1[1]	Balanced diets including typical levels of the isogenic counterpart + ß-carotene (level/s adequate to the transgenic crop)	Depends on the claim of genetic modification: - concentration of converted substances in the target organ (e.g. vitamin A in liver)[2]
2[1]	Balanced diets with adequate amounts of transgenic crop	- metabolic parameters

[1] equal feed amounts for all animals
[2] until a steady state is achieved in the target organs

Dose-response studies (at least three dosages) with the supplemental precursor and the GMP of the second generation (adequate dosages) could improve the assessment, but are more expensive in terms of time, money and feeding material.

Assessment of various substances with beneficial effects (e.g. enhancer of nutrient utilization, substances with surplus effects or sensoric properties) in GMP

Expression of substances which improve nutrient utilization (e.g. enzymes like phytase or non starch polysaccharides degrading enzymes), which influence processes in the digestive tract (e.g. prebiotics such as oligosaccharides, fructans) or essential oils and other substances which improve sensoric properties or palatability is one of the objectives of genetic modification of plants.

The efficacy of these substances should be demonstrated using specific experimental designs (Table 2).

Table 2: Proposal to assess the effects of substances in GMP, usually used as feed additives (e.g. enzymes, prebiotics, essential oils etc.)

Group	Diet composition	Measurements
1	Balanced diet including typical levels of the isogenic counterpart, *ad libitum* feeding	Depends on the claim of genetic modification: - feed intake
2	Diet of Group 1 plus additive adequate to transgenic crop (or dose-response studies), feeding level of Group 1	- digestibility of nutrients - specific parameters (e.g. mineralization, microbial change in the digestive tract etc.)
3	Balanced diet including typical levels of the transgenic crop, feeding level of Groups 1 and 2	- animal`s performances
4	Diet of Group 2, *ad libitum* feeding	- metabolic effects (immune response, etc.)
5	Diet of Group 3, *ad libitum* feeding	- quality of food of animal origin

Further developments

In the future, GMP of the second generation with more than one modified output trait will be available. Experimental designs have to consider these claims and results must demonstrate the intended changes. It is fact that more experimental groups seem to be necessary in such cases as demonstrated in Tables 1 and 2. Case by case studies should be carried out to show the bioavailability/effect of each changed nutrient or of each decreased content of undesirable substances. If isogenic controls are not available, traditional hybrids should be used as comparators supplemented with adequate nutrients (Table 3).

Table 3: Proposals for the nutritive assessment of feeds from GMPs of the second generation with more than one output trait

Group	Diet composition	Measurements
1	Balanced diet including typical levels of isogenic or near isogenic counterpart, *ad libitum* feeding	Depends on the claim of genetic modification:
2	Diet of Group 1 plus nutrients A, B..(adequate amounts of Diet 3), feeding adequate to Group 1	- analysis and *in vitro* measurements - availability/digestibility - indicator values
3	Balanced diet including typical levels of the transgenic crop, feeding adequate to Group 1	- feed intake - animal performances, feed efficacy
4	Diet of Group 2, *ad libitum* feeding	- incorporation in animal tissues
5	Diet of Group 3, *ad libitum* feeding	- quality of food of animal origin

Transgenic animals and fish might be available in the future. One example is the "phytase transgenic pig" as described by Golovan et al. (2001). The saliva of these pigs is intended to contain the enzyme phytase, which allows the pigs to digest phytate-phosphorus. Other enzyme excretions or metabolic processes may be also modified by genetic modification. But there is still a lot of research required to identify useful targets for genetic modification and to increase overall efficiency of the expensive genetic modification methods in food producing animals (Sang, 2003). Special studies are necessary to assess the modification of animals.

Summary

In the future, more feed from GMP will be available for animal nutrition as whole crops, crop components or co-products. Feeds from GMP of the so-called second generation (with output traits) are characterized by intended beneficial nutritive properties, such as increased contents of valuable nutrients or decreased concentrations of anti-nutritive substances. Specific animal feeding studies need to be conducted with the target species to confirm the expected nutritional properties of the modified crops, their components or co-products depending on the type of modification.

Some examples for adequate studies concerning nutritional assessment of such feeds have been presented as the basis for further discussions.

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