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From Plant Tissue Culture to Modern Biotechnology at the National Research and Development Institute for Biotechnology in Horticulture Stefanesti: Achievements and Prospects

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Abstract

The paper is a review of the main achievements of the NRDIBH in the field of applied biotechnology. Over the last two decades, micropropagation and virus elimination techniques were used with the final aim of establishing a virus-free plant collection for grapevine genotypes. Recently, the molecular markers have been used to characterize Romanian grapevine cultivars, to establish their taxonomical relatedness and geographical origin, or to verify genetic similarities or dissimilarities in micropropagation systems. The plant material from core collection obtained by thermotherapy and/or meristem culture is generally used as starting material for propagation and planting, as control plant material in studies concerning genetic variation of characters, and also as parental plants in breeding programmes. The methods used for establishing the core collection (in vitro multiplication of mother plant material, virus elimination procedures applied to infected vines, molecular techniques for testing the genetic stability and integrity of genetic resources) are presented. The main achievements in exploitation of the regenerative potential of various somatic tissues, such as those of the anthers, ovules, petioles, leaves, and zygotic embryos, are also discussed. Since to the Laboratory of Molecular Biology within the NRDIBH has been given recently the responsibility for the identification and detection of GMO products and foods, the paper presents also the main achievements and results in detection, identification and quantification for RR soybean and MON810 maize events.

Keywords: *Vitis vinifera*, genetic resources, *in vitro* culture, plant regeneration

Introduction

The field of plant biotechnologies, which is a top priority for N.I.R.D.B.H Ştefăneşti, was approached as early as 1987, when the Centre for Genetics, Breeding and Plant Production in Grapevine started its activity. In keeping with European legislation, S.R.D.V.V. Ştefăneşti - Argeş has been promoted, within a national programme, technologies for production and propagation of planting material (cultivars and clones) free of viruses and mycoplasms, based on thermotherapy, *in vitro* culture and micropropagation. At the same time, research projects aiming at the regeneration of plants from somatic tissues, were initiated as an essential requirement for the improvement of grapevine by using conventional methods, or by generating genetic variability and somaclone selection.

Starting with 1998, in order to turn to good account the material endowment, the preoccupations relating to the research activity in the field of plant biotechnologies were extended by increasing the range of horticultural species that can be multiplied through non-conventional techniques. In this respect, within the frame of the Department of Horticultural Biotechnologies were established and checked the *in vitro* culture technologies for a large

number of horticultural species (*i.e.* ornamental, vegetable, medicinal and aromatic plants, fruit species), so that at the present moment over 60 genotypes are being worked on.

This article is a brief review of the main scientific results obtained in the domain of applied biotechnology with grapevine genotypes.

Grapevine germplasm collection

One of the most challenging tasks for our country is to replace the old vineyards with grafted-plants from authentic and certificated grapevine cultivars. Replacing almost all the old vineyards with new planting material (pure wine and table grape cultivars) within the next one or two decades represents a national strategy for developing Romanian viticulture. The National Research and Development Institute for Biotechnology in Horticulture was given the responsibility to establish the national collection of grapevine genetic resources for the benefit of present and future generations.

Since 1988, fundamental and applied research in the field of biotechnology, have been promoted for horticultural plants, including both *in vitro* propagation and genetic improvement. NRDIBH was successfully involved in setting up a collection of 225 grapevine cultivars, organized and maintained in accordance with both national and EU requirements. This germplasm collection includes native and worldwide grapevine genotypes as following: 15 seedless table cvs; 47 seeded table cvs; 67 white and rose wine cvs; 35 red wines cvs; 6 flavoured wines cvs; 28 resistant cvs; 26 rootstock cvs.

All of these plants were obtained starting from meristematic tissues derived from one single vine, tested at the reception and, after that, periodically for their sanitary status (M. OŞLOBEANU & al. [1]). The collection is considered not only the source of *Initial* grapevine planting material, but also as source of plant material for research activities promoted within the national programmes aiming at the:

- study of genetic diversity, adaptation to local environment;
- study of variability and stability of horticultural traits;
- control for the authenticity of the cultivars in any vineyard, by comparison with cultivars registered in the collection.

One of the first priorities for research activities was to analyze the Romanian grapevine cultivars. Many of these have been cultivated from ancient times (e.g. Coarna neagra, Feteasca alba, Feteasca neagra, Feteasca regala, Francuse, Tamaioasa romaneasca, Ceaus, Babeasca, Galbena de Odobesti), and were characterized by morphological, biochemical and production parameters. In the last three years, a sustained research activity was promoted for a complete evaluation of the Romanian grapevine genetic resources using methods for genetic evaluation. It has been already established that for a complete evaluation of a germplasm collection, molecular markers provide valuable data about the gene pools, genotypes and genetic diversity, as a complement to morphological, biochemical, ecological and genetic information. Recentlly, RAPD and microsatellites markers were used for characterisation of some varieties from our germplasm collection, aiming to provide useful information about genetic data for each conserved genotype (R.N. GHEORGHE al. [2]), to establish taxonomy relatedness, geographic origin and ecological aspects (genetic diversity) (R.N. GHEORGHE & al. [3]), or to verify genetic similarities or dissimilarities in micropropagation systems (R.N. GHEORGHE & al. [4]).

The guaranty of authenticity for grapevine genotypes from our core collection enforced the use of molecular markers for testing the genetic stability and integrity of genetic resources. In the same time, research activities aiming at identifying duplicates in collection and eliminating redundant material (to maintain only as much as is necessary) were initiated.

For the next one or two years, the genetic characterisation of all cultivars from core collection and use of molecular markers are planned for reaching the following objectives:
- verifying the correct registration of each grape genotype, to be true to type and maintained properly;
- establishing the degree of similarity among genotypes;
- verifying the genetic structure of grapevine populations existing in the collection and the comparison of the results with those obtained in similar collections from other countries.
- establishing the distinctness of the ancient and new grapevine varieties, important requirement for characterization and registration of the Romanian germplasm, and for exchanges of planting material.

Tissue culture and virus elimination

Starting with 1988, research activities for sanitary selection and virus elimination were initiated according to the certification scheme applied in other European countries, in parallel with tissue culture procedures. Since that time, the program for obtaining virus-free grapevine plants was constantly developed due to the increasing number of cultivars and clones needed to be available as healthy material. The plant material within our collection is the result of standard operating procedures currently applied, including thermotherapy and/or tissue culture with periodically tests for sanitary selection and grapevine virus presence diagnostics. Thus, the germplasm resources not only for authenticity (trueness to type), but also for its phytosanitary status is guaranteed.

At the beginning, according to the certification scheme followed by our laboratory, the testing for virus infection was carried out by using herbaceous test plant and indexing on woody plant indicators (I. TIȚA & E. BUCIUMEANU [5]). Since 1993, in the laboratory for virology has been constantly used the Enzyme –Linked Immunosorbent Assay - ELISA (with DAS-, TAS- and DAS- biotin variants) method with commercial reagents, for the detection of the most economically important grapevine viruses: fanleaf virus + arabis mosaic virus (GFLV+ArMV); leafroll associated virus serotypes 1, 2, 3 (GLRaV-1,2,3); fleck virus (GFkV) and virus A (GVA) (E. BUCIUMEANU & al. [6];

The close relationship between grapevine as planting material and the effects of virus infections on the quality and yield potential aroused interests to promote studies for: comparative studies of healthy and virus infected plants, the virus diagnosis and elimination, virus purification, ultrastructural studies.

Remarkable results were obtained for investigating the behaviour of virus infected grapevines comparatively with the healthy material under *in vitro* culture conditions (E.VIŞOIU & al.[7]. Depending on the type of viruses, stage of plant development (or maturation), the season, different methods for virus testing and elimination were used, such as:
- meristematic tissues culture and/or thermotherapy;
- *in vitro* micrografting (E. BUCIUMEANU & al. [8]; E. VIŞOIU & al. [9]; E. VIŞOIU & E. BUCIUMEANU [10]) for diagnosis of leafroll, fleck, vein necrosis and corky bark diseases;
- regeneration by somatic embryogenesis followed by ELISA tests (C.F. POPESCU & al. [11]).

The most dangerous viruses of grapevine, closterovirus-like particle associated with leafroll disease (GLRaV-3) and also isometric viruses (GFLV and GFkV), were purified by routine procedures. The grapevine infected tissues were subjected to electron microscope studies and revealed ultrastructure changes induced by virus particle presence (A. BREZEANU & E. BUCIUMEANU [12].

Depending on local and national needs, the laboratory for Virology of our institute established new research targets meant to raise expectations for future performance. The main objectives to solve are:
- to enlarge the number of diagnosticated intracellular pathogens and their studies (virus, viroid, bacteria, phytoplasma);
- to apply and test new techniques for virus elimination, such as chemotherapy and electrotherapy, followed by verification of their efficiency comparatively with heat treatment and/or *in vitro* culture as classical methods;
- to find solutions for the applied methods to obtain more rapid, with lower costs and higher rate efficiency in virus elimination;
- to apply new technologies (RT-PCR) providing great potential for viruses detection in general.

Plant regeneration from somatic tissues

It is well known that high frequency of plant regeneration is an essential requirement for genetic engineering techniques, or for induction of genetic variation and selection of somaclones. Therefore, in the field of biotechnology were promoted research activities aiming at: 1) testing the competence for plant regeneration by organogenesis and/or embryogenesis of somatic tissues from petiole segments, leaf fragments and anthers; 2) establishing the main factors affecting the viability of the explants and evolution of organogenesis and embryogenesis processes; 3) optimizing the protocols for regeneration from somatic tissues; 4) verifying the genetic stability of regenerants.
The assessment of regeneration potential revealed a large variation among the 33 grapevine cultivars investigated (Table 1). Moreover, *de novo* regeneration of shoots or whole plants was successful with only 18 grapevine cultivars, from leaf, petiole, anther, or ovule tissues. An interesting fact is that in some genotypes plant regeneration from leaves or petioles occurs through organogenesis, while in other genotypes occurs through somatic embryogenesis.

The potential for *de novo* formation of meristematic structures from already differentiated tissues has proven to be dependent by genotype, type of explants, and also by the interaction between genotypes and culture medium components (C.F. POPESCU [13]).
Small amounts of auxin (0.05 mg/l IBA) in combination with moderate amounts of cytokinin (1.2 mg/l BA) were essential for triggering organogenesis and for the formation of normal developed plants, able to adapt to the *ex vitro* conditions. The very important role of the type of growth regulators used for inducing organogenesis was demonstrated by the results of experiments in which plant regeneration was almost impossible to induce in some grapevine genotypes when the leaf and petiole explants were cultured *in vitro* on media containing BA, but occur on the media containing the cytokinin-like compound thidiazuron (TDZ).

The induction of somatic embryogenesis from anther tissues have also proved to be a process strongly dependent by genotype, this making the optimization of the culture condition absolutely necessary for achieving high frequencies of regeneration. The results obtained by us showed that the plant regeneration from *in vitro* cultured anthers is controlled by an assembly of factors, among which the genotype, physiological state of the donor plant, development stage of the microspores, and culture medium, plays in the mentioned order the major roles (C.F. POPESCU [14]).

An important characteristic of the embryogenic cultures proved to be that the number of somatic embryos developed from anthers decrease with the increasing of duration of calli culture. In direct correlation, the results of isoenzyme analyses showed that the regeneration of somatic embryos within intervals shorter than 7-8 months from the initiation of the anther cultures is favourable for genetic uniformity of the regenerated plants, while the regeneration

after a longer interval could be associated with genetic variation. Amplified Fragments Length Polymorphism (AFLP) technique used for the assessment of genetic variation in plants regenerated from anthers by somatic embryogenesis allowed us to make an important advance toward understanding the relationship between the pattern of regeneration and the amplitude of genetic variation, and also toward the control of regeneration process (C.F. POPESCU [15]).

Table 1. The potential of plant regeneration from somatic tissues in some grapevine genotypes

Genotype	Explant	Type of regeneration	Regeneration %	Media
Siegfried Rebe	Antere	Indirect organogenesis	15.1	M&S modificed
Mission	Antere	Indirect embryogenesis	10,3	+ IAA + BA
Valerien	Antere	Indirect embryogenesis	4,5	+ 2% zaharosa
Coarnă neagră sel.	Antere	Direct embryogenesys	7,3	
Aromat de Iaşi	Petiole	organogenesis	7,9	M&S + BA
	Leaf		0,5	
Chardonnay	Petiole	organogenesis	3,6	or
	Leaf		2,0	
Riesling de Rhin	Petiole	organogenesis	8,0	M&S
	Leaf		2,4	+ BA (or TDZ)
Fetească neagră	Petiole	organogenesis	13,8	+ IBA (sau AIA)
	Leaf		1,9	+ 2% zaharoza
Timpuriu	Petiole	organogenesis	15,8	
de Pietroasa	Leaf		0,8	
Aligoté	Petiole	organogenesis	8,8	
	Leaf		0,4	
Chenin Blanc	Petiole	organogenesis	1,6	
	Leaf		0	
Muscat Iantarnâi	Petiole	organogenesis	0	
	Leaf		5,8	
Zghihara	Petiole	organogenesis	0	
	Leaf		3,3	
Augusta	Petiole	organogenesis	0,5	
	Leaf		3,1	
Frumoasă alba	Petiole	organogenesis and	10,5	M&S modified
	Leaf	embryogenesis	3,9	+ BA + AIA
Coarna roşie	Petiole	organogenesis and	12,3	+ 1,5% zaharoza
	Leaf	embryogenesis	0,5	
Bicane	Petiole	organogenesis and	17,7	
	Leaf	embryogenesis	0	
Crăciunel 26	Petiole	organogenesis and	27,6	
	Leaf	embryogenesis	12,0	
Augusta	ovule	Indirect embryogenesis	5,5	M&S modified
Ranâi Magaraci			8,9	+ BA + IBA
Coarnă neagră			10,3	+ 3% zaharoza

Field observations and analyses are underway in order to characterize the main horticultural traits and growth behaviour of grapevines regenerated from somatic tissues. One important objective for the next two years is to subject these grapevines for genetic analysis. Ampelographic characterization of cultivars and molecular techniques will provide valuable information to establish that regeneration protocols developed in our laboratory are appropriate for clonal propagation, or for induction of genetic variability.

Another tissue culture approach was for *in vivo* and *in vitro* tests for salt tolerance. Stable cultures of anther derived somatic embryos (at the same age and phase of development) were used to establish the tolerance potential to salt stress in grapevine tissues.

The results revealed that differences in response to salt-stress observed during *in vitro* testing have a genetic background and could be used for new salt tolerance somaclone regeneration. Also, the threshold of tolerance for viable somatic embryos capable to germinate into plantlets was 70 mM, equivalent to 4 g/l NaCl. The conclusion was that *in vitro* selection for salinity tolerance of somatic embryos proved to be an easy and efficient method. Needing a short time testing in comparison to classic methods (4 months in opposite to 3 years).

Grapevine rootstocks were regenerated from apex cultures grown on media with various concentration of NaCl (between 17 and 43 mM). The plantlets were acclimatized and subjected to daily watered with solution of 0, 51, 68 and 102 mM NaCl (3, 4 and 6 g/l). Among the investigated genotypes, SO 4-4 and Fercal 240 proved to be more tolerant to salt, as revealed by their good *in vitro* growth ability, less necroses of the leaves in acclimatized plants, and the highest differences of K/Na ratio in comparison to control. It is concluded that salt tolerance induced in apex culture does persist in whole plants acclimatized and *in vitro* stress selection could be used for improving salt tolerance (C.F. POPESCU & E.VISOIU [16]).

These results suggest that it is possible to select grapevine rootstocks having increased and stable tolerance to NaCl stress, and to use them in appropriate rootstock-scion combinations. However, several conditions seems to be essential for a reliable selection for salt stress-tolerance: 1) to compare always grapevines having the same use, either as rootstocks, or as scions; 2) to use only the system of regeneration from meristematic tissues, and *in vitro* culture media supplemented with up to 50 mM NaCl; 3) the selection for salt tolerance of grapevine plants regenerated on salt-containing media must be carried out *ex vitro*, by comparing them with control plants originated from the same type of tissue, but regenerated on salt-free media, and subjected to the same level of saline stress only as *ex vitro* plants.

Immature embyo resque of grapevine

The commercial quality of grapevines is an important objective in the improvement of table grapevine cultivars. If this character is associated with the herbaceous consistence of seeds, or even their absence (seedlessness), the value of biological material is enhanced. The *in vitro* culture of ovules containing immature zygotic embryos is the only method allowing to obtain new seedless table grapevine genotypes, with various ploidy level (sometimes originating from interspecific crosses), in a relatively short time (A. PERL & et al., [17]). This technique is used since the ’80 as complementary, or even as alternative to the conventional methods of breeding and selection of grapevine, in numerous countries with long tradition in viticulture (USA, Israel, Australia, Argentine, France, Italy, Spain, South-Africa, Bulgaria, etc).

Since the *in vitro* culture of immature embryos is considered, equally, a method with relatively high efficiency for regenerating plants from crosses between seeded cultivars with early ripening and seedless cultivars, an important objective of our research was to establish the factors with major influence on the regeneration potential: choosing the parental genotypes and the optimum moment for collecting the grapevine clusters, as well as the composition of culture media allowing a high rate of germination *in vitro* (C.F. POPESCU & et al., [18]).

In recent years, by using selected genitors and improved methods for culturing the immature embryos, more than a thousand plants have been obtained from 9 different crosses (Table 2). The large variation of the frequency of immature embryos having the capacity of growing and germinating *in vitro,* depending on the genotypes used in crosses, showed that the identification of cultivars whose immature embryos can be cultivated successfully is an

essential requirement for obtaining large populations of hybrids, as initial material for the selection of new genotypes in which the seedlessness is associated with traits responsible for high yielding, fruit quality and resistance to abiotic and biotic stress.

Table 2. Results of the *in vitro* culture of grapevine immature embryos derived from various crosses

Cross combination	**Germination *in vitro* (%)**	**Culture medium for germination**	**Number of plants transfered in the field**
Beauty Seedless x Sultanina	16.2	M&S + IAA + BA	36
Beauty Seedless x Victoria	14.6	M&S + CH + glicine	26
Augusta x Sultanina	5.0	M&S + IAA + BA	8
Kossuth Lajos x Centennial Seedless	17.0	M&S + IAA + BA	15
Kossuth Lajos x King's Ruby	18.3	M&S + CH + cysteine	16
Kossuth Lajos x Superior Seedless	21.6	M&S + CH + cysteine	55
Muscat Iantarnâi x Centennial Seedless	15.8	M&S + CH + cisteina	6
Muscat Iantarnâi x King's Ruby	19.4	M&S + CH + cysteine	16
Muscat Iantarnâi x Superior Seedless	21.6	M&S + CH + cysteine	34
Nimrang x Centennial Seedless	20.2	M&S + CH + glicine	15
Nimrang x King's Ruby	13.3	M&S + CH + glicine	-
Muscat de Alexandria x 1-48-25	25.8	M&S + IAA + BA	Plants in the *in vitro* multiplication stage, or in the *ex vitro* stage, in nutritive pots
Muscat de Alexandria x Călina	30.4	M&S + IAA + BA	
Coarnă neagră x Călina	48.1	M&S + CH + cysteine	
1-39-53 x Călina	24.3	M&S + IAA + BA	
1-40-7 x Călina	12.9	M&S + IAA + BA	
1-39-53 x 1-48-25	20.0	M&S + IAA + BA	
1-40-7 x 1-48-25	21.0	M&S + IAA + BA	

The *in vitro* germination staggered over a relatively long interval, and the evolution of zygotic embryos was different. Most of the plantlets developed by differentiating apical and root meristems, and only few, and only in certain cross combinations, developed by indirect somatic embryogenesis from the callus formed at the surface of zygotic embryos or ovules. Comparative analysis of the peroxydase isoenzymes in plants regenerated *in vitro* allowed the selection of those formed from zygotic embryos resulted from controlled crosses between seeded and seedless cultivars, and elimination of those regenerated by organogenesis from the somatic tissues of the ovules.

The morphological and ampelographical characteristics of hybrid plants, expressed in field conditions, represents essential criteria for choosing of highly valuable genotypes, possessing the trait of seedlessness, high yielding, and enhanced resistance to specific diseases. The selected genotypes are used in the following breeding stages, mainly in back-crosses with seedless genotypes, aiming to consolidate this trait and maintain the characteristics of productivity and quality.

A short-term research priority is the genetic improvement and selection of new grapevine genotypes with desired features, based on the acquired genetic information, and using both conventional and unconventional methods. Therefore, molecular markers for seedlessness will be tested aiming to predict the cross-compatibility, self-compatibility and self-incompatibility, to reduce the number of backcrosses and to increase the efficiency of selection for desired features.

Detection and quantification of GMOs events

NRDIBH was charged with responsibility for the identification and detection of GMO in seed, plant, and their primary products (feeds). As more genetically modified foods are being produced and marketed and as many different types of genetically modified organisms (GMOs) continue to be released into the environment, before and without to be tested for their genetic stability, the debate on the safety of genetic engineering has intensified in the whole Europe.

Since 2006, INCDBH Stefanesti started to develop the capacities and to implement the technologies required for identifying the GMO risk for all those crops listed by the EU in order to meet the food safety standards. From the beginning was established that the role of the laboratory would not be the production of GMOs but the detection of them. Moreover, NRDIBH has no economic motivation to use or deliver such plants or products and therefore has a neutral position concerning the GMOs. All the obtained results will be useful equally for the producers of planting material, seed producers, owners of variety patents, plant growers, food producers, and all categories of consumers.

In response to the increasing demands of consumers and policy makers to give confidence that food products on the market meet the requirements of the rapidly evolving EU legislation, a range of analytical methods must become available for detecting, identifying and quantifying either the DNA introduced or the protein(s) expressed in transgenic plants or products. The availability of validated methods for the detection and quantification of genetically modified organisms (GMOs) imposes the use of qualitative and quantitative methods to estimate the presence and, respectively, concentration of GMOs, techniques which can be compared to those used in authorized laboratories in other European countries.

One important rule of the scientific and client service staff is to act as a partner for our clients and to find the most cost effective and complete analytical solutions to meet their needs. In this respect, the laboratory for detection, identification and quantification of GMOs of NRDIBH Stefanesti perform services for:

- all farmers who cultivate any possible GM crops - to verify the resulted plant material before putting it on the market;
- all seed producer - to verify the initial seeds used for planting and also to check the resulted seeds after each breeding generation;
- private farms and research units having field trials with GMOs - to check the eventually gene flow to the nearest fields;
- all growers (farmers) of conventional and organic cultures - to verify their plant material;
- all producers of plant derived products - to check the content of GMOs in the final products in order to label.

The laboratory for detection, identification and quantification of GMOs of NRDIBH Stefanesti was organized, equipped and started to work in 2008. At the moment the laboratory is performing two procedures, both being under accreditation:

- DNA qualitative analyses of plant material and derived products from soybean and maize to establish their modified / non-modified status;
- DNA quantity analyses for evaluation the genetic modification events with soy (RR soy, event 40-3-2) and maize (MON810).

The seeds, plant materials, or their primary products (forage, grouts, and flour) are harvested by authorized inspectors and send to laboratory for tests. The first requirements before to start the analyses are: the sample, termed "analytical sample" must be fully representative of the field sample submitted for the analysis and enough large and

homogenised so to assure a certain repeatable proportion of the same number of GM-type DNA molecules.

In order to ensure reliability of our results, the laboratory staff strictly follows the recommendation for GMO testing (Guidance Document on Measurement Uncertainty for GMO testing Laboratories – JRC documents): positive-, negative- and inhibition controls, replications and repetitions, and certified reference materials.

For the qualitative method, the laboratory is using SureFood GMO 35S+NOS Screening Kit aiming to detect the presence of the two typical genetic constructs present in the majority of GMOs currently on the market: CaMV 35S promoter and A. tumefaciens NOS terminator . Similarly, for the quantitative method we are using SureFood GMO MON810 Corn real-time PCR kit, the final result indicating the percentage of GMO DNA value in unknown sample. Both methods are applied following the supplier recommended protocols.

After almost two years of testing, the laboratory performed analyses for 140 samples of RR soy and MON810 maize events for screening and quantification (table 3.). For all samples were emitted Trying Reports and, in case of a positive results with qualitative tests, we continue to perform the quantitative tests to establish the percentage of GM seeds impurity. Since Romania banned the GM soya crops starting with 2007, these results showed that most of the positive samples are impurities either as preserved seeds from previous crops, or as imported seeds.

Table 3. Received soybean and maize samples from different state and private clients

	Number of clients	**Total samples (no)**	**Positive samples (no)**	**% of positive results**
2008				
Soya	12	29	6	20.7
Maize	8	45	25 / 3 more than 0.9%	6.6(GM crops and field tests)
2009				
Soya	19	39	27	69.2
Maize	3	27	10 / 6 more than 0.9%	22.2 (GM crops and field tests)

Recently, Romania adopted the European directives and enforce documents that state the compulsoriness of the distinction between self-pollinating and vegetatively propagated crops and cross-pollinating crops. According to order no. 185 / March 23, 2009 the presence of GM seeds in seed lots of conventional plant varieties is accepted till a threshold of 0.3% in the case of cross-pollination crops, of 0.5 % in the case of self-pollinating and vegetative propagated crops, while for seed crops which germinate after some years the threshold tolerance was set at 0.7 %.

Taking into consideration the more restrictive requirements for the quality of crops, the increasing number of commercially-available transformation events, their more complex structures, the necessity for more accurate results in testing the GMO presence and content becomes obvious. Therefore, the laboratory for detection, identification and quantification of GMOs of NRDIBH Stefanesti established for the nearest future the following targets: a) to complete the acreditation for the two methods (screening and quantification); b) to increase the number of methods for checking and confirmation of samples status; c) to apply the detection, identification and quantification methods also for other types of matrices.

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