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# GENETICALLY MODIFIED SOYBEAN PLANTS AND THEIR ECOSYSTEM

ABSTRACT: Transgenic plants are developed by introgressing new genes using methods of molecular genetics and genetic engineering. The presence of these genes in plant genome is identified on the basis of specific oligonucleotides, primers, and the use of PCR (Polymerase Chain Reaction) and DNA fragments multiplication. Genetically modified plants such as soybean constitute a newly created bioenergetic potential whose gene expression can cause disturbance of the biological balance ecosystem, soil structure and soil microbiological activity.

Genetically modified plants may acquire monogenic or polygenic traits causing genetic and physiological changes in these plants, which may elicit a certain reaction of the environment including changes of microbiological composition of soil rhizosphere.

The aim of introgressing genes for certain traits into a cultivated plant is to enhance its yield and intensify food production. There are more and more genetically modified plant species such as soybean, corn, potato, rice and others and there is a pressure to use them as human food and animal feed.

Genetically modified soybean plants with introgressed gene for resistance to total herbicides, such as Round-up, are more productive than non-modified, herbicide-sensitive soybeans.

KEY WORDS: GMO, PCR, transgenic plants, ecosystem

## INTRODUCTION

Genetically modified organisms (GMO) are created all over the world, which possess new, desirable, previously non-existing traits. Presently, however, GMOs are suspected of possible negative effects on the environment and humans (Kalaitzandonakes, 1999; Metcalfe et al., 1996). Because

of that, biotechnology and genetic engineering are closely controlled and regulated in this and other countries.

Applications of modern biotechnology show a significant potential in improving agricultural productivity, reducing poverty and enhancing food security in developing countries.

Techniques of genetic engineering, such as introgression of gene for new and improved existing traits, are tools used by breeders for development of transgenic plants. These plants are made artificially, without natural pollination. The introgressed genetic sequence may come from another related plant or from a completely different species. An example for the latter case is transgenic Bt corn which produces a protein with insecticidal properties on account of a gene received from a soil bacterium (M i r e l e s, 2000). Plants containing new genes are called genetically modified, or GM crops, although all plants are in fact GM originating from genetic modifications of their wild progenitors. Transgenic plants of soybean and corn contain introgressed genes for tolerance to herbicides, pesticides, insecticides, etc. A soybean variety has been developed which is tolerant to glyphosate, the active substance in the non-selective herbicide „Roundup". Soybean resistance to this herbicide was introduced via *Agrobacterium* sp. gene fragment of cauliflower mosaic virus (promoter) which contains peptide sequence for Roundup resistance (V a n H o e f et al., 1998). This gene fragment produces synthase — the enzyme responsible for soybean tolerance to „Roundup". On the other side, non-transformed soybeans are highly sensitive to this herbicide (D e l a n n a y et al., 1995; P a d g e t t e et al., 1995).

Transgenic soybean lines with high tolerance to this herbicide maintain high and stable yields upon herbicide treatment. Introduction of GM plants into an environment disturbs the existing biological balance. Biological divergence between genotypes in the ecosystem is increased, and so is the divergence between this ecosystem and other ecosystems which originally had been much closer to it.

In EU countries, testing of GM plants during growing season or after it, but in any case prior to the use for processing and food production, is obligatory. These tests serve to protect humans against potential risks coming from genetically modified plants. In the case of non-modified crops, fields that exhibit phenotypic modifications are inspected because such modification may indicate possible tainting with GM plants.

The aim of this investigation was to determine relations between GM plants and ecosystem to which they belong. Soil microbiological activity was analyzed in fields under GM soybeans to determine the effect of these plants on changes of the environment.

## MATERIAL AND METHODS

Modified (+) and unmodified (–) soybean plants with intact roots and soil around them were sampled in 14 locations. Soil samples were taken aseptically from the depth of 0—30 cm. Genetic identification of soybean plants was do-

ne in DNA extracts using the PCR method (polymerase chain reaction) on the basis of specific primers 35S promotor and NOS3" terminator (Van Hoef et al., 1998; Meyer et al., 1996; Wurz and Willmund, 1997). Qualitative presence of the introgressed gene was determined on the basis of the PCR method and amplification of DNA fragments. After identification of plants, soil analyses were done.

Determination of microbial number in the soil was done using plate counts. The following groups of microorganisms were determined: total number of microorganisms on soil agar (Pochon and Tardieux, 1962), number of fungi on the Czapek-Dox agar (Sharlau, 2000), number of azotobacters on the Fiodorov agar (Anderson, 1958), and the number of ammonifiers on the nutritious agar (Torlak).

## RESULTS AND DISCUSSION

The PCR method determines whether a genetic transformation has been made or not. New quality traits obtained by transformation are the result of physiological processes which, in their turn, are controlled by certain genes. The objectives of gene manipulation are increased efficiency of plant production and improved protection of the environment and humans.

The numbers of bacteria and fungi were high in both soil variants (+GM crops, –GM crops), without significant differences between them (Table 1).

Tab. 1 — General microbiological activity of soil

| No. | Soil sample* | Total number of bacteria ($10^7$ $g^{-1}$ abs. dry soil) | Total number of fungi ($10^4$ $g^{-1}$ abs. dry soil) |
|---|---|---|---|
| 1 | 1 +GM | 81.37 | 46.50 |
| 2 | 1 –GM | 108.65 | 20.37 |
| 3 | 10 $km^2$ +GM | 69.36 | 35.87 |
| 4 | 10 $km^2$ –GM | 128.88 | 23.01 |
| 5 | 3 +GM | 57.41 | 12.75 |
| 6 | 3 –GM | 38.70 | 34.40 |
| 7 | 3/1 +GM | 54.30 | 15.20 |
| 8 | 3/1 –GM | 44.68 | 25.53 |
| 9 | 5 +GM | 44.65 | 10.63 |
| 10 | 5 –GM | 53.65 | 12.87 |
| 11 | 6 K +GM | 130.20 | 6.97 |
| 12 | 6 K –GM | 92.48 | 7.11 |
| 13 | 8 +GM | 113.93 | 15.95 |
| 14 | 8 –GM | 226.78 | 24.69 |

* +GM — sample of soil in which GM plants were grown.
* –GM — sample of soil in which non-GM plants were grown.

The presence of azotobacters was high in all samples, which was characteristic for the tested soil type, except that somewhat lower values were determined in samples 8, 9 and 10, but these reductions were too small to indicate any soil disturbance (Table 2).

Tab. 2 — Microorganisms of the nitrogen cycle

| No. | Soil sample* | Azotobacter ($10^2$ $g^{-1}$ abs. dry soil) | Ammonifiers ($10^7$ $g^{-1}$ abs. dry soil) |
|---|---|---|---|
| 1 | 1 +GM | 11.04 | 51.15 |
| 2 | 1 –GM | 54.89 | 24.90 |
| 3 | 10 $km^2$ +GM | 34.08 | 74.14 |
| 4 | 10 $km^2$ –GM | 25.89 | 25.31 |
| 5 | 3 +GM | 34.55 | 48.90 |
| 6 | 3 –GM | 29.03 | 43.01 |
| 7 | 3/1 +GM | 16.29 | 23.89 |
| 8 | 3/1 –GM | 2.12 | 57.44 |
| 9 | 5 +GM | 7.97 | 25.51 |
| 10 | 5 –GM | 6.43 | 34.33 |
| 11 | 6 K +GM | 40.68 | 41.85 |
| 12 | 6 K –GM | 33.19 | 59.28 |
| 13 | 8 +GM | 43.86 | 102.54 |
| 14 | 8 –GM | 20.77 | 44.90 |

* +GM — sample of soil in which GM plants were grown.
* –GM — sample of soil in which non-GM plants were grown.

The number of ammonifiers was optimal in most of the analyzed samples (Table 2).

Interactions between GM and non-GM plants were not found (B e r i n g e r, 2000).

## CONCLUSION

The transgenic DNA or proteins from transgenic crops can be detected in plant material and processed food. Potential effects of GM crops on ecosystems and environment are of special significance and must be observed.

The results obtained in this study showed that for the GM and non GM soybean plants caused no change in the soil microbiological activity. It seems to indicate that GM plants do not cause large changes in the biological balance of the environment in which they grow.

Transgenic DNA has been deemed to be safe for consumption as it is made up of the same building blocks as plant genomic DNA. Transgenes should remain stable from generation to generation and GM plants should not disturb the biological balance of the environment. Some authors have considered the

benefits and risks of transgenic crops in great detail, which can be of importance for further biotechnological advancement.

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# ГЕНЕТСКИ МОДИФИКОВАНЕ БИЉКЕ СОЈЕ И ЊИХОВ ЕКОСИСТЕМ

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Резиме

Методе молекуларне генетике уведене у генетски инжињеринг учествују у програму трансгених биљака, насталих увођењем нових гена, који су у основи тражених особина. Њихово присуство у биљном геному се идентификује на бази специфичних олигонуклеотида, прајмера, коришћењем PCR (Polymerase Chain Reaction), методе умножавања ДНК фрагмената. Генетски модификоване биљке, као што је соја, постају новонастали биоенергетски потенцијал за средину у којој се развијају, чија експресија гена може изазвати поремећај биоравнотеже, екосистема, структуре земљишта, његове микробиолошке активности.

Генетски модификоване биљке могу бити са уведеним моногеним или полигеним својствима, што доводи до њихових генетско-физиолошких промена и одређених реакција у средини у којој се налазе. Сматра се да организми са новонасталом генетском структуром могу утицати на измену био-равнотеже средине у којој се налазе, као што су промене хемијског и микробиолошког састава у ризосфери земљишта, које ће условити низ измена његове биоенергетске способности.

Увођење гена за одређене особине биљака има за циљ повећање њиховог приноса и интензивнију производњу хране. Отуда је све већи број модификованих биљних врста, као што су соја, кукуруз, кромпир, пиринач и друге, које имају велико учешће у исхрани људи и животиња, због чега је неопходна контрола таквих биљних врста и средине у којој се оне налазе.

Генетски модификоване биљке соје, са унетим геном за резистентност на тотални хербицид, као што је Round up, доприноси њиховој већој продуктивности, за разлику од немодификованих биљака, које не подносе овакву врсту хербицида. Међутим, особина новоунета у геном биљке може имати различите последице по околину у којој се налази, што је пре свега у вези са циљем њеног настанка, као што су отпорност соје на тотални хербицид, Round up и њен однос са средином.