Review article

BIOTECHNOLOGY: REALITY OR DREAM

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The development of molecular biology and molecular genetics, especially of the recombinant DNA technology enabled improvement of experimental methods that provide manipulation within a cell-free system, such as cell and tissue cultures. Such methods resulted in the development of different new technologies with specific properties in relation to the conventional definitions. According to PERSLEY and LANTIN (2000) the following components are essential for the contemporary biotechnology: (i) genomics – a molecular characterization of all genes and gene products of an organism; (ii) bioinformatics – the assembly of data from genomic analysis into accessible forms; (iii) transformation – the introduction of genes controlling a trait of interest into a genome of a desired organism (micro organisms, plants, animal systems). By the application of cotemporary biotechnology new methods in the field of diagnostic are developed such as rapid and more accurate identification of the presence and absence of genes in the genome of the organism of interest (identification of pathogens, prenatal diagnostics, molecular markers assisted breeding for plants, etc.). The traits of an organism are determined by its genetic material, i.e. by a molecule of deoxyribonucleic acid (DNA). WATSON and CRICK (1953) were the first scientists to describe the structure of DNA as a double-stranded helix. Higher organisms contain a set of linear DNA molecules - chromosomes and a full set of chromosomes of

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an organism is a genome. Each genome is divided into a series of functional units, i.e. genes. The traits of an organism depend on genes, but their expression depends not only on genes, but also on many other factors, including whether a gene, controlling the trait, expresses, specific cells in which it expresses and specially the mode by which the gene and its product interact with the environment. A special aspect within the application of biotechnology occurs as an interaction of a foreign gene with a genome of an integrated organism. Also, application of biotechnology provides transfer of one or several favorable genes from any evolutionary category into other category of an organism and in such a way it is possible to develop genetically modified organisms (GMO) having expressed desired, target traits. A survey of the application of biotechnology in the world and our country is presented in this paper.

Key words: genomics, bioinformatics, genetic transformations

INTRODUCTION

At the end of the 20th century agriculture entered a new technological era: biotechnology integrated scientific branches such as biology, biochemistry, molecular biology, genetics, chemical engineering and informatics. Discoveries within these fields opened up new horizons in prevention of plant and animal diseases, pesticide-free insect control, livestock efficiency increase, increase of production and improvement of food quality and reduction of environmental degradation.

Functional genomics is one of the essential factors behind this revolution, as it uses a scientific approach for identification, definition of gene functions and determination where and how genes mutually function in control of a particular trait. The cotemporary approach essentially presenting study on a genome structure (e.g. mapping), is generally aimed at traits controlled by one gene or a small number of genes and therefore provided information is often mostly related to a gene location in the genome. Although information on the gene location is a critical point, the determination of a gene function provides possibilities to study relations and interactions among several thousands genes. In such a way, the background for investigation when and why a specific trait expresses, which sets of genes specifically govern the expression of such a trait and under which conditions is set up. Obtained pieces of information enable researches with a possibility to develop economically important plants whose genome encompasses specifically modified and by genomic transformation integrated genes, affecting traits that allow growing under different environments. Among many several traits such as tolerance to high and low temperatures or saline and acid soils, resistance to pathogenic micro organisms or the synthesis of substances of interest for the production of food of a higher quality. At the same time, the application of new biotechnology provides the determination of a potential or actual risk of consumption of food produced from genetically modified plants, which is a prerequisite for biosafety that will provide minimization or complete elimination of risk.

BIOTECHNOLOGY – DEFINITION AND APPLICATION WITHIN PLANT BREEDING

According to the Convention on Biological Diversity (CBD), biotechnology means "the application of any technology that uses biological systems, living organisms or their derivates with the aim to produce - develop - or modify products or processes of specific purposes". Genetic modifications defined by CPB (Cartagena Protocol on Biosafety) acts exclude other old or new techniques that lead to changes of genotypes (CARTAGENA PROTOCOL ON BIOSAFETY, 2000). Interpreted in such a way the definition encompasses a great number of tools and methods that are practically applied within fields of agriculture and food production. According to PERSLEY and LANTIN (2000) the following presents the essential components of contemporary biotechnology:

- molecular characterization of all genes and gene products of an organism
- bioinformatics - the assembly of data from genomic analysis into accessible forms
- transformation - the introduction of a gene conferring potentially useful traits into plant, animal or genome of micro organisms
- molecular breeding - the use of molecular markers for the breeding process improvement
- diagnostics - rapid and more accurate identification of pathogens based on molecular characterization

Genetic modifications of plants, as well as, conventional breeding have the same goal - to develop plants with improved traits. A full set of genes is transferred from parental lines into a progeny by conventional breeding methods. In such a way not only one or two favorable genes, but also hundreds of others, some favorable some unfavorable, are transferred. In addition, the conventional breeding methods are based on transfer of genetic material among individuals of the same or very close species.

The application of biotechnology (genetic engineering and cell and tissue culture) has several advantages over conventional plant breeding and the key advantage is that it is possible to transfer one or several favorable genes from any plant species, micro organisms, animal systems into a target plant and *vice versa* and in such a way it is possible to develop genetically modified plants (GMP).

GENETICALLY MODIFIED PLANTS

The presence of DNA molecules within all cells of organisms is the base for the development of genetically modified organisms including plants. The traits of an organism are determined by its genetic material, i.e. by a molecule of deoxyribonucleic acid (DNA) as a double-stranded helix (WATSON and CRICK, 1953). This molecule encompasses genetic information and governs metabolic processes. Genes are discrete segments of DNA containing information necessary for the formation of a specific protein. Proteins in cells function as enzymes in biochemical reactions or as structural or depot units, contributing to the expression of a spe-

cific trait. Processes of transcription and translation, controlled by a set of regulatory mechanisms in a way that a particular protein is synthesized only where and when it is needed, are included into transfer of information from DNA to a protein. Very divergent species have a similar mechanism on translation of genetic information from a DNA into a protein; hence a DNA sequence can be transferred from bacteria into a plant cell where it can also function.

Results of ARBER *et al*. (1983) obtained within a system of bacterial viruses showed that restriction enzymes, isolated from bacteria, could be used for a precise sectioning of the DNA molecule in specific sites defined as sequences of four, five and six nucleotides, presenting a basis of the development of the new recombinant DNA technology. If DNA ligase, an enzyme that catalyses the joining of two ends of DNA is used, DNA fragments can be joined (stuck together) into a new molecule of the altered structure. If cohesive fragments belong to different organisms, their cohesion results into a recombined DNA molecule, which is used for a gene transfer in the process of transformation to get genetically modified organisms (GMO).

The processes involved in developing genetically modified plants are the following:

- identification and isolation of the desired gene,
- gene cloning,
- development of transgenes,
- gene transfer and
- introduction into breeding processes.

The identification of a single gene is not sufficient - it is necessary to be acquainted with regulatory mechanisms of gene actions, as well as, with their secondary effects and interactions with other genes. Prior to this, it is necessary to identify an organism containing the desired gene in its genome. A little is known about specific genes affecting plant traits such as yield, stress tolerance, disease resistance etc.

Arabidopsis thaliana is a plant of the *Cruciferae* family with only five chromosomes having progenies in only seven weeks. Due to a great similarity among plants, *Arabidopsis* can be used as a model system and sequences of genes of this plant, and known functions could be used as molecular - DNA markers in the identification and localization of homologous sequences in genomes of other plant species. Some of such markers can be used for the identification of chromosomal regions containing genes that code quantitative traits including tolerance to soil salinity (ZHU, 2000).

Tobacco plants, transformed by the use of *Ti* - plasmids of the bacterium *Agrobacterium tumefaciens* as a vector, were the first transgenic plants with active foreign genes introduced by methods of biotechnology (HORSCH *et al*., 1984; DEBLOCK *et al*., 1984). The transformation was confirmed by the presence of a foreign DNA sequence in the primary transgenic plants and their progenies. Beside gene integration and vertical transmission, the expression of the introduced gene, governing resistance of transgenic plants to the antibiotic neomycin, was deter-

mined. This resistance is a result of the action of a marker gene used in transformation which controls the synthesis of the enzyme neomycin phosphotrasferase that inactivates the neomycin activity by phosphorylation. The transformation was performed in the initial experiments by the introduction of genes into protoplasts, while POTRYCUS (1990) reviewed the majority of used efficient methods. In our experiments, the same marker gene controlling activity of the enzyme neomycin phosphotransferase (*NPT* II) was used for maize transformation by the application of three different methods: microinjection into archesporial tissue of several cell divisions prior to meiosis, co-cultivation of dry seed in the solution of plasmid DNA and the use of pollen grain as a carrier of plasmid DNA (KONSTANTINOV *et al*., 1993).

The accessibility of efficient systems of transformation of cultivated plants is important for plant breeding. The initial studies were aimed at the introduction of genes controlling resistance to insects, herbicides and micro organism's disease-producing agents. Progress was very rapid and genes controlling these traits have been already successfully integrated into several economically important plants species, such as maize, soybean, cotton, oil seed rape, sugar beet, tomato, alfalfa that are commercially grown on large areas (JAMES, 2001). Subsequent studies encompassed changes of a trait whose expression depended on the developmental stage and differentiation of tissues and organs.

Dramatic progress in the development of transfer systems of genes into plant organisms and the possibility of regeneration of vital fertile plants established conditions for the introduction of foreign genes, controlling useful plant traits, significant increase of genetic yield potential and therefore the food production increase. Besides, transgenic plants, synthesizing specific substances of importance for pharmaceutical industry, were developed.

GENETICALLY MODIFIED CROPS AND RISKS IN CONSUMPTION OF FOOD PRODUCED FROM THESE CROPS

Food does not only satisfy the elementary physiological needs, but it is also related to contentment and recovery and therefore any indication that any of food components may carry a risk to health has to be avoided.

Genetically modified plants provide food industry with a possibility to produce new products with significantly lower investments. The examples of such production are the following: (i) great quantities of starch of special types or starch with altered structure or chain length providing extended storage; (ii) greater amounts of specific oils or elimination of certain fat acids in plant oil and (iii) proteins with the balanced amino acids content. Enzymes and genes involved into biosynthesis of dyes and scents are of importance for both, food industry and consumers of such produced food.

The effect of products derived from genetically modified plants on human health depends on specific chemical composition of the product itself. Products for instance with the increased content of digestible iron or if substances causing allergy is removed from it can be potentially useful, while it can be harmful if ge-

netic modification introduced new allergen or toxin that remained during the production process (WEKSLER, 2000). Genetic manipulation can cause that a new allergen can be introduced into food not only from the known source of allergenicity, but also from plants, bacteria or viruses whose potential allergenicity is unknown. Due to it, the studies on allergenicity are a compulsory part of the investigation process of safety of products derived by processing of genetically modified plants.

The example of the consumers' respond to potential risks is a movement of consumers in England related to the implication that can arise from a well known study of effects of feeding rats on genetically modified potato. The studies showed that potato modified to synthesize a protein toxic to animals could have adverse effects on some organs and the immune system (RUCKMAN, 2002).

Having sufficient information on specificities of the integrated DNA gene sequence, a detailed comprehension of functions of gene products and a high level of knowledge on biological and chemical traits are stated to be key reasons for low risks for human and animal health in consumption of food obtained by processing of GMO.

There are three levels of the potential risk: i) a promoter sequence, a constituent part of a construct, as it is a key factor in gene expression; ii) a sequence of a marker gene that is a constituent part of the construct and has a role in the selection of a transformant and iii) a coding sequence of a gene that is introduced during transformation.

In recent times intensive studies on risk caused by promoter sequences of bacteria and viruses, a constituent active part encompassing a gene of interest introduced into a heterologous organism, have been performed. A promoter sequence (35S) of a gene of cauliflower mosaic virus was used in experiments of transformation of the majority of plant species commercially grown. HO *et al*. (1999) pointed out to the risk that this sequence could activate or inactivate the expression of genes of a host plant or endogenous viruses then could recombine with mammalian viruses with unpredictable consequences. Genes whose introduction changes existing biochemical processes, including genetic manipulation of regulatory genes, are a special approach. The isolation of large bacteria with regulatory sequences provides reliable gene expressions within specific tissues of transgenic plants.

The possibility of a gene expression reduction in transgenic plants has a significant role in studies on the gene expression and function, as well as, plant breeding. Results of studies on insertion of DNA donors into plant chromosomes by homologous recombination (PASZKOWSKI *et al*., 1988) show that it is possible to use this approach in selective gene inactivation.

In our experiments performed to develop transgenic plants, introduced marker gene (*NPT* II) induced changes in activities of particular loci, controlling isoenzymic activity and complex of soluble proteins. These changes were determined in the germ of mature maize kernel (MLADENOVIĆ *et al*., 1991).

Genetically modified plants used in food production provide consumers not only with satisfaction of the basic function of a nutrient, but also with many

improvements, such as a reduction of the allergen content or the increase of the amounts of vitamins and other vital substances (YE *et al*., 2000; LUCCA *et al*., 2001). One of example is margarine produced from plants having increased plant sterol content which lowers the synthesis of cholesterol. Another good example is cereals with the increased content of iron and vitamin B12. Rice, contrary to barely, is extremely susceptible to iron deficiency. Its transformation was performed by a fragment of barely genomic DNA containing two *naat* genes that are coding the synthesis of a key enzyme involved into phytosiderophore biosynthesis. Phytosiderophore is a substance released by a grass root system into soil that dissolves iron and in such a way makes it accessible for plants. Yield of rice transformed by these genes was four fold-higher than one of the original genotype, and quality also improved (TAKAHHASHI *et al*., 2001).

Transformation methods alongside with methods of the GMO detection either in the original form or in products obtained from GMO were developed. Some restrictions of the detection method have to be emphasized. In Europe, a law related to food includes a regulation of a high protection level of health and safety of consumers, as well as, the requirement to establish the legislation primarily on scientific results and risk estimations. New types of food are defined as food not abundantly used and are estimated and classified within EC (European Commission, 2001) as:

- elementary equivalent to traditional referent food;
- sufficiently similar to traditional referent food and
- insufficiently equivalent or similar to traditional referent food.

In a genetically modified organism (GMO) a transgenic product presents a difference between the original and modified genotype. During the comparative studies on possibilities of occurrence of side effects of genetic modification it is useful to compare GMO with original genotypes under identical conditions. If the comparison proves isogeny of GMO and the original type, except for the foreign gene introduced by transformation, statistically significant differences can be indicative of the existence of secondary effects (ANONYMOUS, 1999).

Genetically modified plants can differ from non-modified ones due to the fact that they contain either a new specific DNA sequence and/or a specific new protein not present in the analogous product. Methods currently applied in the detection of genetic modifications are based on the following: (i) DNA detection by which modification was performed (method of DNA multiplication based on polymerase chain reaction - PCR method) and (ii) detection of the appropriate transgenic protein product by immunological methods (ANONYMOUS, 1999).

An amount of a particular protein present in a sample that can contain many other proteins can be detected by immunological methods (ELISA). The method uses an antibody to bind a specific protein and an antibody conjugate and an enzyme that can metabolize a colored product that can be visualized and quantified. The quantified assay is in the form of a plate or a band.

The PCR method is based on the detection of a DNA fragment introduced into a plant genome. This method allows multiplication of specific DNA sequences

up to the extent that they can be quantitatively and qualitatively analyzed. The majority of the methods based on the PCR is of high quality, sensitive and can detect one or several gene copies. The presence of regulatory sequences (35S promoter, *nos* terminator), marker genes and genes with which transformation was performed can be detected by the use of specific oligonucleotides. The advantage of the PCR-based method over the immunological method is that this method provides the detection of genetically modified DNA in samples which were thermally or chemically treated in their processing. The quantitative PCR-method will be used to determine the level of genetically modified components in food.

Genomic technology, such as genome sequencing and expression, on the whole led biological sciences to the level of informatics and the use of the term bioinformatics. This expression is used to describe the scope of methods and activities within information systems formed for processing and exchange of data obtained under laboratory conditions, interpretation and integration of data, preparation of documents and electronic issues of data related to sequences, mapping and data bases (BOGUSKI, 1994).

The European Community has enacted a law by which a compulsory labeling of food with over 1% of substrates from genetically modified organisms (GMO) has been anticipated. The way of sampling outside of laboratories has become an essential question. One of the examples is seed sampling from containers for seed storage or seed transport. Another example is a variability of GMO : non-GMO ratio in a multiplied "homogenous" sample in a laboratory. These two examples point to the problem of the detection of the GMO presence in food.

The regulation of the use of transgenic plants has to be based on the scientific principles that provide health safety and the acceptable price of food produced from them that also recognize a low risk of gene transfer technologies and clearly define benefits of growers of such plants, food producers and consumers of such food.

COMMERCIALISATION

Commercial cultivation of genetically modified plants directly depends on the following:

- the evaluation of traits under field conditions of cultivation;
- the success of breeding and seed production for cultivation on large areas,
- the choice of optimal growing systems and
- the release of new biotypes and protection of rights to intellectual property.

The improvement of the plant breeding process is achieved so rapidly that commercial cultivation depends on the legislation, protection of rights to intellectual property and public acceptance. Patenting genetically modified organisms is designated as a key step in compensation of expenses related to the development of plants with significantly new traits. According to DIAMOND vs. CHRABARTY (1980) micro organisms could not have been patented because they had been living

organisms. In 1985, the US Patent Office enacted the law, which enabled patenting plants (*Ex parte* HIBBERD, 1985).

The total global area sown with genetically modified crops increased from 1.7 M ha in 1996 to 52.6 M ha in 2001 (JAMES, 2001). The greatest areas are in the USA (35.7 M ha), then Argentina (11.8 M ha), Canada (3.2 M ha) and China (1.5 M ha). In Europe, genetically modified crops are grown in Romania, Bulgaria, Spain, Germany and France. Genetically modified crops were observed in 1668 trials under field conditions in the European Community during 2001 (EUROPEAN COMMISSION, 2001). The largest areas of 33.3 M ha, then 9.8 M ha, 6.8 M ha and 2.7 M ha were sown with genetically modified soy bean, then maize, cotton and oil seed rape, respectively. Genetically modified sweet maize, earth nut, pumpkin and papaya have been grown to a smaller extent (BIO, 1998). Out of global total areas sown with soy bean, i.e. cotton, approximately 46%, i.e. 20%, respectively, are sown with genetically modified plants. Total areas under genetically modified maize and oil seed rape do not differ from those in 2000.

The utilization of genetic modification (GM) technologies in Europe is currently related to many discussions arisen from strong opposing to consumption of food produced from genetically modified plants, especially to the utilization of soybean meal in feed production (BROOKES, 2002). Meal produced from oil seed crops has been intensively used in feed as a source of proteins. Different meals have different nutritive values and soybean meal is considered the essential ingredient of feed. The total soybean meal production in the EC amounts to 27 M tons and the greatest percentage of it is used in feed. The main reason for this is the fact that soybean meal contains 44-50% of proteins. The USA, Brazil and Argentina are the most important sources The majority of soybean crops in two principal sources, USA and Argentina, are genetically modified (GM), hence the greatest part of soybean meal used in the EC for feed production originates from GM genotypes. During this and the following year (BROOKES, 2002), fundamental changes in GM and non GM soybean markets are predicted in several directions: (i) a great support to the movement for utilization of non-modified soybean in England; (ii) amounts of non-modified soybean are expected to be 22-25 M tons, but if requirements of the Asian market are included the necessary quantities will amount to 30 M tons; (iii) due to the growth of genetically modified soybean, by the introduction of the gene that control tolerance to herbicides (Roundup Ready Soybeans), in Argentina and Brazil, a drop of a quantity of non-modified soybean to 20-25 M tons is expected. Based on general forecasts, the increase of requirements for non-modified soybean is expected, meaning the increase of the number of countries-producers with a possibility to apply a strict legislation on growth and utilization of GM organisms and products-based on them is expected. One of certain consequences is the increase of prices of meat and other products originating from animals not fed on genetically modified soybean, while the respond of consumers will be interesting (HMSO, 2000)

BIOTECHNOLOGY APPLICATION WITHIN NON-AGRICULTURAL FIELDS

The utilization of transgenic plants in the production of special chemical products is also a great possibility. Plants were traditionally a raw material for the production of broad spectrum polymers - from starch and cellulose, belonging to carbohydrates, to polyhydrocarbons, such as gum and waxes. During the last decades, many of these products were substituted by the synthesis based on oil derivates. Problems of different types related to the utilization of oil derivates have been causing ever increasing attention and studies on biological polymers. Genetic engineering will significantly increase a spectrum and content of accessible biopolymers. Transgenic plants present a potential of the production of foreign proteins with various applications in medicine. Proteins as neuropeptides, factors of blood or growth promoters can be produced in plant seeds. Several proteins of the animal origin have already been produced in transgenic plants (EICHHOLTZ *et al*., 1987; LEFEBVRE *et al*., 1987; BARTE *et al*., 1986), while the synthesis of pharmaceutical proteins has been performed in oil seed rape. Progress was achieved in the development of transgenic plants performing the synthesis of drugs and medicines: a transgenic plant was used simultaneously as food and vaccine (KONG *et al*., 2001), or in another case, a transgenic plant was an intermediary host used for extraction and filtration of interesting pharmaceutical substances (recombinant proteins such as antibodies, hormones or blood factors, for instance).

It is known that the tomato contains carotenoid lycopene with a protection effect against prostate cancer. Besides, fruit skin contains small amounts of flavonoids, potentially very strong antioxidants. MUIR *et al*. (2001) performed tomato transformation by the introduction of a gene from petunia that controlled the synthesis of the enzyme involved into flavonol biosynthesis. Fruit skins of transgenic plants contain up to 78 fold higher amounts of flavonol in relation to the original genotype and present a good raw material for the pharmaceutical industry in production of specific drugs and medicines.

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REFERENCES

anonymous (1999): Methods for detection of GMO grain in commerce, www.acpa.org/public/issues/biotech/detechment.htlm

ARBER, W., L. ENQUIST, B. HOHN, N.E. MURRAY, and K. MURRAY (1983): Experimental methods for use with lambda. *In*: Lambda II. (Ed. R.W.Hedrix *et al*.) p. 433. Cold Spring Laboratory, Cold Spring Harbor, New York.

BARTE, A. *et al*. (1986): Plant. Mol. Biol., *6*, 347.

BIO (1998): Transgenic products on the market. Biotechnology Industry Organization, Washington DC, available online at http://www.bio.org/food&ag7transgenic products.htlm

BOGUSKI, R. (1994): www.ncbi.ulm.nih.gov.

BROOKES, G. (2002): GM and non GM ingredient market dynamics and implications for the EU feed industry. Agro food Industry hi-tech. January/February 2002, pp. 13 – 16.

CARTAGENA PROTOCOL ON BIOSAFETY (2000): http://www.biodiv.org/biosafe/Protocol.html. *In*: Biotechnology and Development MONITOR, No. 43. pp. 2 – 7, September 2000)

DE BLOCK, M., L. HERRERA-ESTELLA, M. VAN MONTAGU, J. SCHELL, and P. ZAMBRYSKI (1984): Expression of foreign genes in regenerated plants and their progeny. EMBO J., *3*, 1681-1689.

DIAMOND *vs*. CHRACAABARTY (1980): 447, U.S., 303.

EICHHOLTZ, D.A. *et al*. (1987): Somat.Cell. Mol. Genet., *13*, 67.

EUROPEAN COMMISSION (2001): Joint Research Center, www.jrc.org

Ex parte HIBBERD (1985): United States Patents Quarterly *227*, 303.

HO, M., A. RYAN, and J. CUMMINS. (1999): Cauliflower mosaic virus promoter – a recipe for disaster. Microb. Ecol. Health Dis., *10*, 33-59.

HORSCH B.R., E.J. FRY, L.N. HOFFMANN, D. EICHHOLTZ, G.S. ROGERS, and T.R. FRALAY (1985): A simple and general method for transferring genes into plants. Science, *227*, 1229-1231.

JAMES, C. (2001): Global review of commercialized transgenic crops. ISAAA Briefs No.24, preview. ISAAA. Ithaca, NY.

KONG, A. *et al*. (2001): Proc. Natl. Acad. Sci. USA, *98*, 11539-11544.

KONSTANTINOV, K., S. MLADENOVIĆ, G. SARATLIĆ, S. GOŠIĆ, B. TADIĆ, V. ŠUKALOVIĆ, and J. DUMANOVIĆ (1993): Transgenic maize: disturbance of host genome expression by bacterial gene integration. In: breeding and Molecular Biology: accomplishments and future promises. Proc. Of XVIth Conference of maize and Sorghum EUCARPIA Section. June 6 – 9, 1993. Bergamo, Italy.

LEFEVBRE, D.D., B.L. MIKI, and J.F. LALIBERTE (1986): Bio/Technology 5, 1053.

LUCCA, P., R. HURRELL, and I. POTRYCUS (2001): Theor. Appl. Genet., *102*, 392-397.

MLADENOVIĆ, S., K. KONSTANTINOV, M. DENIĆ, and B.TADIĆ (1991): Genetika, *23*, 191.

MUIR, R.S., G.J. COLLINS, S. ROBINSON, S. HUGHES, A. BOVY, C.H.R DE VOS, A.J. VAN TUNEN, and M.E. VERHOEYEN (2001): Overexpression of chalcone isomerase in tomato results in fruit containing increased levels of flavonols. Nature Biotechnology, *19*, 470-474.

PASZKOWSKI, J., M. BAUR, A. BOGUCKI, and I. POTRYKUS (1988) EMBO J., *7*, 4021.

PERSLEY, G.J. and M.M. LANTIN (2000): Agricultural biotechnology and the poor. Proceedings of an international conference, Washington D.C. oct. 21-22, 1999. www.cgiar.org/biotech7rep0100/contents.htm.

POTRYCUS, I. (1990): Physiologia Plant., *79*, 125.

RUCKMAN, S. (2002): Functional Food – are safety and efficacy important. NutraCoc, May/June.

TAKAHASHI, M., H. NAKANISHI, S. KAWASAKI, K.N. NISHIZAVA, and S. MORI (2001): Enhanced tolerance of rice to low iron availability in alkaline soils using barley nicotinamine aminotransferase genes. Nature Biotechnology, *19*, 466-469.

HMSO (2000): UK House of Commons Agriculture Committee: The segregation of Genetically Modified Foods, London, 2000.

WATSON, J.D. and F.H.C. CRICK (1953): Nature, *171*, 737.

WEKSLER, M.E. (2000): GM food opportunities to improve human health. OECD Conference on GM food, Edinburgh, Scotland.

YE, X., S. AL-BABILI, A. KLOTI, J. ZHANG, P. LUCCA, P. BEYER, and I. POTRYCUS (2000): Engineering provitamin A beta-carotine biosynthetic pathway into (carotenoid-free) rice endosperm. Science *287*, 303-305.

ZHU, J.K. (2000): Genetic analysis of plant salt tolerance using Arabidopsis. Plant Physiol., *124*, 941-948.

BIOTEHNOLOGIJA: STVARNOST ILI SAN

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Izvod

Razvoj molekularne biologije i molekularne genetike posebno tehnologije rekombinantne DNK uz istovremeno usavršavanje tehnologija koje omogućavaju manipulacije u bezćelijskom sistemu kao što su kultura ćelija i tkiva rezultirao je u formiranju oblasti biotehnologije sa specifičnim osobinama u odnosu na klasične definicije. Integracijom najpropulzivnijih naučnih disciplina kao što su biologija, biohemija, molekularna biologija, genetika, hemijski inženjering i informatika otvorene su neograničene mogučnosti i izazovi u sprečavanju bolesti humane populacije, biljaka i životinja, kontroli insekata bez pesticida, povećanju produktivnosti stoke, povećanju proizvodnje i poboljšanju kvaliteta hrane i redukciji životne sredine. Osnovne komponente moderne biotehnologije, prema Presley-u i Lantin (2000) su: (i) genomika – molekularna karakterizacija svih gena i produkata gena organizma; (ii) bioinformatika – organizovanje podataka dobijenih iz analize genoma u prihvatljivu formu; (iii) transformacija – unošenje gena koji kontroliše osobinu od interesa u genom željenog organizma (mikroorganizmi, biljke, animalni sistemi) i (iv) dijagnostika – brža i pouzdanija identifikacija prisustva ili odsustva gena u genomu organizma od interesa (identifikacija patogena, prenatalna dijagnostika, molekularno oplemenjivanje biljaka korišćenjem molekularnih markera i sl.). Svojstva organizma su određena njegovim genetičkim materijalom, odnosno molekulom dezoksiribonukleinske kiseline (DNK). Watson i Crick (1953) su prvi objasnili strukturu molekula DNK kao dvolančane zavojnice. Viši organizmi sadrže set linearnih DNK molekula - hromozoma i kompletan set hromozoma organizma je genom. Svaki genom je podeljen u serije funkcionalnih jedinica odnosno gena. Svojstva organizma zavise od gena ali njihovo ispoljavanje zavisi i od mnogih drugih faktora, uključujući da li se gen odgovoran za tu osobinu eksprimira, specifičnih ćelija u kojima se eksprimira a posebno kako gen i njegov produkt intereaguju sa spoljašnjom sredinom. Poseban aspekt u okviru korišćenja biotehnologije se javlja kao interakcija stranog gena sa genomom organizma u koji je integrisan. Primena tehnologije rekombinatne DNK ima nekoliko prednosti u odnosu na tradicionalno oplemenjivanje. Genetičke modifikacije biljaka kao i klasično oplemenjivanje imaju isti cilj, dobijanje biljaka sa poboljšanim svojstvima. Klasičnim metodama oplemenjivanja prenosi se kompletan set gena iz roditeljskih linija u novo potomstvo. Time se ne prenosi samo jedan ili dva željena gena već stotine drugih gena, neki poželjni, a neki ne. Takođe, tradicionalne me-

tode oplemenjivanja se zasnivaju na prenosu genetičkog materijala između jedinki iste ili veoma bliske vrste. Primenom biotehnologije moguće je preneti jedan ili više poželjnih gena iz bilo koje evolucione kategorije u istu ili drugu kategoriju organizma i na taj način stvoriti genetički modifikovane organizme (GMO) sa željenim osobinama. U radu će biti dat pregled stanja u oblasti primene biotehnologije u svetu i kod nas.

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