A PROTOTYPE DECISION SUPPORT SYSTEM FOR GMO TRACEABILITY

Petra Kralj(1), Nada Lavrač(1,2),
Kristina Gruden(3), Ana Rotter(3), Dejan Štebih(3), Dany Morisset(3), Jana Žel(3)
(1) Jožef Stefan Institute, Jamova 39, Ljubljana, Slovenia
(2) University of Nova Gorica, Vipavska 13, Nova Gorica, Slovenia
(3) National Institute of Biology, Večna pot 111, Ljubljana, Slovenia
e-mail: petra.kralj@ijs.si

ABSTRACT

The problem of traceability of genetically modified organisms (GMOs) addresses the detection, identification and quantification of GMOs in food, feed and seed samples. Due to a large number of GMOs in the market, a system for reliable and affordable traceability of GMOs, optimizing the price of testing for a given sample, has to be established. We have defined the input to the future decision support system for assay selection to be of a tabular form, with rows corresponding to GMOs, and columns corresponding to assays they react to. In this paper we present a prototype decision support system for GMO detection and identification.

1 INTRODUCTION

A genetically modified organism (GMO) is an organism whose genetic material (Figure 1) has been altered using modern genetic engineering techniques (known as recombinant DNA technology). The general principle of producing a GMO is to artificially modify the genetic material of an organism's genome in order to give it a new property (a plant's resistance to a disease or insect, improvement of the nutritional value, increased crop productivity, a plant's tolerance to a herbicide, etc.)

Modern biotechnology has many applications in the pharmaceutical and agri-food industries. One example is the use of GMOs in the food production chain. In order to ensure that this development of modern biotechnology, specifically of GMOs, takes place in accordance with precautionary principle [1], the European Union has established a legal framework comprising various acts [1, 2, 3, 4, 5, 6]. In this paper we focus on [4] and [6] on GMO traceability and labeling of food and feed produced from GMOs.

The growing number of GMO allowed in the European Union and in the rest of the world and the need of inspection services to be able to effectively assess whether the regulatives and directives are being respected have lead to a problem of minimizing laboratory costs for testing samples of food or feed for GMO presence, GMO identification and GMO quantification.

In this paper we present a prototype decision support system [7] called GMOtrack. The goal of GMOtrack is to support laboratory work on GMO traceability. It evaluates possible assay sets for a given sample of food of feed and suggesting those that would minimize the cost of testing for GMO presence and GMO identification. It also allows "what if" analysis.

This paper is organized as follows: Section 2 presents the biological background necessary to understand the data and the GMO traceability problem. Section 3 is dedicated to the description of the GMO traceability problem. In Section 5 we present the prototype of the decision support system for GMO traceability. It is followed by conclusions and further work in Section 5.

2 BIOLOGICAL BACKGROUND

Deoxyribonucleic acid, or DNA, is a nucleic acid that contains the genetic instructions used in the development and functioning of all known living organisms. The main role of DNA molecules is the long-term storage of information, since it contains the instructions needed to construct other components of cells, such as proteins and RNA molecules. The DNA segments that carry this genetic information are called genes, but other DNA sequences have structural purposes, or are involved in regulating the use of this genetic information. Since DNA is a very stable molecule, it can be detected in processed food and other products of biological origin.

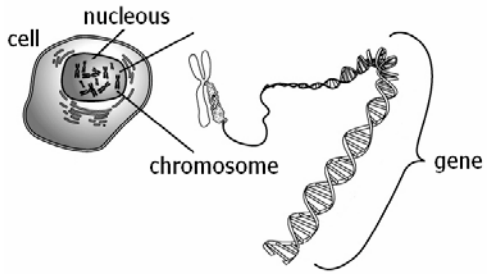

Figure 1: *Schematic representation of a genome in a cell.*

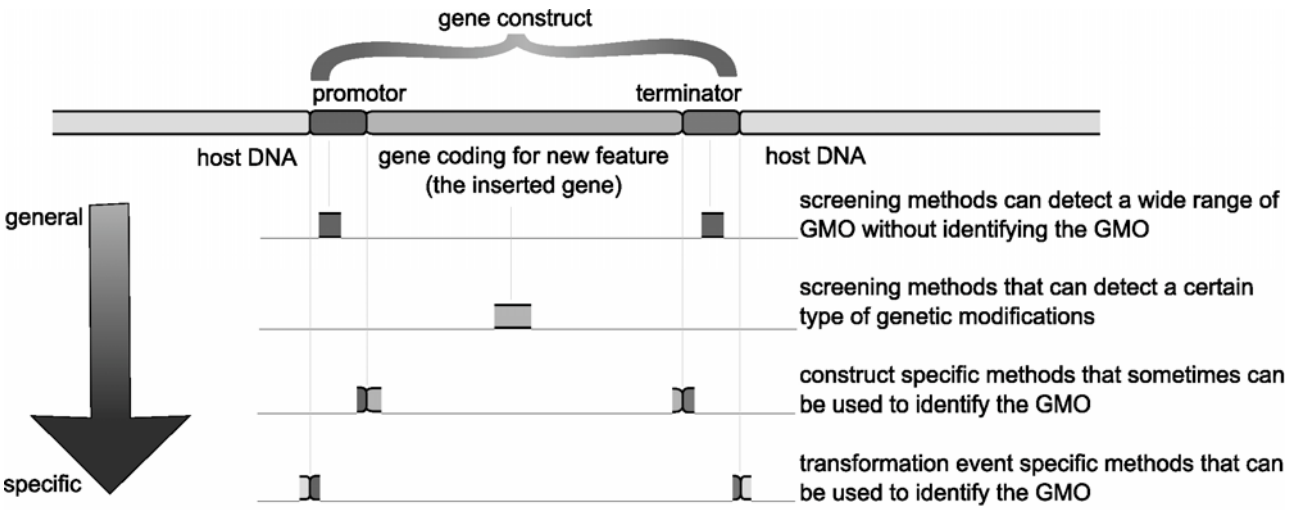

Figure 2: *Targets for PCR based detection of genetic modifications, ordered according to specificity.*

A gene construct is a functional unit necessary for the expression of a gene of interest. Apart from the gene of interest itself, a so-called promoter ("starter") and a terminator ("stop signal") are required for expression. In most cases, additional sequences are included, e.g. marker genes, which are also accompanied by a promoter and a terminator. The name "construct" is used because the sequences normally do not exist in this combination, but must be "put together" (constructed). A schematic representation of a gene construct is depicted in the upper part of Figure 2.

Detection of a GMO or a derivative of a GMO can be done by detecting a DNA molecule that is specifically associated with or derived from the genetic modification of interest. DNA can be purified and multiplied in billions of copies in a few hours with a technique called PCR (polymerase chain reaction) [8]. There is normally a linear correlation between the quantity of GMO and DNA.

The specificity of currently available DNA based methods can be divided into four categories (Figure 2): 1) screening methods that can detect a wide range of GMO without identifying the GMO, 2) screening methods that can detect a certain type of genetic modifications, 3) construct specific methods that sometimes can be used to identify the GMO, 4) transformation event specific methods that can be used to identify the GMO. In addition PCR-based GMO analyses usually include testing for presence of DNA from the particular species of interest, e.g. soybean DNA.

3 THE GMO TRACEABILITY PROBLEM

All foods and feed produced from GMOs, including products that no longer contain detectable traces of genetic modification must be labeled. The threshold for adventitious presence for EU-approved varieties of GMOs for use in food and feed is set at 0.9 percent [4]. Above this level, all products must be labeled.

GMO tracking is a several steps process. After the inspector has acquired the sample and has brought it to a certified laboratory for testing, grinding, homogenization, DNA isolation and DNA quantification need to be done before applying a real-time PCR to qualitatively detect GMO presence and identify the present GMOs. If the presence of GMOs is confirmed, the quantification step is required to ascertain if GMO content is above the allowable adventitious presence level of 0.9 percent. The qualitative detection of GMO presence and identification of the present GMOs are the most expensive steps in the GMO traceability process. The identification of GMOs presence is becoming extremely expensive with increasing number of GMOs on the market. We believe it can be improved (lowered costs) by acquiring and systemizing the data about GMOs gene constructs and by using this data as the basis for an intelligent decision support system.

The decision support system GMOtrack, presented in this paper, helps the user to select a set of assays (real-time PCR tests) to be done for a specific sample. We believe that, by selecting the right set of tests according to the sample, we can lower the cost of the entire analysis.

There are several sources of data about GMOs that are on the market in the European Union and in the rest of the world [9, 10], but there is no unified database describing the gene constructs of these GMOs. We are gathering data about GMOs in a unified table (A small part of this table is shown in Figure 3).

The first line of our unified table is reserved for DNA elements and corresponding assays to detect them. Each following line is one GMO. In the first column we have the GMO names. The second column denotes the species. The following columns are “1” if the corresponding GMO contains the element and “0” if it does not.

Since real-time PCR is time consuming and due to the available technology, one sample is preferably tested for presence of several elements in parallel (at the same time).

In routine practice at most two iterations of assays are required. Our goal is to support the decision making process of deciding which assays to perform in each iteration depending on the sample in order to detect and identify GMOs.

GMOname	species	P35S	P-ract	P-4AS1	P-ubiZM1	P-TA29	P-NOS	P-CMoVb	P-FMV	P-PEPC	PSsuAra	P-PCDK
MON810	maize	1	0	0	0	0	0	0	0	0	0	0
MON 863	maize	1	0	1	0	0	0	0	0	0	0	0
RRS	soja	1	0	0	0	0	0	0	0	0	0	0
NK 603	maize	1	1	0	0	0	0	0	0	0	0	0
DAS1507	maize	1	0	0	1	0	0	0	0	0	0	0
GT 73 / RT	oilseed	0	0	0	0	0	0	0	1	0	0	0
Mon 1445	cotton	1	0	0	0	0	0	1	0	0	0	0
MON 531	cotton	1	0	0	0	0	1	0	0	0	0	0
T25	maize	1	0	0	0	0	0	0	0	0	0	0
GA 21	maize	0	1	0	0	0	0	0	0	0	0	0
Bt 11	maize	1	0	0	0	0	0	0	0	0	0	0
Bt176	maize	1	0	0	0	0	0	0	0	1	0	1
MS8	oilseed	0	0	0	0	1	0	0	0	0	1	0
RF3	oilseed	0	0	0	0	1	0	0	0	0	1	0
MS1	oilseed	0	0	0	0	1	1	0	0	0	1	0
RF1	oilseed	0	0	0	0	1	1	0	0	0	1	0

Figure 3: *A part of the unified GMO table.*

The cost is defined as follows: all the assays have the same price. Doing more assays in parallel lowers the costs. The chart depicting cost grows related to the number of assays is presented in Figure 4.

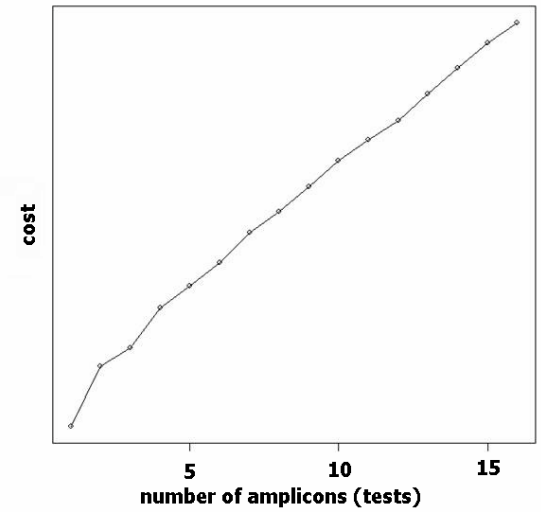

Figure 4: *A chart depicting costs relative to the number of assays.*

4 A PROTOTYPE DECISION SUPPORT SYSTEM FOR GMO TRACEABILITY

We have implemented a prototype decision support system program for GMO traceability called GMOtrack. After loading the data, the user selects the species contained in his sample. In the screenshot in Figure 5, oilseed rape and potato are the selected species. The minimum coverage constraint is by default set to 100 percent meaning that the user is interested in only those assay sets that detect the presence of all GMOs of the selected species. The user could also set a lower coverage constraint. The user then sets the maximum number of assays at the first level constraint and starts the computation by pressing the "Compute" button. In the screenshot in Figure 5 this constraint is set to 4. The program generates assay sets and evaluates them according to their GMO detection and GMO identification power. It then outputs the assay tests sorted from the best to the worst.

The user can then browse through the proposed assay sets. In Figure 5 the user has chosen the assay set {P35S, PSsuAra, gox247 and EH92-527-1} (highlighted). The program then shows what different outcomes of these assays would mean.

Example 1:

- The outcome P35S=0, PSsuAra=0, gox247=0 and EH92-527-1=0 would mean that the analyzed sample does not contain any GMO. No further analysis is needed.

Example 2:

- The outcome P35S=0, PSsuAra=0, gox247=1 and EH92-527-1=0 would mean that the sample contains exactly one GMO which has been identified. No further analysis is needed.

Example 3:

- The outcome P35S=1, PSsuAra=0, gox247=1 and EH92-527-1=1 would mean that two GMOs have been identified in the sample and that seven other GMOs could be present in the sample. In this case, further analysis is needed to asses the presence of other GMOs.

The user could then further investigate which GMOs are identified with every outcome of and which tests, if any, need to be done in the second phase.

5 CONCLUSIONS AND FURTHER WORK

GMO traceability is a very important issue in the modern world. Besides allowing the consumer to choose the products based on GMO content, there are several other applications of GMO traceability, like environmental impact.

In this paper we have presented a prototype program GMOtrack which is a decision support system for optimization of two of the most expensive steps in GMO tracking - GMO detection and GMO identification.

Acknowledgement

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non-GM suppy chains: their CO-EXistence and TRAceability".

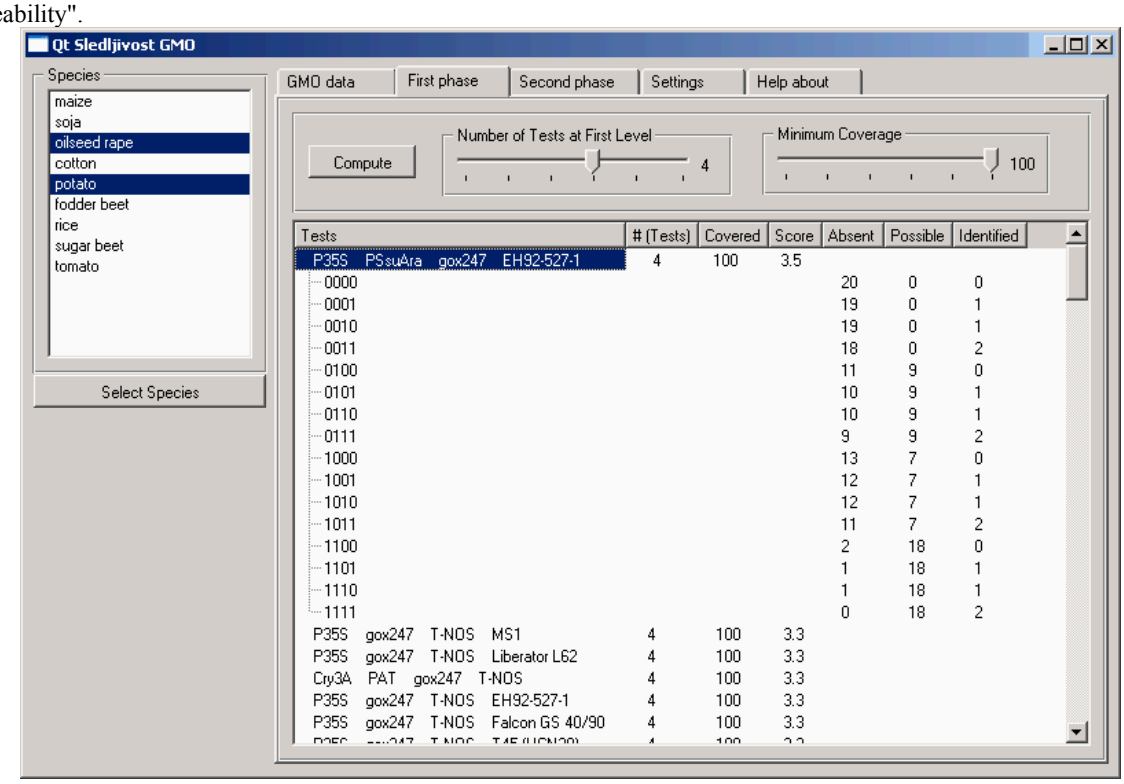

Figure 5: *Screenshot of the GMOtrack prototype decision support system program.*

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