

Detection of Genetically Modified Foods Existence in Egypt Markets

Fatma Elzahraa, I. Abdel rahman, M. M. M. Bekhit¹; M. S. Abdel Sabour¹; T. A. Alakkad¹ and S. D. Ibrahim²

1. Dept. of Genetics and Genetic engineering, Faculty of agriculture, Benha University.
2. Agricultural Genetic Engineering Research Institute (AGERI), Agriculture Research Center, Giza, Egypt

Corresponding author: Makhoulf.bakhit@fagr.bu.edu.eg

Abstract

Analysis for some field crops, feed and food products found in the local markets of Egypt was applied for GMOs detection using molecular technique based on PCR. DNA was extracted from samples including three groups italic fresh samples (egg plant, guava, olive, mango, tomato, potato and orange), cereals and seed samples (wheat, barley, broad bean, chickpea, corn, lentils, lima bean, soybean) and food products (bechmel, beef, chicken soup, chips, feta cheese, indomie, jelly, popcorn and powder milk), followed by tests to ensure purity of DNA samples using specific primers. GMO detection in the studied samples was conducted using specific primers for the most commonly used genes in genetic transformation such as 35S promoter from Cauliflower mosaic virus and Nopaline synthase terminator (T-Nos), using the following plasmids as positive control: PGIIMH35S-g2ps1 and pBI221. Tests revealed existence of GMOs in some imported products such as pudding, Jelly, popcorn and sauce due to the presence of 35S promoter and NOS terminator in soybean, while tests showed negative results in the local crops tested.

Key words: Genetically modified foods, GMO, 35S promoter, Egypt

Introduction

With the advent of the age of plant genetic engineering and with the first experience to production Flavr-Savr tomatoes which released commercially in 1994, The start of global and research interest in increasing crops produced by genetic engineering to overcome many of the problems faced some crops as well as increasing the productive efficiency of many crops (Yuan *et al.*, 2018). Currently, we can say that the increase in the area planted with GMOs was in the last few years similar to the explosion. The area planted with GMOs today reached about 189.8 million hectares worldwide in 2017. About 80% of GMOs represent the main part of food and feedstuff, such as soybean, corn and rapeseeds (Becker and Ulrich, 2018). According to some statistics, more than 40% of corn, more than 50% of cotton, more than 45% of soybean and at least two-thirds of all U.S.A food products contain GMOs (Qian *et al.*, 2018). Until November 2018 there are 498 GMOs are approved in 30 crops in 44 nations for cultivation with 46 varieties, 229 maize cultivars, 61 cotton cultivars, 40 soybean cultivars and 48 potato cultivars (ISAAA, 2018 and Gao *et al.*, 2019).

Globally, there are more than 50 nations and regions have fortified control and management of GMOs and release GMOs labeling regulations (Li *et al.*, 2019). GMOs were allowed in the United States, Canada and Argentina widely. By contrast, the situation was completely different in the European Union (EU) and there a conclusive rejection and strict official legislation to the GMOs (Briefs, 2017 and Sánchez-Paniagua *et al.*, 2018). This indicates that the safety of GMOs is still indecisive, so

different threshold labelling levels were set in different countries to vary from 0 to 5 %. Some of them were obligatory (e.g., Australia, Brazil, Chile, China, EU, India, etc.), others were optionally (e.g., Canada, Argentina, U.S, etc) (Qian *et al.*, 2018). The EU is one of the first to show clear opposition to GMOs globally and the consequent development of strict regulations and laws to track GMOs from the field to the counter. To protect the consumers freedom of choice, the European Union has approved obligatory labeling of food or feed to denote it as GMOs (; Regulation, 2003a; Regulation, 2003b and Gao *et al.*, 2019). While GMOs bring us great benefits, their prospective risks stay controversial. These arguments can be summarized as two parts of life health and environmental safety, including whether it is toxic, whether it will cause gene drift, whether it is sensitized, whether it will produce antibiotic resistance and whether it will destroy biodiversity, etc (Fraiture *et al.*, 2015 and Fraiture *et al.*, 2017). It should be noted that EU and US legislation and regulations are the yardsticks for developing countries that do not have specific regulations that define a specific approach to dealing with genetically modified foods.

On a local scale, as reported by James (2015) the situation of the production of genetically engineered crops in Egypt is still in the research phase and experimental and has not yet reached the adoption and generalization GMOs area in Egypt does not exceed 1% in comparison with USA 43% and Brazil 19% they are the largest countries to production GMOs globally.

Therefore, the biggest challenge for researchers is to find and update accurate methods for detecting GMOs. Recently, there are many complex multistage

which based on PCR have been established for detection and scanning of GMO. Initially, screening experiments target specific DNA sequences which studies have referred to that use often in plant transformation. If the results are positive, after screening must be some construct-specific assays are done, and then event-specific identification and quantification of the GMO (Holst-Jensen *et al.*, 2003; Waiblinger *et al.*, 2010 and Becker and Ulrich, 2018;).

At present, many studies mentioned that the most regulatory elements which widely used are the 35S promoter and NOS terminator have been used considerably to regulate expression of targeted transgenes with current commercialized GMOs by more than 86.3%. The 35S promoter derived from cauliflower mosaic virus (CaMV) as a primary target in GMOs screening which is found in approved GMOs by at least 65% and higher than that in commercially important transgenic crops (about 126 GMOs events). and the NOS terminator which isolated from nopaline synthase gene (NOS) of *Agrobacterium tumefaciens*, and newly used with about 90 GMOs events. So, these elements are used often to screening GMOs and routine screening strategies for GMOs (Hull *et al.*, 2000; Oraby *et al.*,

2005;Kok *et al.*, 2005;Wu *et al.*, 2014; Datukishvili *et al.*, 2015; Gao *et al.*, 2019 and Li *et al.*, 2019).

Therefore, the study was planned to detect the genetically modified foods directly obtained from the markets in Egypt.

Materials and methods:

Samples collection:

In our study, there was a great challenge for the team to develop a sampling plan due to the great variety of food samples in the Egyptian market of foreign origin or imported foodstuffs from abroad which are likely to be genetically engineered. Therefore, the sampling strategy was developed to include the division of the samples to be grouped into three groups, each of which had similar characteristics: the fresh food group which included 30 samples, the grains and seeds group which included 35 samples and finally the processed food group which included 12 samples. So that samples are collected from three governorates, namely, Cairo, Qalyubia and Menoufia governorate. This is due to the fact that these three provinces are a major center for the production and distribution of fresh and processed foodstuffs to the rest of the Egypt.

Table 1. Samples of three groups (G1, G2 and G3) collected from different location in Egypt.

G1. Fresh samples	G2. Cereals and seeds	G3. Food products
Berry	American wheat	Bechamel
Courgette	Arugula	Beef
Dates	Barley seeds	Chicken Soup
Eggplant	Broad bean seeds	Chips
Guava	Bulgarian wheat	Feta cheese
Lemon 1	Celery seeds	Indomie
Lemon 2	Chickpea	Jelly
Loquat	Coriander seeds	popcorn
Mallow	Corn	Powder milk
Mango	Courgette seeds	Powder milk
Moringa	Cowpea seeds	Pudding
Olive 1	Cucumber seeds	Sauce
Olive 2	Dill seeds	
Orang 1	fenugreek seeds	
Orange 2	Leek seeds	
Orange 3	Lentils	
Papper 1	lima bean	
Papper 2	Luffa seeds	
Papper 3	Lupin	
Plum	Mallow	
Pomegranate	mallow seeds	
Potato	Parsley seeds	
Purslane	peas seeds	
Radish	Polish wheat	
Squash	Popcorn	
Sweet potato	Radish	
Tangerine	Russian wheat	
Tomato	Soy bean 1	
Watercress	Soy bean 2	
Watermelon	Spinach beet seeds	
	Spinach seeds	

Tomato seeds
Turnip seeds
Ukrainian wheat
Watermelon seeds

Table 2. Plasmid names which used as positive in this study

Plasmid name	The genetical constitution of the plasmid
PGIIMH35-2PS1	35S pro+T-NOS+....+....
pBI-221	35S pro+T-NOS+....+....

DNA Extraction method:

Despite there are many methods used in the separation of DNA, we adopted in this research the method according to **Porebski et al. (1997)** with some modifications. Taking into consideration the respect the scientific requirements in terms of standardization of the weight of the samples in proportion to the total solids of the sample, good homogenize and the unify all factors that may affect the accuracy of the results obtained.

Estimation of DNA concentration:

The DNA concentration was estimated after extraction by depending on a Nanodrop UV/Vis spectrophotometer and agarose gel methods. All DNA samples were quantified using Nano-drop method according **Healey et al. (2014)**. the quality was also measured by running 2 µl of the DNA extracted from each sample understudied on 0.7% agarose gel in comparison to 5µl of a DNA size marker (100bp DNA ladder). To estimate DNA concentration, compare the degree of fluorescence of the DNA sample with the different bands in DNA size marker (**Oraby et al., 2005 and Wang and Fang, 2005**).

Reproduction of DNA by using PCR device.

PCR reactions: the PCR amplification reactions were carried out as mentioned by **Oraby et al. (2005)**. Reactions were performed in 25 µl volume composed of 1x reaction buffer, 0.2 mM of dNTPs,

1.5 mM MgCl₂, 0.2 µM of each primer, 1 unit of Taq polymerase and 40 ng of template DNA, in sterile distilled water.

Thermocycling profile and detection of the PCR products: PCR amplification of the DNA was performed in a Perkin Elmer thermal cycler 9700 programmed to fulfill 42 cycles. The temperature profile in the different cycles was as follows: an initial strand separation cycle at 94°C for 5 min followed by 40 cycles comprised of a denaturation step at 94°C for 1min, an annealing step at 36°C for 1 min and an extension step at 72°C for 2 min. The final cycle was a polymerization cycle for 7 min at 72°C.

GMO scanning: PCR products were loading by electrophoresis in a 1.5% agarose gel containing ethidium bromide (0.5 mg/ml) in 1 x TBE buffer at 120 volts. A 100bp DNA ladder was used as molecular size standard. PCR products were visualized under UV light and documented using a TMXR+ Gel Documentation System (Bio-Rad).

GMO screening methods:

In this study, we depend on the most favorable candidates for screening GMO method according to **Ruttink et al. (2010)**, promoter (35S) and terminator (NOS) sequence. The cauliflower mosaic virus (CaMV) 35S promoter and the nonpalin synthase (nos) terminator are the common genetic elements in most genetically modified foods worldwide (**Alasaad et al., 2016**)

Table 3. Primer sequences and size of amplification products

Primer	Sequence 5'-3'	Amplicon length (bp)	Annealing temp (°C)	Reference
35S promoter	AAAGATGGACCCCCACCCAC GAGGAAGGGTCTTGCGAAGG	195 or 390	54	Blake et al. (1991) Oraby et al. (2005)
NOS terminator	CTGTTGCCGGTCTTGCGATGAT CCGCGCGGATAATTTATCCTAG	180	54	Hemmer (1997) Oraby et al. (2005)

Results and discussion

So far, Egypt has not effectively acceded to international conventions that criminalize and prevent the circulation of genetically engineered foods that are not known to consumers. It has not been so far just letters from the Egyptian government to international organizations and recommendations from the latter to Egypt's accession to the World

Trade Organization as a state that prohibits the trade of genetically engineered foods without a statement of GMO products to maintain its consumers and provide greater safety. Egypt is one of the countries importing food and crops with 60- 70% of its annual consumption. Despite the growing global interest in the detection of genetically engineered foods and the development of standards and legislation to regulate the world trade market accordingly, the entry of

imported food in Egypt depends only on the food nutrition content and limits allowed for fungal and toxins without concern that the food is a genetic engineer or Not (**Khidr *et al.*, 2018**).

Extraction and amplification of DNA:

In order to ensure the realistic results of the samples used depending on the DNA fraction, this requires high precision and careful care in extracting, processing and purifying DNA molecules and making them suitable for complete experiment. The DNA fragment is affected by many factors that may lead to DNA amplification failure, such as compounds associated in the sample such as proteins, sugars, fat, polyphenols and chemicals such as CTAB. Not only that, there are other challenges that are the common food processing steps like grinding, mixing, extraction, refining or heating which can lead to an obstacle in getting the desired results optimally. Due to the large differences in the target samples, it is necessary to determine the optimal strategy for extracting and purifying DNA according to the type of sample. As it is no doubt that the best strategy for fresh samples may not suitable with the samples manufactured and so on. The CTAB protocol has a qualitative advantage with fresh samples compared to manufactured samples to obtain the best DNA properties. This protocol needs to be some modifications with processed foods to achieve the required properties in the extracted DNA.

DNA concentration ($ng\mu l^{-1}$):

In previous studies, there are many methods that studied to extraction of DNA (**Rogers and Bendich, 1985; Hemmer, 1997; Porebski et al., 1997; Jankiewicz et al., 1999; Son et al., 2009 and Sönmezoğlu and Keskin, 2015**), but in our study we were focus of the most accurate method to decrease the variation factors. DNA concentration and yield as in **Table (4)** showed that if we take an overview of the three groups, we will notice that the fresh sample was the highest amount of DNA generally, then the cereals and seeds and the lowest group was food products group. The fresh sample group give average $675.77ng\mu l^{-1}$ despite the extreme contrast between samples. The highest five concentration were

Courgette ($326 ng\mu l^{-1}$), Mango ($1409 ng\mu l^{-1}$), Moringa ($1385 ng\mu l^{-1}$), pomegranate ($1164 ng\mu l^{-1}$) and Watermelon ($1099 ng\mu l^{-1}$). While, the lowest five concentration were Dates ($13 ng\mu l^{-1}$), Guava ($75 ng\mu l^{-1}$), Olive1 ($110 ng\mu l^{-1}$), Purslane ($119 ng\mu l^{-1}$) and Olive2 ($123 ng\mu l^{-1}$).

In second group (cereals and seeds samples) showed that the average of DNA concentrate was $544.60 ng\mu l^{-1}$, it is significantly less than first group (fresh samples). The highest samples were rapeseeds ($1909 ng\mu l^{-1}$), Soy bean2 ($1740 ng\mu l^{-1}$), Broad bean seeds ($1733 ng\mu l^{-1}$), Cowpea seeds ($1646 ng\mu l^{-1}$) and Mallow ($1581 ng\mu l^{-1}$). On the other hand, the lowest samples were Lentils ($43 ng\mu l^{-1}$), Coriander seeds ($46 ng\mu l^{-1}$), Watermelon seeds ($102 ng\mu l^{-1}$), lima bean ($108 ng\mu l^{-1}$) and Celery seeds ($136 ng\mu l^{-1}$). Quite differently, the third group (food products) gave the lowest results, the third group average was $78.97ng\mu l^{-1}$. The highest samples were Feta cheese ($350 ng\mu l^{-1}$), Sauce ($121 ng\mu l^{-1}$) and Indomie ($114 ng\mu l^{-1}$). While, the lowest sample showed a significant decrease, Pudding ($0.2 ng\mu l^{-1}$), Jelly ($2.4 ng\mu l^{-1}$) and Bechamel ($22 ng\mu l^{-1}$). From previous results we note that, there are many reasons which lead to decrease of DNA concentration after amplification in PCR, by other meaning the extracted protein not able to amplification. In view of the previous studies (**Anklam *et al.*, 2002 and Gryson, 2010**), we find that many researchers touched on this subject explaining the reasons that may lead to this case. Perhaps one of the most striking reasons and most influential is heat treatments for samples during dehydration for cereals and seeds after harvested or heat processing for food products. These results are consistent with many previous studies such as **Oraby *et al.* (2005)** who mentioned that the concentration of the DNA extracted by the CTAB dependent method ranged from 86 to $1650 ng\mu l^{-1}$ in the tested samples. Concentration of the DNA extracted by using the kits ranged from 300 to $3100 ng\mu l^{-1}$. There are various factors that may contribute to the quality of the PCR products, such as the purity of the DNA extracted from the sample and the size and homogeneity of the sample. A representative sampling of any lot is also vital in attaining accurate results.

Table 4. DNA concentration

G1. Fresh samples		G2. Cereals and seeds		G3. Food products	
Sample	Conc.($ng\mu l^{-1}$)	Sample	Conc. ($ng\mu l^{-1}$)	Sample	Conc. ($ng\mu l^{-1}$)
Berry	598	American wheat	834	Bechamel	22
Courgette	326	Arugula	502	Beef	30
Dates	13	Barley seeds	138	Chicken Soup	51
Eggplant	902	Broad bean seeds	1733	Chips	32
Guava	75	Bulgarian wheat	165	Feta cheese	350
Lemon	483	Celery seeds	136	Indomie	114
Lemon 2	602	Chickpea	1209	Jelly	2.4
Loquat	461	Coriander Seeds	46	popcorn	65

Table 4 Cont.

Mallow	831	Corn	347	Powder milk	76
Mango	1409	Courgette seeds	358	Powder milk	84
Moringa	1385	Cowpea seeds	1646	Pudding	0.2
Olive 1	110	Cucumber seeds	262	Sauce	121
Olive 2	123	Dill seeds	265		
Orang	819	Fenugreek seeds	393		
Orange 2	379	Leek seeds	147		
Orange 3	466	Lentils	43		
Papper 1	834	Lima bean	108		
Papper 2	466	Luffa seeds	165		
Papper 3	551	Lupin	509		
Plum	178	Mallow	1581		
Pomegranate	1164	Mallow seeds	154		
Potato	412	Parsley seeds	162		
Purslane	119	Peas seeds	597		
Radish	558	Polish wheat	242		
Squash	154	Popcorn	404		
Sweet potato	978	Radish	706		
Tangerine	274	Russian wheat	281		
Tomato	967	Soy bean 1	870		
Watercress	603	Soy bean 2	1740		
Watermelon	1099	Spinach beet	314		
		Seeds			
		Spinach seeds	357		
		Tomato seeds	334		
		Rapeseeds	1909		
		Ukrainian wheat	302		
		Watermelon	102		
		seeds			
Average	675.77		544.60		78.97

Screening of Genetically Modified Foods in the Egypt Market:

Screening of first group (fresh food samples):

Vegetables and fruits are grown worldwide and make up a major portion of the diet of humans in many parts of the world. They play a significant role in human nutrition, especially as sources of vitamins (C, A, B1, B6, B9, E), minerals, dietary fiber and phytochemicals. Vegetables and fruits in the daily diet have been strongly associated with improvement of gastrointestinal health, good vision, and reduced risk of heart disease, stroke, chronic diseases such as diabetes, and some forms of cancer (Keatinge et al., 2010 and Ryder, 2011).

A world vegetable survey showed that 402 vegetable crops are cultivated worldwide, representing 69 families and 230 genera. Leafy vegetables of which the leaves or young leafy shoots are consumed were the most often utilized (53% of the total), followed by vegetable fruits (15%), and vegetables with below ground edible organs comprised 17%. Many vegetable crops have more than one part used. Most of the vegetables are marketed fresh with only a small proportion processed because most vegetables are perishable. Consumption shortly after harvest guarantees optimal vegetable quality (Dias and Ortiz Rios, 2014).

Through the results obtained for the first group, which includes the fresh samples of a random group of fruits and vegetables collected from several different places and after repeated tests several times. The results indicated the absence of any positive results with NOS terminator as in Figure (1) or 35S promoter as in Figure (2). To the best of our knowledge and reading, the number of researches conducted to detect genetically modified fresh vegetables and fruit is Very limited. Although, the earliest genetic modification experiments were for vegetables such as tomatoes and potatoes. According to (Kramer and Redenbaugh, 1994 nd Dias and Ortiz Rios, 2014;), the first commercially grown transgenic crop was FlavrSavr tomato, which was released by Calgene in 1994. This tomato contains an antisense version of the polygalacturonase (PG) gene. The use of this gene ensued after many years of research on several genes involved in fruit development and tomato ripening. They were identified, cloned, and characterized to breed transgenic tomato cultivars. However, FlavrSavr tomato failed in the market since this cultivar was considered inferior by growers and was rapidly withdrawn from the market. Plant genetic engineers learned an important lesson after this failure: the importance of cooperation with plant breeders. But here we should point out that when we examined

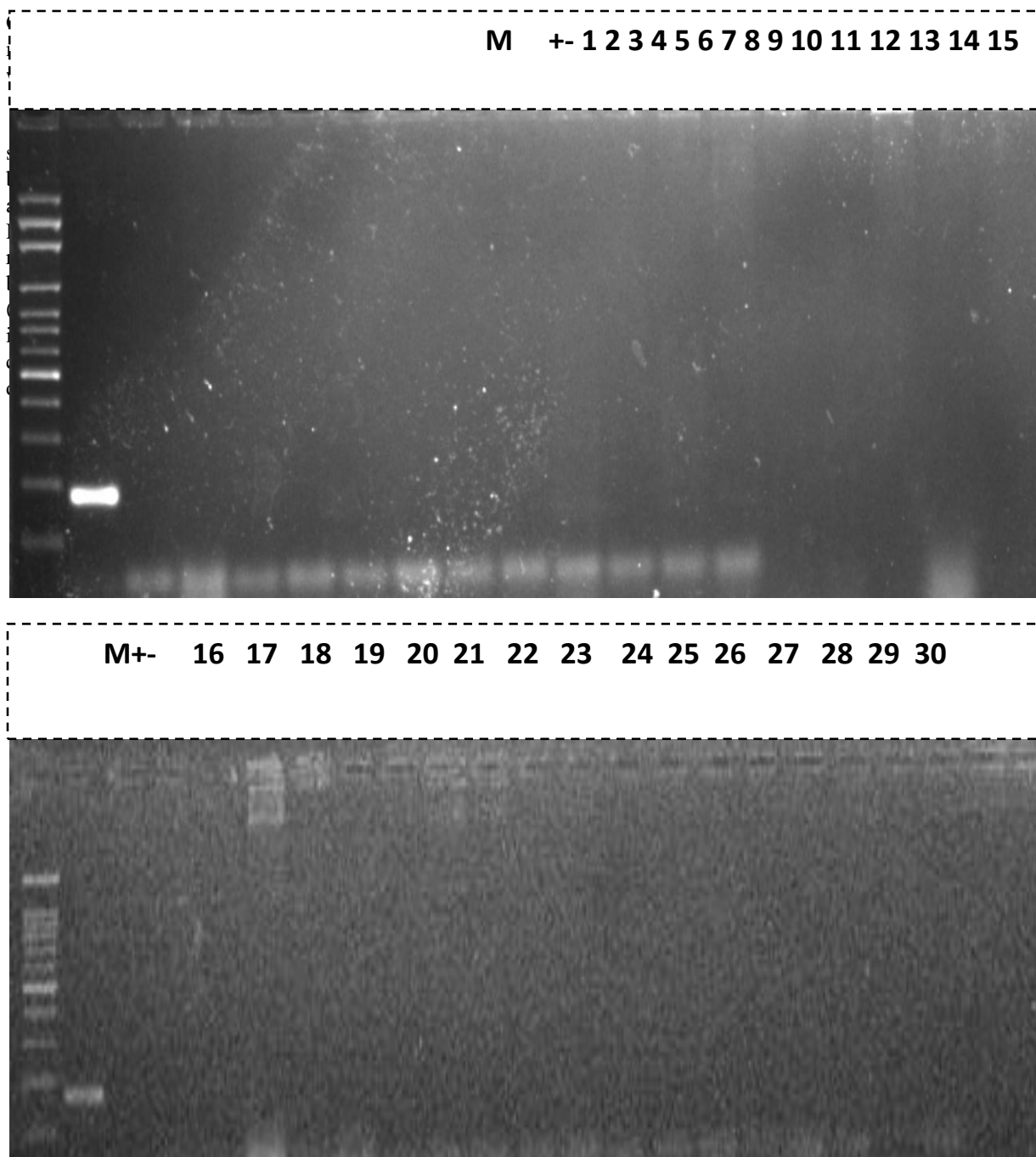


Fig (1): Detection of NOS terminator segment in fresh samples; with positive indicator(+), negative indicator(-) and M (Ladder 100 bp).

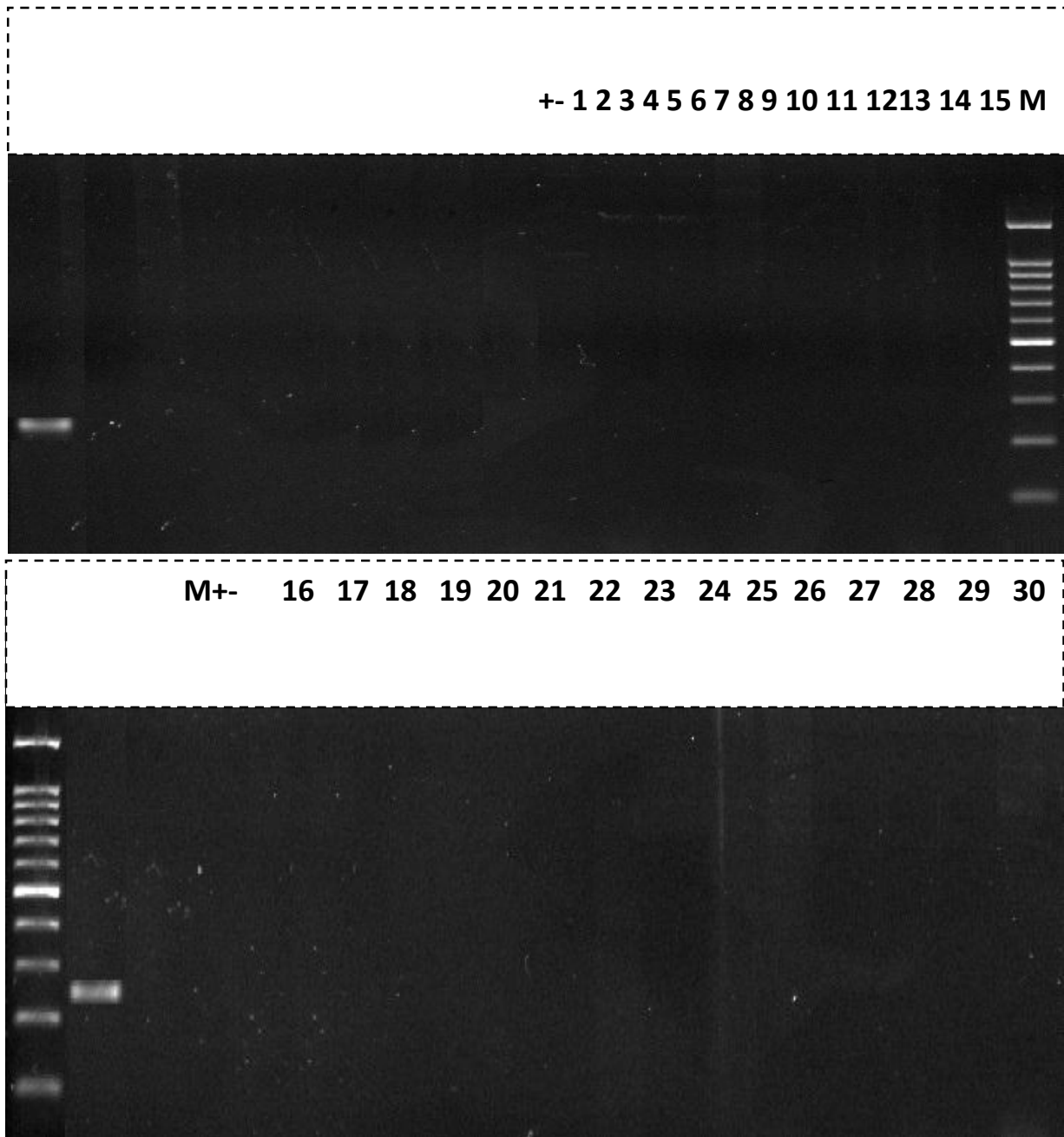


Fig (2): Detection of 35S promoter segment in fresh samples; with positive indicator (+) , negative indicator (-) and M (Ladder 100 bp).

Screening of second group (cereals and seed samples):

The second group aimed to study a group of the most common food materials in the diet for all age groups whether directly used or as raw or secondary raw material in many food products and processed foodstuffs. In order to achieve the purpose of this study, the samples of this group were collected from the largest grain and seed stores in Cairo and a larger number of the other two groups. Although the beginning of genetic engineering targeted vegetables as already mentioned, cereals and seeds have taken

the greatest interest in the following stages and now some genetically engineered cereals and seeds are grown in some countries by 100%.

The results indicated the absence of any positive results with NOS terminator as in **Figure (3)** or 35S promoter as in **Figure (4)**, this is despite the widespread proliferation of genetically engineered crops, which account for cereals and seeds with the largest share. According to **Viljoen *et al.* (2006)** GM crops accounted for 29% of global crop production, it is estimated globally that 56% of soybean, 28% of cotton, 19% of canola and 14% of maize is GM.

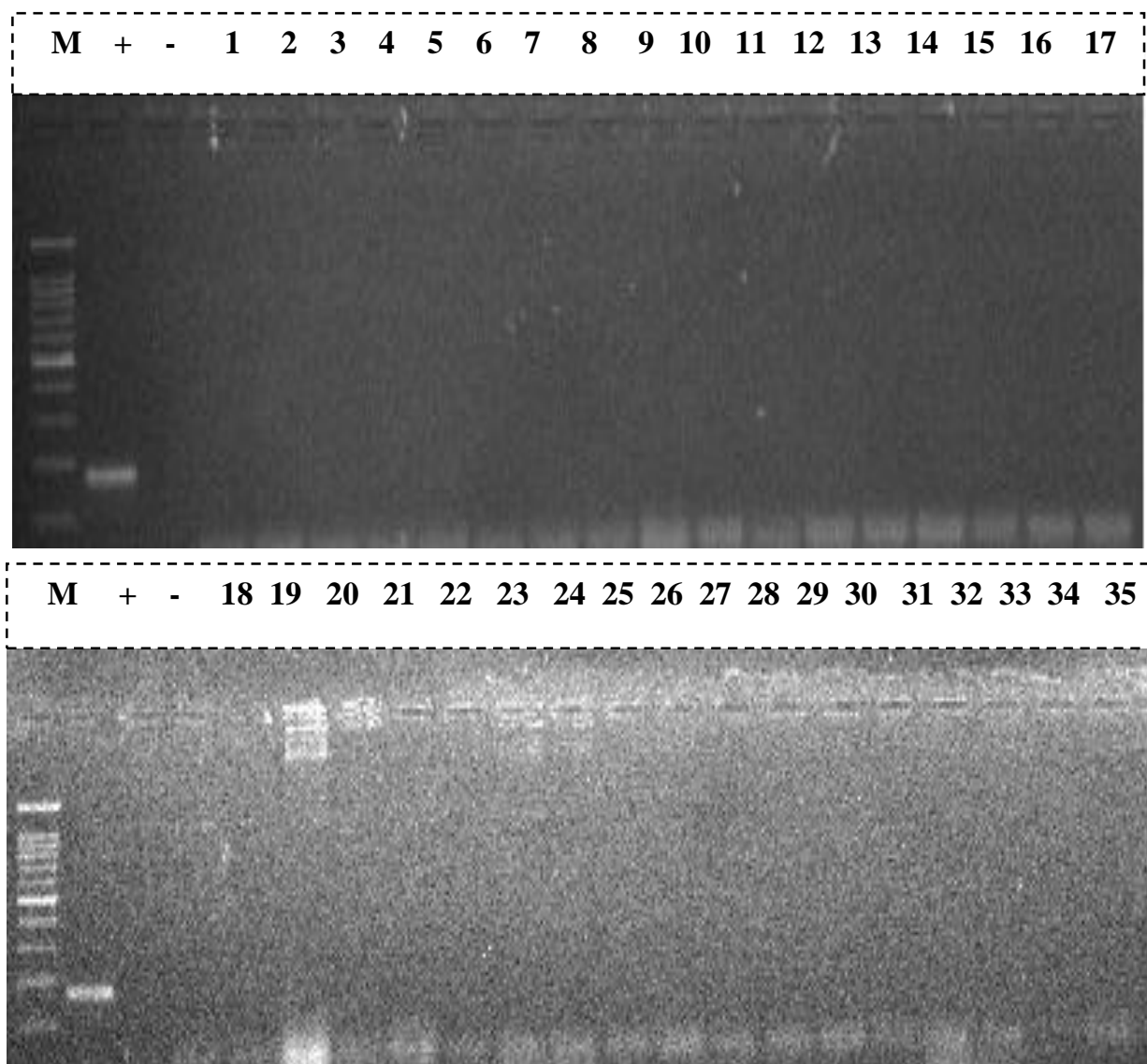


Fig (3): Detection of NOS terminator segment in cereals and seed samples; with positive indicator (+), negative indicator (-) and M (Ladder 100 bp).

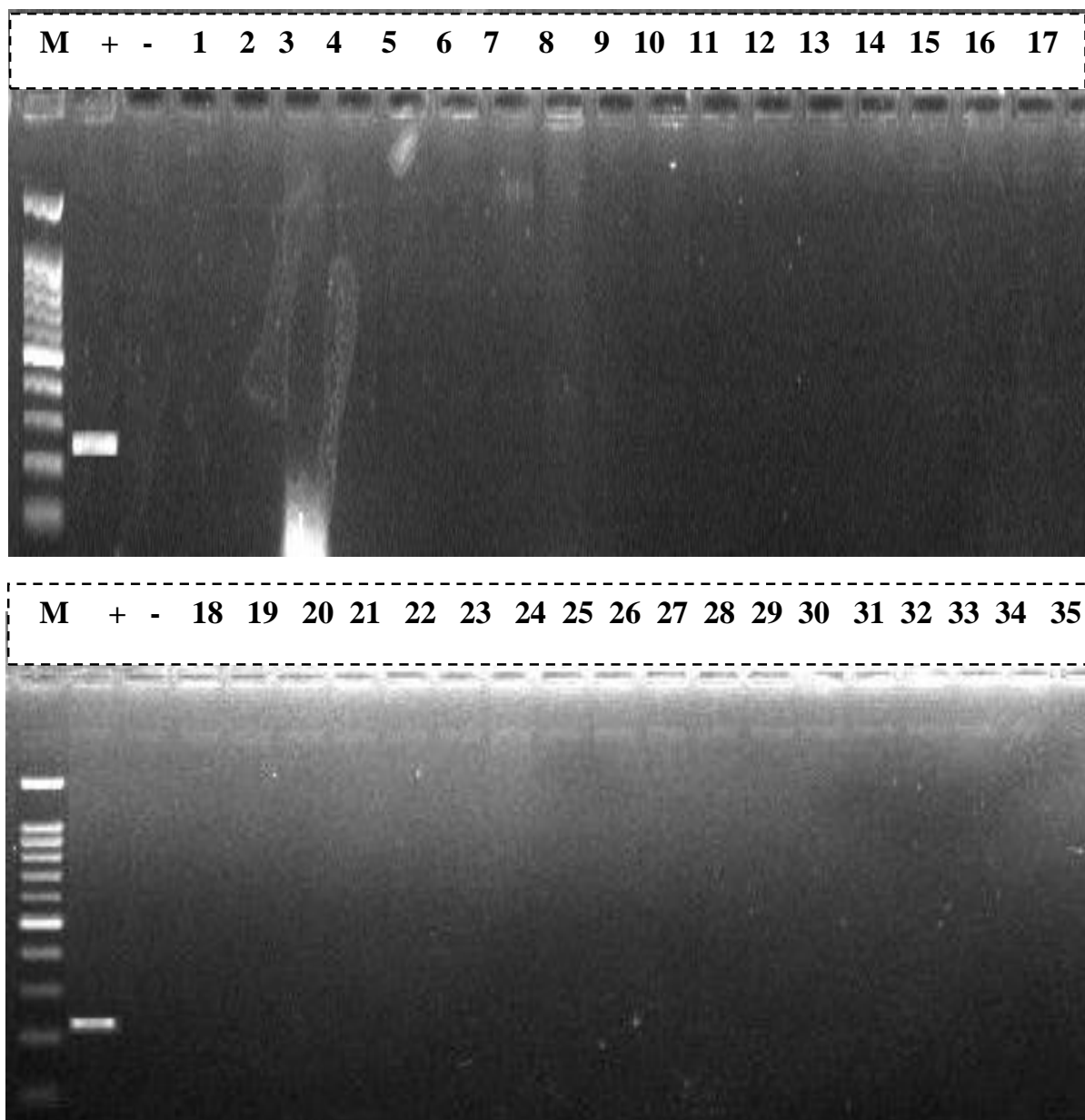


Fig (4): Detection of 35S promoter segment in cereals and seed samples; with positive indicator (+), negative indicator(-) and M (Ladder 100 bp).

Screening of third group (food products):

The availability of genetically engineered products in manufactured products increases because many products enter into their manufacture and thus increase the chance of having a more genetically modified gene component. In this group, the research team selected the imported food products to detect the genetically modified sequence. to provide a kind of safety and tranquility to the consumer for consumed the imported products.

The results as in **Figure (5 and 6)** which showed that there are **four** samples given a positive result with **NOS terminator** as in **Figure (5 and 7)** and **Table (4)** which represent 33% of the samples tested in third group. NOS terminator test showed a clear visible band at 118 pb with some imported samples like Pudding (Turkey), Jelly (United Arab Emirates) and popcorn (European Commission), which means that these samples are genetically modified using NOS terminator. On the other hand, there is one product which manufactured in Egypt (Tomato Sauce) is given a positive result with NOS terminator which due to there are some imported material or raw seeds used in producing this product. Whereas, there are some other samples which not have been shown any visible bands in NOS terminator range (118 pb), which mean that these samples are not genetically modified using NOS terminator. These samples contain some samples manufactured in Egypt like Powder milk, Chicken Soup, Indomie and Feta cheese, also there are some imported sample like Chips (Malaysia) and Beef (Brazil). Egypt is considered one of the countries that does not commit to the development of mandatory data on food packages that indicate whether they are genetically engineered or not. Despite the many risks expected from the use of genetically engineered materials, as has been mentioned in many studies around the world, which led to the adoption by some countries of reservations on the use of genetically engineered food or at least mandatory labeling indicating that food is a genetic engineer to ensure the right of the consumer to determine the selection of the use of genetically engineered foods or not to use them (**Fraiture et al., 2017; Gao et al., 2019 and Sánchez-Paniagua et al., 2018**). therefore, it was expected to find a lot of imported food genetically engineered. As our results NOS terminator (nopaline synthase gene of *Agrobacterium tumefaciens*) is

widely used as a genetically engineering element for one or more of food product ingredients for the products which manufactured in Egypt by using imported ingredients, there are two products that give positive result with NOS. Indomie product which have brand (Indomie) it is made in Egypt by Indofood Indonesian Company. This product is manufactured mainly from wheat flour, in our country, only about 40% of the annual consumption of wheat is produced domestically and is entirely directed to subsidized bread production, while private sector companies rely on imported wheat, including pasta and indomie producers (**Veninga and Ihle, 2018**).according to this result, there are about 45-55% of wheat and wheat flour in Egyptian markets are GMO by a certain percentage. The second positive GMO product manufactured in Egypt by some imported ingredients was Bechamel mix which manufactured by Helou El Sham Company. This product made by wheat flour, corn starch, soy protein, vegetable oil, salt and flavours. As it is known both of wheat flour, corn starch and soy protein are imported from different countries, therefore, these three ingredients are responsible for giving this positive result of GMO. The third positive GMO product manufactured in Egypt was Sauce product. On the other hand, the imported products which, given a positive GMO with NOS terminator were as following; i. Pudding product which manufactured in Turkey by Basak company, Basak company exports to Egypt more than 10 kinds of different products (by examining products in the market during sampling). In this study, we examined one product of the company's products and gave a positive result, and this raises a kind of concern about the other products produced by this company in terms of containing genetically modified materials. ii. Jelly product which manufactured by Greens UK company, imported from United Arab Emirates (UAE) and these products are available in the Egyptian market in many types of most types of instant sweets such as Semolina, Cake mix, White outs, Corn flour, Cheese cake, Carmella and dessert whip. iii. Popcorn product, which made by Bright star company in European Commission (EC) and imported by Nahrain for food Industries Company, this product is written free GMO. The main ingredient in this product is corn which, is the reason why there is a positive result of GMO.

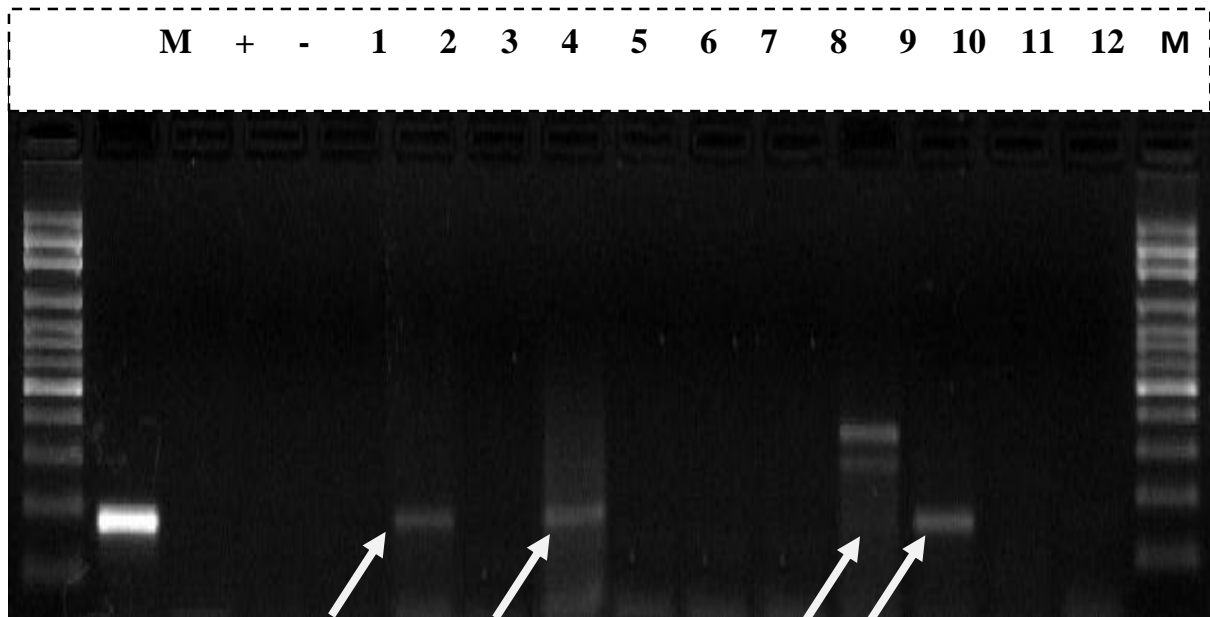
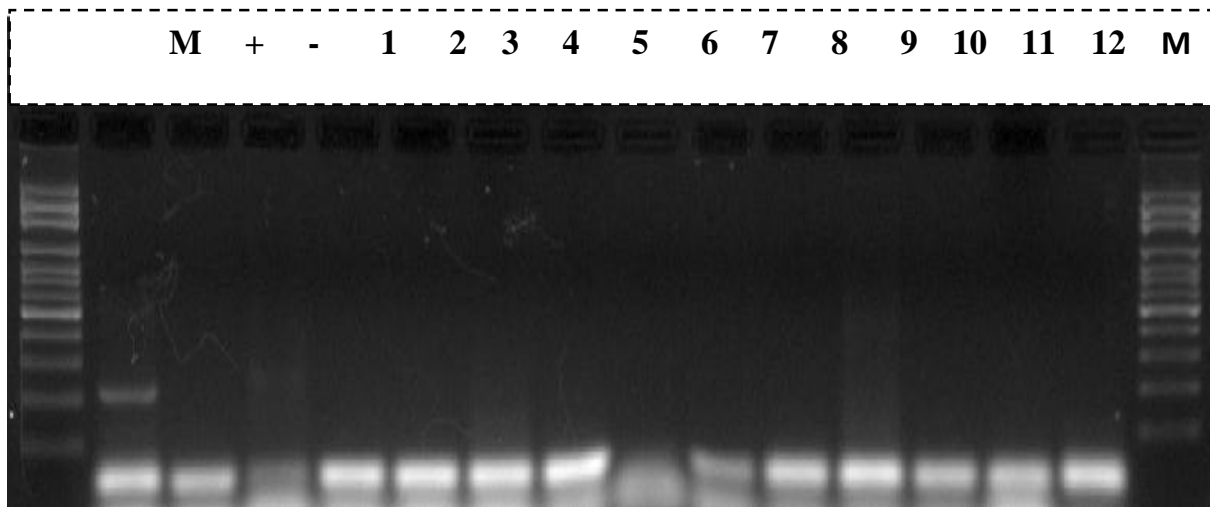


Figure (6): Detection of NOS terminator segment in food products samples; with positive indicator(+), negative indicator(-) and M (Ladder 100 bp).



Fig(5): Detection of 35S promoter segment in food products samples; with positive indicator (+), negative indicator (-) and M (Ladder 100 bp).

Table 4. Revealed the detected GMOs in the local markets of Egypt.

Sample code	Sample	Brand	Origin state	Detection	
				NOS	35S
PR 1	Powder milk	Milky gold	Egypt	Not detect	Not detect
PR2	Powder milk	Mirro	Egypt	Not detect	Not detect
PR3	Pudding	Custard	Turkey	Positive	Not detect
PR4	Bechamel	Ethery Queen	Egypt	Not detect	Not detect
PR5	Jelly	Jelly	UAE	Positive	Not detect
PR6	Chicken Soup	Chicken Soup	Egypt	Not detect	Not detect
PR7	Indomie	Indomie	Egypt	Not detect	Not detect
PR8	Chips	Chips	Malaysia	Not detect	Not detect
PR9	popcorn	Popcorn	EC	Positive	Not detect
PR10	Sauce	Sauce	Egypt	Positive	Not detect
PR11	Feta cheese	Cheesa	Egypt	Not detect	Not detect
PR12	Beef	Beef	Brazil	Not detect	Not detect

References

- Alasaad, N.; Alzubi, H. and Kader, A. A. (2016).** Data in support of the detection of genetically modified organisms (GMOs) in food and feed samples. *Data in brief*, 7: 243-252.
- Anklam, E.; Gadani, F.; Heinze, P.; Pijnenburg, H. and Van Den Eede, G. (2002).** Analytical methods for detection and determination of genetically modified organisms in agricultural crops and plant-derived food products. *European Food Research and Technology*, 214: 3-26.
- Becker, R. and Ulrich, A. (2018).** Improved detection and quantification of cauliflower mosaic virus in food crops: assessing false positives in GMO screening based on the 35S promoter. *European Food Research and Technology*, 1-11.
- Blake, N. K.; Ditterline, R. L. and Stout, R. G. (1991).** Polymerase chain reaction used for monitoring multiple gene integration in *Agrobacterium*-mediated transformation. *Crop science*, 31: 1686-1688.
- Briefs, I. (2017).** Global status of commercialized biotech/GM crops in 2017: biotech crop adoption surges as economic benefits accumulate in 22 years.
- Datukishvili, N.; Kutateladze, T.; Gabriadze, I.; Bitskinashvili, K. and Vishnepolsky, B. (2015).** New multiplex PCR methods for rapid screening of genetically modified organisms in foods. *Frontiers in microbiology*, 6: 757.
- Dias, J. S. and Ortiz Rios, R. O. (2014).** Advances in transgenic vegetable and fruit breeding.
- Fraiture, M. A.; Herman, P.; De Loose, M.; Debode, F. and Roosens, N. H. (2017).** How can we better detect unauthorized GMOs in food and feed chains? *Trends in biotechnology*, 35: 508-517.
- Fraiture, M. A.; Herman, P.; Taverniers, I.; De Loose, M.; Deforce, D. and Roosens, N. H. (2015).** Current and new approaches in GMO detection: challenges and solutions. *BioMed research international* 2015.
- Gao, W.; Tian, J.; Huang, K.; Yang, Z.; Xu, W. and Luo, Y. (2019).** Ultrafast, universal and visual screening of dual genetically modified elements based on dual super PCR and a lateral flow biosensor. *Food chemistry*, 279: 246-251.
- Gryson, N. (2010).** Effect of food processing on plant DNA degradation and PCR-based GMO analysis: a review. *Analytical and bioanalytical chemistry*, 396: 2003-2022.
- Healey, A.; Furtado, A.; Cooper, T. and Henry, R. J. (2014).** Protocol: a simple method for extracting next-generation sequencing quality genomic DNA from recalcitrant plant species. *Plant methods*, 10: 21.
- Hemmer, W. (1997).** "Foods derived from genetically modified organisms and detection methods," Agency BATS Basel.
- Holst-Jensen, A.; Rønning, S. B.; Løvseth, A. and Berdal, K. G. (2003).** PCR technology for screening and quantification of genetically modified organisms (GMOs). *Analytical and Bioanalytical Chemistry*, 375: 985-993.
- Hull, R.; Covey, S. and Dale, P. (2000).** Genetically modified plants and the 35S promoter: assessing the risks and enhancing the debate. *Microbial Ecology in Health and Disease*, 1: 1-5.
- ISAAA (2018).** Global status of commercialized biotech/GM crops in 2017: Biotech crop adoption surges as economic benefits accumulate in 22 Years. ISAAA Brief No. 53Ithaca, NY: ISAAA.
- James, C. (2015).** Global status of commercialized biotech/GM crops: 2014. ISAAA brief 49.
- Jankiewicz, A.; Broll, H. and Zagon, J. (1999).** The official method for the detection of genetically modified soybeans (German Food Act LMBG § 35): a semi-quantitative study of sensitivity limits with glyphosate-tolerant soybeans (Roundup Ready) and insect-resistant Bt maize (Maximizer). *European Food Research and Technology*, 209: 77-82.
- Keatinge, J. D.; Waliyar, F.; Jamnadas, R. H.; Moustafa, A.; Andrade, M.; Drechsel, P.; Hughes, J. d. A.; Kadirvel, P. and Luther, K. (2010).** Relearning old lessons for the future of

- food—by bread alone no longer: diversifying diets with fruit and vegetables. *Crop Science*, 50: S-51-S-62.
- Khidr, Y.; Arafa, M.; ELDEMERY, S. M. and Elsanhoty, R. (2018).** detection of genetically modified potato in pota-to tubers cultivated in egypt. *Egyptian Journal of Genetics And Cytology*, 47.
- Kok, E.; Scholtens, I.; Van Hoef, A. and Aarts, H. (2005).** Detection of genetically modified DNA in food. 2000. Capturado em 10.
- Kramer, M. G. and Redenbaugh, K. (1994).** Commercialization of a tomato with an antisense polygalacturonase gene: The FLAVR SAVR™ tomato story. *Euphytica*, 79: 293-297.
- Li, R.; Shi, J.; Liu, B.; Wang, C.; Zhang, D.; Zhao, X. and Yang, L. (2019).** Inter-laboratory validation of visual loop-mediated isothermal amplification assays for GM contents screening. *Food chemistry*, 274: 659-663.
- Oraby, H. A.; Hassan, A. A., and Abou Mossallam, A. A. (2005).** Screening food products for the presence of CaMV 35S promoter and NOS 3' terminator. *Journal of the Science of Food and Agriculture*, 85: 1974-1980.
- Porebski, S.; Bailey, L. G. and Baum, B. R. (1997).** Modification of a CTAB DNA extraction protocol for plants containing high polysaccharide and polyphenol components. *Plant molecular biology reporter*, 15: 8-15.
- Qian, C.; Wang, R.; Wu, H.; Ping, J. and Wu, J. (2018).** Recent advances in emerging DNA-based methods for genetically modified organisms (GMOs) rapid detection. *TrAC Trends in Analytical Chemistry*.
- Regulation, C. (2003).** No 1830/2003 of the European Parliament and of the Council of 22 September 2003 concerning the traceability and labelling of genetically modified organisms and the traceability of food and feed products produced from genetically modified organisms and amending Directive 2001/18/EC. *Official Journal* 50, 268.
- Regulation, E. (2003b).** No. 1829/2003 of the European Parliament and of the Council of 22nd September (2003) on genetically modified food and feed. *Official J Eur Union Lt* 268, 1-23.
- Rogers, S. O. and Bendich, A. J. (1985).** Extraction of DNA from milligram amounts of fresh, herbarium and mummified plant tissues. *Plant molecular biology*, 5: 69-76.
- Ruttink, T.; Demeyer, R.; Van Gulck, E.; Van Droogenbroeck, B.; Querci, M.; Taverniers, I. and De Loose, M. (2010).** Molecular toolbox for the identification of unknown genetically modified organisms. *Analytical and Bioanalytical Chemistry*, 396: 2073-2089.
- Ryder, E. (2011).** World vegetable industry: production, breeding, trends. *Hortic Rev*, 38: 299.
- Sánchez-Paniagua López, M.; Manzaneres-Palenzuela, C. L. and López-Ruiz, B. (2018).** Biosensors for GMO testing: nearly 25 years of research. *Critical reviews in analytical chemistry*, 48: 391-405.
- Son, R.; Raha, A. R.; Oi, M. L. and Wong, C. M. V. L. (2009).** Comparison of DNA extraction efficiencies using various methods for the detection of genetically modified organisms (GMOs). *International Food Research Journal* 16.
- Sönmezoğlu, Ö. A. and Keskin, H. (2015).** Determination of genetically modified corn and soy in processed food products. *Journal of Applied Biology & Biotechnology*, 3: 032-037.
- Veninga, W., and Ihle, R. (2018).** Import vulnerability in the Middle East: effects of the Arab spring on Egyptian wheat trade. *Food Security*, 10: 183-194.
- Viljoen, C.; Dajee, B. and Botha, G. (2006).** Detection of GMO in food products in South Africa: Implications of GMO labelling. *African journal of biotechnology*, 5: 73-82.
- Waiblinger, H.U.; Grohmann, L.; Mankertz, J.; Engelbert, D.; and Pietsch, K. (2010).** A practical approach to screen for authorised and unauthorised genetically modified plants. *Analytical and Bioanalytical Chemistry*, 396: 2065-2072.
- Wang, W.Y. and Fang, T. J. (2005).** Development of multiplex and quantitative PCR assay to detect genetically modified roundup ready soybean in foods. *Journal of Food and Drug Analysis* 13.
- Wu, Y.; Wang, Y.; Li, J.; Li, W.; Zhang, L.; Li, Y.; Li, X.; Zhu, L. and Wu, G. (2014).** Development of a general method for detection and quantification of the P35S promoter based on assessment of existing methods. *Scientific reports*, 4: 7358.
- Yuan, X.; Zhang, Y. Y.; Palma, M. A. and Ribeca, L. A. (2018).** Understanding Consumer response to GMO Information. In "2018 Annual Meeting, February 2-6, 2018, Jacksonville, Florida". Southern Agricultural Economics Association.

الكشف عن وجود اغذية معدلة وراثيا في الاسواق المصرية

ا.د. مخلوف محمد محمود بخيت - ا.د. محمد سراج الدين عبد الصبور - د. تامر أحمد العقاد د. شفيق درويش ابراهيم
 قسم الوراثة والهندسة الوراثية. كلية الزراعة. جامعة بنها قسم الوراثة والهندسة الوراثية. كلية الزراعة. جامعة بنها
 قسم الوراثة والهندسة الوراثية. كلية الزراعة. جامعة بنها
 معهد بحوث الهندسة الوراثية الزراعية (اجيرى), الجيزة. مصر

في السنوات الأخيرة ، تعرضت الكائنات المعدلة وراثيًا (GMOs) للتدقيق لأنها مرتبطة بالإمداد الغذائي البشري. يؤكد العلماء المشجعون للكائنات المحورة وراثياً على إمكانات الهندسة الوراثية كقائدتها المهمة للمجتمع ، على سبيل المثال ، زيادة المحصول أو تحسين الجودة واستحداث المقاومة للأمراض الفطرية والبكتيرية والفيروسية. ومع ذلك ، فإن تحذير الآخرين من استخدام الكائنات المعدلة وراثيا ، يمكن أن يسلط الضوء على صحة الإنسان وخاصة الحساسية ومقاومة المضادات الحيوية. كان هناك أيضا قلق بشأن القوانين الأخلاقية والاجتماعية للهندسة الوراثية. مقارنة بأوروبا ، لا تزال مصر خالية من الأغذية المعدلة وراثيا باستثناء المنتجات الغذائية المصنعة المستوردة. اليوم ، تم اختبار عينات من الغذاء الآن بشكل روتيني لمحتوى الكائنات المعدلة وراثيا. وتبين الدراسة نتائج اختبار أكثر من مائة من المنتجات الغذائية باستخدام PCR لمحتوى الكائنات المعدلة وراثيا. تم فحص محتوى الكائنات المعدلة وراثيا من الأطعمة المستوردة المختلفة ، مثل مجموعة الأغذية الطازجة بما في ذلك 30 عينة ، مجموعة الحبوب والبقول والبذور بما في ذلك 35 عينة وأخيرا مجموعة الأطعمة المصنعة (12 عينة) بما في ذلك خلال الفترة من 2016/2017 الى 2017/2018. من بين 77 عينة من جميع المنتجات التي تم اختبارها ، كانت أربع عينات إيجابية بالنسبة للكائنات المعدلة وراثيا (محتوى أكبر من 5٪). بالنسبة لدولة مثل مصر التي تستورد كميات نسبية من المنتجات الغذائية ، لا بد من اختبار جودة وسلامة منتجاتها وكذلك إجراء دراسات مراقبة صحية.