РОЗДІЛ 2. Благополуччя довкілля та екологічна безпека
SECTION 2. Environmental well-being and safety

DOI: 10.31073/onehealthjournal2026-II-04
UDC 619:604.6.001.11:633.002.6

STUDY OF SOYBEAN AND SOYBEAN PROCESSING PRODUCTS FOR GMO CONTENT USING REAL TIME PCR METHOD FOR 2022-2025

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Abstract. *The paper presents the results of research on soybeans and their processed products for the content of genetically modified organisms (GMOs) of plant origin using the real-time polymerase chain reaction (Real-time PCR) method for 2022-2025. The aim of the work was to analyze the results of research on soybean meal, soybean grain and soybean oil for the presence of GMO DNA using the Real-time PCR method. The research was conducted in the Research Department of Biochemical and Molecular Research of Food, Feed and Water of the State Scientific and Research Institute for Laboratory Diagnostics and Veterinary and Sanitary (SSRILDVSE) Examination in accordance with DSTU ISO 21569:2008, DSTU ISO 24276:2008, DSTU ISO 21570:2008, DSTU ISO 21571:2008, DSTU 5021.1:2008. The Real-time PCR was performed on an Applied Biosystems Quant Studio 5 amplifier using diagnostic kits (R-Biopharm, Germany): for screening – Sure Food GMO Screen 35S/NOS/FMV+IAC, Sure Food Screen 4 Plex BAR/NPTII/PAT/CTP2:CP4 EPSPS; for identification of GM soybean lines – Sure Food GMO ID Soya I (MON 87708 Soya, MON 87701, DP 305423, MON 87769), Sure Food GMO ID Soya II (A 5547-127, A 2704-12, MON 40-3-2, MON 89788); for quantitative determination of GM soybean lines GTS (MON) 40-3-2 and MON 89788 – Sure Food GMO Quant Roundup Ready Soya, Sure Food GMO Quant 35S Soya, Sure Food GMO Quant RR2Y Soya. Certified reference materials were used as positive controls: BF410cp, BF410dp, BF410ep (ERM, Belgium).*

During 2022-2025, 1832 soybean and soybean product specimens were tested using the real-time PCR method for the presence of GMO DNA-markers of plant origin, of which 1050 positive results were obtained, which in percentage terms is 57.3% for the studied period. The number of positive results in soybean meal samples varied from 55.2% to 62.1%, in soybean grains this indicator was from 56.7% to 60.0%, and in soybean oil from 33.3% to 75.0%.

Analysis of the results of studies conducted on soybeans and soybean processed products using the Real-time PCR method for the presence of GMO DNA indicates the circulation of GM soybean lines in Ukraine, among which the most common are GTS (MON) 40-3-2 and MON 8978. Further research will allow for constant monitoring of the situation regarding GMOs in Ukraine, the prevalence and circulation of GM lines of plant origin, and control over compliance with legislative requirements regarding labeling "with GMOs" if the content exceeds 0.9%.

Keywords: genetically modified organisms, Real-time PCR, soybean meal, soybean grain, soybean oil, GM soybean lines

Agricultural production technologies are being improved, adapted to the latest requirements and international standards, and the appearance of such products on the consumer market causes interest and concern at the same time (Martyniuk, 2017, Buhera, 2011). The production of genetically modified products is a natural consequence of the development of modern agriculture using the achievements of scientific and technological progress, which changes the structure of global production, consumption and trade in food products (Kozytska, 2010). According to Article 3 of the Law of Ukraine “On the State System of Biosafety in the Creation, Testing, Transportation and Use of Genetically Modified Organisms”, the main principles of state policy in the field of genetic engineering activities and handling of GMOs are: priority of preserving human health and environmental protection in comparison with obtaining economic benefits from the use of GMOs, control over import into the customs territory of Ukraine, state support of genetic engineering research and scientific and practical developments in the field of biological and genetic safety (Zakon Ukrainy No. 1103-V, 2007).

The issue of circulation and regulation of the use of GMOs is the labeling of goods containing them. The Law of Ukraine “On Information for Consumers on Food Products” states that if the presence of genetically modified organisms in a food product exceeds 0.9%, the labeling must include the mark

"with GMO" (Shevtsova, 2014, Zakon Ukrainy No. 2639-VIII, 2018). The use of GMOs in agricultural production and the production of agricultural products based on this technology requires an extremely balanced approach, given the existing potential risks to the environment and human health (Shevtsova, 2014, Yu, 2012, Zhao, 2025). The careful and strict regulation of the circulation of GMO products in the EU is due to the lack of research on the long-term impact of GMOs on human health, environmental protection and biodiversity conservation. Like any modern technology, GMOs have both their supporters and opponents. Therefore, in some countries the use of GMOs is prohibited, while in others it is permitted or partially permitted. The cultivation of GMOs is prohibited or severely restricted in more than 30 countries around the world, mainly in Europe, as well as in some countries in Asia, Africa and America (ISAAA, 2014). Most EU countries have banned the cultivation of GMOs on their territory (although imports are permitted in many). These include: Italy, Germany, France, Austria, Greece, Hungary, the Netherlands, Latvia, Lithuania, Poland, Luxembourg, Serbia. The widespread introduction of genetically modified organisms into the agricultural sector necessitates the creation of a reliable system for the control and identification of transgenic inserts. The real-time PCR method is the "gold standard" for the detection of GMOs due to its high specificity and the ability to quantitatively determine the content of modified DNA in a sample. An important advantage of real-time PCR is its high sensitivity, which allows the detection of even minimal amounts (less than 0.01%) of GM lines in complex multi-component food products, ensuring compliance with product labeling requirements (Smith, 2025, Gomes, 2025).

The aim of the study. To analyze the results of real-time PCR studies of soybean meal, soybean grain, soybean oil, regarding the presence of GMO DNA, for 2022-2025.

Materials and methods. The studies were conducted in the Research Department of Biochemical and Molecular Studies of Food Products, Feed and Water of the State Scientific and Research Institute for Laboratory Diagnostics and Veterinary and Sanitary Expertise, in accordance with: DSTU ISO 21569:2008. Food products. Methods for detecting genetically modified organisms and products containing them; DSTU ISO 24276:2008 Food products. Methods for detecting genetically modified organisms and products containing them. Basic requirements, terms and definitions of concepts; DSTU ISO 21570:2008 Methods for detecting genetically modified organisms and products containing them. Quantitative methods based on nucleic acid analysis; DSTU ISO 21571:2008 Food products. Methods for detecting genetically modified organisms and products containing them. Extraction of nucleic acid; DSTU 5021.1:2008 Soybean. Identification of genetically modified organisms. Part 1. Sampling method and rules for sample preparation. The real-time PCR method is sensitive, specific and universal, which allows detecting DNA even at very low concentrations by qualitative and quantitative analysis methods. The qualitative method (screening) consists in detecting GMO DNA determined by regulatory sequences (35S promoter, NOS-terminator, 35S promoter FMV) and genes (PAT, BAR, NPTII, CTP2:CP4 EPSPS). Quantitative determination by PCR-RF method consists in determining the ratio of the amount of DNA of a certain GM line to the total amount of DNA of a given species (object of study). Real-time PCR was performed on an Applied Biosystems Quant Studio 5 amplifier using diagnostic kits (R-Biopharm, Germany): for screening – Sure Food GMO Screen 35S/NOS/FMV+IAC, Sure Food Screen 4 Plex BAR/NPTII/PAT/CTP2:CP4 EPSPS; for identification of GM soybean lines – Sure Food GMO ID Soya I (MON 87708 Soya, DP305423 Soya, MON 87701 Soya, MON 87769 Soya), Sure Food GMO ID Soya II (RR Soya, RR2Y Soya, A2704-12 Soya, A5547-127 Soya); for quantitative determination of GM soybean lines GTS (MON) 40-3-2 and MON 89788 - Sure Food GMO Quant Roundup Ready Soya, Sure Food GMO Quant 35S Soya, Sure Food GMO Quant RR2Y Soya (Haydey, 2021). Certified reference material of GM soybean lines: BF410cp, BF410dp, BF410ep (ERM, Belgium) of various concentrations was used as a positive control (Shevtsova, 2014, Haydey, 2021, JRC, 2024). The main stages of the research included: sample preparation of the studied samples, DNA extraction, preparation of PCR mixtures, amplification, interpretation and analysis of the research results.

Results. In 2022-2025, 254 samples of soybeans and soybean processed products were studied by real-time PCR in the Research Department of Biochemical and Molecular Research of Food, Feed and Water of the SSRILDVSE. The research was conducted by qualitative analysis (detection of common transgenic inserts and genes), identification of GM soybean lines (MON 87708, MON 87701, DP 305423, MON 87769, A 5547-127, A 2704-12, MON 40-3-2, MON 89788) and quantitative determination of GM soybean lines GTS (MON) 40-3-2, MON 89788.

The results of research on soybeans and soybean processed products are presented in Table 1.

Table 1

Information on the number of studies on soy and soy products

Research object	Research conducted											
	Total research	Qualitative screening	Identification of GM soybean lines								Quantitative determination	
			MON 87708	MON 87701	DP305423	MON 87769	A5547-127	A2704-12	MON 40-3-2	MON 89788	MON 40-3-2	MON 89788
2022												
Soybean meal	**396**	84	16	16	16	16	42	42	42	42	40	40
Soybean grain	**10**	4	-	-	-	-	-	-	2	2	2	-
2023												
Soybean meal	**290**	52	21	21	21	21	26	26	26	26	25	25
Soybean grain	**127**	79	-	-	-	-	-	-	9	15	9	15
2024												
Soybean meal	**508**	94	33	33	33	33	47	47	47	47	47	47
Soybean oil	**3**	3	-	-	-	-	-	-	-	-	-	-
2025												
Soybean meal	**494**	98	31	31	31	31	46	46	46	46	45	43
Soybean oil	**4**	4	-	-	-	-	-	-	-	-	-	-

Research object	Number of positive results										
	Total	Qualitative screening	Identification of GM soybean lines			Quantitative determination					
			A2704-12	MON 40-3-2	MON 89788	GTS(MON) 40-3-2 (RR) Soya			MON 89788 (RR2Y) Soya		
						Total	Of these:		Total	Of these:	
							≤ 0,9 %	≥ 10 %		≤ 0,9 %	≥ 10 %
2022											
Soybean meal	**246**	84	2	40	40	40	1	39	40	0	17
Soybean grain	**6**	2	-	2	0	2	1	1	-	-	-
2023											
Soybean meal	**160**	52	4	26	26	26	4	22	26	6	20
Soybean grain	**72**	24	-	9	15	9	9	0	15	15	0
2024											
Soybean meal	**284**	94	2	47	47	47	6	32	47	9	14
Soybean oil	**1**	1	-	-	-	-	-	-	-	-	-
2025											
Soybean meal	**278**	98	0	46	46	45	11	31	43	15	15
Soybean oil	**3**	3	-	-	-	-	-	-	-	-	-

In 2022, 42 samples of soybean meal and 4 - soybean grains were tested. A total of 406 studies were conducted using the real-time polymerase chain reaction method to detect GMO DNA of plant origin: 88 studies using the qualitative method, 238 - identification of GM soybean lines, 80 - quantitative determination of GM soybean lines GTS (MON) 40-3-2 and MON 89788. During the studies, 252 positive results were obtained, of which 86 were for qualitative detection of GMO DNA, 84 - identification of GM soybean lines and 82 - quantitative determination. When determining the quantitative content of DNA of GM soybean lines: GTS (MON) 40-3-2 (Roundup Ready), 39 results were obtained with a content of more than 10% and one - less than 0.9%; MON 89788 - 17 results more than 10% and 23 - less than 10%. 2 positive results were obtained from 16 studies of DNA identification of GM soybean line A 2704-12. When detecting GM soybean lines: MON 87708, MON 87701, DP 305423, MON 87769 and A 5547-127, no positive results were obtained. In 2022, 406 studies of soybean meal (396) and soybean grain (10) were conducted. As shown in Fig. 1, 62.1% GMOs were detected in soybean meal, and 60.0% in soybean grain.

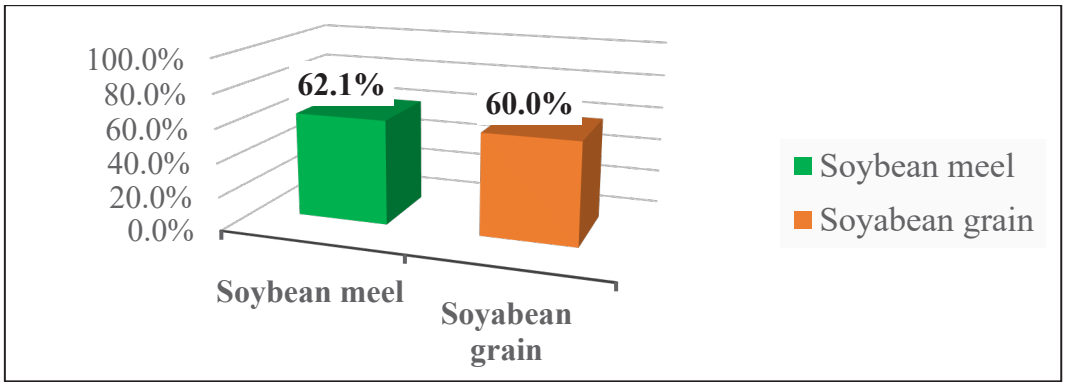

Fig. 1. Number of positive results in percentage obtained in 2022 when testing soybean meal and soybean grain

During 2023, 417 studies were conducted to detect GMO DNA of plant origin: 131 studies by qualitative method, 212 - identification of GM soybean lines, 74 - quantitative determination of GM soybean lines GTS (MON) 40-3-2 and MON 89788. 26 samples of soybean meal and 79 samples of soybean grain were examined. 232 positive results were obtained for qualitative detection of GMOs, of which: 76 by qualitative analysis, 80 - identification of GM soybean lines and 76 - determination of quantitative content. During studies to determine the quantitative content of DNA of GM soybean lines: GTS (MON) 40-3-2 (Roundup Ready 40-3-2), 22 results were obtained above 10% and 4 - below 0.9%; MON 89788 - 20 more than 10% and 21 - less than 0.9%. GM soybean line A 2704-12 was detected in 4 samples out of 21 tested. As a result of identification, no DNA of GM soybean lines was detected: MON 87708, MON 87701, DP 305423, MON 87769, A 5547-127 and MON 87701. In 2023, 160 positive results were obtained from 290 conducted studies of soybean meal, which was 55.2% and 72 (56.7%) - from 127 studies of soybean grain (Fig. 2).

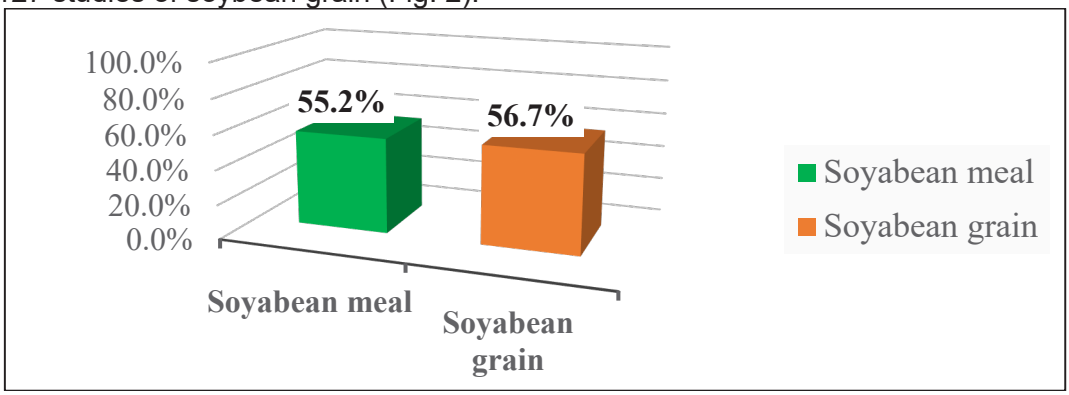

Fig. 2. Number of positive results in percentage obtained in 2023 when testing soybean meal and soybean grain

50 samples (47 soybean meal and 3 soybean oil) were received for testing in 2024. 511 tests were conducted: 97 - qualitative analysis, 320 - identification of DNA of GM soybean lines, and 94 - quantitative determination. During the research, 285 positive results were obtained: qualitative detection - 95, identification of GM lines - 96 and quantitative detection - 94. With a quantitative content of more than 10% of the GM line GTS (MON) 40-3-2 (Roundup Ready 40-3-2), 32 results were obtained and less than 0.9% - 6, MON 89788 with a content of more than 10% - 14, and less than 0.9% - 9. During the identification of the GM line of soybean A 2704-12, 2 positive results out of 33 were obtained. During the research, no GM lines MON 87708, MON 87701, DP 305423, MON 87769 and A 5547-127 were detected. In 2024, 508 studies of soybean meal were conducted, with 284 (55.9%) positive results and soybean oil - 1 (33.3%) out of 3 (Fig. 3).

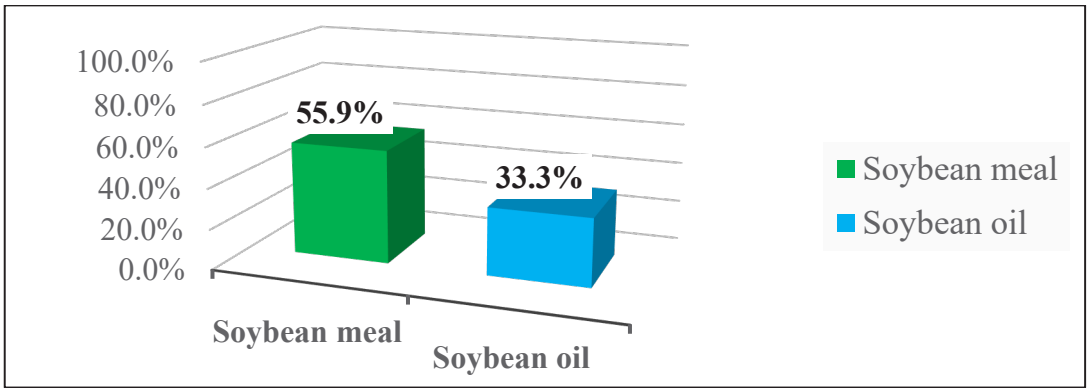

Fig. 3. Number of positive results in percentage obtained in 2024 when testing soybean meal and soybean oil

In 2025, we conducted 498 (qualitative detection of GMO DNA – 102, identification of GM soybean lines – 308 and quantitative determination – 88) studies of 49 samples of soybean meal and 4 samples of soybean oil. 281 positive results were obtained, of which 101 were during qualitative detection, 92 - identification of GM soybean lines and 88 - quantitative detection. 31 results were obtained for the detection of the GM soybean line GTS (MON) 40-3-2 (Roundup Ready) in an amount of more than 10%, 11 - less than 0.9%. MON 89788 more than 10% - 15 results and less than 0.9% - 11. No positive results were obtained during the identification of GM soybean lines: MON 87708, MON 87701, DP 305423, MON 87769, A 5547-127 and A 2704-12. In 2025, 494 studies of soybean meal were conducted with 278 (56.3%) positive results and soybean oil with 4 - 3 (75.0%), as indicated in Fig. 4.

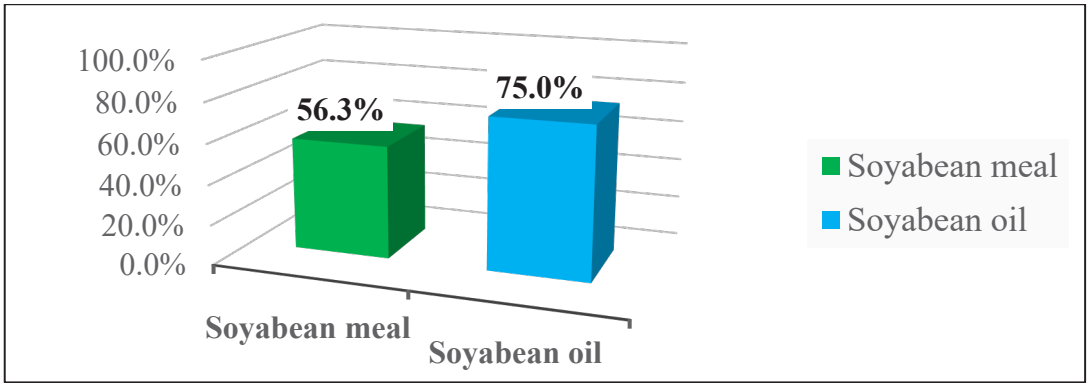

Fig. 4. Number of positive results in percentage obtained in 2025 when testing soybean meal and soybean oil

In total, during 2022-2025, 1832 studies of soybeans and products of its processing were conducted using real-time PCR (2022 - 406, 2023 - 417, 2024 - 511, 2025 - 498) for the presence of GMO DNA of plant origin, of which 1050 positive results were obtained (2022 - 252, 2023 - 232, 2024 - 285, 2025 - 281), which is 57.3% in percentage terms for the period under study.

Discussion. The growing threat of a global food crisis due to the increase in the world population has prompted the artificial breeding of genetically modified organisms by growing less capricious plants with higher yields, shorter growing season, drought resistance and disease resistance (Labzhynska, Volodchenkova, 2017).

According to the literature, real-time PCR remains important in the study of deep-processed soybean products, such as meal, oil and lecithin, due to the difficulty of extracting DNA from them. This method allows you to minimize the influence of inhibitors and DNA degradation, which is typical for thermally processed raw materials (Zhao, 2024).

Smith (2025) confirmed that real-time PCR (qPCR) remains the main, cost-effective tool for routine detection and quantification of the soybean genome, especially for regulatory compliance (Smith, 2025).

The detected lines GTS (MON) 40-3-2 and MON 89788 indicate the dominance of glyphosate-resistant soybean, which requires increased labeling control.

Conclusions. Analysis of the results indicates the circulation of GM soybean lines: GTS (MON) 40-3-2, MON 8978 and A 2704-12 in Ukraine. During 2022-2025, 1832 studies of soybean and its processed products for the presence of GMO DNA of plant origin were conducted, of which 1050

positive results (57.3%) were obtained. Summarizing the results of the research, we note that out of the total number of soybean and soybean processed products tested, 62.1% of positive results were obtained in 2022, 55.6% in 2023, 55.8% in 2024, and 57.3% in 2025.

The prospect of further research. Is to monitor the situation regarding GMOs in Ukraine, in particular the prevalence and circulation of GM lines of plant origin and control over compliance with the requirements of the legislation on labeling "with GMOs" in the presence of more than 0.9%.

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ДОСЛІДЖЕННЯ СОЇ ТА ПРОДУКТІВ ПЕРЕРОБКИ СОЇ НА ВМІСТ ГМО МЕТОДОМ ПЛР-РЧ ЗА 2022-2025 РР.

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***Резюме**. В статті наведено результати досліджень сої та продуктів її переробки щодо вмісту генетично-модифікованих організмів (ГМО) рослинного походження методом полімеразно-ланцюгової реакції в режимі реального часу (ПЛР-РЧ) за 2022-2025 рр. Метою роботи було провести аналіз результатів досліджень шроту соєвого, зерна сої та олії соєвої щодо присутності ДНК ГМО методом ПЛР-РЧ. Дослідження проводились в науково-дослідному відділі біохімічних і молекулярних досліджень харчових продуктів, кормів та води Державного науково-дослідного інституту з лабораторної діагностики та ветеринарно-санітарної експертизи (ДНДІЛДВСЕ) згідно ДСТУ ISO 21569:2008,* ДСТУ ISO 24276:2008, *ДСТУ ISO 21570:2008, ДСТУ ISO 21571:2008, ДСТУ 5021.1:2008. Постановку ПЛР-РЧ здійснювали на ампліфікаторі Applied Biosystems Quant Studio 5 з використанням діагностичних наборів (R-Biopharm, Німеччина): для скринінгу – Sure Food GMO Screen 35S/NOS/FMV+IAC, Sure Food Screen 4 Plex BAR/NPTII/PAT/CTP2:CP4 EPSPS; для ідентифікації ГМ-ліній сої – Sure Food GMO ID Soya I* (*MON 87708 Soya, MON 87701, DP 305423, MON 87769), Sure Food GMO ID Soya II (A 5547-127, A 2704-12, MON 40-3-2, MON 89788); для кількісного визначення ГМ-ліній сої GTS (MON) 40-3-2 та MON 89788 – Sure Food GMO Quant Roundup Ready Soya, Sure Food GMO Quant 35S Soya, Sure Food GMO Quant RR2Y Soya. В якості позитивного контролю використовували сертифіковані референтні матеріали: BF410cp, BF410dp, BF410ep (ERM, Бельгія).*

Впродовж 2022 – 2025 рр проведено 1832 дослідження сої та продуктів переробки сої методом ПЛР-РЧ щодо присутності ДНК ГМО рослинного походження, з них отримано 1050 позитивних результатів, що у відсотковому співвідношенні складає 57,3 % за досліджуваний період. Кількість позитивних результатів у зразках шроту соєвого варіювала від 55,2 % до 62,1 %, у зерні сої цей показник становив від 56,7 % до 60,0 %, а в олії соєвій від 33,3 % до 75,0 %.

Аналіз результатів проведених досліджень сої та продуктів переробки сої методом ПЛР-РЧ, щодо присутності ДНК ГМО, свідчить про циркуляцію на території України ГМ-лінії сої, серед яких, найбільш поширені GTS (MON) 40-3-2 та MON 8978. Проведення й надалі досліджень дасть змогу постійного відстежування ситуації щодо ГМО на території України, поширеності і циркуляції ГМ-ліній рослинного походження та контроль за дотриманням вимог законодавства щодо маркування "з ГМО" за наявності понад 0,9%.

Ключові слова: генно-модифіковані організми, ПЛР-РЧ, шрот соєвий, зерно сої, олія соєва, ГМ-лінії сої

DOI: 10.31073/onehealthjournal2026-II-04