COMMISSIE
COGEM
GENETISCHE
MODIFICATIE

BEZOEKADRES:
A. VAN LEEUWENHOEKLAAN 9
3721 MA BILTHOVEN

POSTADRES:
POSTBUS 578
3720 AN BILTHOVEN

TEL.: 088 689 2777
INFO@COGEM.NET
WWW.COGEM.NET

Aan de staatssecretaris van Openbaar Vervoer en Milieu
Ministerie van Infrastructuur en Waterstaat
De heer A.A. Aartsen
Postbus 20901
2500 EX Den Haag

DATUM 19 december 2025
KENMERK CGM/251219-01
ONDERWERP Advice on the import and processing of GM soybean MON 94115

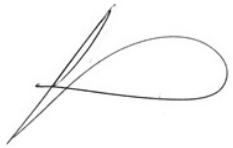
Geachte heer Aartsen,

Naar aanleiding van een adviesvraag over de milieurisico's van import en verwerking van gg-soja MON 94115 (MA 250009_001; GMFF-2025-35442/AP201), ontwikkeld door Bayer CropScience LP, deelt de COGEM u het volgende mee. Op verzoek van het Food-Feed loket voor ggo-markttoelatingen is het advies in het Engels geschreven.

Samenvatting:
De COGEM is gevraagd om te adviseren over eventuele milieurisico's van import en verwerking van de genetisch gemodificeerde (gg-) sojalijn MON 94115. Deze sojalijn brengt het *H_N90 PPO* gen tot expressie en is daardoor tolerant voor PPO-remmende herbiciden.
Hoewel het klimaat niet optimaal is, wordt soja op kleine schaal in Nederland geteeld. Opslagplanten worden hier zelden waargenomen en hebben nooit geleid tot verwilderde populaties. Er zijn geen wilde verwanten van soja aanwezig in Europa, waardoor de ingebrachte herbicidetolerantie zich niet naar andere soorten kan verspreiden. De moleculaire karakterisering van gg-soja MON 94115 voldoet aan de eisen van de COGEM. Er zijn geen redenen om aan te nemen dat expressie van het ingebrachte gen ervoor zorgt dat deze gg-sojalijn zich in Nederland zou kunnen vestigen.
Alle aspecten in overweging nemende, is de COGEM van oordeel dat de milieurisico's voor Nederland bij import en verwerking van gg-soja MON 94115 verwaarloosbaar klein zijn. Omdat andere instanties een voedselveiligheidsbeoordeling uitvoeren, heeft de COGEM bij deze vergunningaanvraag de risico's van incidentele consumptie niet beoordeeld.

De door de COGEM gehanteerde overwegingen en het hieruit voortvloeiende advies treft u hierbij aan als bijlage.

Hoogachtend,



Prof. dr. ing. Sybe Schaap
Voorzitter COGEM

c.c.

- Drs. Y. de Keulenaar, Hoofd Bureau ggo
- Ministerie van IenW, Directie Omgevingsveiligheid en milieurisico's, DG Milieu en Internationaal
- Ing. M.A.C. Möllers, Food-Feed loket

Advice on the import and processing of GM soybean MON 94115

COGEM advice CGM/251219-01

- The present application (MA 250009_001; GMFF-2025-35442/AP201) concerns the authorisation for import and processing for use in food and feed of genetically modified (GM) soybean MON 94115;
- MON 94115 carries the *H_N90 PPO* gene, which encodes the PPO protein that confers tolerance to PPO-inhibiting herbicides;
- The molecular characterisation of MON 94115 meets the criteria of COGEM;
- In the Netherlands, feral soybean populations do not occur;
- Hybridisation of soybean with other species is impossible in the Netherlands;
- There are no indications that the introduced trait allow GM soybean MON 94115 to survive in the Dutch environment;
- COGEM is of the opinion that import and processing of GM soybean MON 94115 poses a negligible risk to the environment in the Netherlands;
- COGEM abstains from giving advice on the potential risks of incidental consumption since a food/feed assessment is carried out by other organisations.

1. Introduction

The present application (MA 250009_001; GMFF-2025-35442/AP201), filed by Bayer Agriculture BV on behalf of Bayer CropScience LP, concerns the import and processing of genetically modified (GM) soybean MON 94115. MON 94115 carries the *H_N90 PPO* gene, which encodes protoporphyrinogen IX oxidase (PPO) that confers tolerance to PPO-inhibiting herbicides. The applicant notes that this event will be used as a parental line to generate stacked events but will not be commercialised as a single product.

2. Previous COGEM advice

COGEM has not previously advised on GM soybean that expresses a protein conferring tolerance to PPO-inhibiting herbicides, but has issued positive opinions on GM soybean that is tolerant to other herbicides.[1,2,3,4,5]

3. Environmental risk assessment

The objective of an environmental risk assessment (ERA) is to identify and evaluate potential adverse effects of the genetically modified organism (GMO), direct or indirect, immediate, or delayed, on human health and the environment. This ERA involves the import and processing of GM soybean. Any concerns relating to cultivation, management or harvesting practices are beyond the scope of this

advice. When assessing the environmental risk of incidental spillage of GM soybean COGEM first considers the likelihood that the event could establish itself in the Netherlands or could hybridise with related species. Other so-called 'areas of concern' (e.g. effects on non-target organisms) are addressed only if there is a possibility that the event could establish itself or if gene flow to other species might occur.

3.1 Characteristics of soybean

Soybean (*Glycine max*) belongs to the *Leguminosae* (*Fabaceae*) family and is cultivated from equatorial to temperate zones. The optimum temperature for soybean growth is between 25 °C and 30 °C. Soybean is sensitive to frost and therefore does not survive freezing conditions.[6,7,8] The soybean plant is not weedy in character.[7,8] To reduce yield losses during harvest, soybean plants with minimal seed scattering were selected for breeding. Soybean seeds rarely display dormancy, poorly survive in soil, and do not form a persistent soil seed bank.[7,9] Soybean volunteers are rarely observed throughout the world and do not compete effectively with other cultivated plants or primary colonisers.[7,8] In addition, volunteers are easily controlled mechanically or chemically.[8] Soybean is a predominantly self-pollinating species. The anthers mature in the bud and directly pollinate the stigma of the same flower.[7,8] The cross-pollination rate of soybean is low and on average between 1 to 3%.[7,8,10,11,12,13,14] Soybean pollen disperses almost only over short distances.

3.2 Receiving environment

As mentioned previously, soybean is sensitive to frost. Frost is common in the Netherlands, with an average of 51 days a year of minimum temperatures below 0 °C.[15] Although the Dutch climate is not optimal, soybean is cultivated on a small scale (101 hectares or approximately 250 acres in 2025, preliminary data).[16] Soybean volunteers are very uncommon in the Netherlands and have never resulted in establishment of wild populations.[17,18] To the best of COGEM's knowledge, there are no reports of feral soybean populations in Europe. Additionally, in Europe, hybridisation with other species is not possible because there are no wild relatives of soybean.[7,8]

Conclusion: In the Netherlands feral soybean populations do not occur and hybridisation of soybean with other species is not possible.

3.3 Description of the introduced genes and traits

The GM soybean MON 94115 expresses the *H_N90 PPO* gene. This gene encodes protoporphyrinogen IX oxidase (PPO) and is derived from the bacterium *Enterobacter cloacae*.[19]

PPO is crucial for the production of chlorophyll, which the soybean plants need for photosynthesis. PPO catalyses the oxidation of protoporphyrinogen IX to protoporphyrin IX. Inhibition of PPO by PPO-inhibiting herbicides causes accumulation of toxic intermediates of the chlorophyll biosynthesis pathway. These intermediates, when exposed to light, cause the formation of reactive oxygen species (ROS) that damage cell membranes, leading to cell leakage and ultimately plant death. This process is often visible as rapid leaf spotting, browning, and tissue desiccation.[20]

MON 94115 was generated by *Agrobacterium tumefaciens* (strain AB30) mediated transformation with the PV-GMHT533023 plasmid. This plasmid is 21.1 kb and contains one T-DNA region (approximately 15.5 kb), consisting of the *H_N90 PPO* expression cassette, the *aadA* selectable marker cassette, *cre* cassette,

and the Site-Directed Integration cassette. The plasmid was designed to enable both site-directed integration (SDI) of the T-DNA into a specific location in the soybean genome and Cre/*lox*-mediated auto-excision to generate selectable marker-free events, along with the removal of the machinery that enables SDI.[21]

SDI uses a guide RNA (gRNA)-directed nuclease to enable targeted integration of the transgene at a specific genomic location. In this process, the gRNA guides the Cas12a (CRISPR-associated protein 12a) enzyme to the target site in the genome, where Cas12a introduces a double-stranded DNA break. This break facilitates the integration of T-DNA via the endogenous DNA repair mechanisms. Here, SDI was used to insert T-DNA from the PV-GMHT533023 plasmid into a specific locus within the soybean genome.

To remove the *aad4* selectable marker cassette and the SDI-enabling components, Cre/*lox* recombination technology was employed. In the SDI method, transgenic plants are generated with a single T-DNA insertion, where the selectable marker, Cre recombinase, and SDI machinery are all flanked by two *loxP* sites. Upon activation, the Cre enzyme catalyses recombination between the *loxP* sites, excising all intervening sequences.[21] As a result, the genome of MON 94115 retains only the *H_N90 PPO* expression cassette, along with the left and right border regions.

Introduced genes	**Encoded proteins**	**Regulatory elements**	**Traits**
H_N90 PPO	protoporphyrinogen IX oxidase (PPO) originating from *Enterobacter cloacae*	Ubiquitin promoter sequence from *Medicago truncatula*, sequence from *Adansonia digitata* encoding a peptide that directs PPO to the chloroplast, *Zea mays* terminator sequence	Tolerance to PPO-inhibiting herbicides

3.4 Molecular characterisation

The applicant performed a next generation sequencing analysis of the MON 94115 genome to determine the sequence of the insertion site, insert and flanking regions. The sequencing results demonstrated a single integration site on chromosome 13 in the MON 94115 soybean genome without disruption of any endogenous features. No other unintended sequences such as plasmid backbone sequences were detected.

Comparison of the MON94115 insert (3,201 bp) and its flanking regions (1,000 bp on both ends) final consensus sequence with the sequence of the transformation plasmid demonstrated that the insert in the MON 94115 genome only contains a single *loxP* site and the *H_N90 PPO* cassette. This confirms the successful Cre/*lox*-mediated auto-excision. A sequence comparison between the control soybean genome and the sequence generated from the 5′ and 3′ flanking sequences of MON 94115 indicates that 10 bases of soybean genomic DNA were deleted during integration of the T-DNA.

The applicant screened the 3′ and 5′ junctions of the insert and its flanking regions, as well as the entire insert, for potential newly created open reading frames (ORFs). According to the applicant, the putative

products of the identified ORFs did not generate any protein sequence similarity with known allergens, toxins, or other biologically active proteins.

Overall, the molecular characterisation was conducted according to the criteria previously laid down by COGEM.[22] The results from the molecular characterisation do not provide indications that MON 94115 soybean could pose a risk to the environment.

Conclusion: The molecular characterisation of soybean MON 94115 is adequate and no indications for potential environmental risks were identified.

3.5 Phenotypic and agronomic characteristics

The applicant evaluated the phenotypic and agronomic characteristics of MON 94115 at multiple field sites across the United States during the 2023 growing season, comparing it to both the conventional counterpart (soybean A3555) and several commercial reference varieties.

Phenotypic traits were assessed for MON 94115, the conventional control, and four commercial reference varieties, both in the absence and presence of a PPO-inhibiting herbicide. Statistical analysis of these field trials revealed no phenotypic changes in MON 94115 that would suggest increased weediness or pest potential compared to conventional soybean.

Seed germination was also compared among MON 94115, the conventional control, and the reference varieties. Parameters measured included percent germinated seed, percent dead seed, percent viable firm swollen seed, and percent viable hard seed. All values for MON 94115 were within the range observed in the reference varieties, with no statistically significant differences from the conventional control.

Additionally, the applicant analysed the grain composition of MON 94115 – measuring moisture, nutrients, anti-nutrients, and isoflavones – and compared these results to the conventional control. Although some minor differences were observed, the applicant concluded that MON 94115 is compositionally similar to conventional soybean varieties.

Based on these findings, COGEM concludes that, aside from the introduced herbicide tolerance trait, the agronomic and phenotypic characteristics of MON 94115 are comparable to those of conventional soybean varieties. There is no evidence to suggest that MON 94115 would have an increased ability to survive or establish in the Dutch environment.

Conclusion: There are no indications that soybean MON 94115 would be able to survive or establish in the Netherlands.

4. Food/feed assessment

This application is submitted under Regulation (EC) 1829/2003, therefore a food/feed assessment is carried out by EFSA and national organisations involved in the assessment of food safety. In the Netherlands, WFSR carries out a food and/or feed assessment for Regulation (EC) 1829/2003 applications. The outcome of the assessment by other organisations (EFSA, WFSR) was not known when this advice was completed.

5. Post-market environmental monitoring

The applicant supplied a post-market environmental monitoring (PMEM) plan. The general surveillance (GS) plan differs in specific details, but is overall comparable to the GS plans from previous applications of GM crops. COGEM has published several recommendations for further improvement of GS plans,[23,24] but considers the current GS and PMEM plan adequate for the import and processing of soybean MON 94115.

Conclusion: The current PMEM plan of gg-soybean MON 94115 is sufficient for import and processing.

6. Overall conclusion

Conclusion: COGEM is of the opinion that import and processing of soybean MON 94115 poses a negligible risk to the environment in the Netherlands. COGEM abstains from giving advice on the potential risks of incidental consumption since other organisations carry out a food/feed assessment.

7. Additional remark regarding hypothetical import

COGEM notes that although an application for import and processing of MON 94115 was filed, the applicant states that it will not be commercialised as a single event and will only be used to create stacked events. This situation results from the procedures followed by EFSA, i.e. that an application for import and processing of a stacked GM line can only be filed if the parental GM lines have been assessed.[25] COGEM points out that in this particular case it is not relevant to assess the single event MON 94115, because it will never be commercialised as a standalone product. COGEM considers the request for authorisation of MON 94115 for import and processing and use in food and feed as an example of following unnecessary procedures.

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