It must be noted that, regarding the minimum requirements for the scope of testing in Chapter J of the VLOG Standard, not all GMOs were taken into account that are authorised in the EU or tolerated for feed within the meaning of EU Regulation No. 619/2011. Furthermore, GMOs not authorised in the EU are not part of the minimum requirements. In the event of an examination of the marketability and proper labelling of a feed, other GMOs would be taken into account (this includes other GMOs authorised in the EU, GMOs tolerated in feed pursuant to EU Regulation No. 619/2011, and GMOs not authorised in the EU).

In consultation with laboratories, VLOG regularly checks and updates the following minimum requirements in Chapter J of the VLOG Standard concerning the scope of testing of raw materials and feed. In the event of developments that other GMOs become relevant (e.g. RASFF reports), VLOG will provide its members and VLOG-certified companies with corresponding testing requirements/guidelines in a timely manner.

This does not mean, however, that the companies participating in the VLOG system are dispensed from their own due diligence obligations to regularly check and, if necessary, update the scope of testing.

1. Minimum requirements for raw materials/single-component feed

1.1. Minimum requirements for raw soy materials/soy-based single-component feed

**Determination and assessment of the summation value of the most relevant soy GMOs:**

- Quantification of GTS 40-3-2 (RRS-1)
- Quantification of MON89788 (RRS-2)
- Qualitative detection of A2704-12 and A5547
  
  In the event of a positive result for A2704 and A5547, the quantity of this GMO can, for example, be estimated using the \( \Delta \Delta C_t \) method or similar method ensuring that sufficient quantities of species DNA are present. For estimated values over 0.1%, a post-quantification must be carried out.

Alternately, the laboratory may work with screening parameters that detect at least the GMOs mentioned. *When using the *pat* gene (or LibertyLink construct), A5547 (or other single copy material) must be used as the reference material for estimation.* In subsequent identification/quantification of positive findings, at least all GMOs (if corresponding elements are positive) mentioned here must be quantified.

1.2. Minimum requirements for raw corn materials/corn-based single-component feed

1. **Screening for 35S Promoter (p35S) and NOS Terminator (tNOS).**

   Other screening elements can be implemented to narrow the corresponding GMO down.

2. **If positive:**

   *In the event of a positive result for 35S Promotor (p35S) and/or NOS Terminator (tNOS), the quantity of these screening elements can, for example, be estimated using the \( \Delta \Delta C_t \) method or a similar method. For estimated values (MON89034 or a suitable reference material for maize/corn that contains 35S in single copy) over 0.1%, identification and subsequent quantification must be carried out. If there are several positive results, the estimated individual values must be added together.*

Testing at least for NK603, TC1507, MON810, MON89034.
3. If using the positive screening parameters, one or more of these GM corn types can be ruled out, then the same number of commercialised GM corn types that come into question must be searched for instead.

Positive screening results for values over 0.1% must be clarified; if no GM corn types can be found, other GM types must be analysed, e.g. RRS1.

4. **Determining the summation value of the corn GMO**
   Identified varieties must be quantified if the estimation of the concentration, when using, for example, the ΔΔct method or another similar method ensuring that sufficient quantities of species DNA are present, leads to values over 0.1%.

RRS-1 positive:
Estimating the soy mass and assessing the amount of soy: Is it a relevant amount or minimal traces? If a botanical contamination containing GMO is determined, an assessment according to the official guideline must take place.

1.3. **Minimum requirements for raw canola materials/canola-based single-component feed**

*There are two possible testing procedures.*

**First testing procedure:**

1. **Triple screening** that detects all current relevant GM canola varieties and botanical contaminations (e.g. tNOS, pat gene (or LibertyLink construct), CTP2-CP4epsps (or pFMV))

2. **ID depending on positive screening results**
   - tNOS positive: at least RRS + bar gene or MS8 / RF3 directly
   - CTP2-CP4epsps / pFMV positive: at least GT73

If no canola GMO is detected, the presence of a botanical contaminant containing GMO with other plant varieties must be clarified (estimation and assessment of masses). Is it a relevant quantity or minimal traces? If botanical contamination with GM soy above the 0.5% limit is detected, the GM soy content must be determined (see minimum requirements for feed containing soy). For botanical contamination with maize/corn, the GMO summation value for maize/corn with maize/corn contamination over 0.9% must be determined and a new evaluation pursuant to official guidelines must take place.

**Second testing procedure:**

1. **Estimating the soy mass:**

   For quantities over 0.5%, the quantity of soy GM must be determined (cf. Minimum requirements for feed containing soy).

2. **Qualitative evidence of canola GT73 + canola MS8 or canola RF3 (or bar gene)**

3. **Determining the summation value of GM canola**
   Identified GM canola varieties must be quantified if the estimation of the quantity, when using, for
example, the ΔΔct method or another method ensuring that sufficient quantities of species DNA are present, leads to values over 0.1%.

Positive screening results must be clarified.

**1.4. Minimum requirements for rice and rice products**

1. **Preparation of laboratory samples**
   Two subsamples of at least 250 g each are to be created from the laboratory sample sent, and each is to be analysed separately (1 extraction, 2 PCRs per subsample).

2. **Element-specific screening**
   p35S + tNOS + cry1Ab/cry1Ac sequence

3. **Design-specific proof**
   Identification, by agreement between the company and the laboratory, of GMO events that cause a positive screening result (see 1).

4. **Exclusion of botanical impurities (GMO carryovers from other plant species) from corn, soy, cotton and (naturally occurring) Cauliflower Mosaic Virus.**

   If the element-specific screening yields a positive result, design-specific proof is to be provided as the next step. In combination with the exclusion of botanical impurities and the Cauliflower Mosaic Virus, an investigation is to be made of whether the sample contains genetically modified rice.

5. **Evaluation of the PCR results**
   If the targeted sequence of genetically modified rice is proven for at least one of the subsamples analysed, this result is to apply to the entire sample and the batch. The batch cannot be marketed in the EU and cannot be labelled with the “Ohne GenTechnik” seal.

**1.5 Minimum requirements for salmon and salmon products**

1. **Design-specific proof**
   AquAdvantage® Atlantic salmon (Salmo salar).

2. **Evaluation of the PCR results:**
   If the targeted sequence of genetically modified salmon is proven for at least one of the subsamples analysed, this result is to apply to the entire sample and the batch. The batch cannot be marketed in the EU and cannot be labelled with the “Ohne GenTechnik” seal.
2. Minimum requirements for compound feed

2.1. Minimum requirements for compound feed containing soy

Determinand and assessment of the summation value of the most relevant GMOs:

Soy:

- Quantification of GTS 40-3-2 (RRS-1)
- Quantification of MON89788 (RRS-2)
- Qualitative detection of A2704-12 and A5547
  In the event of positive result for A2704 and A5547, the quantity of this GMO can, for example, be estimated using the ΔΔct method or similar method ensuring that sufficient quantities of species DNA are present. For values over 0.1%, a post-quantification must be carried out.

In case of limited analysability of the soy ingredient, the practical LOD must be indicated.

For corn ingredient:

Additional qualitative detection of the 4 commercialised corn varieties: NK603, TC1507, MON810, MON89034.

In case of a positive result, the quantity of this GMO can, for example, be estimated using the ΔΔct method or a similar method ensuring that sufficient quantities of species DNA are present. For values over 0.1%, a regular quantification of the GMOs detected must be carried out.

In the event of limited analysability of the corn ingredient, the practical detection limit (LOD) must be indicated.

For canola ingredient:

Additional qualitative detection of GT73.

In case of positive identification, quantification of GT73 must take place if the estimation of the quantity using, for example, the ΔΔct method or another similar method ensuring that sufficient quantities of species DNA are present, leads to values over 0.1%.

In case of limited analysability of the canola ingredient, the practical LOD must be indicated.

Alternately, the laboratory may also work with screening parameters that detect at least the GMOs mentioned (soy, canola, corn). In subsequent identification/quantification of positive results, at least all GMOs (if corresponding elements are positive) mentioned here must be identified and, if necessary, quantified.
2.2. Minimum requirements for soy-free compound feed

**Determination and assessment of the summation value of the most relevant GMOs:**

**Estimating the soy mass:**

In a first step, the mass of soy in the feed is estimated. For quantities over 0.9%, the quantity of GM soy must be determined (cf. Minimum requirements for feed containing soy) and an assessment according to the official guideline⁴ must take place.

**For canola ingredient:**

Qualitative evidence of canola GT73 + canola MS8 or canola RF3 (or bar gene).

In the event of positive identification, quantification of GMO or GMOs found must take place if the estimation of the quantity when using, for example, the ΔΔct method or another similar method ensuring that sufficient quantities of species DNA are present, leads to values over 0.1%.

In case of limited analysability of the canola ingredient, the practical LOD must be indicated.

**For corn ingredient:**

Qualitative evidence of 4 corn varieties used commercially: NK603, TC1507, MON810, MON89034

In the event of positive identification, quantification of GMO or GMOs found must take place if the estimation of the quantity when using, for example, the ΔΔct method or another similar method ensuring that sufficient quantities of species DNA are present, leads to values over 0.1%.

In the event of limited analysability of the corn ingredient, the practical LOD must be indicated.

Alternately, the laboratory may work with screening parameters that detect at least the GMOs mentioned (concerning canola and corn). In subsequent identification/quantification of positive results, at least all GMOs (if corresponding elements are positive) mentioned here must be identified and, if necessary, quantified.

2.3. Other products/raw materials

The strategies for analysing GMOs in other single-component feed, raw materials, (food) ingredients, intermediate products or food must continue to be agreed upon with the commissioned laboratory, taking into account the composition and origin of the products.

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⁴ Guideline for the Control of GMOs in Feed (German: Leitfaden zur Kontrolle von GVO in Futtermitteln – version of November 2011). Monitoring of the production, of treatment, of use and of bringing to the market of feed in connection with genetically modified organisms (GMOs). ... Compiled by the project group ‘GMOs in Feed’ of the Feed Working Group within the Working Group for Consumer Protection of the German Länder (German: LAV – Länderarbeitsgemeinschaft Verbraucherschutz) with participation of the Federal Government and the Association of German Agricultural Analytic and Research Institutes (VDLUFA), http://www.ohnegentechnik.org/Leitfaden_Futtermittel