ANNEX

Testing scheme necessary to ascertain the achievement of the Union target for the reduction of the relevant *Salmonella* serotypes in adult breeding flocks of *Gallus gallus*

3. EXAMINATION OF THE SAMPLES

3.1. **Transport and preparation of the samples**

3.1.1. *Transport*

Samples shall preferably be sent by express mail or courier to the laboratories referred to in Articles 11 and 12 of Regulation (EC) No 2160/2003, within 24 hours after collection. If not sent within 24 hours, they shall be stored refrigerated. Transportation can be at ambient temperature as long as excessive heat (over 25 °C) and exposure to sunlight are avoided. At the laboratory samples shall be kept refrigerated until examination, which shall be started within 48 hours following receipt and within 96 hours after sampling.

3.1.2. *Hatcher basket liners:*

(a) Place the sample in 1 litre of buffered peptone water (BPW) which has been pre-warmed at room temperature and mix gently.

(b) Continue the culture of the sample by using the detection method described in point 3.2.

3.1.3. *Boot swabs and dust samples:*

(a) The pair(s) of boot/sock swabs and dust sample (fabric swab) shall be carefully unpacked to avoid dislodging adherent faecal material or loose dust material and placed in 225 ml of BPW which has been pre-warmed to room temperature.

(b) The boot/socks and fabric swab shall be fully submersed in BPW to provide sufficient free liquid around the sample for migration of *Salmonella* away from the sample and therefore more BPW may be added, if necessary.

Separate preparations must be made of the boot swabs and the fabric swab.

(c) Where five pairs of boot/sock swabs are pooled into two samples, each pooled sample must be placed in of 225 ml of BPW, or more if necessary, to fully submerge the sample and provide sufficient free liquid around the sample for migration of *Salmonella* away from the sample.

(d) Swirl to fully saturate the sample and continue the culture by using the detection method described in point 3.2.

3.1.4. *Other faecal material samples:*

(a) The faeces samples shall be pooled and thoroughly mixed and a 25 g sub-sample shall be collected for culture.

(b) The 25 g sub-sample shall be added to 225 ml of BPW which has been pre-warmed to room temperature.

(c) The culture of the sample shall be continued by using the detection method described in point 3.2.

If ISO standards on the preparation of relevant samples for the detection of *Salmonella* are agreed on, they shall be applied and replace those referred to in points 3.1.2, 3.1.3 and 3.1.4 on sampling preparation.
[[F1]3.1.5.

In case of collection of fabric swabs in accordance with point 2.2.2.1(d) or one pair of boot swabs and 2 moistened fabric swabs in accordance with 2.2.2.1(e), pooling shall occur in accordance with point 3.1.3(b).**]**

3.2. Detection method

[[F2]The detection of *Salmonella* spp. shall be carried out according to EN ISO 6579-1.**]**

Textual Amendments

F2 Substituted by Commission Regulation (EU) 2019/268 of 15 February 2019 amending Regulations (EU) No 200/2010, (EU) No 517/2011, (EU) No 200/2012 and (EU) No 1190/2012 as regards certain methods for Salmonella testing and sampling in poultry (Text with EEA relevance).

As regards the boot swabs samples, dust samples and other faecal material samples referred to in point 3.1, the incubated BPW enrichment broth for future culture may be pooled. To do so, incubate both samples in BPW as referred to in point 3.1.3. Take 1 ml of incubated broth from each sample and mix thoroughly, then take 0,1 ml of the mixture and inoculate the modified semi-solid Rappaport-Vassiliadis (MSRV) plates.

The samples in BPW must not be shaken, swirled or otherwise agitated after incubation as this releases inhibitory particulates and reduces subsequent isolation in MSRV.

3.3. Serotyping

At least one isolate from each sample showing a positive reaction shall be typed, following the Kaufmann-White scheme.

[[F2]3.4. Alternative methods

Alternative methods may be used instead of the methods for detection and serotyping provided for in points 3.1, 3.2 and 3.3 of this Annex, if validated in accordance with EN ISO 16140-2 (for alternative detection methods).**]**

3.5. Storage of strains

It shall be guaranteed that at least one isolated strain of the relevant *Salmonella* serotypes from sampling as part of official controls per house and per year are stored for possible future phagetyping or antimicrobial susceptibility testing, using the normal methods for culture collection, which must ensure integrity of the strains for a minimum period of two years. If the competent authority decides so, isolates from sampling by food business operators shall also be stored for these purposes.

Changes to legislation:
There are outstanding changes not yet made to Commission Regulation (EU) No 200/2010. Any changes that have already been made to the legislation appear in the content and are referenced with annotations.
View outstanding changes

Changes and effects yet to be applied to the whole legislation item and associated provisions

- Signature words omitted by S.I. 2019/740 reg. 7(5)
- Art. 1A inserted by S.I. 2019/740 reg. 7(3)
- Art. 1A words substituted in earlier amending provision S.I. 2019/740, reg. 7(3) by S.I. 2020/1463 reg. 7(4)(a)