SPECIFIC PROTOCOL

19th European Proficiency Test on Pesticides in Food of Animal Origin and Commodities with High Fat Content

„Pesticides in beef meat (EUPT-AO19)“

(22.04.2024)

➢ Introduction

This protocol is complementary to the valid version of the General Protocol for EU Proficiency Tests for Pesticide Residues in Food and Feed, Ed. 11[1]. The current proficiency test covers pesticides that can be determined by multi-residue methods. This EUPT is to be performed by all National Reference Laboratories for Pesticides in Food of Animal Origin and Commodities with High Fat Content (NRL-AO) as well as by all official EU laboratories (OfLs) responsible for official pesticide residue controls on food of animal origin, as far as their scope overlaps with that of the EUPT-AO19. The commodity chosen to prepare the test material is beef meat and it is considered as a representative commodity for “meat (muscle) and seafood, Group 7” (see Annex A of document SANTE 11312/2021v2)[2].

➢ PT item (test material)

This proficiency test concerns the analysis of pesticide residues in beef meat. The matrix contains only spiked pesticides.

Since 2020, the EURLs for pesticide residues do not provide blank PT items. Each laboratory is asked to use “pesticide free” representative beef meat for recovery experiments as well as for the preparation of matrix-matched or procedural calibration standards.

The organisers will check the PT items for sufficient homogeneity and for stability using conditions that reproduce sample shipment and storage for the duration of the proficiency test.

All these tests will be conducted by the EURL-AO which is accredited according to ISO 17043 for organising proficiency tests.

The participants will receive ~ 100 g of beef meat PT item (one can) containing spiked pesticides.

1 http://www.eurl-pesticides.eu/docs/public/tmplt_article.asp?CntID=821&LabID=100&Lang=EN

2 https://food.ec.europa.eu/system/files/2023-11/pesticides_mrl_guidelines_wrkdoc_2021-11312.pdf

- **Analytical parameters**

The PT item contains several pesticides from the target pesticide lists given in Annex 1 and 2 of this document.

It is mandatory to analyse all pesticides included in Annex 1. Pesticides included in Annex 2 can be analysed on voluntary basis.

Laboratories should carefully read the target pesticide lists, where important information about reporting of results, as well as the Minimum Required Reporting Levels (MRRLs), are given. The target pesticide lists contain only individual compounds and results should only be reported for individual compounds, no matter how the residue definitions are set.

During EUPT AO 12 - 16 a quite number of laboratories had problems to report spinosyn A and D individually. Therefore, a possibility for reporting the result of spinosad (sum of spinosyn A and D) was added. Please, report results for spinosyn A and D only, if you use the individual standards for calibration.

The MRRL values will be used to identify false positive and false negative results and for the calculation of z-scores for false negatives.

Please, report important observations during analysis in the EUPT Webtool in the special field for comments (e.g. damaged can or losses, additional pesticides not listed in the target lists). Please consider therefore the EUPT Webtool Guideline.

- **Shipment of PT item**

Dispatch of the PT item is planned on **06 May 2024**.

PT items will be shipped in cans in a normal box with no further cooling. The organisers will aim to ensure that all participating laboratories will receive their PT items as soon as possible. PT items will be shipped with TNT/FedEx. Prior to shipment an e-mail will be sent to the participating laboratories from the shipper.

Laboratories must make their own arrangements for the receipt of the package. They should inform the organiser of any public holidays in their country/city during the week of the shipment, and must make all necessary arrangements to receive the shipment, even if the laboratory is closed.

- **Instructions on PT item handling**

Once received, the PT item shall be stored cooled (4°C) to avoid any deterioration/spoilage and to minimise possible pesticide losses.

Bring the content of the PT item to room temperature and homogenise the whole sample thoroughly before analysis, including any liquid that may be present due to the technical processing.

It is recommended to divide the whole sample amount into analytical test portions and weigh them into the tubes used for extraction. All analytical test portions not used for analysis should be stored chilled. This procedure helps to avoid possible losses caused by several thawing steps of the test material.

All participants should use their own routine standard operating procedures for extraction, clean-up and analytical measurement and their own reference standards for identification and quantification purposes. Considering the available amount of PT item, laboratories employing methods requiring large analytical portions are advised to scale them down.

The homogeneity tests were conducted using 10 g of the PT item and automated EN1528 method[3] (for GC-amenable pesticides) and 5 g of the PT item for QuEChERS-AO method[4] (for LC-amenable pesticides). As sub-sampling variability increases with decreasing analytical portion size, sufficient homogeneity can only be guaranteed where participants employ analytical portions that are equal or larger than those stipulated in the previous sentence.

- **EUPT Webtool and Deadlines**

Sample receipt acknowledgement, scope selection, analytical results and method information are to be submitted via the EUPT Webtool (open in incognito or private window). Please consider the guideline on how to use the EUPT Webtool: EUPT Webtool Guideline. To access the EUPT Webtool participants must use their unique login data (username and password), which was sent to you from the colleagues from DTU Denmark. The password can be retrieved via https://guest.dtu.dk/Sites/GuestLogin/RetrievePassword.aspx using your email address or your username.

1. PT item receipt and acceptance

Once the laboratory has received the PT item, it must report to the organiser, via the EUPT Webtool (Result Submission Website EUPT-AO19), the date of receipt, the condition of the PT item, and its acceptance. The deadline for acceptance is **13 May 2024**. If the laboratory does not respond by this deadline the organiser will assume that the PT item has been received and accepted. If participants have not received the PT items by the **13 May 2024 at noon**, they must inform the organiser immediately by e-mail (eurl-pesticides@cvuafr.bwl.de).

[3] EURL_AO_Validation_report_2021-06_GC-Orbitrap_GC-MS_MS_quantification_fish_offals

[4] EURL_AO_Validation_report_2021-07_LC-MSMS_quantification_offal_and_fish

2. Scope selection

The analytical scope must be selected prior to the shipment of the samples. This is done via the EUPT Webtool. The scope selection subpage will be opened from **22 April to 03 May 2024**. As default all mandatory pesticides are preselected. Results can only be reported for analytes that have been selected during the scope selection procedure.

Important: If you did not select your scope in time, all analytes of the mandatory pesticide list (see Annex 1) will be selected for your scope!

3. Results and method submission

After shipment of the samples, from 06 May 2024 onwards, it is possible to submit the results by logging into the EUPT Webtool. **The deadline for result submission is 07 June 2024 at 23:00 CEST.** Method information can be added within the next 7 days (until 14 June 2024).

Important: After the final submission it will NOT be possible to edit the results. The website will not be accessible after this date and any results reported after the deadline will not be included in the statistical treatment, or in the final report. Participants will receive an email confirming the submission of their results. Attached to the email will be an excel file with all their submitted data and a pdf of the pesticide and concentration submitted.

4. Reporting of qualitative and quantitative results

Results shall NOT be reported where a pesticide

a) was not detected,

b) was detected below the RL (Reporting Limit) of the laboratory or

c) was detected below the MRRL.

Results reported as "< RL" will be considered as „not detected".

Significant figures:

Residue levels shall be expressed to three significant figures, e.g. 0.0581, 0.251 or 1.35 mg/kg.

5. Reporting information on analytical methodology

All laboratories are requested to provide information on the analytical method(s) they have used via EUPT Webtool (Result Submission Website EUPT-AO19). The laboratories are requested to fill-in this important information in order to minimise the administrative burden of collecting this information at a later date. Submission of method information is only possible until 7 working days after result submission deadline (until **14 June 2024**).

6. Reporting of supplementary information in case of false negative results

In case of false negative results, the affected laboratories will be asked via e-mail to provide details of the methodology used after the deadline for results submission. This can be done by accessing the

EUPT Webtool (Result Submission Website EUPT-AO19) until 7 working days after result submission deadline (until **14 June 2024**).

Important: If no sufficient information on the methodology used is provided, the organiser reserves the right not to accept the analytical results reported by the participant.

- **Follow-up actions**

According to article 94 2c of Regulation (EU) No 2017/625, underperformance of any NRL-AO in comparative testing will be followed by EURL-AO.

- **Documents**

In the EURL-document repository (CIRCA BC) all documents relating to EUPT AO19 can be found. Links to most of the documents are also available on the EUPT-AO19 website.

- **Subcontracting**

The following tasks were subcontracted:

1. Generation of login credentials for EUPT webtool (EURL-CF, Lyngby, Denmark)
2. Programming and administration of EUPT-AO19 result submission website (EURL-CF, Lyngby, Denmark)
3. Purchase of blank beef meet (Gertrud-Luckner-Gewerbeschule, Freiburg, Germany)
4. Provision of equipment for preparation of the PT items (Gertrud-Luckner-Gewerbeschule, Freiburg, Germany)

- **Time schedule**

Actor	Activity	Date
EURL	Preliminary announcement matrix beef meat at Joint Workshop in Stuttgart	20 October 2023
EURL	First information supplied to laboratories and call for participation	Beginning of February 2024
Participant	**Registration** via EUPT website	**15 December 2023 – 01 April 2024**
Participant	**Scope selection** via EUPT webtool	**22 April – 03 May 2024**
Participant	Proof of shipment address in EURL-Datapool	Ending 26 April 2024
EURL	**Dispatch of test material**	**06 May 2024**
Participant	Confirmation of test material receipt	07 – 13 May 2024
Participant	**Deadline for reporting of test results**	**07 June 2024***
Participant	Deadline for reporting of additional method information (no changes of reported results possible)	14 June 2024
EURL	Deadline for preliminary report	09 August 2024
EURL	Dispatch of the final report as pdf-file	Approx. end of 2024

*) Please make sure to report your results on time as there will be no extension of the deadline.

- **Participation fee**

There is a **fee of EUR 200.00** for shipping and handling to participants within the European Union and EFTA countries (**including NRLs**). Fees for participants from **other countries** are **EUR 400.00**. An invoice will be sent as pdf-file via e-mail.

- **Delays in Payment**

The participants will receive an **invoice as pdf-file via e-mail** to the corresponding e-mail address given during registration. Laboratories wishing to additionally receive an invoice in paper form should write the request to eurl-pesticides@cvuafr.bwl.de before **26 April 2024**. Please make sure that the payment is made before the stipulated deadline stated on the invoice (**28 June 2024**). If the invoice is not paid within the stipulated time, reminders will be sent within a four week period.

From the second reminder onwards an administration fee of 25 € will be charged per reminder. Based on Reg. (EC) 625/2017, OfLs not paying the EUPT sample delivery fee will be initially warned that their participation in subsequent EUPTs could be denied. In case of a repetitive non-payment, the EUPT organisers will inform the corresponding NRL or the competent authority to take action.

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Annex 1

EUPT-AO19 Pesticide target list of mandatory analytes

Table A1: List of 56 **mandatory** analytes and minimum required reporting levels (MRRL) in EUPT-AO19 (PT item beef meat). Results shall be rounded to three significant figures (e.g. 0.0581, 0.251 or 1.35). Version 1.1, changes are marked in red

Analyte	MRRL (mg/kg)	Analyte	MRRL (mg/kg)
Aldrin	0.005	Fipronil	0.005
Azinphos-methyl	0.010	Fipronil sulfone	0.005
Bifenthrin (sum of isomers)	0.010	Heptachlor	0.005
Chlordane, cis-	0.005	Heptachlorepoxid, cis-	0.005
Chlordane, trans-	0.005	Heptachlorepoxid, trans-	0.005
Chlorfenvinphos	0.010	Hexachlorcylcohexane (HCH), alpha-isomer	0.005
Chlorpyrifos(-ethyl)	0.010	Hexachlorcylcohexane (HCH), beta-isomer	0.005
Chlorpyrifos-methyl	0.010	Hexachlorcylcohexane (HCH), gamma-isomer (Lindane)	0.005
Cyfluthrin (sum of isomers)	0.010	Hexachlorobenzene (HCB)	0.005
Cypermethrin (sum of isomers)	0.010	Indoxacarb (sum of isomers)	0.010
DDD, p,p´- (TDE)	0.005	lambda-Cyhalothrin (sum of isomers)	0.010
DDE, p,p´-	0.005	Malathion (parent only)	0.010
DDT, o,p´-	0.005	Methoxychlor, 4,4´-	0.010
DDT, p,p´-	0.005	Nitrofen	0.005
Deltamethrin (cis-isomer)	0.010	Oxychlordane	0.005
Diazinon	0.010	Parathion(-ethyl)	0.010
Dieldrin	0.005	Parathion-methyl (parent only)	0.010
Endosulfan, alpha-	0.005	Pendimethalin	0.010
Endosulfan, beta-	0.005	Permethrin (sum of isomers)	0.010
Endosulfan-sulfate	0.005	Phosmet (parent only)	0.010
Endrin	0.005	Phoxim	0.010
Famoxadone	0.010	Pirimiphos-methyl	0.010
Fenthion oxon	0.010	Profenofos	0.010
Fenthion oxon sulfone	0.010	Pyrazophos	0.010
Fenthion oxon sulfoxide	0.010	Quintozene (parent only)	0.005
Fenthion sulfone	0.010	Resmethrin (sum of isomers)	0.010
Fenthion sulfoxide	0.010	Tecnazene	0.005
Fenvalerate/Esfenvalerate (sum of RR, SS, RS & SR isomers)	0.010	Vinclozolin (parent only)	0.005

Annex 2

EUPT-AO19 Pesticide target list of voluntary analytes

Table A2: List of 48 **voluntary** analytes and minimum required reporting levels (MRRL) in EUPT-AO19 (PT item beef meat). Results shall be rounded to three significant figures (e.g. 0.0581, 0.251 or 1.35). Version 1.1, changes are marked in red

Analyte	MRRL (mg/kg)	Analyte	MRRL (mg/kg)
Benzovindiflupyr	0.010	BTS 44595 (Prochloraz metabolite)	0.010
Bixafen (parent only)	0.010	BTS 44596 (Prochloraz metabolite)	0.005
Bixafen-desmethyl	0.010	Prothioconazole-desthio	0.010
Boscalid (parent only)	0.010	Spinosad(1)	0.010
Boscalid (hydroxy metabolite 2-chloro-N-(4′-chloro-5-hydroxybiphenyl-2-yl)nicotinamide, free phenol, only)	0.010	Spinosyn A(2)	0.010
Carbendazim (Carbendazim only)	0.010	Spinosyn D(2)	0.010
Coumatetralyl	0.010	Spiroxamine (parent only, sum od isomers)	0.010
Chlordecone	0.005	Sulfoxaflor (sum of isomers)	0.010
Chlorpropham (parent only)	0.010	tau-Fluvalinate (sum of isomers)	0.010
Cyproconazole	0.010	Tebuconazole	0.010
Didecyldimethylammonium chloride (DDAC-C10)	0.010	Hydroxy-Tebuconazole (free phenol, only)	0.010
Epoxiconazole	0.010	Tetraconazole (sum of isomers)	0.010
Etofenprox	0.010	Thiacloprid	0.010
Fenpropidin (parent only)	0.010	Thiophanate methyl	0.010
Fenpyrazamine	0.010	Mefentrifluconazole	0.010
Fenpropimorph (parent only)	0.010	Metconazole (sum of isomers)	0.010
Fluopyram	0.010	Molinate	0.010
Fluopyram benzamide	0.010	Oxadiargyl	0.010
Fluquinconazole	0.010	Oxasulfuron	0.010
Flusilazole (parent only)	0.010	Oxyfluorfen	0.010
Metaflumizone (sum of isomers)	0.010	Picolinafen	0.010
Penflufen (sum of isomers)	0.010	Propaquizafop (parent only)	0.010
Penthiopyrad	0.010	Pyraclostrobin	0.010
Prochloraz (parent only)	0.010	Quinoclamine	0.010

(1) Results for Spinosad should be reported either if individual standards for Spinosyn A and D or a mixture of Spinosyn A and D are used for quantification.

(2) Results for Spinosyn A or D should be reported, if the individual standards were used for quantification.